Crude Drugs and Related Drugs

Acacia

Gummi Arabicum

アラビアゴム

Acacia is the secretions obtained from the stems and branches of *Acacia senegal* Willdenow or other species of the same genus (*Leguminosae*).

Description Colorless or light yellow-brown, translucent or somewhat opaque spheroidal tears, or angular fragments with numerous fissures on the surface; very brittle; the fractured surface glassy and occasionally iridescent.

Odorless; tasteless, but produces a mucilaginous sensation on the tongue.

Pulverized Acacia (1.0 g) dissolves almost completely in 2.0 mL of water, and the solution is acid.

It is practically insoluble in ethanol (95).

Identification To 1 g of pulverized Acacia add 25 mL of water and 1 mL of sulfuric acid, and heat under a reflux condenser for 60 minutes. After cooling, add gently 2.0 g of anhydrous sodium carbonate. To 1 mL of this solution add 9 mL of methanol, mix well, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 10 mg each of D-galactose, L-arabinose and L-rhamnose monohydrate in 1 mL water separately, add methanol to make 10 mL, and use these solutions as the standard solutions (1), (2) and (3), respectively. Perform the test with these solutions as directed under Thin-layer chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solutions (1), (2) and (3) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, acetic acid (100) and water (12:3:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, and heat the plate at 105°C for 2 minutes: the three spots obtained from the sample solution are the same with each spot from the standard solutions (1), (2) and (3) in the color tone and the *R*f value, respectively.

Purity **(1)** Insoluble residue—To 5.0 g of pulverized Acacia add 100 mL of water and 10 mL of dilute hydrochloric acid, and dissolve by gentle boiling for 15 minutes with swirling. Filter the warm mixture through a tared glass filter (G3), wash the residue thoroughly with hot water, and dry at 105°C for 5 hours: the mass of the residue does not exceed 10.0 mg.

(2) Tannin-bearing gums—To 10 mL of a solution of Acacia (1 in 50) add 3 drops of iron (III) chloride TS: no dark green color is produced.

(3) Glucose—Use the sample solution obtained in the Identification as the sample solution. Separately, dissolve 10 mg of glucose in 1 mL of water, add methanol to make 10 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of acetone and water (9:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: no spot obtained from the sample solution is observed at the position corresponding to the spot from the standard solution.

Loss on drying <*5.01*> Not more than 17.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Powdered Acacia

Gummi Arabicum Pulveratum

アラビアゴム末

Powdered Acacia is the powder of Acacia.

Description Powdered Acacia occurs as a white to light yellowish white powder. It is odorless, tasteless, but produces a mucilaginous sensation on the tongue.

Under a microscope <*5.01*>, Powdered Acacia, immersed in olive oil or liquid paraffin, reveals colorless, angular fragments or nearly globular grains. Usually starch grains or vegetable tissues are not observed or very trace, if any.

Powdered Acacia (1.0 g) dissolves almost completely in 2.0 mL of water, and the solution is acid.

It is practically insoluble in ethanol (95).

Identification To 1 g of Powdered Acacia add 25 mL of water and 1 mL of sulfuric acid, and heat under a reflux condenser for 60 minutes. After cooling, add gently 2.0 g of anhydrous sodium carbonate. To 1 mL of this solution add 9 mL of methanol, mix well, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 10 mg each of D-galactose, L-arabinose and L-rhamnose monohydrate in 1 mL water, add methanol to make 10 mL, and use these solutions as the standard solutions, (1), (2) and (3), respectively. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solutions (1), (2) and (3) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, acetic acid (100) and water (12:3:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, and heat the plate at 105°C for 2 minutes: the three spots obtained from the sample solution are the same with each spot from the standard solutions (1), (2) and (3) in the color tone and the *R*f value, respectively.

Purity **(1)** Insoluble residue—To 5.0 g of Powdered Acacia add 100 mL of water and 10 mL of dilute hydrochloric acid, and dissolve by gentle boiling for 15 minutes with swirling. Filter the warm mixture through a tared glass filter (G3), wash the residue thoroughly with hot water, and dry at 105°C for 5 hours: the mass of the residue does not exceed 10.0 mg.

(2) Tannin-bearing gums—To 10 mL of a solution of Powdered Acacia (1 in 50) add 3 drops of iron (III) chloride TS: no dark green color is produced.

(3) Glucose—Use the sample solution obtained in the Identification as the sample solution. Separately, dissolve 10 mg of glucose in 1 mL of water, add methanol to make 10 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of acetone and water (9:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: no spot obtained from the sample solution is observed at the position corresponding to the spot from the standard solution.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Tight containers.

Achyranthes Root

Achyranthis Radix

ゴシツ

Achyranthes Root is the root of *Achyranthes bidentata* Blume or *Achyranthes fauriei* H. Léveillé et Vaniot (*Amaranthaceae*).

Description Main root or main root with some lateral roots, with or without short remains of rhizome at the crown; main root, long cylindrical and sometimes somewhat tortuous, 15 – 90 cm in length, 0.3 – 0.7 cm in diameter; externally grayish yellow to yellow-brown, with numerous longitudinal wrinkles, and with scattering scars of lateral roots. Fractured surface is flat; grayish white to light brown on the circumference, and with yellow-white xylem in the center. Hard and brittle, or flexible.

Odor, slight; taste, slightly sweet, and mucilaginous.

Under a microscope <*5.01*>, a transverse section reveals a rather distinct cambium separating the cortex from the xylem; small protoxylem located at the center of the xylem, and surrounded by numerous vascular bundles arranged on several concentric circles; parenchyma cells containing sand crystals of calcium oxalate; starch grains absent.

Identification Shake vigorously 0.5 g of pulverized Achyranthes Root with 10 mL of water: a lasting fine foam is produced.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Achyranthes Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Achyranthes Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Stem—When perform the test of foreign matter <*5.01*>, the amount of stems contained in Achyranthes Root does not exceed 5.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than stems contained in Achyranthes Root does not exceed 1.0%.

Loss on drying <*5.01*> Not more than 17.0% (6 hours).

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Agar

Agar

カンテン

Agar is the solid residue obtained by freezing dehydration of a mucilage derived from *Gelidium elegans* Kuetzing, other species of the same genus (*Gelidiaceae*), or other red algae (*Rhodophyta*).

Description White, translucent rectangular column, string or flakes. Rectangular column about 26 cm in length, 4 cm square in cross section; a string of about 35 cm in length and about 3 mm in width; flakes about 3 mm in length; externally, with wrinkles and somewhat lustrous, light and pliable.

Odorless; tasteless and mucilaginous.

It is practically insoluble in organic solvents.

A boiling solution of Agar (1 in 100) is neutral.

Identification **(1)** To a fragment of Agar add dropwise iodine TS: a dark blue to reddish purple color develops.

(2) Dissolve 1 g of Agar in 65 mL of water by boiling for 10 minutes with constant stirring, and add a sufficient amount of hot water to make up the water lost by evaporation: the solution is clear. Cool the solution between 30°C and 39°C: the solution forms a firm, resilient gel, which does not melt below 85°C.

Purity **(1)** Sulfuric acid—Dissolve 1.0 g of Agar in 100 mL of water by boiling: the solution is not acidic.

(2) Sulfurous acid and starch—To 5 mL of the solution obtained in (1) add 2 drops of iodine TS: the solution is not decolorized immediately, and does not show a blue color.

(3) Insoluble matter—To 7.5 g of Agar add 500 mL of water, boil for 15 minutes, and add water to make exactly 500 mL. Measure exactly 100 mL of the solution, add 100 mL of hot water, heat to boiling, filter while hot through a tared glass filter (G3), wash the residue with a small amount of hot water, and dry the residue at 105°C for 3 hours: the mass of the residue is not more than 15.0 mg.

(4) Water absorption—To 5.0 g of Agar add water to make 100 mL, shake well, allow to stand at 25°C for 24 hours, and filter through moistened glass wool in a 100-mL graduated cylinder: the volume of the filtrate is not more than 75 mL.

Loss on drying <*5.01*> Not more than 22.0% (6 hours).

Total ash <*5.01*> Not more than 4.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Powdered Agar

Agar Pulveratum

カンテン末

Powdered Agar is the powder of Agar.

Description Powdered Agar appears as a white powder, is odorless, and is tasteless and mucilaginous.

Under a microscope <*5.01*>, Powdered Agar, immersed in olive oil or liquid paraffin, reveals angular granules with striations or nearly spheroidal granules 5 to 60 μm in diameter.

It becomes transparent in chloral hydrate TS.

It is practically insoluble in organic solvents.

A boiling solution of Powdered Agar (1 in 100) is neutral.

Identification **(1)** To a part of Powdered Agar add dropwise iodine TS: a dark blue to reddish purple color develops.

(2) Dissolve 1 g of Powdered Agar in 65 mL of water by boiling for 10 minutes with constant stirring, and add a sufficient amount of hot water to maintain the original volume lost by evaporation: the solution is clear. Cool the solution between 30°C and 39°C: the solution forms a firm, resilient gel, which does not melt below 85°C.

Purity **(1)** Sulfuric acid—Dissolve 1.0 g of Powdered Agar in 100 mL of water by boiling: the solution is not acid.

(2) Sulfurous acid and starch—To 5 mL of the solution obtained in (1) add 2 drops of iodine TS: the solution is not decolorized immediately, and does not show a blue color.

(3) Insoluble matter—To 7.5 g of Powdered Agar add 500 mL of water, boil for 15 minutes, and add water to make exactly 500 mL. Take exactly 100 mL of the solution, add 100 mL of hot water, heat to boiling, filter while hot through a tared glass filter (G3), wash the residue with a small amount of hot water, and dry the residue at 105°C for 3 hours: the mass of the residue is not more than 15.0 mg.

(4) Water absorption—To 5.0 g of Powdered Agar add water to make 100 mL, shake well, allow to stand at 25°C for 24 hours, and filter through moistened glass wool in a 100-mL graduated cylinder: the volume of the filtrate is not more than 75 mL.

Loss on drying <*5.01*> Not more than 22.0% (6 hours).

Total ash <*5.01*> Not more than 4.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Tight containers.

Akebia Stem

Akebiae Caulis

モクツウ

Akebia Stem is the climbing stem of *Akebia quinata* Decaisne or *Akebia trifoliata* Koidzumi (*Lardizabalaceae*), usually cut transversely.

Description Circular or ellipsoidal sections 0.2 – 0.3 cm in thickness, and 1 – 3 cm in diameter; phloem on both fractured surfaces is dark grayish brown; xylem reveals light brown vessel portions and grayish white medullary rays lined alternately and radially; pith light grayish yellow, and distinct; flank grayish brown, and with circular or transversely elongated elliptical lenticels.

Almost odorless; slightly acrid taste.

Under a microscope <*5.01*>, a transverse section reveals ring layers mainly consisting of fiber bundles with crystal cells and stone cell groups and surrounding the outside of the phloem in arc shape. Medullary rays of the cortex consisting of sclerenchyma cells containing solitary crystals; portion near cambium is distinct; cells around the pith remarkably thick-walled; xylem medullary rays and parenchyma cells around the pith contain solitary crystals of calcium oxalate and starch grains less than 8 μm in diameter.

Identification To 0.5 g of pulverized Akebia Stem add 10 mL of water, boil, allow to cool, and shake vigorously: lasting fine foams are produced.

Total ash <*5.01*> Not more than 10.0%.

Containers and storage Containers—Well-closed containers.

Alisma Tuber

Alismatis Tuber

タクシャ

Alisma Tuber is the tuber of *Alisma orientale* Juzepczuk (*Alismataceae*), from which periderm has been usually removed.

Description Spherical or conical tubers, 3 – 8 cm in length, 3 – 5 cm in diameter, sometimes a 2- to 4-branched irregular tuber; externally light grayish brown to light yellow-brown, and slightly annulate; many remains of root appearing as small warty protrusions; fractured surface nearly dense, the outer portion grayish brown, and the inner part white to light yellow-brown in color; rather light in texture and difficult to break.

Slight odor and slightly bitter taste.

Identification To 1.0 g of pulverized Alisma Tuber add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Use alisma tuber triterpenes TS for identification as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with a spot among the three spots from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Alisma Tuber according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Alisma Tuber according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed contain-

ers.

Powdered Alisma Tuber

Alismatis Tuber Pulveratum

タクシャ末

Powdered Alisma Tuber is the powder of Alisma Rhizome.

Description Powdered Alisma Tuber occurs as a light grayish brown powder, and has a slight odor and a slightly bitter taste.

Under a microscope <*5.01*>, Powdered Alisma Tuber reveals mainly starch grains, fragments of parenchyma containing them, parenchyma cells containing yellow contents, and fragments of vascular bundles. Starch grains, spheroidal to ellipsoidal simple grains, 3 – 15 μm in diameter.

Identification To 1.0 g of Powdered Alisma Tuber add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Use alisma tuber triterpenes TS for identification as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with a spot among the three spots from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of Powdered Alisma Tuber according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Alisma Tuber according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Aloe

Aloe

アロエ

Aloe is the dried juice of the leaves mainly of *Aloe ferox* Miller, or of interspecific hybrids of the species with *Aloe africana* Miller or *Aloe spicata* Baker (*Liliaceae*).

It contains not less than 4.0% of barbaloin, calculated on the basis of dried material.

Description Aloe occurs as blackish brown to dark brown, irregular masses; sometimes the external surface covered with a yellow powder; the fractured surface smooth and glassy.

Odor, characteristic; taste, extremely bitter.

Identification **(1)** Dissolve 0.5 g of pulverized Aloe in 50 mL of water by warming. After cooling, add 0.5 g of siliceous earth, and filter. Perform the following tests using the filtrate as the sample solution.

(i) Dissolve 0.2 g of sodium tetraborate decahydrate in 5 mL of the sample solution by warming in a water bath. Add a few drops of this solution into 30 mL of water, and shake: a green fluorescence is produced.

(ii) Shake 2 mL of the sample solution with 2 mL of nitric acid: a yellow-brown color which changes gradually to green is produced. Then warm this colored solution in a water bath: the color of the solution changes to red-brown.

(2) To 0.2 g of pulverized Aloe add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of barbaloin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone, water and acetic acid (100) (20:5:2:2) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution and a red fluorescent spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Resin—Warm 0.5 g of pulverized Aloe with 10 mL of diethyl ether on a water bath, and filter. Wash the residue and the filter paper with 3 mL of diethyl ether. Combine the filtrate and the washing, and evaporate the solvent: the mass of the residue is not more than 5.0 mg.

(2) Ethanol-insoluble substances—Heat 1.0 g of pulverized Aloe with 50 mL of ethanol (95) for 30 minutes under a reflux condenser. Filter the warm mixture through a tared glass filter (G4), and wash the residue on the filter with ethanol (95) until the last washing becomes colorless. Dry the residue at 105°C for 5 hours, and weigh: the mass of the residue is not more than 0.10 g.

Loss on drying <*5.01*> Not more than 12.0%.

Total ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Water-soluble extract: not less than 40.0%.

Assay Weigh accurately about 0.1 g of pulverized Aloe, add 40 mL of methanol, and heat under a reflex condenser for 30 minutes. After cooling, filter, and add methanol to the filtrate to make exactly 50 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of barbaloin for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide) for 24 hours, add 40 mg of oxalic acid dihydrate, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of barbaloin in each solution.

$$\text{Amount (mg) of barbaloin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of barbaloin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 360 nm).

Column: A stainless steel column 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (74:26:1).

Flow rate: Adjust so that the retention time of barbaloin is about 12 minutes.

System suitability—

System performance: Dissolve 10 mg of barbaloin for assay add 40 mg of oxalic acid dihydrate, in methanol to make 100 mL. To 5 mL of the solution add 1 mL of a solution of ethenzamide in methanol (1 in 2000) and methanol to make 10 mL. When the procedure is run with 5 μL of this solution under the above operating conditions except the wavelength of 300 nm, barbaloin and ethenzamide are eluted in this order with the resolution between these peaks being not less than 2.0.

System repeatability: When the test is repeated 6 times with 5 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of barbaloin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Aloe

Aloe Pulverata

アロエ末

Powdered Aloe is the powder of Aloe.

It contains not less than 4.0% of barbaloin, calculated on the basis of dried material.

Description Powdered Aloe occurs as a dark brown to yellowish dark brown powder. It has a characteristic odor and an extremely bitter taste.

Under a microscope <*5.01*>, Powdered Aloe, immersed in olive oil or liquid paraffin, reveals greenish yellow to reddish brown, angular or rather irregular fragments.

Identification **(1)** Dissolve 0.5 g of Powdered Aloe in 50 mL of water by warming. After cooling, add 0.5 g of siliceous earth, and filter. Perform the following tests with the filtrate as the sample solution.

(i) Dissolve 0.2 g of sodium tetraborate decahydrate in 5 mL of the sample solution by warming in a water bath. Add a few drops of this solution into 30 mL of water, and shake: a green fluorescence is produced.

(ii) Shake 2 mL of the sample solution with 2 mL of nitric acid: a yellow-brown color which changes gradually to green is produced. Then warm this colored solution in a water bath: the color of the solution changes to red-brown.

(2) To 0.2 g of Powdered Aloe add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of barbaloin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone, water and acetic acid (100) (20:5:2:2) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and the same *R*f value with the red fluorescent spot from the standard solution.

Purity **(1)** Resin—Warm 0.5 of Powdered Aloe with 10 mL of diethyl ether on a water bath, and filter. Wash the residue and the filter paper with 3 mL of diethyl ether. Combine the filtrate and the washing, and evaporate the solvent: the mass of the residue does not exceed 5.0 mg.

(2) Ethanol-insoluble substances—Heat 1.0 g of Powdered Aloe with 50 mL of ethanol (95) for 30 minutes under a reflux condenser. Filter the warm mixture through a tared glass filter (G4), and wash the residue on the filter with ethanol (95) until the last washing becomes colorless. Dry the residue at 105°C for 5 hours, and weigh: the mass of the residue is not more than 0.10 g.

Loss on drying <*5.01*> Not more than 12.0%.

Total ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Water-soluble extract: not less than 40.0%.

Assay Weigh accurately about 0.1 g of Powdered Aloe, add 40 mL of methanol, and heat under a reflex condenser for 30 minutes. After cooling, filter, and add methanol to the filtrate to make exactly 50 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of barbaloin for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide) for 24 hours, add 40 mg of oxalic acid dihydrate, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of barbaloin in each solution.

$$\text{Amount (mg) of barbaloin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of barbaloin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 360 nm).

Column: A stainless steel column about 6 mm in inside diameter and about 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (74:26:1).

Flow rate: Adjust so that the retention time of barbaloin is about 12 minutes.

System suitability—

System performance: To about 10 mg of barbaloin for assay add 40 mg of oxalic acid dihydrate, and dissolve in methanol to make 100 mL. To 5 mL of the solution add 1 mL of a solution of ethenzamide in methanol (1 in 2000) and methanol to make 10 mL. When the procedure is run with 5 μL of this solution under the above operating conditions except the wavelength of 300 nm, barbaloin and ethenzamide are eluted in this order with the resolution between these

peaks being not less than 2.0.

System repeatability: When the test is repeated 6 times with 5 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of barbaloin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Alpinia Officinarum Rhizome

Alpiniae Officinarum Rhizoma

リョウキョウ

Alpinia Officinarum Rhizome is the rhizome of *Alpinia officinarum* Hance (*Zingiberaceae*).

Description Alpinia Officinarum Rhizome is a slightly curved and cylindrical rhizome, sometimes branched; 2 – 8 cm in length, 0.6 – 1.5 cm in diameter; externally red-brown to dark brown with fine striped lines, grayish white nodes and several traces of rootlet; hard to break; fracture surface, light brown in color and thickness of cortex is approximately the same as that of stele.

Odor, characteristic; taste, extremely pungent.

Under a microscope <*5.01*>, a transverse section reveals epidermal cells often containing oil-like substances; cortex, endodermis and stele present beneath the epidermis; cortex and stele divided by endodermis; vascular bundles surrounded by fibers, scattered throughout the cortex and stele, cortex and stele composed of parenchyma interspersed with oil cells; parenchyma cells containing solitary crystals of calcium oxalate and starch grains, starch grains generally simple (sometimes 2- to 8-compound), narrowly ovate, ellipsoidal or ovate, 10 – 40 μm in diameter and with an eccentric navel.

Identification To 0.5 g of pulverized Alpinia Officinarum Rhizome add 5 mL of acetone, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography, develop the plate with a mixture of cyclohexane, ethyl acetate and acetic acid (100) (12:8:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two spots appear at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Alpinia Officinarum Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Alpinia Officinarum Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-extract: not less than 14.0%.

Containers and storage Containers—Well-closed containers.

Aluminum Silicate Hydrate with Silicon Dioxide

Kasseki

カッセキ

Aluminum Silicate Hydrate with Silicon Dioxide is a mineral substance, mainly composed of aluminum silicate hydrate and silicon dioxide.

It is not the same substance with the mineralogical talc.

Description Aluminum Silicate Hydrate with Silicon Dioxide occurs as white to light red powdered crystalline masses, which becomes easily fine powder on crushing. The powder is roughish and easily adheres to skin, and becomes slightly darken and obtains plasticity when moisten with water.

It has a characteristic odor and almost tasteless. It feels like as sand of fine grains by chewing.

Under a microscope <*5.01*>, the powder of Aluminum Silicate Hydrate with Silicon Dioxide, thoroughly grained between a slide glass and a cover glass together with mounting medium, shows numbers of round to polygonal crystals not smaller than 10 μm in diameter.

Identification To 0.5 g of powdered Aluminum Silicate Hydrate with Silicon Dioxide add 3 mL of diluted sulfuric acid (1 in 3), heat until white vapors evolve, then after cooling add 20 mL of water, and filter. The filtrate neutralized to be a weak acidity with ammonia TS responds to Qualitative Tests <*1.09*> (1), (2) and (4) for aluminum salt.

Purity **(1)** Heavy metals <*1.07*>—To 1.5 g of Aluminum Silicate Hydrate with Silicon Dioxide add 50 mL of water and 5 mL of hydrochloric acid, and boil gently for 20 minutes while thorough shaking. After cooling, centrifuge, and separate the supernatant liquid. Wash the precipitate twice with 10 mL portions of water, centrifuging each time, and combine the supernatant liquids. Add ammonia solution (28) dropwise to the combined liquid until a slight precipitate form, then add, while shaking vigorously, dilute hydrochloric acid dropwise to dissolve the precipitate. Add 0.45 g of hydroxylammonium chloride to this solution, heat, then after cooling add 0.45 g of sodium acetate trihydrate and 6 mL of dilute acetic acid, and add water to make 150 mL. Perform the test with 50 mL of this solution as the test solution. Prepare the control solution by adding to 2.0 mL of Standard Lead Solution, 0.15 g of hydroxylammonium chloride, 0.15 g of sodium acetate trihydrate and 2 mL of dilute acetic acid, and add water to make 50 mL (not more than 40 ppm).

(2) Arsenic <*1.11*>—To 1.0 g of Aluminum Silicate Hydrate with Silicon Dioxide add 5 mL of dilute hydrochloric acid, heat gently until boiling begins while shaking thoroughly, then cool quickly, and centrifuge. To the precipitate add 5 mL of dilute hydrochloric acid, shake thoroughly, centrifuge, and take the supernatant liquid. Repeat this operation with 10 mL of water, combine all the extracts, and concentrate the extract to make 5 mL by heating on a water bath. Perform the test using this solution as the test solution (not more than 2 ppm).

Containers and storage Containers—Well-closed containers.

Amomum Seed

Amomi Semen

シュクシャ

Amomum Seed is the seed mass of *Amomum villosum* Loureiro var. *xanthioides* T. L. Wu et S. J. Chen, *Amomum villosum* Loureiro var. *villosum* or *Amomum longiligulare* T. L. Wu (*Zingiberaceae*).

Description Approximately spherical or ellipsoidal mass, 1 – 1.5 cm in length, 0.8 – 1 cm in diameter; externally grayish brown to dark brown, and with white powder in those dried by spreading lime over the seeds; the seed mass is divided into three loculi by thin membranes, and each loculus contains 10 to 20 seeds joining by aril; each seed is polygonal and spherical, 0.3 – 0.5 cm in length, about 0.3 cm in diameter, externally dark brown, with numerous, fine protrusions; hard tissue; under a magnifying glass, a longitudinal section along the raphe reveals oblong section, with deeply indented hilum and with slightly indented chalaza; white perisperm covering light yellow endosperm and long embryo.

Characteristic aroma when cracked, and taste acrid.

Identification To 1.0 g of coarse powdered Amomum Seed add 20 mL of hexane, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use a mixture of hexane and borneol acetate (1000:1) as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Total ash <*5.01*> Not more than 9.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Essential oil content <*5.01*> Perform the test with 30.0 g of pulverized Amomum Seed: the volume of essential oil is not less than 0.6 mL.

Containers and storage Containers—Well-closed containers.

Powdered Amomum Seed

Amomi Semen Pulveratum

シュクシャ末

Powdered Amomum Seed is the powder of Amomum Seed.

Description Powdered Amomum Seed occurs as a grayish brown powder, and has a characteristic aroma and an acrid taste.

Under a microscope <*5.01*>, Powdered Amomum Seed reveals fragments of wavy perisperm cells filled with starch grains and containing in each cell a calcium oxalate crystal; yellow and long epidermal cells of seed coat and fragments of thin-walled tissue perpendicular to them; fragments of groups of brown, thick-walled polygonal stone cells.

Identification To 2.0 g of Powdered Amomum Seed add 20 mL of hexane, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use a mixture of hexane and borneol acetate (1000:1) as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Total ash <*5.01*> Not more than 9.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Essential oil content <*5.01*> Perform the test with 30.0 g of Powdered Amomum Seed: the volume of essential oil is not less than 0.4 mL.

Containers and storage Containers—Tight containers.

Anemarrhena Rhizome

Anemarrhenae Rhizoma

チモ

Anemarrhena Rhizome is the rhizome of *Anemarrhena asphodeloides* Bunge (*Liliaceae*).

Description Rather flat and cord-like rhizome, 3 – 15 cm in length, 0.5 – 1.5 cm in diameter, slightly bent and branched; externally yellow-brown to brown; on the upper surface, a longitudinal furrow and hair-like remains or scars of leaf sheath forming fine ring-nodes; on the lower surface, scars of root appearing as numerous round spot-like hollows; light and easily broken. Under a magnifying glass, a light yellow-brown transverse section reveals an extremely narrow cortex; stele porous, with many irregularly scattered vascular bundles.

Odor, slight; taste, slightly sweet and mucous, followed by bitterness.

Identification **(1)** Shake vigorously 0.5 g of pulverized Anemarrhena Rhizome with 10 mL of water in a test tube: a lasting fine foam is produced. Filter the mixture, and to 2 mL of the filtrate add 1 drop of iron (III) chloride TS: a dark green precipitate is produced.

(2) To 1 g of pulverized Anemarrhena Rhizome add 10 mL of 1 mol/L hydrochloric acid TS, and heat under a reflux condenser for 30 minutes. After cooling, centrifuge, and remove the supernatant liquid. To the residue add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of sarsasapogenin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate

of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Anemarrhena Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Anemarrhena Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter <*5.01*>—The amount of fiber, originating from the dead leaves, and other foreign matters contained in Anemarrhena Rhizome is not more than 3.0%.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Containers and storage Containers—Well-closed containers.

Angelica Dahurica Root

Angelicae Dahuricae Radix

ビャクシ

Angelica Dahurica Root is the root of *Angelica dahurica* Bentham et Hooker filius ex Franchet et Savatier (*Umbelliferae*).

Description Main root from which many long roots are branched out and nearly fusiform and conical in whole shape, 10 – 25 cm in length; externally grayish brown to dark brown, with longitudinal wrinkles, and with numerous scars of rootlets laterally elongated and protruded. A few remains of leaf sheath at the crown and ring-nodes closely protruded near the crown. In a transverse section, the outer region is grayish white in color, and the central region is sometimes dark brown in color.

Odor, characteristic; taste, slightly bitter.

Identification To 0.2 g of pulverized Angelica Dahurica Root add 5 mL of ethanol (95), shake for 5 minutes, and filter. Examine the filtrate under ultraviolet light (main wavelength: 365 nm): a blue to blue-purple fluorescence develops.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Angelica Dahurica Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Angelica Dahurica Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Leaf sheath—When perform the test of foreign matter <*5.01*>, the amount of leaf sheath contained in Angelica Dahurica Root does not exceed 3.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than leaf sheath contained in Angelica Dahurica Root is not more than 1.0%.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 25.0%.

Containers and storage Containers—Well-closed containers.

Apricot Kernel

Armeniacae Semen

キョウニン

Apricot Kernel is the seed of *Prunus armeniaca* Linné, *Prunus armeniaca* Linné var. *ansu* Maximowicz or *Prunus sibirica* Linné (*Rosaceae*).

It contains not less than 2.0% of amygdalin, calculated on the basis of dried material.

Description Flattened, somewhat asymmetric ovoid seed, 1.1 – 1.8 cm in length, 0.8 – 1.3 cm in width, 0.4 – 0.7 cm in thickness; sharp at one end and rounded at the other end where chalaza situated; seed coat brown and its surface being powdery with rubbing easily detachable stone cells of epidermis; numerous vascular bundles running from chalaza throughout the seed coat, appearing as thin vertical furrows; seed coat and thin semitransparent white albumen easily separate from cotyledon when soaked in boiling water; cotyledon, white in color.

Almost odorless; taste, bitter and oily.

Under a microscope <*5.01*>, surface of epidermis reveals stone cells on veins protruded by vascular bundles, forming round polygon to ellipse and approximately uniform in shape, with uniformly thickened cell walls, and 60 – 90 μm in diameter; in lateral view, stone cell appearing obtusely triangular and its cell wall extremely thickened at the apex.

Identification **(1)** When Apricot Kernel is knocked and ground together with water, the odor of benzaldehyde is produced.

(2) To 1.0 g of ground Apricot Kernel add 10 mL of methanol, immediately heat under a reflux condenser for 10 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:5:4) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a spot with a bluish white fluorescence appears at an *R*f value of about 0.7. Spray evenly thymol-sulfuric acid-methanol TS for spraying upon the plate, and heat the plate at 105°C for 5 minutes: one of the several spots from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Rancidity—Grind Apricot Kernel with hot water: no unpleasant odor of rancid oil is perceptible.

(2) Foreign matter <*5.01*>—When perform the test with not less than 250 g of Apricot Kernel, it contains not more than 0.10% of fragments of endocarp.

Loss on drying <*5.01*> Not more than 7.0% (6 hours).

Assay Weigh accurately 0.5 g of ground Apricot Kernel, add 40 mL of diluted methanol (9 in 10), heat immediately

under a reflux condenser for 30 minutes, and cool. Filter the mixture, add diluted methanol (9 in 10) to make exactly 50 mL. Pipet 5 mL of this solution, add water to make exactly 10 mL, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

$$\text{Amount (mg) of amygdalin} = M_S \times A_T/A_S \times 2$$

M_S: Amount (mg) of amygdalin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Apricot Kernel Water

キョウニン水

Apricot Kernel Water contains not less than 0.09 w/v% and not more than 0.11 w/v% of hydrogen cyanide (HCN: 27.03).

Method of preparation Prepare by one of the following methods.

(1) To Apricot Kernels, previously crushed and pressed to remove fixed oils as much as possible, add a suitable amount of Water, Purified Water or Purified Water in Containers, and carry out steam distillation. Determine the amount of hydrogen cyanide in the distillate by the method as directed in the Assay, and carry on the distillation until the content of hydrogen cyanide in the distillate is about 0.14 w/v%. To the distillate add Ethanol in about 1/3 of the volume of the distillate, and dilute with a mixture of Purified Water or Purified Water in Containers and Ethanol (3:1) until the content of hydrogen cyanide meets the specification.

(2) Dissolve 7.5 mL of freshly prepared mandelonitrile in 1000 mL of a mixture of Purified Water or Purified Water in Containers and Ethanol (3:1), mix well, and filter. Determine the amount of hydrogen cyanide in the solution as directed in the Assay, and, if the amount is more than that specified above, dilute the solution to the specified concentration by the addition of the mixture of Purified Water or Purified Water in Containers and Ethanol (3:1).

Description Apricot Kernel Water is a clear, colorless or pale yellow liquid. It has an odor of benzaldehyde and a characteristic taste.

pH: 3.5 – 5.0

Identification To 2 mL of Apricot Kernel Water add 1 mL of ammonia TS, and allow to stand for 10 minutes: a slight turbidity is produced. Allow to stand for 20 minutes: the turbidity is intensified.

Specific gravity <*2.56*> d_{20}^{20}: 0.968 – 0.978

Purity **(1)** Sulfate <*1.14*>—Add a few drops of 0.1 mol/L sodium hydroxide VS to 5.0 mL of Apricot Kernel Water to make slightly alkaline, evaporate on a water bath to dryness, and ignite between 450°C and 550°C. Dissolve the residue in 1.0 mL of dilute hydrochloric acid, and add water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution with 0.50 mL of 0.005 mol/L sulfuric acid VS (not more than 0.005%).

(2) Heavy metals <*1.07*>—Evaporate 50 mL of Apricot Kernel Water on a water bath to dryness, ignite between 450°C and 550°C, dissolve the residue in 5 mL of dilute acetic acid with warming, add water to make exactly 50 mL, and filter. Remove the first 10 mL of the filtrate, dilute the subsequent 20 mL to 50 mL with water, and perform the test using this solution as the test solution. Prepare the control solution as follows: to 2.0 mL of Standard Lead Solution add 2 mL of dilute acetic acid and water to make 50 mL (not more than 1 ppm).

(3) Free hydrogen cyanide—To 10 mL of Apricot Kernel Water add 0.8 mL of 0.1 mol/L silver nitrate VS and 2 to 3 drops of nitric acid at 15°C, filter, and add 0.1 mol/L silver nitrate VS to the filtrate: no change occurs.

(4) Residue on evaporation—Evaporate 5.0 mL of Apricot Kernel Water to dryness, and dry the residue at 105°C for 1 hour: the mass of the residue is not more than 1.0 mg.

Assay Measure exactly 25 mL of Apricot Kernel Water, add 100 mL of water, 2 mL of potassium iodide TS and 1 mL of ammonia TS, and titrate <*2.50*> with 0.1 mol/L silver nitrate VS until a yellow turbidity persists.

$$\text{Each mL of 0.1 mol/L silver nitrate VS} = 5.405 \text{ mg of HCN}$$

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Aralia Rhizome

Araliae Cordatae Rhizoma

ドクカツ

Aralia Rhizome is usually the rhizome of *Aralia cordata* Thunberg (*Araliaceae*).

Description Aralia Rhizome is curved, irregular cylindrical to masses occasionally with remains of short roots. 4 – 12 cm in length, 2.5 – 7 cm in diameter, often cut crosswise or

lengthwise. 1 to several, enlarged dents by remains of stems on the upper part or rarely 1.5 – 2.5 cm in diameter, remains of short stem. The outer surface is dark brown to yellow-brown, with longitudinally wrinkles, bases or dents of root. The transverse section of rhizome reveals dark brown to yellow-brown, scattered brownish small spots with oil canals, and with numerous splits.

Odor, characteristic; taste, slightly bitter.

Under a microscope ⟨*5.01*⟩, a transverse section of rhizome reveals the outermost layer to be cork layer, rarely composed of cork stone cells, followed these appeared several cellular layers of collenchyma. Vascular bundle and medullary rays is distinct, pith broad. Phloem fibre bundles are sometimes observed at the outer portion of phloem. Oil canals composed of schizogenous intercellular space in cortex and pith. Cortex composed of vessels, xylem fibres, and occasionally thick-wall xylem parenchyma. Vascular bundles scattered on the pith. And, parenchymatous cells observed rosette aggregates of calcium oxalate. Starch grains composed of simple grains, 2- to 6- compound grains.

Identification To 1 g of pulverized Aralia Rhizome add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane, ethyl acetate and acetic acid (100) (30:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a purple spot appears at an *R*f value of about 0.5.

Purity Heavy metals ⟨*1.07*⟩—Proceed with 1.0 g of pulverized Aralia Rhizome according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 12.0%.

Total ash ⟨*5.01*⟩ Not more than 9.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Areca

Arecae Semen

ビンロウジ

Areca is the seed of *Areca catechu* Linné (*Palmae*).

Description Rounded-conical or flattened nearly spherical seed 1.5 – 3.5 cm high and 1.5 – 3 cm in diameter; hilum at the center of its base and usually forming a dent; externally grayish red-brown to grayish yellow-brown, with a network of pale lines; hard in texture; cross section dense in texture, exhibiting a marbly appearance of grayish brown seed coat alternating with white albumen; center of the seed often hollow.

Odor, slight; taste, astringent and slightly bitter.

Identification To 1.0 g of pulverized Areca add 5 mL of 0.01 mol/L hydrochloric acid TS and 5 mL of ethyl acetate, shake for 15 minutes, centrifuge, and remove the ethyl acetate layer. To the aqueous layer add 1 mL of sodium hydroxide TS and 5 mL of ethyl acetate, shake for 15 minutes, centrifuge, and use the ethyl acetate layer as the sample solution. Separately, dissolve 1 mg of arecoline hydrobromide for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, water and acetic acid (100) (10:6:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS, air-dry, then spray evenly sodium nitrite TS: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution. The color of this spot fades immediately and then disappears after air-drying.

Purity **(1)** Pericarp—When perform the test of foreign matter ⟨*5.01*⟩, the amount of pericarp contained in Areca is not more than 2.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than the pericarp contained in Areca does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 2.5%.

Containers and storage Containers—Well-closed containers.

Artemisia Capillaris Flower

Artemisiae Capillaris Flos

インチンコウ

Artemisia Capillaris Flower is the capitulum of *Artemisia capillaris* Thunberg (*Compositae*).

Description Capitulum, of ovoid to spherical, about 1.5 – 2 mm in length, about 2 mm in diameter, with linear leaves and pedicels. Outer surface of capitulum, light green to light yellow-brown in color; outer surface of leaf, green to green-brown; outer surface of pedicel, green-brown to dark brown. Under a magnifying glass, at the capitulum, involucral scale in 3 – 4 succubous rows; outer scale, of ovate with obtuse; inner scale, of elliptical, 1.5 mm in length, longer than outer one, with keel midrib and thin membranous margin. Floret, tubular; marginal flower, of female; disk flower, of hermaphrodite. Achene, of obovoid, 0.8 mm in length. Light in texture.

Odor, characteristic, slight; taste, slightly acrid, which gives slightly numbing sensation to the tongue.

Identification To 0.5 g of pulverized Artemisia Capillaris Flower add 10 mL of methanol, shake for 3 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a principal spot with a blue fluorescence appears at an *R*f value of about 0.5.

Purity Stem—When perform the test of foreign matter ⟨*5.01*⟩, Artemisia Capillaris Flower does not contain any

stem more than 2 mm in diameter.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 9.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Artemisia Leaf

Artemisiae Folium

ガイヨウ

Artemisia leaf is the leaf and twig of *Artemisia princeps* Pampanini or *Artemisia montana* Pampanini (*Compositae*).

Description Wrinkled leaves and their fragments, frequently with thin stems. The upper surface of leaf dark green, the lower surface covered densely with grayish white cotton-like hairs. When smoothed by immersion in water, unfolded laminas 4 – 15 cm long, 4 – 12 cm wide, 1- to 2-pinnately cleft or pinnately parted. Segments in 2 to 4 pairs, oblong-lanceolate to oblong, apex acuminate sometimes obtuse, margins irregularly lobed or entire. Small sized leaves tri-cleft or entire, lanceolate.

Order, characteristic; taste, slightly bitter.

Under a microscope ⟨*5.01*⟩, a transverse section of leaf reveals several-cells-layered collenchyma beneath epidermis of midvein; vascular bundles at the central portion of midvein, occasionally fiber bundles adjacent to phloem and xylem; laminas composed of upper epidermis, palisade tissue, spongy tissue and lower epidermis, long soft hairs, T-shaped hairs and glandular hairs on epidermis of laminas; epidermal cells contain tannin-like substances, parenchyma cells contain oil-like substances and tannin-like substances.

Identification To 0.5 g of pulverized Artemisia Leaf (the parts like a floccose substance which are not easily pulverized may be removed) add 5 mL of a mixture of methanol and water (3:2), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg each of umbelliferone for thin-layer chromatography and scopoletin for thin-layer chromatography in 10 mL each of methanol, and use these solutions as the standard solution (1) and the standard solution (2), respectively. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 5 μL each of the standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (20:10:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): two of the several spots obtained from the sample solution have the same color tone and *R*f value with the corresponding blue-white fluorescent spots from the standard solutions (1) and (2).

System suitability—(Ultraviolet light (main wavelength: 365 nm)).

To 1 mL of the standard solution (1) add methanol to make 10 mL. Confirm that when perform the test with 1 μL of this solution under the above conditions, a bluish white fluorescent spot is detectable.

Purity *Artemisia argyi*—To 0.5 g of powdered Artemisia Leaf (the parts like a floccose substance which are not easily pulverized may be removed) add 5 mL of a mixture of methanol and water (3:2), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, to 0.5 g of artemisia·argyi for purity test add 5 mL of a mixture of methanol and water (3:2), shake for 10 minutes, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (20:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): no spot appears from the sample solution at the position of the green fluorescent spot (*R*f value of about 0.5) obtained from the standard solution.

Loss on drying ⟨*5.01*⟩ Not more than 14.0%.

Total ash ⟨*5.01*⟩ Not more than 13.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 3.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 16.0%.

Containers and storage Containers—Well-closed containers.

Asiasarum Root

Asiasari Radix

サイシン

Asiasarum Root is the root and rhizome of *Asiasarum heterotropoides* F. Maekawa var. *mandshuricum* F. Maekawa or *Asiasarum sieboldii* F. Maekawa (*Aristolochiaceae*).

Description Asiasarum Root is a nearly cylindrical rhizome with numerous thin and long roots, externally light brown to dark brown. The root, about 15 cm in length, about 0.1 cm in diameter, with shallow longitudinal wrinkles on the surface, and brittle. The rhizome, 2 – 4 cm in length, 0.2 – 0.3 cm in diameter, often branched, with longitudinal wrinkles on the surface; internode short; each node has several scars of petiole and peduncle, and several thin and long roots.

Odor, characteristic; taste, acrid, which gives slightly numbing sensation to the tongue.

Identification To 1 g of pulverized Asiasarum Root add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of asarinin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample

solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Asiasarum Root according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Asiasarum Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Terrestrial part—When perform the test of foreign matter <*5.01*>, any terrestrial parts are not found.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than terrestrial part contained in Asiasarum Root is not more than 1.0%.

(5) Aristolochic acid I—To exactly 2.0 g of pulverized Asiasarum Root add exactly 50 mL of diluted methanol (3 in 4), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve exactly 1.0 mg of aristolochic acid I for crude drugs purity test in diluted methanol (3 in 4) to make exactly 100 mL. Pipet 1 mL of this solution, add diluted methanol (3 in 4) to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*>, according to the following conditions: the sample solution shows no peak at the retention time corresponding to aristolochic acid I from the standard solution. If the sample solution shows such a peak, repeat the test under different conditions to confirm that the peak in question is not aristolochic acid I.

Operating conditions—

Detector: An ultraviolet or visible absorption photometer (wavelength: 400 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of a solution prepared by dissolving 7.8 g of sodium dihydrogen phosphate dihydrate and 2 mL of phosphoric acid in water to make 1000 mL and acetonitrile (11:9).

Flow rate: Adjust so that the retention time of aristolochic acid I is about 15 minutes.

System suitability—

Test for required detectability: Measure exactly 1 mL of the standard solution, and add diluted methanol (3 in 4) to make exactly 10 mL. Confirm that the ratio, S/N, of the signal (S) and noise (N) of aristolochic acid I obtained with 20 μL of this solution is not less than 3.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of aristolochic acid I is not more than 5.0%.

(6) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Essential oil content <*5.01*> Perform the test with 30.0 g of pulverized Asiasarum Root: the volume of essential oil is not less than 0.6 mL.

Containers and storage Containers—Well-closed containers.

Asparagus Root

Asparagi Radix

テンモンドウ

Asparagus Root is the root of *Asparagus cochinchinensis* Merrill (*Liliaceae*), from which most of the velamen is removed, after being passed through hot water or steamed.

Description Fusiform to cylindrical tubers, 5 – 15 cm in length, 5 – 20 mm in diameter; externally light yellow-brown to light brown, translucent and often with longitudinal wrinkles; flexible, or hard and easily broken in texture; fractured surface, grayish yellow, glossy and horny.

Odor, characteristic; taste, sweet at first, followed by a slightly bitter aftertaste.

Under a microscope <*5.01*>, a transverse section reveals stone cells and their groups scattered on outer layer of cortex; mucilaginous cells containing raphides of calcium oxalate in the parenchyma cells of cortex and stele; no starch grains.

Identification To 1 g of the coarse cutting of Asparagus Root add 5 mL of a mixture of 1-butanol and water (40:7), shake for 30 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of 1-butanol, water and acetic acid (100) (10:6:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 2 minutes: the spot of a red-brown at first then changes to brown color appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Asparagus Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Asparagus Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 18.0% (6 hours).

Total ash <*5.01*> Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Astragalus Root

Astragali Radix

オウギ

Astragalus Root is the root of *Astragalus membranaceus* Bunge or *Astragalus mongholicus* Bunge (*Leguminosae*).

Description Nearly cylindrical root, 30 – 100 cm in length, 0.7 – 2 cm in diameter, with small bases of lateral root dispersed on the surface, twisted near the crown; externally light grayish yellow to light yellow-brown, and covered with irregular, dispersed longitudinal wrinkles and horizontal

lenticel-like patterns; difficult to break; fractured surface fibrous. Under a magnifying glass, a transverse section reveals an outer layer composed of periderm; cortex light yellowish white, xylem light yellow, and zone near the cambium somewhat brown in color; thickness of cortex from about one-third to one-half of the diameter of xylem; white medullary ray from xylem to cortex in thin root, but often appearing as radiating cracks in thick root; usually pith unobservable.

Odor, slight; taste, sweet.

Identification To 1 g of pulverized Astragalus Root add 5 mL of potassium hydroxide TS and 5 mL of acetonitrile in a glass-stoppered centrifuge tube. After shaking this for 10 minutes, centrifuge, and use the acetonitrile layer as the sample solution. Separately, dissolve 1 mg of astragaloside IV for thin-layer chromatography in 10 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown fluorescent spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Astragalus Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Astragalus Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Root of *Hedysarum* species and others—Under a microscope <*5.01*>, a vertical section of Astragalus Root reveals no crystal fiber containing solitary crystals of calcium oxalate outside the fiber bundle.

(4) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Atractylodes Lancea Rhizome

Atractylodis Lanceae Rhizoma

ソウジュツ

Atractylodes Lancea Rhizome is the rhizome of *Atractylodes lancea* De Candolle, *Atractylodes chinensis* Koidzumi or their interspecific hybrids (*Compositae*).

Description Irregularly curved, cylindrical rhizome, 3 – 10 cm in length, 1 – 2.5 cm in diameter; externally dark grayish brown to dark yellow-brown; a transverse section nearly orbicular, with light brown to red-brown secretes as fine points.

Often white cotton-like crystals produced on its surface.

Odor, characteristic; taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section usually reveals periderm with stone cells; parenchyma of cortex, usually without any fiber bundle; oil sacs, containing light brown to yellow-brown substances, located at the end region of medullary rays; xylem exhibits vessels surrounded by fiber bundles and arranged radially on the region adjoining the cambium; pith and medullary rays exhibit the same oil sacs as in the cortex; parenchyma cells contain spherocrystals of inulin and small needle crystals of calcium oxalate.

Identification To 2.0 g of pulverized Atractylodes Lancea Rhizome add 5 mL of hexane, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and acetic acid (100) (10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a grayish green spot appears at an *R*f value of about 0.5.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Atractylodes Lancea Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Atractylodes Lancea Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Atractylodes Lancea Rhizome: the volume of essential oil is not less than 0.7 mL.

Containers and storage Containers—Well-closed containers.

Powdered Atractylodes Lancea Rhizome

Atractylodis Lanceae Rhizoma Pulveratum

ソウジュツ末

Powdered Atractylodes Lancea Rhizome is the powder of Atractylodes Lancea Rhizome.

Description Powdered Atractylodes Lancea Rhizome occurs as a yellow-brown powder. It has a characteristic odor, and a slightly bitter taste.

Under a microscope <*5.01*>, Powdered Atractylodes Lancea Rhizome reveals mainly parenchyma cells, spherocrystals of inulin, fragments of parenchyma cells containing small needle crystals of calcium oxalate as their contents; and further fragments of light yellow thick-walled fibers, stone cells and cork cells; a few fragments of reticulate and scalariform vessels, and small yellow-brown secreted masses or oil drops; starch grains absent.

Identification To 2.0 g of Powdered Atractylodes Lancea Rhizome add 5 mL of hexane, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test

with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and acetic acid (100) (10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a grayish green spot appears at an *R*f value of about 0.5.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Atractylodes Lancea Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Atractylodes Lancea Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Essential oil content <*5.01*> Perform the test with 50.0 g of Powdered Atractylodes Lancea Rhizome: the volume of essential oil is not less than 0.5 mL.

Containers and storage Containers—Tight containers.

Atractylodes Rhizome

Atractylodis Rhizoma

ビャクジュツ

Atractylodes Rhizome is the rhizome of 1) *Atractylodes japonica* Koidzumi ex Kitamura (*Compositae*) (Wa-byakujutsu) or 2) *Atractylodes macrocephala* Koidzumi (*Atractylodes ovata* De Candolle) (*Compositae*) (Kara-byakujutsu).

Description **1)** Wa-byakujutsu—Periderm-removed rhizome is irregular masses or irregularly curved cylinder, 3 – 8 cm in length, 2 – 3 cm in diameter; externally light grayish yellow to light yellowish white, with scattered grayish brown parts. The rhizome covered with periderm is externally grayish brown, often with node-like protuberances and coarse wrinkles. Difficult to break, and the fractured surface is fibrous. A transverse section, with fine dots of light yellow-brown to brown secrete.

Odor, characteristic; taste, somewhat bitter.

Under a microscope <*5.01*>, a transverse section reveals periderm with stone cell layers; fiber bundles in the parenchyma of the cortex, often adjoined to the outside of the phloem; oil sacs containing light brown to brown substances, situated at the outer end of medullary rays; in the xylem, radially lined vessels, surrounding large pith, and distinct fiber bundle surrounding the vessels; in pith and in medullary rays, oil sacs similar to those in cortex, and in parenchyma, crystals of inulin and small needle crystals of calcium oxalate.

2) Kara-byakujutsu—Irregularly enlarged mass, 4 – 8 cm in length, 2 – 5 cm in diameter; externally grayish yellow to dark brown, having sporadic, knob-like small protrusions. Difficult to break; fractured surface has a light brown to dark brown xylem remarkably fibrous.

Odor, characteristic; taste, somewhat sweet, but followed by slight bitterness.

Under a microscope <*5.01*>, a transverse section usually reveals periderm with stone cells, absence of fibers in the cortex; oil sacs containing yellow-brown contents in phloem ray and also at the outer end of it; xylem with radially lined vessels surrounding large pith, and distinct fiber bundle surrounding the vessels; pith and medullary ray exhibit oil sacs as in cortex; parenchyma contains crystals of inulin and small needle crystals of calcium oxalate.

Identification To 2.0 g of pulverized Atractylodes Rhizome add 5 mL of hexane, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetic acid (100) (10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.6.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Atractylodes Rhizome according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Atractylodes Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Atractylodes lancea rhizome—When proceed as directed in the Identification, using exactly 5 mL of hexane, any grayish green spot does not appear at an *R*f value of about 0.5, immediately below the red-purple spot appeared at an *R*f value of about 0.6.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Atractylodes Rhizome: the volume of essential oil is not less than 0.5 mL.

Containers and storage Containers—Well-closed containers.

Powdered Atractylodes Rhizome

Atractylodis Rhizoma Pulveratum

ビャクジュツ末

Powdered Atractylodes Rhizome is the powder of Atractylodes Rhizome.

Description Powdered Atractylodes Rhizome occurs as a light brown to yellow-brown powder, and has a characteristic odor and a slightly bitter or slightly sweet taste, followed by a slightly bitter aftertaste.

Under a microscope <*5.01*>, Powdered Atractylodes Rhizome reveals mainly parenchyma cells, crystals of inulin and fragments of parenchyma cells containing small needle crystals of calcium oxalate; fragments of light yellow thick-walled fibers, stone cells and cork cells; a few fragments of reticulate and scalariform vessels; small yellow-brown secrete masses or oil droplets; starch grains absent.

Identification To 2.0 g of Powdered Atractylodes Rhizome add 5 mL of hexane, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatogra-

phy <2.03>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetic acid (100) (10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.6.

Purity **(1)** Heavy metals <1.07>—Proceed with 1.0 g of Powdered Atractylodes Rhizome according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.40 g of Powdered Atractylodes Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Atractylodes lancea rhizome—When proceed as directed in the Identification, using exactly 5 mL of hexane, any grayish green spot does not appear at an *R*f value of about 0.5, immediately below the red-purple spot appeared at an *R*f value of about 0.6.

Total ash <5.01> Not more than 7.0%.

Acid-insoluble ash <5.01> Not more than 1.0%.

Essential oil content <5.01> Perform the test with 50.0 g of Powdered Atractylodes Rhizome: the volume of essential oil is not less than 0.4 mL.

Containers and storage Containers—Tight containers.

Bakumondoto Extract

麦門冬湯エキス

Bakumondoto Extract contains not less than 1.2 mg of ginesenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), and not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)
Ophiopogon Root	10 g
Pinellia Tuber	5 g
Brown Rice	5 g
Jujube	3 g
Ginseng	2 g
Glycyrrhiza	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1), using the crude drugs shown above.

Description Bakumondoto Extract occurs as a light yellow to light brown powder or black-brown viscous extract. It has a slight odor, and a sweet taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 5 mL of 1-butanol, shake, centrifuge, remove the 1-butanol layer, and use the aqueous layer as the sample solution. Separately, heat 3.0 g of pulverized ophiopogon root in 50 mL of water under a reflux condenser for 1 hour. After cooling, shake 20 mL of the extract with 5 mL of 1-butanol, centrifuge, remove the 1-butanol layer, and use the aqueous layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 2 μL of the sample solution and 5 μL of the standard solution as bands on the original line on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethanol (99.5), water and acetic acid (100) (120:80:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark blue-green spot (*R*f value: about 0.3) from the standard solution (Ophiopogon Root).

(2) Shake 5.0 g of the dry extract (or 15 g of the viscous extract) with 15 mL of water, add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of cycloartenyl ferulate for thin-layer chromatography in 1 mL of ethyl acetate, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 30 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (50:20:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution. Or examine under ultraviolet light (main wavelength: 365 nm) after spraying evenly a mixture of sulfuric acid and ethanol (99.5) (1:1) on the plate, and heating the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Brown Rice).

(3) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 7.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Ginsenoside Rb_1—Weigh accurately about 2 g of the dry extract (or an amount of the viscous extract, equivalent to about 2 g of dried substance), add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 15 mL of diluted methanol (3 in 5), and repeat the same procedure. Combine all of the supernatant liquid, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, then add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 5 mL of this solution to a column [about 10 mm in inside diameter, packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), and washed just before using with methanol and then diluted methanol (3 in 10)], and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S \times 1/5$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl group bound silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (400:100:1).

Flow rate: 1.0 mL per minute (the retention time of ginsenoside Rb_1 is about 16 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Bear Bile

Fel Ursi

ユウタン

Bear Bile is the dried bile of *Ursus arctos* Linné or allied animals (*Ursidae*).

Description Indefinite small masses; externally yellow-brown to dark yellow-brown; easily broken; fractured surface has a glassy luster, and is not wet.

Usually in a gall sac, occasionally taken out, the gall sac consists of a fibrous and strong membrane, 9 – 15 cm in length and 7 – 9 cm in width; externally dark brown and translucent.

Odor, slight and characteristic; taste, extremely bitter.

Identification To 0.1 g of pulverized Bear Bile, add 5 mL of methanol, warm in a water bath for 10 minutes. After cooling, filter, and use the filtrate as the sample solution. Separately, dissolve 10 mg of sodium tauroursodeoxycholate for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetic acid (100), toluene and water (10:10:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly diluted sulfuric acid on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity Other animal biles—Use the sample solution obtained in the Identification as the sample solution. Separately, dissolve 10 mg of sodium glycocholate for thin-layer chromatography and 20 mg of powdered porcine bile for thin-layer chromatography in 5 mL each of methanol, and use these solutions as the standard solution (1) and (2), respectively. Perform the test with these solutions as directed in the Identification: no spot obtained from the sample solution appears at the position of the spot from the standard solution (1) and no grayish brown to black spot appears at the position of the spot at an *R*f value of about 0.3 from the standard solution (2).

Containers and storage Containers—Well-closed containers.

Bearberry Leaf

Uvae Ursi Folium

ウワウルシ

Bearberry Leaf is the leaf of *Arctostaphylos uva-ursi* Sprengel (*Ericaceae*).

It contains not less than 7.0% of arbutin.

Description Obovate to spatulate leaves, 1 – 3 cm in length, 0.5 – 1.5 cm in width; upper surface yellow-green to dark green; lower surface light yellow-green; margin entire; apex obtuse or round, sometimes retuse; base cuneate; petiole very short; lamina thick with characteristic reticulate venation, and easily broken.

Odor, slight; taste, slightly bitter and astringent.

Under a microscope <*5.01*>, the transverse section reveals thick cuticule; parenchyma cells of palisade tissue and sponge tissue being similar in form; in the vascular bundle, medullary ray consisting of 2 to 7 rows of one-cell line, appearing as bones of Japanese fan; polygonal solitary crystals and clustered crystals of calcium oxalate present sparsely in cells on both outer and inner sides of the vascular bundle, but no crystals in mesophyll.

Identification **(1)** Macerate 0.5 g of pulverized Bearberry Leaf with 10 mL of boiling water, shake the mixture for a few minutes, allow to cool, and filter. Place 1 drop of the filtrate on filter paper, and add 1 drop of iron (III) chloride TS: a dark purple color appears.

(2) To 0.2 g of pulverized Bearberry Leaf add 10 mL of a mixture of ethanol (95) and water (7:3), shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of arbutin for thin-layer chromatography in 1 mL of a mixture of ethanol (95) and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, water and formic acid (8:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid upon the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and the *R*f value.

Purity **(1)** Twig—When perform the test of foreign matter <*5.01*>, the amount of twigs contained in Bearberry Leaf does not exceed 4.5%.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than twigs contained in Bearberry Leaf does not exceed 2.0%.

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Assay Weigh accurately about 0.5 g of pulverized Bearberry Leaf in a glass-stoppered centrifuge tube, add 40 mL of water, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 40 mL of water, and proceed in the same manner. To the combined extracts add water to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 40 mg of arbutin for assay, previously dried for 12 hours (in vacuum, silica gel), dissolve in water to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_T and A_S, of arbutin in each solution.

$$\text{Amount (mg) of arbutin} = M_S \times A_T/A_S$$

M_S: Amount (mg) of arbutin for assay taken

Operating conditions—

Detector: An ultraviolet spectrophotometer (wavelength: 280 nm).

Column: A stainless steel column 4 – 6 mm in inside diameter and 15 – 25 cm in length, packed with octadecylsilanized silica gel (5 – 10 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, methanol and 0.1 mol/L hydrochloric acid TS (94:5:1).

Flow rate: Adjust so that the retention time of arbutin is about 6 minutes.

System suitability—

System performance: Dissolve 50 mg each of arbutin for assay, hydroquinone and gallic acid in water to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, arbutin, hydroquinone and gallic acid are eluted in this order with the resolutions among these peaks being not less than 1.5.

System repeatability: When the test is repeated 5 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of arbutin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Beef Tallow

Sevum Bovinum

牛脂

Beef Tallow is a purified fat obtained by wet steam rendering from the fresh fatty tissues of *Bos taurus* Linné var. *domesticus* Gmelin (*Bovidae*).

Description Beef Tallow occurs as a white, uniform mass. It has a characteristic odor and a mild taste.

It is freely soluble in diethyl ether and in petroleum ether, very slightly soluble in ethanol (95), and practically insoluble in water.

It is breakable at a low temperature, but softens above 30°C.

Melting point: 42 – 50°C

Acid value ⟨*1.13*⟩ Not more than 2.0.

Saponification value ⟨*1.13*⟩ 193 – 200

Iodine value ⟨*1.13*⟩ 33 – 50 (When the sample is insoluble in 20 mL of cyclohexane, dissolve it by shaking a glass-stoppered flask in warm water. Then, if insoluble, increase the volume of solvent.)

Purity **(1)** Moisture and coloration—Beef Tallow (5.0 g), melted by heating on a water bath, forms a clear liquid, from which no water separates. In a 10-mm thick layer of the liquid, it is colorless or slightly yellow.

(2) Alkalinity—To 2.0 g of Beef Tallow add 10 mL of water, melt by heating on a water bath, and shake vigorously. After cooling, add 1 drop of phenolphthalein TS to the separated aqueous layer: no color develops.

(3) Chloride—To 1.5 g of Beef Tallow add 30 mL of ethanol (95), heat for 10 minutes under a reflux condenser, and filter after cooling. To 20 mL of the filtrate add 5 drops of a solution of silver nitrate in ethanol (95) (1 in 50): the turbidity of the mixture does not exceed that of the following control solution.

Control solution: To 1.0 mL of 0.01 mol/L hydrochloric acid VS add ethanol (95) to make 20 mL, then add 5 drops of an ethanolic solution of silver nitrate (1 in 50).

Containers and storage Containers—Well-closed containers.

White Beeswax

Cera Alba

サラシミツロウ

White Beeswax is bleached Yellow Beeswax.

Description White Beeswax occurs as white to yellowish white masses. It has a characteristic odor. It is comparatively brittle when cooled, and the fractured surface is granular, and non-crystalline.

It is slightly soluble in diethyl ether, and practically insoluble in water and in ethanol (99.5).

Acid value ⟨*1.13*⟩ 5 – 9 or 17 – 22 Weigh accurately about 6 g of White Beeswax, place in a glass-stoppered 250-mL flask, and add 50 mL of ethanol (99.5). Warm the mixture to dissolve the wax, add 1 mL of phenolphthalein TS, and proceed as directed in the Acid value. Perform a blank determination using solvent which is not previously neutralized, and make any necessary correction.

Saponification value ⟨*1.13*⟩ 80 – 100 Weigh accurately about 3 g of White Beeswax, place in a glass-stoppered 250-mL flask, and add exactly 25 mL of 0.5 mol/L potassium hydroxide-ethanol VS and 50 mL of ethanol (95), heat under a reflux condenser for 4 hours, and proceed as directed in the Saponification value.

Melting point ⟨*1.13*⟩ 60 – 67°C

Purity Paraffin, fat, Japan wax or resin—Melt White Beeswax at the lowest possible temperature, drip the liquid into a vessel containing ethanol (95) to form granules, and allow them to stand in air for 24 hours. Drop the granules into two mixtures of ethanol (95) and water, one adjusted so as to have a specific gravity of 0.95 and the other 0.97: the granules sink or are suspended in the mixture with the specific gravity of 0.95, and float or are suspended in the other mixture.

Containers and storage Containers—Well-closed containers.

Yellow Beeswax

Cera Flava

ミツロウ

Yellow Beeswax is the purified wax obtained from honeycombs such as those of *Apis mellifera* Linné or *Apis cerana* Fabricius (*Apidae*).

Description Yellow Beeswax occurs as light yellow to brownish yellow masses. It has a characteristic odor, which is not rancid.

It is comparatively brittle when cooled, and the fractured surface is granular, and non-crystalline.

Acid value ⟨*1.13*⟩ 5 – 9 or 17 – 22 Weigh accurately about 6 g of Yellow Beeswax, place in a glass-stoppered 250-mL flask, and add 50 mL of ethanol (99.5). Warm the mixture to dissolve the wax, add 1 mL of phenolphthalein TS, and proceed as directed in the Acid value. Perform a blank determination using solvent which is not previously neutralized, and make any necessary correction.

Saponification value ⟨*1.13*⟩ 80 – 100 Weigh accurately about 3 g of Yellow Beeswax, place in a 250-mL glass-stoppered flask, and add 25 mL of 0.5 mol/L potassium hydroxide-ethanol and 50 mL of ethanol (95), heat under a reflux condenser for 4 hours, and proceed as directed in the Saponification value.

Melting point ⟨*1.13*⟩ 60 – 67°C

Purity Paraffin, fat, Japan wax or resin—Melt Yellow Beeswax at the lowest possible temperature, drip the liquid into a glass vessel containing ethanol (95) to form granules, and allow them to stand in air for 24 hours. Drop the granules into two mixtures of ethanol (95) and water, one adjusted so as to have a specific gravity of 0.95 and the other 0.97: the granules sink or are suspended in the mixture with the specific gravity of 0.95, and float or are suspended in the other mixture.

Containers and storage Containers—Well-closed containers.

Belladonna Root

Belladonnae Radix

ベラドンナコン

Belladonna Root is the root of *Atropa belladonna* Linné (*Solanaceae*).

When dried, it contains not less than 0.4% of hyoscyamine ($C_{17}H_{23}NO_3$: 289.37).

Description Cylindrical root, usually 10 – 30 cm in length, 0.5 – 4 cm in diameter; often cut crosswise or lengthwise; externally grayish brown to grayish yellow-brown, with longitudinal wrinkles; periderm often removed; fractured surface is light yellow to light yellow-brown in color and is powdery.

Almost odorless.

Identification Place 2.0 g of pulverized Belladonna Root in a glass-stoppered centrifuge tube, add 30 mL of ammonia TS, and centrifuge after irradiation of ultrasonic waves for 5 minutes. Transfer the supernatant liquid to a separator, add 40 mL of ethyl acetate, and shake. Drain off the ethyl acetate layer, add 3 g of anhydrous sodium sulfate to the ethyl acetate, shake, and filter after the ethyl acetate becomes clear. Evaporate the solvent of the filtrate to dryness under low pressure (in vacuo), dissolve the residue in 1 mL of ethanol (95), and use this solution as the sample solution. Separately, dissolve 2 mg of Atropine Sulfate RS or atropine sulfate hydrate for thin-layer chromatography in 1 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solutions on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, water and ammonia water (28) (90:7:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS on the plate: the principal spot obtained from the sample solution is the same in color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Stem and crown—When perform the test of foreign matter ⟨*5.01*⟩, the amount of stem and crowns contained in Belladonna Root does not exceed 10.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than stems and crowns contained in Belladonna Root does not exceed 2.0%.

Total ash ⟨*5.01*⟩ Not more than 6.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 4.0%.

Assay Weigh accurately about 0.7 g of pulverized Belladonna Root, previously dried at 60°C for 8 hours, place in a glass-stoppered centrifuge tube, and moisten with 15 mL of ammonia TS. To this add 25 mL of diethyl ether, stopper the centrifuge tube tightly, shake for 15 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 25 mL of diethyl ether, proceed in the same manner, and repeat this procedure twice. Combine all the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 5 mL of the mobile phase, add exactly 3 mL of the internal standard solution, and add the mobile phase to make 25 mL. Filter this solution through a filter of a porosity of not more than 0.8 μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 25 mg of Atropine Sulfate RS (previously determine the loss on drying ⟨*2.41*⟩ under the same conditions as Atropine Sulfate Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as the standard stock solution. Pipet 5 mL of the standard stock solution, add exactly 3 mL of the internal standard solution, then add the mobile phase to make 25 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Calculate the ratios, Q_T and Q_S, of the peak area of hyoscyamine (atropine), to that of the internal standard.

$$\text{Amount (mg) of hyoscyamine } (C_{17}H_{23}NO_3) = M_S \times Q_T/Q_S \times 1/5 \times 0.855$$

M_S: Amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

Internal standard solution—A solution of brucine dihydrate in the mobile phase (1 in 2500).

Operating conditions—

Detector: An ultraviolet absorption spectrometer (wavelength: 210 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: Dissolve 6.8 g of potassium dihydrogen phosphate in 900 mL of water, add 10 mL of triethylamine, adjust with phosphoric acid to pH 3.5, and add water to make 1000 mL, and mix this solution with acetonitrile (9:1).

Flow rate: Adjust so that the retention time of atropine is about 14 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, atropine and the internal standard are eluted in this order with the resolution between these peaks being not less than 4.

Containers and storage Containers—Well-closed containers.

Belladonna Extract

ベラドンナエキス

Belladonna Extract contains not less than 0.85% and not more than 1.05% of hyoscyamine ($C_{17}H_{23}NO_3$: 289.37).

Method of preparation To 1000 g of a coarse powder of Belladonna Root add 4000 mL of 35 vol% Ethanol, and digest for 3 days. Press the mixture, add 2000 mL of 35 vol% Ethanol to the residue, and digest again for 2 days. Combine all the extracts, and allow to stand for 2 days. Filter, and prepare the viscous extract as directed under Extracts. An appropriate quantity of Ethanol and Purified Water or Purified Water in Containers may be used in place of 35 vol% Ethanol.

Description Belladonna Extract has a dark brown color, a characteristic odor and a bitter taste.

Identification Mix 0.5 g of Belladonna Extract with 30 mL of ammonia TS in a flask, transfer the mixture to a separator, then add 40 mL of ethyl acetate, and shake the mixture. Drain off the ethyl acetate layer, add 3 g of anhydrous sodium sulfate to the ethyl acetate, shake, and filter after the ethyl acetate becomes clear. Evaporate the solvent to dryness under low pressure (in vacuo), dissolve the residue in 1 mL of ethanol (95), and use this solution as the sample solution. Proceed as directed in the Identification under Belladonna Root.

Purity Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Belladonna Extract as directed under the Extracts (4), and perform the test (not more than 30 ppm).

Assay Weigh accurately about 0.4 g of Belladonna Extract, place in a glass-stoppered centrifuge tube, add 15 mL of ammonia TS, and shake. Add 25 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge, and separate the diethyl ether layer. Repeat this procedure twice with the aqueous layer, using 25 mL each of diethyl ether. Combine the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 5 mL of the mobile phase, add exactly 3 mL of the internal standard solution, and add the mobile phase to make exactly 25 mL. Proceed as directed under Belladonna Root.

$$\text{Amount (mg) of hyoscyamine } (C_{17}H_{23}NO_3) = M_S \times Q_T/Q_S \times 1/5 \times 0.855$$

M_S: Amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

Internal standard solution—A solution of brucine dihydrate in the mobile phase (1 in 2500).

Containers and storage Containers—Tight containers.
Storage—Light-resistant, and in a cold place.

Belladonna Total Alkaloids

ベラドンナ総アルカロイド

Belladonna Total Alkaloids contains not less than 95.0% and not more than 99.0% of hyoscyamine ($C_{17}H_{23}NO_3$: 289.37), not less than 1.3% and not more than 3.9% of scopolamine ($C_{17}H_{21}NO_4$: 303.35), and not less than 99.0% and not more than 102.0% of the total alkaloids (hyoscyamine and scopolamine), calculated on the dried basis.

Method of preparation Belladonna Total Alkaloids is prepared by purification of the extract from Belladonna Root with water or aqueous ethanol.

Description Belladonna Total Alkaloids occurs as white, crystals or crystalline powder.
It is very soluble in methanol, freely soluble in ethanol (99.5), and slightly soluble in water.

Identification Dissolve 2 mg of Belladonna Total Alkaloids in 1 mL of ethanol (95), and use this solution as the sample solution. Then proceed as directed in the Identification under Belladonna Root.

Optical rotation <*2.49*> $[\alpha]_D^{20}$: -18.5 – $-22.0°$ (after drying, 1 g, ethanol (99.5), 25 mL, 100 mm).

Purity **(1)** Heavy metals <*1.07*>—Place 1.0 g of Belladonna Total Alkaloids in a porcelain crucible, and mix with 1.2 mL of dilute hydrochloric acid. Mix with 10 mL of a solution of magnesium nitrate hexahydrate in ethanol (95) (1 in 10), and after evaporating the solvent on a boiling water bath, carbonize by gradual heating. Then proceed according to Method 4, and perform the test. The control solution is prepared as follows: Mix 1.2 mL of dilute hydrochloric acid with 10 mL of a solution of magnesium nitrate hexahydrate in ethanol (95) (1 in 10), and evaporate the solvent on a boiling water bath. After cooling, add 1 mL of sulfuric acid, then proceed according to Method 4, and add 2.0 mL of Standard Lead Solution and water to make 50 mL (not more than 20 ppm).
(2) Arsenic <*1.11*>—Prepare the test solution with 2.0 g of Belladonna Total Alkaloids according to Method 4, and perform the test (not more than 1 ppm).

Loss on drying <*2.41*> Not more than 1.0% (1 g, in vacuum, 60°C, 6 hours).

Residue on ignition <*2.44*> Not more than 0.2% (0.5 g).

Assay Weigh accurately about 25 mg of Belladonna Total Alkaloids, and dissolve in methanol to make exactly 25 mL. Pipet 5 mL of this solution, add exactly 3 mL of the internal standard solution and the mobile phase to make 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 25 mg of Atropine Sulfate RS (separately determine the loss on drying <*2.41*> under the same conditions as Atropine Sulfate Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as the standard stock solution (1). Also, weigh accurately about 25 mg of Scopolamine Hydrobromide RS (separately determine the loss on drying <*2.41*> under the same conditions as Scopolamine Hydrobromide Hydrate), and dissolve in the mobile phase to make exactly 25 mL. Pipet 3 mL of this solution, add the mobile phase to make exactly 25 mL, and use this solution as the standard stock solution (2). Take exactly 5 mL of standard stock solution (1), add exactly 2 mL of the

standard stock solution (2), and add exactly 3 mL of the internal standard solution. To this solution add the mobile phase to make 25 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and calculate the ratios, Q_{TA} and Q_{SA}, of the peak area of hyoscyamine (atropine) to that of the internal standard and the ratios, Q_{TS} and Q_{SS}, of the peak area of scopolamine to that of the internal standard. Then calculate the amounts of hyoscyamine and scopolamine using the following equations. The amount of the total alkaloids is obtained as the sum of them.

The amount (mg) of hyoscyamine ($C_{17}H_{23}NO_3$)
$= M_{SA} \times Q_{TA}/Q_{SA} \times 0.855$

The amount (mg) of scopolamine ($C_{17}H_{21}NO_4$)
$= M_{SS} \times Q_{TS}/Q_{SS} \times 6/125 \times 0.789$

M_{SA}: The amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

M_{SS}: The amount (mg) of Scopolamine Hydrobromide RS taken, calculated on the dried basis

Internal standard solution: A solution of brucine *n*-hydrate in the mobile phase (1 in 2500).

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of around 20°C.

Mobile phase: Dissolve 6.8 g of potassium dihydrogen phosphate in 900 mL of water, add 10 mL of triethylamine, adjust to pH 3.5 with phosphoric acid, and add water to make 1000 mL. To 900 mL of this solution add 100 mL of acetonitrile.

Flow rate: Adjust so that the retention time of atropine is about 14 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, scopolamine, atropine and the internal standard are eluted in this order, and the resolutions between scopolamine and atropine, and atropine and the internal standard are not less than 11 and not less than 4, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the ratio of the peak area of scopolamine to that of the internal standard is not more than 1.5%.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Benincasa Seed

Benincasae Semen

トウガシ

Benincasa seed is the seed of 1) *Benincasa cerifera* Savi or 2) *Benincasa cerifera* Savi forma *emarginata* K. Kimura et Sugiyama (*Cucurbitaceae*).

Description **1)** *Benincasa cerifera* origin—Flattened, ovate to orbicular ovate seed, 10 – 13 mm in length, 6 – 7 mm in width, about 2 mm in thickness; slightly acute at base; hilum and germ pore form two protrusions; externally light grayish yellow to light yellowish brown; prominent band along with marginal edge of seed; under a magnifying glass, surface of the seed is with fine wrinkles and minute hollows.

Odorless; bland taste and slightly oily.

Under a microscope <5.01>, a transverse section reveals the outermost layer of seed coat composed of a single-layered and palisade like epidermis, the epidermis obvious at prominent band along with marginal edge of seed; hypodermis composed of slightly sclerified parenchyma beneath epidermis; inside of the parenchyma several layers of stone cells lie; the innermost layer of seed coat composed of parenchyma several cells thick; perisperm coated with cuticle, composed of parenchyma several cells thick; endosperm composed of a row of compressed cells; cotyledon contains oil drops and aleurone grains, occasionally starch grains.

2) *Benincasa cerifera* forma *emarginata* origin—Flattened, ovate to ellipsoidal seed, 9 – 12 mm in length, 5 – 6 mm in width, about 2 mm in thickness; hilum and germ pore form two protrusions as in 1); externally light grayish yellow, smooth, no prominent band along with marginal edge of seed.

Odorless; bland taste and slightly oily.

Under a microscope <5.01>, a transverse section reveals the outermost layer composed of a single-layered epidermis coated with cuticle, often detached; hypodermis composed of slightly sclerified parenchyma beneath epidermis; inside of the parenchyma several layers of stone cells lie; the innermost layer of seed coat composed of parenchyma several cells thick; perisperm coated with cuticle, composed of parenchyma several cells thick; endosperm composed of a row of compressed cells; cotyledon contains oil drops and aleurone grains, occasionally starch grains.

Identification To 0.5 g of pulverized Benincasa Seed add 10 mL of a mixture of methanol and water (4:1), shake for 10 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of 1-butanol, water and acetic acid (100) (8:6:3) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): two blue-white fluorescent spots appear at an *R*f value of about 0.4, and the spot having the smaller *R*f value shows more intense fluorescence.

Purity Foreign matter <5.01>—It contains not more than 2.0%.

Loss on drying <5.01> Not more than 11.0% (6 hours).

Total ash <5.01> Not more than 5.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 3.0%.

Containers and storage Containers—Well-closed containers.

Benzoin

Benzoinum

アンソッコウ

Benzoin is the resin obtained from *Styrax benzoin* Dryander or other species of the same genus (*Styracaceae*).

Description Benzoin occurs as grayish brown to dark red-brown blocks varying in size; the fractured surface exhibiting whitish to light yellow-red grains in the matrix; hard and brittle at ordinary temperature but softened by heat.

Odor, characteristic and aromatic; taste, slightly pungent and acrid.

Identification **(1)** Heat a fragment of Benzoin in a test tube: it evolves an irritating vapor, and a crystalline sublimate is produced.

(2) Digest 0.5 g of Benzoin with 10 mL of diethyl ether, decant 1 mL of the diethyl ether into a porcelain dish, and add 2 to 3 drops of sulfuric acid: a deep red-brown to deep red-purple color develops.

Purity Ethanol-insoluble substances—Boil gently 1.0 g of Benzoin with 30 mL of ethanol (95) for 15 minutes under a reflux condenser. After cooling, collect the insoluble substances through a tared glass filter (G3), and wash with three 5-mL portions of ethanol (95). Dry the residue at 105°C for 4 hours: the mass of the residue does not exceed 0.30 g.

Total ash ⟨*5.01*⟩ Not more than 2.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Bitter Cardamon

Alpiniae Fructus

ヤクチ

Bitter Cardamon is the fruit of *Alpinia oxyphylla* Miquel (*Zingiberaceae*).

Description Spherical to fusiform fruit, with both ends somewhat pointed; 1 – 2 cm in length, 0.7 – 1 cm in width; externally brown to dark brown, with numerous longitudinal, knob-like protruding lines; pericarp 0.3 – 0.5 mm in thickness, closely adhering to the seed mass, and difficult to separate; inside divided vertically into three loculi by thin membranes, each loculus containing 5 to 8 seeds adhering by aril; seeds irregularly polygonal, about 3.5 mm in diameter, brown to dark brown in color, and hard in texture.

Odor, characteristic; taste, slightly bitter.

Total ash ⟨*5.01*⟩ Not more than 10.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 50.0 g of pulverized Bitter Cardamon: the volume of essential oil is not less than 0.4 mL.

Containers and storage Containers—Well-closed containers.

Bitter Orange Peel

Aurantii Pericarpium

トウヒ

Bitter Orange Peel is the pericarp of the ripe fruit of *Citrus aurantium* Linné or *Citrus aurantium* Linné var. *daidai* Makino (*Rutaceae*).

Description Usually quartered sections of a sphere, sometimes warped or flattened, 4 – 8 cm in length, 2.5 – 4.5 cm in width and 0.5 – 0.8 cm in thickness; the outer surface is dark red-brown to grayish yellow-brown, with numerous small dents associated with oil sacs; the inner surface is white to light grayish yellow-red, with irregular indented reticulation left by vascular bundles; light and brittle in texture.

Odor, characteristic aroma; taste, bitter, somewhat mucilaginous and slightly pungent.

Identification To 1.0 g of Bitter Orange Peel add 10 mL of ethanol (95), allow to stand for 30 minutes with occasional shaking, filter, and use the filtrate as the sample solution. Separately, dissolve 10 mg of naringin for thin-layer chromatography in 10 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly dilute 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in ammonia gas: one of the several spots obtained from the sample solution and a grayish green spot from the standard solution show the same color tone and the same *R*f value.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 50.0 g of pulverized Bitter Orange Peel provided that 1 mL of silicon resin is previously added to the test sample in the flask: the volume of essential oil is not less than 0.2 mL.

Containers and storage Containers—Well-closed containers.

Bitter Tincture

Tinctura Amara

苦味チンキ

Method of preparation

Bitter Orange Peel, in coarse powder	50 g
Swertia Herb, in coarse powder	5 g
Japanese Zanthoxylum Peel, in coarse powder	5 g
70 vol% Ethanol	a sufficient quantity
	To make 1000 mL

Prepare as directed under Tinctures, with the above ingredients. An appropriate quantity of Ethanol and Purified Water or Purified Water in Containers may be used in place of 70 vol% Ethanol.

Description Bitter Tincture is a yellow-brown liquid. It has a characteristic aroma and a bitter taste.

Specific gravity d_{20}^{20}: about 0.90

Identification **(1)** To 1 mL of Bitter Tincture add 5 mL of methanol, then add 0.1 g of magnesium in ribbon form and 1 mL of hydrochloric acid, and allow to stand: the solution is red-purple in color.

(2) Use Bitter Tincture as the sample solution. Separately, to 0.5 g of pulverized Bitter Orange Peel add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the standard solution (1). Proceed with 0.5 g each of pulverized Swertia Herb and Japanese Zanthoxylum Peel in the same manner, and use the solutions so obtained as the standard solution (2) and the standard solution (3). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL each of standard solutions (1), (2) and (3) on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Examine the plate under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same *R*f value with the clear spot at an *R*f value of about 0.7 from the standard solution (3). Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: two of the several spots from the sample solution show the same color tone and *R*f value with the clear spot at an *R*f value of about 0.4 from the standard solution (1), and the clear spot at an *R*f value of about 0.35 from the standard solution (2).

Alcohol number <*1.01*> Not less than 6.9 (Method 2).

Containers and storage Containers—Tight containers.

Bofutsushosan Extract

防風通聖散エキス

Bofutsushosan Extract contains not less than 9 mg and not more than 36 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), not less than 4 mg and not more than 12 mg of total alkaloids (ephedrine and pseudoephedrine), not less than 54 mg and not more than 162 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 13 mg and not more than 39 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)	5)	6)
Japanese Angelica Root	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Peony Root	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Cnidium Rhizome	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Gardenia Fruit	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Forsythia Fruit	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Mentha Herb	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Ginger	0.3 g	0.3 g	0.4 g	0.4 g	1.2 g	0.3 g
Schizonepeta Spike	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Saposhnikovia Root and Rhizome	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	—
Glehnia Root and Rhizome	—	—	—	—	—	1.2 g
Ephedra Herb	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g	1.2 g
Rhubarb	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g
Sodium Sulfate	—	1.5 g	—	1.5 g	—	—
Anhydrous Sodium Sufate	0.7 g	—	0.75 g	—	1.5 g	0.75 g
Atractylodes Rhizome	2 g	2 g	2 g	2 g	2 g	2 g
Platycodon Root	2 g	2 g	2 g	2 g	2 g	2 g
Scutellaria Root	2 g	2 g	2 g	2 g	2 g	2 g
Glycyrrhiza	2 g	2 g	2 g	2 g	2 g	2 g
Gypsum	2 g	2 g	2 g	2 g	2 g	2 g
Aluminum Silicate Hydrate with Silicon Dioxide	3 g	3 g	3 g	3 g	3 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 6), using the crude drugs shown above.

Description Bofutsushosan Extract is a yellow-brown to brown powder or black-brown viscous extract. It has a slightly odor and a sweet and slightly bitter taste.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, add 10 mL of sodium hydroxide TS, shake, centrifuge, and use the diethy ether layer as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of butyl acetate and hexane (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main

wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root; Cnidium Rhizome).

(2) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 1 minute: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple to purple spot from the standard solution (Peony Root).

(3) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfric acid TS on the plate, and heat the plate at 105°C for 1 minute: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple to purple spot from the standard solution (Gardenia Fruit).

(4) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 1.0 g of pulverized forsythia fruit add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution as bands on the original line on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (10:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple spot (*R*f value: about 0.4) from the standard solution (Forsythia Fruit).

(5) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of diluted phosphoric acid (1 in 30), shake, then add 15 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the sample solution. Separately, shake 0.2 g of pulverized mentha herb with 10 mL of diluted phosphoric acid (1 in 30), add 15 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate, water, and acetic acid (100) (10:10:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, heat the plate at 105°C for 5 minutes: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the red-brown spot (*R*f value: around 0.4) from the standard solution (Mentha Herb).

(6) Perform the test according to the following (i) or (ii) (Ginger).

(i) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution.

(ii) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution.

(7) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of 0.1 mol/L hydrochloric acid TS, shake, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use the solution as the sample solution. Separately, dissolve 1 mg of rosmarinic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and acetic acid (100) (60:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the greenish brown spot from the standard solution (Schizonepeta Spike; Mentha Herb).

(8) For preparation prescribed Saposhnikovia Root and Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of 4′-*O*-glycosyl-5-*O*-methylvisamminol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 2 minutes, then examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Saposhnikovia Root and Rhizome).

(9) For preparation prescribed Glehnia Root and Rhizome—To 0.5 g of the dry extract (or 1.5 g of the viscous extract) add 5 mL of ethyl acetate, and heat under a reflux condenser for 30 minutes. After cooling, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of scopoletin for thin-layer chromatography in 10 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Glehnia Root and Rhizome).

(10) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 15 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (4:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot is observed at about 0.5 of *R*f value (Ephedra Herb).

(11) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Rhubarb).

(12) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air- dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the red to red-purple spot from the standard solution (Atractylodes Rhizome).

(13) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium carbonate TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 2.0 g of pulverized platycodon root add 10 mL of sodium carbonate TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate and water (4:4:3) to a distance of about 10 cm, and air-dry the plate. Spray evenly 1,3-naphthalenediol TS on the plate, heat the plate at 105°C for 5 minutes: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot (*R*f value: about 0.4) from the standard solution (Platycodon Root).

(14) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, centrifuge, then add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown to grayish brown spot from the standard solution (Scutellaria Root).

(15) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and stand-

ard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(16) Place 2.0 g of the dry extract (or 6.0 g of the viscous extract) in a porcelain crucible, ignite to incinerate at 550°C, then to the residue add 60 mL of water, shake, centrifuge, and use the supernatant as the sample solution. Add ammonium oxalate TS to the sample solution: a white precipitate is formed. The precipitate does not dissolve in diluted acetic acid, but dissolve on the addition of diluted hydrochloric acid (Gypsum).

(17) Place 2.0 g of the dry extract (or 6.0 g of the viscous extract) in a porcelain crucible, ignite to incinerate at 550°C. To the residue add 60 mL of water, shake well, centrifuge, and use the supernatant as the sample solution. The sample solution responds to the Qualitative Tests <*1.09*> (1) for sulfate (Gypsum; Sodium Sulfate or Anhydrous Sodium Sulfate).

(18) Place 2.0 g of the dry extract (or 6.0 g of the viscous extract) in a crucible, and ignite at 550°C for 5 hours to incinerate. To the residue add 3 mL of diluted sulfuric acid (1 in 3), and heat until white fumes are evolved. After cooling, add 20 mL of water, shake, and filter. To 5 mL of the filtrate add ammonia TS until a white gelatinous precipitate is formed, centrifuge, and remove the supernatant liquid. To the residue add 5 mL of water, shake, centrifuge, and remove the supernatant liquid. Then, to the residue add 5 mL of water, shake, centrifuge, and remove the supernatant liquid. To the obtained residue add 5 drops of alizarin red S TS, and shake occasionally in lukewarm water: the residue shows red to red-brown in color (Kasseki).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not less than 10.0% and more than 22.0%, calculated on the dried basis.

Assay **(1)** Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, elute through a column packed with 2 g of polyamide for column chromatography using 20 mL of water, then add 1 mL of acetic acid (100), add water to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 5/8$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(2) Total alkaloids (ephedrine and pseudoephedrine)—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as above, and remove the diethyl ether layer. To the aqueous layer add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. In addition, repeat twice in the same manner for the aqueous layer using 1.0 mL of ammonia TS and 20 mL of diethyl ether. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of ephedrine hydrochloride for assay of crude drug, previously dried at 105°C for 3 hours, dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine obtained with the sample solution, and the peak area, A_S, of ephedrine obtained with the standard solution.

$$\text{Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)} = M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drug taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, then add 650 mL of water and 1 mL of phosphoric acid.

Flow rate: 1.0 mL per minute (the retention time of ephedrine is about 27 minutes).

System suitability—

System performance: Dissolve 1 mg each of ephedrine hydrochloride for assay of crude drug and pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

(3) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(4) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Boiogito Extract

防已黄耆湯エキス

Boiogito Extract contains not less than 4 mg and not more than 16 mg of sinomenine, and not less than 10 mg and not more than 30 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Sinomenium Stem and Rhizome	5 g	5 g	5 g
Astragalus Root	5 g	5 g	5 g
Atractylodes Rhizome	3 g	3 g	—
Atractylodes Lancea Rhizome	—	—	3 g
Ginger	0.8 g	1 g	1 g
Jujube	3 g	3 g	3 g
Glycyrrhiza	1.5 g	1.5 g	1.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 3), using the crude drugs shown above. Or, prepare a dry extract by adding Light Anhydrous Silicic Acid to an extractive, prepared as directed under Extracts, according to the prescription 3), using the crude drugs shown above.

Description Boiogito Extract is a light yellow-brown to reddish brown powder or black-brown viscous extract. It has a slightly odor, and a sweet taste at first and then a slight hot and bitter taste later.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 15 mL of sodium hydroxide TS, shake, centrifuge, and separate the supernatant liquid. To this liquid add 10 mL of 1-butanol, shake, centrifuge, and separate 1-butanol layer. To this liquid add 10 mL of water, shake, centrifuge, separate the 1-butanol layer, then evaporate the solvent under low pressure (in vacuo), dissolve the residue in 1 mL of methanol, and use the solution as the sample solution. Separately, dissolve 1 mg of sinomenine for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red to red-brown spot from the standard solution (Sinomenium Stem and Rhizome).

(2) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 15 mL of sodium hydroxide TS, shake, centrifuge, and separate the supernatant liquid. To this liquid add 10 mL of 1-butanol, shake, centrifuge, and separate 1-butanol layer. To the aqueous layer add 10 mL of 1-butanol, and proceed in the same manner as above. Combine the 1-butanol layers, add 10 mL of water, shake, centrifuge, separate the 1-butanol layer, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue in exactly 1 mL of methanol, and use this solution as the sample solution. Separately, dissolve 1.0 mg of astragaloside IV for thin-layer chromatography in exactly 10 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-brown spot obtained from the standard solution, and the spot is larger and more intense than the spot from the standard solution (Astragalus Root).

(3) For preparation prescribed Atractylodes Rhizome—To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of Atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red to red-purple spot from the standard solution (Atractylodes Rhizome).

(4) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), then add 0.5 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(5) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-

dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(6) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 11.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not less than 8.0%, calculated on the dried basis. However, for the dry extract prepared by adding Light Anhydrous Silicic Acid, between 9.0% and 18.0%.

Assay **(1)** Sinomenine—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, then add 5.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes, centrifuge, and remove the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, and proceed in the same manner as described above. To the aqueous layer add 5.0 mL of diluted sodium hydroxide TS (1 in 10) and 10 mL of methanol, shake for 15 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and take the supernatant liquid. Combine all the supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of sinomenine for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of sinomenine in each solution.

$$\text{Amount (mg) of sinomenine} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of sinomenine for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: To 3 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, then add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of sinomenine is about 18 minutes).

System suitability—

System performance: When the procedure is run with 10 μL each of the sample solution, the sinomenine standard solution and the glycyrrhizic acid standard solution obtained in Assay (2) under the above operating conditions, peaks of sinomenine and glycyrrhizic acid are observed in the sample solution, glycyrrhizic acid and sinomenine are eluted in this order with the resolution between these peaks being not less than 4.5. Furthermore, except for the peak of glycyrrhizic acid, distinct peaks are observed before and after the peak of sinomenine, and the resolutions between sinomenine and these peaks are respectively not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sinomenine is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solu

tion under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Brown Rice

Oryzae Fructus

コウベイ

Brown Rice is the fruit of *Oryza sativa* Linné (*Gramineae*).

Description Brown Rice occurs as ellipsoidal, slightly flattened, 4 – 6 mm in length; externally translucent, light yellowish white to light brown. Slightly cave in and a white embryo at one end; a brown small dent of scar of style at the other end; few longitudinally striates on the surface.

Odor, slight; taste, slightly sweet.

Under a microscope <*5.01*>, a transverse section of the caryopsis reveals the outermost layer composed of pericarp; vascular bundles in the pericarp; seed coat adhering closely to the pericarp; in the interior, 1 or 2 cellular layered aleuron layers; parenchymatous cells of endosperm contain simple or compound starch grains.

Identification **(1)** To 0.1 g of pulverized Brown Rice add 50 mL of water, and heat in a water bath for 5 minutes. After cooling, add 1 drops of iodine TS, and shake: a blue-purple color develops.

(2) To 1 g of pulverized Brown Rice add 5 mL of ethyl acetate, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of cycloartenyl ferulate for thin-layer chromatography in 1 mL of ethyl acetate, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (5:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple fluorescent spot obtained from the standard solution.

Total ash <*5.01*> Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Bupleurum Root

Bupleuri Radix

サイコ

Bupleurum Root is the root of *Bupleurum falcatum* Linné (*Umbelliferae*).

It contains not less than 0.35% of the total saponin (saikosaponin a and saikosaponin d), calculated on the basis of dried material.

Description Long cone or column shape, single or branched root, 10 – 20 cm in length, 0.5 – 1.5 cm in diameter; occasionally with remains of stem on the crown; externally light brown to brown and sometimes with deep wrinkles; easily broken, and fractured surface somewhat fibrous. Under a magnifying glass, a transverse section reveals the thickness of cortex reaching 1/3 – 1/2 of the radius and tangentially extended clefts in cortex.

Odor, characteristic, and taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section reveals the cortex scattered with a good many oil canals 15 – 35 μm in diameter; in xylem, vessels lined radially or in a staircase pattern, and fiber bundles scattered; in the pith at the crown, the same oil canals as in the cortex; parenchyma cells containing starch grains and oil droplets. Starch grains composed of simple grains, 2 – 10 μm in diameter, or compound grains.

Identification **(1)** Shake vigorously 0.5 g of pulverized Bupleurum Root with 10 mL of water: lasting fine foams are produced.

(2) To 1.0 g of the pulverized Bupleurum Root, add 10 mL of methanol, and boil gently under a reflux condenser for 15 minutes. After cooling, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of saikosaponin a for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution, accompanied by the adjacent yellow-red spot above.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Bupleurum Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Bupleurum Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Stem and leaf—When perform the test of foreign matter <*5.01*>, the amount of the stems and leaves contained in Bupleurum Root does not exceed 10.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than stems and leaves contained in Bupleurum Root does not exceed 1.0%.

Loss on drying <*5.01*> Not more than 12.5% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 11.0%.

Assay Weigh accurately about 1 g of pulverized Bupleurum Root, transfer in a glass-stoppered centrifuge tube, add 20 mL of diluted methanol (9 in 10), shake for 15 minutes, centrifuge, and separate the supernatant liquid. Perform the same procedure with the residue using two 15-mL potions of diluted methanol (9 in 10), combine all the extracts, and add diluted methanol (9 in 10) to make exactly 50 mL. Pipet 5 mL of this solution, add 2.5 mL of dilute sodium hydroxide TS, heat in a water bath at 50°C for 1 hour, and add 7.5 mL of phosphate buffer solution for assay of bupleurum root. Allow this solution to flow through a chromatographic column [about 10 mm inside diameter containing 0.36 g of octadecylsilanized silica gel for pretreatment (55 to 105 μm in particle diameter), conditioned with 10 mL of methanol then 10 mL of water just before use]. Wash the column with 10 mL of diluted methanol (7 in 20), then flow with methanol to get exactly 10 mL of effluent solution, and use this as the sample solution. Use saikosamponins a and d standard TS for assay as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TA} and A_{SA}, of saikosaponin a and A_{TD} and A_{SD}, of saikosaponin d in each solusion. Calculate the amount of saikosaponin a and saikosaponin d by the following equation, and designate the total as the amount of total saponin.

Amount (mg) of saikosaponin a = $M_{SA} \times A_{TA}/A_{SA} \times 1/2$

M_{SA}: Amount (mg) of saikosaponin a for assay taken

Amount (mg) of saikosaponin d = $M_{SD} \times A_{TD}/A_{SD} \times 1/2$

M_{SD}: Amount (mg) of saikosaponin d for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 206 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water and acetonitrile (3:2).

Flow rate: Adjust so that the retention time of saikosaponin a is about 8 minutes.

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, saikosaponin a and saikosaponin d are eluted in this order, and the numbers of theoretical plates and the symmetry factors of their peaks are not less than 4000 and not more than 1.4, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviations of the peak area of saikosaponin a and saikosaponin d are not more than 1.5%, respectively.

Containers and storage Containers—Well-closed containers.

Burdock Fruit

Arctii Fructus

ゴボウシ

Burdock Fruit is the fruit of *Arctium lappa* Linné (*Compositae*).

Description Burdock Fruit is slightly curved, long obovate achene, 5 – 7 mm in length, 2.0 – 3.2 mm in width, 0.8 to 1.5 mm in thickness; externally grayish brown to brown, with black spots; hollow about 1 mm in diameter at one broad end; flat, indistinct, longitudinal ridge at the other narrow end. 100 fruits weigh 1.0 – 1.5 g.

Practically odorless; taste, bitter and oily.

Under a microscope ⟨*5.01*⟩, transverse section reveals an exocarp composed of an epidermis, mesocarp of slightly sclerified parenchyma, and endocarp of a single cellular layer of stone cells; seed coat composed of radially elongated, sclerified epidermis, and parenchyma of several cellular layers; parenchymatous cells of the mesocarp contain a brown substance; stone cells of endocarp contain solitary, discrete crystals of calcium oxalate; cotyledons with starch grains, oil drops, aleurone grains, and minute crystals of calcium oxalate.

Identification To 0.5 g of pulverized Burdock Fruit add 20 mL of methanol, shake for 10 minutes, filter, and use filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of acetone, ethyl acetate and water (15:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.4.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Byakkokaninjinto Extract

白虎加人参湯エキス

Byakkokaninjinto Extract contains not less than 9 mg and not more than 36 mg of mangiferin, not less than 13 mg and not more than 39 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), and not less than 0.9 mg (for preparation prescribed 1.5 g of Ginseng) or not less than 1.8 mg (for preparation prescribed 3 g of Ginseng) of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Anemarrhena Rhizome	5 g	5 g
Gypsum	15 g	15 g
Glycyrrhiza	2 g	2 g
Brown Rice	8 g	8 g
Ginseng	1.5 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Byakkokaninjinto Extract occurs as a very pale yellow-brown to light brown powder or blackish brown viscous extract. It has a slight odor, and has a slightly sweet and slightly bitter taste.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 1 g of pulverized Anemarrhena Rhizome add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 2 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellowish red to dark red spot (at an *R*f value of about 0.3) from the standard solution (Anemarrhena Rhizome).

(2) Place 2.0 g of the dry extract (or 6.0 g of the viscous extract) in a porcelain crucible, and ignite to incinerate at 500 – 550°C. To the residue add 60 mL of water, shake, centrifuge, and use the supernatant as the sample solution. Add ammonium oxalate TS to the sample solution: a white precipitate is formed. The precipitate does not dissolve in diluted acetic acid, but dissolves on the addition of diluted hydrochloric acid (Gypsum).

(3) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(4) To 5.0 g of the dry extract (or 15 g of the viscous extract) add 15 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of cycloartenyl ferulate for thin-layer chromatography in 1 mL of ethyl acetate, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 30 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (50:20:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly a mixture of sulfuric acid and ethanol (99.5) (1:1), heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the light yellow-white to yellow fluorescent spot from the standard solution. (Brown Rice).

(5) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rg_1 RS or ginsenoside Rg_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple to dark purple spot from the standard solution (Ginseng).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 20.0%, calculated on the dried basis.

Assay **(1)** Mangiferin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and use the supernatant as the sample solution. Separately, weigh accurately about 10 mg of mangiferin for assay, dissolve in diluted methanol (1 in 2) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of mangiferin in each solution.

$$\text{Amount (mg) of mangiferin} = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of mangiferin for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 367 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (1780:220:1).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of mangiferin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of mangiferin is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

(3) Ginsenoside Rb_1—Weigh accurately about 1 g of the dry extract (or an amount of the viscous extract, equivalent to about 1 g of the dried substance), add 25 mL of diluted methanol (3 in 5), shake for 30 minutes, then allow to stand, and separate the supernatant liquid. To the residue add 8 mL of water, shake for 15 minutes, then add 12 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. Combine all the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, then add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 10 mL of this solution to a column [about 10 mm in inside diameter, packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), and washed just before using with methanol and then diluted methanol (3 in 10)], and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S \times 1/10$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

For the prescription 1)

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with carbamoyl group bound silica gel for liquid chromatography (3.5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (1700:300:1).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

For the prescription 2)

Operating conditions—

Detector, column temperature, and flow rate: Proceed as directed in the operating conditions in the prescription 1).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl group bound silica gel for liquid chromatography (5 μm in particle diameter).

Mobile phase: A mixture of acetonitrile, water and phos-

phoric acid (400:100:1).
System suitability—
System performance and system repeatability: Proceed as directed in the system suitability in the prescription 1).

Containers and storage Containers—Tight containers.

Cacao Butter

Oleum Cacao

カカオ脂

Cacao Butter is the fat obtained from the seed of *Theobroma cacao* Linné (*Sterculiaceae*).

Description Cacao Butter occurs as a yellow-white, hard, brittle mass. It has a slight, chocolate-like odor, and has no odor of rancidity.

It is freely soluble in diethyl ether and in petroleum ether, soluble in boiling ethanol (99.5), and very slightly soluble in ethanol (95).

Congealing point of the fatty acids: 45 – 50°C

Melting point 31 – 35°C (Cram the sample into a capillary tube without melting the sample).

Specific gravity ⟨*1.13*⟩ d_{20}^{40}: 0.895 – 0.904

Acid value ⟨*1.13*⟩ Not more than 3.0.

Saponification value ⟨*1.13*⟩ 188 – 195

Iodine value ⟨*1.13*⟩ 35 – 43

Containers and storage Containers—Well-closed containers.

Calumba

Calumbae Radix

コロンボ

Calumba is the cross-sectioned root of *Jateorhiza columba* Miers (*Menispermaceae*).

Description Disk-like slices, 0.5 – 2 cm in thickness, 3 – 8 cm in diameter; mostly with concave center and slightly waved; side surface grayish brown in color, with irregular wrinkles; cut surface light yellow and powdery, with pale and dark radiating stripes; cortex rather yellowish; cambium and its neighborhood light grayish brown, warty protrusions in the center; hard in texture, but brittle.

Odor characteristic; taste, bitter.

Identification To 3 g of pulverized Calumba add 30 mL of water, allow to stand for 5 minutes with occasional shaking, and filter. To 2 mL of the filtrate add gently 1 mL of sulfuric acid, and after cooling, add carefully chlorine TS to make two layers: a light red to red color develops at the zone of contact.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 2.0 g of pulverized Calumba according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Calumba according to Method 4, and perform the test (not more than 5 ppm).

Total ash ⟨*5.01*⟩ Not more than 7.5%.

Containers and storage Containers—Well-closed containers.

Powdered Calumba

Calumbae Radix Pulverata

コロンボ末

Powdered Calumba is the powder of Calumba.

Description Powdered Calumba occurs as a grayish yellow powder, and has a characteristic odor and a bitter taste.

Under a microscope ⟨*5.01*⟩, Powdered Calumba reveals numerous starch grains, fragments of parenchyma cells containing them; fragments of cork cells, stone cells, fibers, substitute fibers, vessels, tracheids, and also solitary crystals of calcium oxalate; starch grains consisting of solitary grains or 2- to 3-compound grains; hilum, unevenly scattered, usually 25 – 50 μm, but up to 90 μm in diameter.

Identification To 3 g of Powdered Calumba add 30 mL of water, allow to stand for 5 minutes with occasional shaking, and filter. To 2 mL of the filtrate add gently 1 mL of sulfuric acid, and after cooling, add carefully chlorine TS to make two layers: a light red to red color develops at the zone of contact.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 2.0 g of Powdered Calumba according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of Powdered Calumba according to Method 4, and perform the test (not more than 5 ppm).

Total ash ⟨*5.01*⟩ Not more than 7.5%.

Containers and storage Containers—Well-closed containers.

Camellia Oil

Oleum Camelliae

ツバキ油

Camellia Oil is the fixed oil obtained from the peeled seeds of *Camellia japonica* Linné (*Theaceae*).

Description Camellia Oil is a colorless or pale yellow, clear oil. It is nearly odorless and tasteless.

It is miscible with diethyl ether and with petroleum ether.

It is slightly soluble in ethanol (95).

It congeals partly at −10°C, and completely at −15°C.

Specific gravity d_{25}^{25}: 0.910 – 0.914

Identification To 2 mL of Camellia Oil add dropwise 10 mL of a mixture of fuming nitric acid, sulfuric acid, and water (1:1:1), previously cooled to room temperature: a bluish green color develops at the zone of contact.

Acid value ⟨*1.13*⟩ Not more than 2.8.

Saponification value ⟨*1.13*⟩ 188 – 194

Unsaponifiable matters ⟨*1.13*⟩ Not more than 1.0%.

Iodine value ⟨*1.13*⟩ 78 – 83

Containers and storage Containers—Tight containers.

Capsicum

Capsici Fructus

トウガラシ

Capsicum is the fruit of *Capsicum annuum* Linné (*Solanaceae*).

It contains not less than 0.10% of total capsaicins ((*E*)-capsaicin and dihydrocapsaicin), calculated on the basis of dried material.

Description Elongated conical to fusiform fruit, often bent, 3 – 10 cm in length, about 0.8 cm in width; outer surface lustrous and dark red to dark yellow-red; interior of pericarp hollow and usually divided into two loculi, containing numerous seeds nearly circular and compressed, light yellow-red, about 0.5 cm in diameter.

Usually it remains of calyx and peduncle.

Odor, slight and characteristic; taste, hot and acrid.

Identification To 1.0 g of pulverized Capsicum add 5 mL of ethanol (95), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of (*E*)-capsaicin for thin-layer chromatography in 1 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate and formic acid (10:9:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and expose to an ammonia vapor: a spot obtained from the sample solution and a blue spot from the standard solution show the same color tone and the same *R*f value.

Purity Foreign matter ⟨*5.01*⟩—The amount of foreign matter contained in Capsicum does not exceed 1.0%.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.2%.

Assay Weigh accurately about 0.5 g of moderately fine powder of Capsicum in a glass-stoppered centrifuge tube, add 30 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 10 mL of methanol, shake for 5 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure again, combine all the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-capsaicin for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 40°C) for 5 hours, and dissolve in methanol to make exactly 50 mL. Pipet 2 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TC} and A_{TD}, of (*E*)-capsaicin and dihydrocapsaicin (the relative retention time to (*E*)-capsaicin is about 1.3) obtained with the sample solution, and the peak area, A_S, of (*E*)-capsaicin obtained with the standard solution.

$$\text{Amount (mg) of total capsaicins} = M_S \times (A_{TC} + A_{TD})/A_S \times 0.08$$

M_S: Amount (mg) of (*E*)-capsaicin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 281 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with phenylated silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (3:2).

Flow rate: Adjust so that the retention time of (*E*)-capsaicin is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of (*E*)-capsaicin for assay and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and (*E*)-capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of (*E*)-capsaicin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Capsicum

Capsici Fructus Pulveratus

トウガラシ末

Powdered Capsicum is the powder of Capsicum.

It contains not less than 0.10% of total capsaicins ((*E*)-capsaicin and dihydrocapsaicin), calculated on the basis of dried material.

Description Powdered Capsicum occurs as a yellow-red powder. It has a slight, characteristic odor and a hot, acrid taste.

Under a microscope ⟨*5.01*⟩, Powdered Capsicum reveals fragments of parenchyma containing oil droplets and yellow-red chromoplasts; fragments of epidermis from outer surface of pericarp with thick cuticle; fragments of stone cells from inner surface of pericarp, with wavy curved side walls; fragments of thin vessels; fragments of seed coat with thick wall, and fragments of parenchyma consisting of small cells of endosperm containing fixed oil and aleuron grains.

Identification To 1.0 g of Powdered Capsicum add 5 mL of ethanol (95), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of (*E*)-capsaicin for thin-layer chromatography in 1 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each

of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate and formic acid (10:9:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and expose to an ammonia vapor: a spot obtained from the sample solution and blue spot obtained from the standard solution show the same in color tone and *R*f value.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.2%.

Assay Weigh accurately about 0.5 g of Powdered Capsicum in a glass-stoppered centrifuge tube, add 30 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 10 mL of methanol, shake for 5 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure again, combine all the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-capsaicin for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 40°C) for 5 hours, and dissolve in methanol to make exactly 50 mL. Pipet 2 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TC} and A_{TD}, of (*E*)-capsaicin and dihydrocapsaicin (the relative retention time to (*E*)-capsaicin is about 1.3) obtained with the sample solution, and the peak area, A_S, of (*E*)-capsaicin obtained with the standard solution.

$$\text{Amount (mg) of total capsaicins} = M_S \times (A_{TC} + A_{TD})/A_S \times 0.08$$

M_S: Amount (mg) of (*E*)-capsaicin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 281 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with phenylated silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (3:2).

Flow rate: Adjust so that the retention time of (*E*)-capsaicin is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of (*E*)-capsaicin for assay and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and (*E*)-capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of (*E*)-capsaicin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Capsicum Tincture

トウガラシチンキ

Capsicum Tincture contains not less than 0.010 w/v% of total capsaicins ((*E*)-capsaicin and dihydrocapsaicin).

Method of preparation

Capsicum, in moderately fine cutting	100 g
Ethanol	a sufficient quantity
To make	1000 mL

Prepare as directed under Tinctures, with the above ingredients.

Description Capsicum Tincture is a yellow-red liquid. It has a burning, pungent taste.

Specific gravity d_{20}^{20}: about 0.82

Identification Proceed as directed in the Identification under Capsicum, using Capsicum Tincture as the sample solution. Spot 20 μL each of the sample solution and standard solution.

Alcohol number ⟨*1.01*⟩ Not less than 9.7 (Method 2).

Assay Pipet 2 mL of Capsicum Tincture, add methanol to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-capsaicin for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 40°C) for 5 hours, dissolve in methanol to make exactly 50 mL. Pipet 2 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TC} and A_{TD}, of (*E*)-capsaicin and dihydrocapsaicin (the relative retention time to (*E*)-capsaicin is about 1.3) obtained with the sample solution, and the peak area, A_S, of (*E*)-capsaicin obtained with the standard solution.

$$\text{Amount (mg) of total capsaicins} = M_S \times (A_{TC} + A_{TD})/A_S \times 0.032$$

M_S: Amount (mg) of (*E*)-capsaicin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 281 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with phenylated silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (3:2).

Flow rate: Adjust so that the retention time of (*E*)-capsaicin is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of (*E*)-capsaicin for assay and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and (*E*)-capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times

with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of (*E*)-capsaicin is not more than 1.5%.

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Capsicum and Salicylic Acid Spirit

トウガラシ·サリチル酸精

Method of preparation

Capsicum Tincture	40 mL
Salicylic Acid	50 g
Liquefied Phenol	20 mL
Castor Oil	100 mL
aromatic substance	a suitable quantity
Ethanol	a sufficient quantity
	To make 1000 mL

Prepare as directed under Spirits, with the above ingredients.

Description Capsicum and Salicylic Acid Spirit is a light brown-yellow liquid.
Specific gravity d_{20}^{20}: about 0.84

Identification (1) Shake 10 mL of Capsicum and Salicylic Acid Spirit with 15 mL of sodium hydrogen carbonate TS and 10 mL of diethyl ether, and separate the aqueous layer. To 1 mL of the solution add hydrochloric acid-potassium chloride buffer solution (pH 2.0) to make 200 mL, and to 5 mL of this solution add 5 mL of a solution of iron (III) nitrate enneahydrate (1 in 200): a red-purple color is produced (salicylic acid).

(2) To 0.5 mL of Capsicum and Salicylic Acid Spirit add 20 mL of water and 5 mL of dilute hydrochloric acid, extract with 20 mL of diethyl ether, wash the diethyl ether extract with two 5-mL portions of sodium hydrogen carbonate TS, and then extract with 20 mL of dilute sodium hydroxide TS. To 1 mL of the extract add 1 mL of sodium nitrite TS and 1 mL of dilute hydrochloric acid, shake, and allow to stand for 10 minutes. Add 3 mL of sodium hydroxide TS: a yellow color is produced (phenol).

(3) To 0.2 mL of Capsicum and Salicylic Acid Spirit add 5 mL of dilute hydrochloric acid, extract with 5 mL of chloroform, and use the extract as the sample solution. Dissolve 0.01 g of salicylic acid and 0.02 g of phenol in 5 mL and 25 mL of chloroform, respectively, and use both solutions as the standard solution (1) and the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and standard solutions (1) and (2) on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of chloroform, acetone and acetic acid (100) (45:5:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two spots of the several spots obtained from the sample solution exhibit the same *R*f values as those from standard solution (1) and standard solution (2). Spray evenly iron (III) chloride TS upon the plate: one of the several spots from the sample solution has the same color tone and *R*f value with the spot from the standard solution (1).

Alcohol number <*1.01*> Not less than 8.1 (Method 2). Prepare the sample solution as follows: Pipet 5 mL of Capsicum and Salicylic Acid Spirit at 15 ± 2°C into a glass-stoppered, conical flask containing exactly 45 mL of water while shaking vigorously, allow to stand, and filter the lower layer. Discard the first 15 mL of the filtrate. Pipet 25 mL of the subsequent filtrate, add exactly 10 mL of the internal standard solution, and add water to make exactly 100 mL.

Containers and storage Containers—Tight containers.

Cardamon

Cardamomi Fructus

ショウズク

Cardamon is the fruit of *Elettaria cardamomum* Maton (*Zingiberaceae*). The capsules are removed from the seeds before use.

Description Nearly ellipsoidal, 1 – 2 cm in length, 0.5 – 1 cm in diameter; externally, light yellow with three blunt ridges and many longitudinal lines; 0.1 – 0.2-cm beak at one end; pericarp thin, light and fibrous; interior longitudinally divided into three loculi by thin membranes, each loculus containing 3 to 7 seeds joining by aril; seed irregularly angular ovoid, 0.3 – 0.4 cm in length, dark brown to blackish brown; the dorsal side convex, the ventral side longitudinally grooved; external surface coarsely tuberculated.

Seed has a characteristic aroma, and pungent, slightly bitter taste; pericarp, slight characteristic odor and practically tasteless.

Total ash <*5.01*> Not more than 6.0% (seed).

Acid-insoluble ash <*5.01*> Not more than 4.0% (seed).

Essential oil content <*5.01*> Perform the test with 30.0 g of the pulverized seeds of Cardamon: the volume of essential oil is not less than 1.0 mL.

Containers and storage Containers—Well-closed containers.

Carnauba Wax

Cera Carnauba

カルナウバロウ

Carnauba Wax is the wax obtained from the leaves of *Copernicia cerifera* Martius (*Palmae*).

Description Carnauba Wax occurs as light yellow to light brown, hard and brittle masses or white to light yellow powder. It has a slight, characteristic odor. It is tasteless.

It is practically insoluble in water, in ethanol (95), in diethyl ether and in xylene.
Specific gravity d_{20}^{20}: 0.990 – 1.002
Melting point: 80 – 86°C

Acid value <*1.13*> Not more than 10.0. Use a mixture of xylene and ethanol (95) (2:1) as solvent.

Saponification value <*1.13*> 78 – 95 Weigh accurately about 3 g of Carnauba Wax in a 300-mL flask, add 25 mL of xylene, and dissolve by warming. To this solution add 50 mL of ethanol (95) and exactly 25 mL of 0.5 mol/L potassium hydroxide-ethanol VS, and proceed as directed in the Saponification value. The time of heating should be 2 hours

and the titration should be done by warming.

Iodine value ⟨*1.13*⟩ 5 – 14 (Dissolve the sample by shaking a glass-stoppered flask in warm water.)

Containers and storage Containers—Well-closed containers.

Cassia Seed

Cassiae Semen

ケツメイシ

Cassia Seed is the seed of *Cassia obtusifolia* Linné or *Cassia tora* Linné (*Leguminosae*).

Description Short cylindrical seed, 3 – 6 mm in length, 2 – 3.5 mm in diameter; acuminate at one end and flat at the other; externally green-brown to brown and lustrous, with light yellow-brown longitudinal lines or bands on both sides; hard in texture; cross section round or obtuse polygonal; under a magnifying glass, albumen enclosing a bent, dark-colored cotyledon.

When ground, characteristic odor and taste.

Identification To 1.0 g of pulverized Cassia Seed add 10 mL of diluted methanol (4 in 5), heat on a water bath for 5 minutes, and filter. Evaporate the solvent of the filtrate, dissolve the residue in 5 mL of water, add 2 mL of ethyl acetate, and shake for 10 minutes. Centrifuge this solution, and use the ethyl acetate layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of diethyl ether, cyclohexane and formic acid (5:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly potassium hydroxide-ethanol TS on the plate: an orange to yellow-brown spot appears at an *R*f value of about 0.35.

Purity Foreign matter ⟨*5.01*⟩—The amount of foreign matter contained in Cassia Seed does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Containers and storage Containers—Well-closed containers.

Castor Oil

Oleum Ricini

ヒマシ油

Castor Oil is the fixed oil obtained by compression from the seeds of *Ricinus communis* Linné (*Euphorbiaceae*).

Description Castor Oil is a colorless or pale yellow, clear, viscous oil. It has a slight, characteristic odor, and has a bland at first, and afterwards slightly acrid taste.

It is miscible with ethanol (99.5) and with diethyl ether.

It is freely soluble in ethanol (95), and practically insoluble in water.

When cooled to 0°C, it becomes more viscous, and turbidity is gradually formed.

Identification To 3 g of Castor Oil add 1 g of potassium hydroxide, and heat the mixture carefully to fuse: a characteristic odor is perceptible. Dissolve the fused matter in 30 mL of water, add an excess of magnesium oxide, and filter. Acidify the filtrate with hydrochloric acid: white crystals is produced.

Specific gravity ⟨*1.13*⟩ d_{25}^{25}: 0.953 – 0.965

Acid value ⟨*1.13*⟩ Not more than 1.5.

Saponification value ⟨*1.13*⟩ 176 – 187

Hydroxyl value ⟨*1.13*⟩ 155 – 177

Iodine value ⟨*1.13*⟩ 80 – 90

Purity Adulteration—Shake to mix 1.0 g of Castor Oil with 4.0 mL of ethanol (95): it dissolves clearly. Add 15 mL of ethanol (95): no turbidity is produced.

Containers and storage Containers—Tight containers.

Aromatic Castor Oil

加香ヒマシ油

Method of preparation

Castor Oil	990 mL
Orange Oil	5 mL
Mentha Oil	5 mL
To make	1000 mL

Mix the above ingredients.

Description Aromatic Castor Oil is a colorless or yellowish, clear, viscous liquid. It has an aromatic odor.

Identification To 3 g of Aromatic Castor Oil add 1 g of potassium hydroxide, and heat the mixture carefully to fuse: a characteristic odor is perceptible. Dissolve the fused matter in 30 mL of water, add an excess of magnesium oxide, and filter. Acidify the filtrate with hydrochloric acid: white crystals are produced.

Containers and storage Containers—Tight containers.

Catalpa Fruit

Catalpae Fructus

キササゲ

Catalpa Fruit is the fruit of *Catalpa ovata* G. Don or *Catalpa bungei* C. A. Meyer (*Bignoniaceae*).

Description Slender stick-like fruit, 30 – 40 cm in length and about 0.5 cm in diameter; externally, dark brown; inner part contains numerous seeds; seed compressed or semitubular, about 3 cm in length and about 0.3 cm in width, externally grayish brown; hairs, about 1 cm in length, attached to both ends of seed; pericarp, thin and brittle.

Odor, slight; taste, slightly astringent.

Identification To 1.0 g of pulverized Catalpa Fruit add 20 mL of water, warm on a water bath for 5 minutes, and filter immediately. Transfer the filtrate to a separator, and extract with two 20-mL portions of 1-butanol. Combine the extracts, evaporate the solvent under low pressure (in vacuo) on a water bath, dissolve the residue in 1 mL of methanol,

and use this solution as the sample solution. Separately, dissolve 1 mg of parahydroxybenzoic acid in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (20:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultra-violet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a dark purple spot from the standard solution show the same color tone and the same *R*f value. Prescribe that the moving distance of the spot corresponding to parahydroxybenzoic acid from the sample solution is 1: a dark purple spot develops at the relative moving distance of about 0.3.

Purity Peduncle—When perform the test of foreign matter <*5.01*>, the amount of peduncles contained in Catalpa Fruit does not exceed 5.0%.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Cherry Bark

Pruni Cortex

オウヒ

Cherry Bark is the bark of *Prunus jamasakura* Siebold ex Koidzumi or *Prunus verecunda* Koehne (*Rosaceae*).

Description Flat or semi-tubular pieces of bark; 3 – 6 mm thick, externally light brown to brown, internal surface smooth, grayish brown to brown, occasionally periderm peeled off; the bark with periderm externally rough and lenticels observed; internal surface with many fine longitudinal lines; transversely cut surface grayish brown to brown, fibrous.

Odor, slightly characteristics; taste, slightly bitter and astringent.

Under a microscope <*5.01*>, a transverse section reveals cork layer containing solitary crystals and rosette aggregates of calcium oxalate in the bark with periderm; in secondary cortex many stone cells and idioblasts arranged irregularly and parenchyma cells containing solitary crystals and rosette aggregates of calcium oxalate dotted; groups of phloem fibers lined alternately with the other tissue of phloem between rays.

Identification Shake 1 g of pulverized Cherry Bark with 10 mL of dilute hydrochloric acid, and heat in a boiling water bath for 10 minutes. After cooling, add 5 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the diethyl ether layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (20:20:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a crimson spot appears at an *R*f value of about 0.5.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Chotosan Extract

釣藤散エキス

Chotosan Extract contains not less than 24 mg and not more than 72 mg of hesperidin, not less than 6 mg and not more than 18 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), and not less than 0.3 mg of the total alkaloid (rhyncophylline and hirsutine), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Uncaria Hook	3 g	3 g
Citrus Unshiu Peel	3 g	3 g
Pinellia Tuber	3 g	3 g
Ophiopogon Root	3 g	3 g
Poria Sclerotium	3 g	3 g
Ginseng	2 g	3 g
Saposhnikovia Root and Rhizome	2 g	3 g
Chrysanthemum Flower	2 g	3 g
Glycyrrhiza	1 g	1 g
Ginger	1 g	1 g
Gypsum	5 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Chotosan Extract is a light brown to yellow-brown powder or black-brown viscous extract. It has a slight odor, and has a pungent and slightly sweet first, then bitter taste.

Identification (1) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 20 mL of water and 2 mL of ammonia TS, add 20 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg each of rhyncophylline for thin-layer chromatography and hirsutine for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with one of the two dark purple spots

from the standard solution (Uncaria Hook).

(2) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of hesperidin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone, water and acetic acid (100) (10:6:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in an ammonia gas: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (Citrus Unshiu Peel).

(3) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 5 mL of 1-butanol, shake, centrifuge, remove the 1-butanol layer, and use the aqueous layer as the sample solution. Separately, heat 3.0 g of pulverized ophiopogon root in 50 mL of water under a reflux condenser for 1 hour. After cooling, shake 20 mL of the extract with 5 mL of 1-butanol, centrifuge, remove the 1-butanol layer, and use the aqueous layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL of the sample solution and 5 μL of the standard solution as bands on the original line on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethanol (99.5), water and acetic acid (100) (120:80:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark blue-green spot (around *R*f value 0.3) from the standard solution (Ophiopogon Root).

(4) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(5) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of 4′-*O*-glycosyl-5-*O*-methylvisamminol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (Saposhnikovia Root and Rhizome).

(6) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 20 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of luteolin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 3 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and formic acid (5:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Chrysanthemum Flower).

(7) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(8) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple spot from the standard solution (Ginger).

(9) Shake 1.0 g of a dry extract (or 3.0 g of the viscous extract) with 30 mL of methanol, centrifuge, and separate the supernatant liquid. Shake the residue with 30 mL of water, centrifuge, and separate the supernatant liquid. Add ammonium oxalate TS to this solution: a white precipitate is formed, and it does not dissolve by addition of dilute acetic acid, but it dissolve by addition of dilute hydrochloric acid. (Gypsum)

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 7.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 15.0%, calculated on the dried basis.

Assay **(1)** Hesperidin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of dried substance), add exactly 50 mL of diluted tetrahydrofuran (1 in 4), shake for 30 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of hesperidin for assay, previously dried in a desiccator (silica gel) for 24 hours, dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add diluted tetrahydrofuran (1 in 4) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of hesperidin in each solution.

$$\text{Amount (mg) of hesperidin} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of hesperidin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (82:18:1).

Flow rate: 1.0 mL per minute (the retention time of hesperidin is about 15 minutes).

System suitability—

System performance: Dissolve 1 mg each of hesperidin for assay and naringin for thin-layer chromatography in diluted methanol (1 in 2) to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, naringin and hesperidin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of hesperidin is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the resultant aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

(3) Total alkaloid (rhyncophylline and hirsutine)—Weigh accurately about 1 g of the dry extract (or an amount of the viscous extract, equivalent to about 1 g of dried substance), add 20 mL of diethyl ether, shake, add 3 mL of 1 mol/L hydrochloric acid TS and 7 mL of water, shake for 10 minutes, centrifuge, and remove the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, and repeat the above process. To the aqueous layer add 10 mL of sodium hydroxide TS and 20 mL of diethyl ether, shake for 10 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 20 mL of diethyl ether, proceed in the same manner, and repeat the procedure twice. Combine all the extracts, evaporate the solvent under low pressure (in vacuo) at not more than 40°C, and dissolve the residue in the mobile phase to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of rhyncophylline for assay and about 5 mg of hirsutine for assay, and dissolve in a mixture of methanol and dilute acetic acid (7:3) to make exactly 100 mL. Pipet 10 mL of this solution, add a mixture of methanol and dilute acetic acid (7:3) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each

of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas of rhyncophylline and hirsutine, A_{TR} and A_{TH}, and A_{SR} and A_{SH}, in each solution.

Amount (mg) of the total alkaloid (rhyncophylline and hirsutine)
$= M_{SR} \times A_{TR}/A_{SR} \times 1/50 + M_{SH} \times A_{TH}/A_{SH} \times 1/50$

M_{SR}: Amount (mg) of rhyncophylline for assay taken
M_{SH}: Amount (mg) of hirsutine for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 245 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 5 g of sodium lauryl sulfate in 1150 mL of acetonitrile and 1350 mL of water, mix with 1 mL of phosphoric acid.

Flow rate: 1.0 mL per minute (the retention time of rhyncophylline is about 12 minutes and that of hirsutine is about 27 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of rhyncophylline and hirsutine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviations of the peak area of rhyncophylline and hirsutine are not more than 1.5%, respectively.

Containers and storage Containers—Tight containers.

Chrysanthemum Flower

Chrysanthemi Flos

キク力

Chrysanthemum Flower is the capitulum of 1) *Chrysanthemum indicum* Linné or 2) *Chrysanthemum morifolium* Ramatuelle (*Compositae*).

Description **1)** *Chrysanthemum indicum* origin—Capitulum, 3 – 10 mm in diameter; involucre, consisting of 3 to 5 rows of involucral scales, often with peduncle; the outer involucral scale, linear to lanceolate; inner involucral scale, narrow ovate to ovate; ligulate flower, in a single circle, yellow to light yellow-brown in color; tubular flowers, numerous, light yellow-brown; outer surface of involucre, yellow-brown to brown; light in texture and easy to break.

Odor, characteristic; taste, slightly bitter.

2) *Chrysanthemum morifolium* origin—Capitulum, 15 – 40 mm in diameter; involucre, consisting of 3 to 4 rows of involucral scales, often with peduncle; the outer involucral scale, linear to lanceolate; inner involucral scale, narrow ovate to ovate; ligulate flowers, numerous, white to yellow in color; tubular flowers, small in number, light yellow-brown, occasionally degenerate; outer surface of involucre, green-brown to brown; light in texture and easy to break.

Odor, characteristic; taste, slightly bitter.

Identification To 1 g of pulverized Chrysanthemum Flower add 20 mL of methanol, shake for 10 minutes, and filter. Evaporate the filtrate to dryness, dissolve the residue in 1 mL of methanol, and use this solution as the sample solution. Separately, dissolve 1 mg of luteolin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, 2-butanone, water and formic acid (25:3:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying <5.01> Not more than 15.0% (6 hours).

Total ash <5.01> Not more than 8.5%.

Acid-insoluble ash <5.01> Not more than 1.0%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 30.0%.

Containers and storage Containers—Well-closed containers.

Cimicifuga Rhizome

Cimicifugae Rhizoma

ショウマ

Cimicifuga Rhizome is the rhizome of *Cimicifuga dahurica* Maximowicz, *Cimicifuga heracleifolia* Komarov, *Cimicifuga foetida* Linné or *Cimicifuga simplex* Turczaninow (*Ranunculaceae*).

Description Knotted, irregularly shaped rhizome, 6 – 18 cm in length, 1 – 2.5 cm in diameter; externally dark brown to blackish brown, with many remains of roots, often with scars of terrestrial stems; the center of the scar dented, and the circumference being pale in color and showing a radial pattern; fractured surface fibrous; pith dark brown in color and often hollow; light and hard in texture.

Almost odorless; taste, bitter and slightly astringent.

Identification Dissolve 1 g of pulverized Cimicifuga Rhizome add 5 mL of dilute hydrochloric acid and 5 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the diethy ether layer as the sample solution. Use (*E*)-isoferulic acid-(*E*)-ferulic acid TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (30:10:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the spot among the several spots obtained from the sample solution has the same color tone and *R*f value with the blue fluorescent spot from the standard solution.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of pulverized Cimicifuga Rhizome according to Method 3, and

perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Cimicifuga Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Rhizome of *Astilbe thunbergii* Miquel—Under a microscope <*5.01*>, pulverized Cimicifuga Rhizome does not contain crystal druses in the parenchyma.

Total ash <*5.01*> Not more than 9.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 18.0%.

Containers and storage Containers—Well-closed containers.

Cinnamon Bark

Cinnamomi Cortex

ケイヒ

Cinnamon Bark is the bark of the trunk of *Cinnamomum cassia* J. Presl (*Lauraceae*), or such bark from which a part of the periderm has been removed.

Description Usually semi-tubular or tubularly rolled pieces of bark, 0.1 – 0.5 cm in thickness, 5 – 50 cm in length, 1.5 – 5 cm in diameter; the outer surface dark red-brown, and the inner surface red-brown and smooth; brittle; the fractured surface is slightly fibrous, red-brown, exhibiting a light brown, thin layer.

Characteristic aroma; taste, sweet and pungent at first, later rather mucilaginous and slightly astringent.

Under a microscope <*5.01*>, a transverse section of Cinnamon Bark reveals a primary cortex and a secondary cortex divided by an almost continuous ring consisting of stone cells; nearly round bundles of fibers in the outer region of the ring; cell wall of each stone cell often thickened in a U-shape; secondary cortex lacking stone cells, and with a small number of sclerenchymatous fibers coarsely scattered; parenchyma scattered with oil cells, mucilage cells and cells containing starch grains; medullary rays with cells containing fine needles of calcium oxalate.

Identification To 2.0 g of pulverized Cinnamon Bark add 10 mL of diethyl ether, shake for 3 minutes, filter, and use the filtrate as the sample solution. Perform the test with this solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a purple spot develops at an *R*f value of about 0.4. Spray evenly 2,4-dinitrophenylhydrazine TS upon the spot: a yellow-orange color develops.

Purity Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 15.5% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Cinnamon Bark provided that 1 mL of silicon resin is previously added to the sample in the flask: the volume of essential oil is not less than 0.5 mL.

Containers and storage Containers—Well-closed containers.

Powdered Cinnamon Bark

Cinnamomi Cortex Pulveratus

ケイヒ末

Powdered Cinnamon Bark is the powder of Cinnamon Bark.

Description Powdered Cinnamon Bark is red-brown to brown in color. It has a characteristic aroma and a sweet, pungent taste with a slightly mucilaginous and astringent aftertaste.

Under a microscope <*5.01*>, Powdered Cinnamon Bark reveals starch grains, fragments of parenchyma cells containing them; fragments of fibers, oil cells containing yellow-brown oil droplets, stone cells, cork stone cells, cork tissue, and fine crystals of calcium oxalate. Starch grains are simple and compound grains 6 to 20 μm in diameter.

Identification To 2.0 g of Powdered Cinnamon Bark add 10 mL of diethyl ether, shake for 3 minutes, filter, and use the filtrate as the sample solution. Perform the test with this solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a purple spot develops at an *R*f value of about 0.4. Spray 2,4-dinitrophenylhydrazine TS upon the spot: a yellow orange color develops.

Purity **(1)** Petiole—Under a microscope <*5.01*>, Powdered Cinnamon Bark does not reveal epidermal cells, hairs, cells containing chlorophyll granules, and fragments of vascular bundle.

(2) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of Powdered Cinnamon Bark provided that 1 mL of silicon resin is previously added to the sample in the flask: the volume of essential oil is not less than 0.35 mL.

Containers and storage Containers—Tight containers.

Cinnamon Oil

Oleum Cinnamomi

ケイヒ油

Cinnamon Oil is the essential oil distilled with steam from the leaves and twigs or bark of *Cinnamomum cassia* J. Presl or from the bark of *Cinnamomum zeylanicum* Nees (*Lauraceae*).

It contains not less than 60 vol% of the total aldehydes.

Description Cinnamon Oil is a yellow to brown liquid. It has a characteristic, aromatic odor and a sweet, pungent taste.

It is clearly miscible with ethanol (95) and with diethyl ether.

It is practically insoluble in water.

It is weakly acidic. Upon aging or long exposure to air, it darkens and becomes viscous.

Specific gravity d_{20}^{20}: 1.010 – 1.065

Identification Shake 4 drops of Cinnamon Oil with 4 drops of nitric acid: the mixture forms white to light yellow crystals at a temperature below 5°C.

Purity **(1)** Rosin—Mix 1.0 mL of Cinnamon Oil with 5 mL of ethanol (95), then add 3 mL of freshly prepared, saturated ethanol solution of lead (II) acetate trihydrate: no precipitate is produced.

(2) Heavy metals <*1.07*>—Proceed with 1.0 mL of Cinnamon Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

Assay Pipet 5.0 mL of Cinnamon Oil into a cassia flask, add 70 mL of sodium hydrogensulfite TS, and heat the mixture in a water bath with frequent shaking to dissolve completely. To this solution add sodium hydrogensulfite TS to raise the lower level of the oily layer within the graduate portion of the neck. Allow to stand for 2 hours, and measure the volume (mL) of the separated oily layer.

Total aldehydes (vol%)
= {5.0 − (volume of separated oily layer)} × 20

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Cistanche Herb

Cistanchis Herba

ニクジュヨウ

Cistanche Herb is stout stem of 1) *Cistanche salsa* G. Beck, 2) *Cistanche deserticola* Y. C. Ma or 3) *Cistanche tubulosa* Wight (*Orobanchaceae*), spadix removed in case flowers open.

Description **1)** *Cistanche salsa* origin—Flatly cylindrical, 5 – 25 cm in length, 1 – 2.5 cm in diameter; the one end mostly slightly narrow and curved; external surface brown to blackish brown, covered with thick scales; fleshy and solid, slightly soft and oily, hardly broken; fractured surface yellow-brown to brown, vascular bundles light brown and arranged in a wavy ring.

Odor, characteristic; taste, slightly sweet, followed by slight bitterness.

Under a microscope <*5.01*> a transverse section of middle part reveals the outermost part is an epidermis coated with cuticle; cortex composed of parenchyma; collateral vascular bundles fusiform or rhombic and arranged in a wavy ring in the inner portion of cortex; groups of cells with slightly thickened cell walls sometimes attached outside of phloem of collateral vascular bundles, and exhibit tail like form; pith composed of parenchyma; parenchyma contains starch grains or gelatinized starch.

2) *Cistanche deserticola* origin—Flatly cylindrical, and approximate to 1), but large in size, 5 – 50 cm in length, 1 – 8 cm in diameter.

Odor, characteristic; taste, slightly sweet, followed by slight bitterness.

Under a microscope <*5.01*> a transverse section of middle part reveals, approximate to 1).

3) *Cistanche tubulosa* origin—Flatly fusiform to cylindrical, slightly curved, 5 – 25 cm in length, 2 – 9 cm in diameter; external surface brown to blackish brown, covered with thick scales; solid in texture and firm, hardly broken; fractured surface light grayish brown to yellow-brown, vascular bundles yellow-white and scattered throughout the surface.

Odor, characteristic; taste, slightly sweet, followed by slight bitterness.

Under a microscope <*5.01*> a transverse section of middle part reveals, approximate to 1) and 2), but collateral vascular bundles distributed throughout the parenchyma from marginal region to the center of transverse section; cells with slightly thickened cell walls observed sometimes around collateral vascular bundles, but exhibit no tail like form;

Identification To 1 g of pulverized Cistanche Herb add 5 mL of water and 5 mL of 1-butanol, shake for 15 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of verbascoside for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chlolo-1,4-benzoquinone monoimine TS on the plate, and allow to stand in an ammonia gas: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Cistanche Herb according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Cistanche Herb according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 20.0%.

Total ash <*5.01*> Not more than 11.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 35.0%.

Containers and storage Containers—Well-closed containers.

Citrus Unshiu Peel

Citri Unshiu Pericarpium

チンピ

Citrus Unshiu Peel is the pericarp of the ripe fruit of *Citrus unshiu* Marcowicz or *Citrus reticulata* Blanco (*Rutaceae*).

It contains not less than 4.0% of hesperidin, calculated on the basis of dried material.

Description Irregular pieces of pericarp, about 2 mm in thickness; externally yellow-red to dark yellow-brown, with numerous small dents associated with oil sacs; internally white to light grayish yellow-brown; light and brittle in texture.

Odor, characteristic aroma; taste, bitter and slightly pungent.

Identification To 0.5 g of pulverized Citrus Unshiu Peel add 10 mL of methanol, warm on a water bath for 2 minutes, and filter. To 5 mL of the filtrate add 0.1 g of magnesium in ribbon-form and 1 mL of hydrochloric acid, and allow to stand: a red-purple color develops.

Purity Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Citrus Unshiu Peel provided that 1 mL of silicon resin is previously added to the sample in the flask: the volume of essential oil is not less than 0.2 mL.

Assay Weigh accurately about 0.1 g of pulverized Citrus Unshiu Peel, add 30 mL of methanol, heat under a reflux condenser for 15 minutes, centrifuge after cooling, and separate the supernatant liquid. To the residue add 20 mL of methanol, and proceed in the same manner. Combine the extracts, and add methanol to make exactly 50 mL. Pipet 5 mL of this solution, add water to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of hesperidin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add water to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of hesperidin in each solution.

$$\text{Amount (mg) of hesperidin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of hesperidin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (82:18:1).

Flow rate: 1.0 mL per minute (the retention time of hesperidin is about 15 minutes).

System suitability—

System performance: Dissolve 1 mg each of hesperidin for assay and naringin for thin-layer chromatography in 10 mL of methanol, and add water to make 20 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, naringin and hesperidin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of hesperidin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Clematis Root

Clematidis Radix

イレイセン

Clematis Root is the root with rhizome of *Clematis mandshurica* Ruprecht, *Clematis chinensis* Osbeck or *Clematis hexapetala* Pallas (*Ranunculaceae*).

Description Clematis Root consists of short rhizome and numerous slender roots. The root, 10 – 20 cm in length, 1 – 2 mm in diameter, externally brown to blackish brown, with fine longitudinal wrinkles, brittle. The cortex easily separable from central cylinder; root, grayish white to light yellow-brown in the transverse section, light grayish yellow to yellow in the central cylinder; under a magnifying glass, central cylinder almost round, slight 2 – 4 sinuses on xylem. The rhizome, 2 – 4 cm in length, 5 – 20 mm in diameter, externally light grayish brown to grayish brown; cortex peeled off and fibrous, often with rising node; apex having the residue of lignified stem.

Odor, slight; practically tasteless.

Under a microscope, <*5.01*> transverse section of root reveals an epidermis in the outermost layer; with exodermis lying just inside of the epidermis; cortex and stele divided by endodermis; cortex composed of parenchymatous tissue; xylem with 2 – 4 small concavities where phloem is present; parenchymatous cells contain both simple and 2- to 8-compound starch grains.

Identification **(1)** To 0.5 g of pulverized Clematis Root add 10 mL of water, and boil for 2 to 3 minutes. After cooling, shake vigorously: lasting fine foams appear.

(2) To 0.5 g of pulverized Clematis Root add 3 mL of acetic anhydride, warm on a water bath for 2 minutes, and filter. To the filtrate add 1 mL of sulfuric acid gently: a brown color appears at the zone of contact.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Clematis Root according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Clematis Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 3.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Clove

Caryophylli Flos

チョウジ

Clove is the flowering bud of *Syzygium aromaticum* Merrill et Perry (*Eugenia caryophyllata* Thunberg) (*Myrtaceae*).

Description Dark brown to dark red buds, 1 – 1.8 cm in length, consisting of slightly compressed and four-sided receptacle, crowned by 4 thick sepals and 4 nearly spherical, membranous, imbricated petals, enclosing numerous stamens and a single style.

Odor, strong and characteristic; taste, pungent, which gives numbing sensation to the tongue.

Identification To 1.5 g of pulverized Clove add 20 mL of ethyl acetate, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 5 mg of eugenol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Stem—When perform the test of foreign matter ⟨*5.01*⟩, the amount of the stem contained in Clove does not exceed 5.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than the stem contained in Clove does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 10.0 g of pulverized Clove: the volume of essential oil is not less than 1.6 mL.

Containers and storage Containers—Well-closed containers.

Powdered Clove

Caryophylli Flos Pulveratus

チョウジ末

Powdered Clove is the powder of Clove.

Description Powdered Clove occurs as a dark brown powder. It has a strong, characteristic odor and a pungent taste, leaving a sensation of numbness on the tongue.

Under a microscope ⟨*5.01*⟩, Powdered Clove reveals epidermal tissue with stomata, collenchyma, parenchyma with oil sacs, and spongy parenchyma or its fragments; furthermore, a few fusiform thick-walled fibers, spiral vessels 6 – 10 μm in diameter, anther and pollen grains, and rosette aggregates of calcium oxalate 10 – 15 μm in diameter. Epidermis of anther shows characteristically reticulated walls; pollen grains tetrahedral 10 – 20 μm in diameter; rosette aggregates of calcium oxalate arranged in crystal cell rows, or contained in collenchyma cells and parenchyma cells.

Identification To 1.5 g of Powdered Clove add 20 mL of ethyl acetate, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 5 mg of eugenol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity Foreign matter ⟨*5.01*⟩—Under a microscope, Powdered Clove does not contain stone cells or starch grains.

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 10.0 g of Powdered Clove: the volume of essential oil is not less than 1.3 mL.

Containers and storage Containers—Tight containers.

Clove Oil

Oleum Caryophylli

チョウジ油

Clove Oil is the volatile oil distilled with steam from the flower buds or leaves of *Syzygium aromaticum* Merrill et Perry (*Eugenia caryophyllata* Thunberg) (*Myrtaceae*).

It contains not less than 80.0 vol% of total eugenol.

Description Clove Oil is a colorless or light yellow-brown, clear liquid. It has a characteristic aroma and a burning taste.

It is miscible with ethanol (95) and with diethyl ether.

It is slightly soluble in water.

It acquires a brown color upon aging or by air.

Identification **(1)** To 5 drops of Clove Oil add 10 mL of calcium hydroxide TS, and shake vigorously: the oil forms a flocculent mass, and a white to light yellow color develops.

(2) Dissolve 2 drops of Clove Oil in 4 mL of ethanol (95), and add 1 to 2 drops of iron (III) chloride TS: a green color is produced.

Refractive index <*2.45*> n_D^{20}: 1.527 – 1.537

Specific gravity <*1.13*> d_{20}^{20}: 1.040 – 1.068

Purity **(1)** Clarity of solution—Dissolve 1.0 mL of Clove Oil in 2.0 mL of diluted ethanol (7 in 10): the solution is clear.

(2) Water-soluble phenols—To 1.0 mL of Clove Oil add 20 mL of boiling water, shake vigorously, filter the aqueous layer after cooling, and add 1 to 2 drops of iron (III) chloride TS: a yellow-green, but no blue or violet, color develops.

(3) Heavy metals <*1.07*>—Proceed with 1.0 mL of Clove Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

(4) Optical rotation <*2.49*> α_D^{20}: 0 – −1.5° (100 mm).

Assay Take 10.0 mL of Clove Oil in a Cassia flask, add 70 mL of sodium hydroxide TS, shake for 5 minutes and warm for 10 minutes in a water bath with occasional shaking, add sodium hydroxide TS to the volume after cooling, and allow to stand for 18 hours. Measure the volume (mL) of the separated oily layer.

Total eugenol (vol%)
= {10 − (volume of separated oily layer)} × 10

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Cnidium Monnieri Fruit

Cnidii Monnieris Fructus

ジャショウシ

Cnidium Monnieri Fruit is the fruit of *Cnidium monnieri* Cusson (*Umbelliferae*).

Description Elliptical cremocarp, often each mericarp separated; 2 – 3 mm in length, 1 – 2 mm in width; externally light brown to brown, each mericarp usually with five winged longitudinal ridges; inner surface of mericarp almost flat.

Odor, characteristic; it gives characteristic aroma, later a slight sensation of numbness on chewing.

Under a microscope <*5.01*>, a transverse section reveals one oil canal between longitudinal ridges, usually two oil canals in the inner part of mericarp facing to gynophore; longitudinal ridges composed of slightly lignified parenchymatous cells, with vascular bundles in the base; epidermal cells and parenchymatous cells of longitudinal ridges contain solitary crystals of calcium oxalate; parenchymatous cells of albumen contain oil drops and aleurone grains, and occasionally starch grains.

Identification To 1 g of pulverized Cnidium Monnieri Fruit add 10 mL of ethyl acetate, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of osthole for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and the *R*f value with the blue-white fluorescent spot from the standard solution.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 17.0%.

Acid-insoluble ash <*5.01*> Not more than 6.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Cnidium Rhizome

Cnidii Rhizoma

センキュウ

Cnidium Rhizome is the rhizome of *Cnidium officinale* Makino (*Umbelliferae*), usually passed through hot water.

Description Irregular massive rhizome, occasionally cut lengthwise; 5 – 10 cm in length, and 3 – 5 cm in diameter; externally grayish brown to dark brown, with gathered nodes, and with knobbed protrusions on the node; margin of the vertical section irregularly branched; internally grayish white to grayish brown, translucent and occasionally with hollows; dense and hard in texture.

Odor, characteristic; taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section reveals cortex and pith with scattered oil canals; in the xylem, thick-walled and lignified xylem fibers appear in groups of various sizes; starch grains usually gelatinized, but rarely remaining as grains of 5 – 25 μm in diameter; crystals of calcium oxalate not observable.

Identification To 1 g of pulverized Cnidium Rhizome add 5 mL of methanol and 0.1 mL of sodium hydroxide TS, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution (1). Dissolve 1 mg of (*E*)-ferulic acid in 2 mL of methanol and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL each of the standard solution (1) and the standard solution (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (30:25:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (1). Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes

and allow to cool: one of the several spots from the sample solution has the same color tone and *R*f value with the spot from the standard solution (2).

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Cnidium Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Cnidium Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Powdered Cnidium Rhizome

Cnidii Rhizoma Pulveratum

センキュウ末

Powdered Cnidium Rhizome is the powder of Cnidium Rhizome.

Description Powdered Cnidium Rhizome occurs as a gray to light grayish brown powder. It has a characteristic odor and a slightly bitter taste.

Under a microscope <*5.01*>, Powdered Cnidium Rhizome reveals colorless and gelatinized starch masses, and fragments of parenchyma containing them; fragments of scalariform and reticulate vessels 15 – 30 μm in diameter; fragments of thick-walled and lignified xylem fibers 20 – 60 μm in diameter; fragments of yellow brown cork tissue; fragments of secretory tissue.

Identification To 1 g of Powdered Cnidium Rhizome add 5 mL of methanol and 0.1 mL of sodium hydroxide TS, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution (1). Dissolve 1 mg of (*E*)-ferulic acid in 2 mL of methanol and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL each of the standard solution (1) and the standard solution (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (30:25:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (1). Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes and allow to cool: one of the several spots from the sample solution has the same color tone and *R*f value with the spot from the standard solution (2).

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Cnidium Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Cnidium Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Cnidium Rhizome does not contain a large quantity of starch grains, stone cells, crystals of calcium oxalate or other foreign matter.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Coconut Oil

Oleum Cocois

ヤシ油

Coconut oil is the fixed oil obtained from the seeds of *Cocos nucifera* Linné (*Palmae*).

Description Coconut Oil is a white to light yellow mass or a colorless or light yellow, clear oil. It has a slight, characteristic odor and a mild taste.

It is freely soluble in diethyl ether and in petroleum ether. It is practically insoluble in water.

At a temperature below 15°C, it congeals to a hard and brittle solid.

Melting point: 20 – 28°C

Acid value <*1.13*> Not more than 0.2.

Saponification value <*1.13*> 246 – 264

Unsaponifiable matter <*1.13*> Not more than 1.0%.

Iodine value <*1.13*> 7 – 11

Containers and storage Containers—Tight containers.

Codonopsis Root

Codonopsis Radix

トウジン

Codonopsis Root is the root of *Codonopsis pilosula* Nannfeldt or *Codonopsis tangshen* Oliver (*Campanulaceae*).

Description Codonopsis Root nearly cylindrical, 8 – 30 cm in length, 0.5 – 2.5 cm in diameter; gradually slender to the apex, often branched; outer surface light yellow to grayish brown; from the base to central part with ring-like wrinkles, and longitudinal wrinkles entirely obvious; numerous projections composed of scars of stems at the crown, with a round dent at the distal end; blackish brown and tremellose secretion often at the scars of lateral roots; flexible and easily bendable or hard and easily breakable in texture; in cut surface yellow-white to light brown in cortex, light yellow in xylem, sometimes with slit in cortex.

Odor, slight and characteristic; taste, slightly sweet.

Under a microscope <*5.01*>, a transverse section reveals cork layer at the outermost portion, outer 1- to 10-layer consisting of cork stone cells; groups of laticifers containing light yellow substances arranged radially in phloem, intercellular spaces usually observed; vessels of xylem arranged radially; starch grains and crystals of inulin usually contained in phloem parenchyma cells.

Identification To 2.0 g of pulverized Codonopsis Root add 50 mL of water, and heat in a water bath for 1 hour. After cooling, filter, and wash the filtrate with two 20-mL portions of ethyl acetate. Separate the aqueous layer, extract with two 30-mL portions of water saturated 1-butanol. Combine the 1-butanol layers, and evaporate the solvent in a water bath under low pressure (in vacuo). Dissolve the residue in 1 mL of methanol, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, water and ethyl acetate (6:5:2) to a distance of about 10 cm, and air-dry the plate. Spray evenly naphthoresorcin-phosphoric acid TS on the plate, and heat the plate at 105°C for 10 minutes: an orange to red-purple spot at an *R*f value of about 0.5 is observed.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Codonopsis Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Codonopsis Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 23.0% (6 hours).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 25.0%.

Containers and storage Containers—Well-closed containers.

Coix Seed

Coicis Semen

ヨクイニン

Coix Seed is the seed of *Coix lachryma-jobi* Linné var. *mayuen* Stapf (*Gramineae*), from which the seed coat has been removed.

Description Ovoid or broad ovoid seed, about 6 mm in length, and about 5 mm in width; with a slightly hollowed apex and base; dorsal side distended; ventral side longitudinally and deeply furrowed in the center; dorsal side mostly white in color and powdery; in the furrow on the ventral surface, attached brown, membranous pericarp and seed coat. Under a magnifying glass, the cross section reveals light yellow scutellum in the hollow of the ventral side. Hard in texture.

Odor, slight; taste, slightly sweet; adheres to the teeth on chewing.

Identification To a transverse section of Coix Seed add iodine TS dropwise: a dark red-brown color develops in the endosperm, and a dark gray color develops in the scutellum.

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Powdered Coix Seed

Coicis Semen Pulveratum

ヨクイニン末

Powdered Coix Seed is the powder of Coix Seed.

Description Powdered Coix Seed occurs as a brownish, grayish white to grayish yellow-white powder, and has a slight odor and a slightly sweet taste.

Under a microscope <*5.01*>, Powdered Coix Seed reveals starch grains, and fragments of endosperm containing them; fragments of tissue accompanied with epidermal cells of pericarp composed of yellowish and oblong cells, and fragments of parenchyma cells containing fixed oil, aleuron grains and starch grains; a very few fragments of spiral vessels. Starch grains are simple and 2-compound grains, simple grain nearly equidiameter to obtuse polygon, 10 – 20 μm in diameter, and have a stellate cleft-like hilum in the center. Spherical starch grains, coexisting with aleuron grains, are spherical simple grains, 3 – 7 μm in diameter.

Identification Place a small amount of Powdered Coix Seed on a slide glass, add dropwise iodine TS, and examine under a microscope <*5.01*>: nearly equidiameter and obtuse polygonal simple starch grains, usually 10 – 15 μm in diameter, and compound starch grains have a reddish brown color. Small spheroidal starch grains, coexisting with fixed oil and with aleuron grains in parenchymatous cells, have a blue-purple color.

Purity Foreign matter—Under a microscope <*5.01*>, Powdered Coix Seed reveals no fragments of tissue having silicified cell wall, no stone cells, no fragments of other thick-walled and lignified cells, no fragments of reticulate, scalariform and pitted vessels, no fragments of fibers and hairs, and no large starch grains, more than 10 μm in diameter, appearing blue-purple upon addition of iodine TS.

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 3.0%.

Containers and storage Containers—Tight containers.

Condurango

Condurango Cortex

コンズランゴ

Condurango is the bark of the trunk of *Marsdenia cundurango* Reichenbach filius (*Asclepiadaceae*).

Description Tubular or semi-tubular pieces of bark, 0.1 – 0.6 cm in thickness, 4 – 15 cm in length; outer surface grayish brown to dark brown, nearly smooth and with numerous lenticels, or more or less scaly and rough; inner surface light grayish brown and longitudinally striate; fractured surface fibrous on the outer region and generally granular in the inner region.

Odor, slight; taste, bitter.

Under a microscope <*5.01*>, a transverse section reveals a cork layer composed of several cellular layers of thin-walled cells; primary cortex with numerous stone cell groups; secondary cortex with phloem fiber bundles scattered inside the

starch sheath consisting of one-cellular layer; articulate latex tubes scattered in both cortices; parenchyma cells containing starch grains or rosette aggregates of calcium oxalate; starch grain 3 – 20 μm in diameter.

Identification Digest 1 g of pulverized Condurango in 5 mL of water, and filter: the clear filtrate becomes turbid on heating, but becomes clear again upon cooling.

Purity Foreign matter <*5.01*>—The xylem and other foreign matter contained in Condurango do not exceed 2.0%.

Total ash <*5.01*> Not more than 12.0%.

Containers and storage Containers—Well-closed containers.

Condurango Fluidextract

コンズランゴ流エキス

Method of preparation Take moderately fine powder of Condurango, and prepare the fluidextract as directed under Fluidextracts using a suitable quantity of a mixture of Purified Water or Purified Water in Containers, Ethanol and Glycerin (5:3:2) as the first solvent, and a suitable quantity of a mixture of Purified Water or Purified Water in Containers and Ethanol (3:1) as the second solvent.

Description Condurango Fluidextract is a brown liquid. It has a characteristic odor and a bitter taste.

Identification Mix 1 mL of Condurango Fluidextract with 5 mL of water, filter, if necessary, and heat the clear solution: turbidity is produced. However, it becomes almost clear upon cooling.

Purity Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Condurango Fluidextract as direct under the Fluidextracts (4), and perform the test (not more than 30 ppm).

Containers and storage Containers—Tight containers.

Coptis Rhizome

Coptidis Rhizoma

オウレン

Coptis Rhizome is the rhizome of *Coptis japonica* Makino, *Coptis chinensis* Franchet, *Coptis deltoidea* C. Y. Cheng et Hsiao or *Coptis teeta* Wallich (*Ranunculaceae*), from which the roots have been removed practically.

It contains not less than 4.2% of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], calculated on the basis of dried material.

For Coptis Rhizome used only for extracts or infusions and decoctions, the label states the restricted utilization forms.

Description Irregular, cylindrical rhizome, 2 – 4 cm, rarely up to 10 cm in length, 0.2 – 0.7 cm in diameter, slightly curved and often branched; externally grayish yellow-brown, with ring nodes, and with numerous remains of rootlets; generally remains of petiole at one end; fractured surface rather fibrous; cork layer light grayish brown, cortex and pith are yellow-brown to reddish yellow-brown, xylem is yellow to reddish yellow in color.

Odor, slight; taste, extremely bitter and lasting; it colors the saliva yellow on chewing.

Under a microscope <*5.01*>, a transverse section of Coptis Rhizome reveals a cork layer composed of thin-walled cork cells; cortex parenchyma usually exhibiting groups of stone cells near the cork layer and yellow phloem fibers near the cambium; xylem consisting chiefly of vessels, tracheids and xylem fibers; medullary ray distinct; pith large; in pith, stone cells or stone cells with thick-walled and lignified cells are sometimes recognized; parenchyma cells contain minute starch grains.

Identification **(1)** To 0.5 g of pulverized Coptis Rhizome add 10 mL of water, allow to stand for 10 minutes with occasional shaking, and filter. To 2 to 3 drops of the filtrate add 1 mL of hydrochloric acid and 1 to 2 drops of hydrogen peroxide TS, and shake: a red-purple color develops.

(2) To 0.5 g of pulverized Coptis Rhizome add 20 mL of methanol, shake for 2 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberin chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution and a yellow to yellow-green fluorescence spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Coptis Rhizome according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm). When the decision is difficult by this method, perform the test as directed under Atomic Absorption Spectrophotometry <*2.23*>. Put 5.0 g of pulverized Coptis Rhizome in a platinum, quartz or porcelain crucible, heat gently, and then incinerate by ignition between 450°C and 550°C. After cooling, add a small amount of 2 mol/L nitric acid TS, filter if necessary, and wash the crucible and filter several times with small portions of 2 mol/L nitric acid TS. Combine the filtrate and the washings, add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the sample solution. Separately, to 2.5 mL of Standard Lead Solution add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the standard solution. Perform the test with the sample solution and the standard solution according to the following conditions: the absorbance of the sample solution is not more than that of the standard solution (not more than 5 ppm).

Gas: Combustible gas—Acetylene or hydrogen.
Supporting gas—Air.
Lamp: A lead hollow-cathode lamp.
Wavelength: 283.3 nm.

The procedure and permissible limit for Coptis Rhizome labeled to be used for extracts or infusions and decoctions are as follows.

To 4.0 g of moderately fine cuttings of Coptis Rhizome add 80 mL of water, and heat until the amount becomes about 40 mL with occasional stirring. After cooling, filter, and proceed with the filtrate according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of

Standard Lead Solution (not more than 5 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Coptis Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 11.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Assay Weigh accurately about 0.5 g of pulverized Coptis Rhizome, add 30 mL of a mixture of methanol and dilute hydrochloric acid (100:1), heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure twice with the residue, using 30-mL and 20-mL portions of a mixture of methanol and dilute hydrochloric acid (100:1). To the last residue add 10 mL of methanol, shake well, and filter. Combine the whole filtrates, add methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (previously determine the water <*2.48*> in the same manner as Berberine Chloride Hydrate), dissolve in methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$)]
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogenphosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: Adjust so that the retention time of berberine is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in methanol to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 5 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Coptis Rhizome

Coptidis Rhizoma Pulveratum

オウレン末

Powdered Coptis Rhizome is the powder of Coptis Rhizome.

It contains not less than 4.2% of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], calculated on the basis of dried material.

Description Powdered Coptis Rhizome occurs as a yellow-brown to grayish yellow-brown powder. It has a slight odor and an extremely bitter, lasting taste, and colors the saliva yellow on chewing.

Under a microscope <*5.01*>, almost all elements are yellow in color; it reveals mainly fragments of vessels, tracheids and xylem fibers; parenchyma cells containing starch grains; polygonal cork cells. Usually, round to obtuse polygonal stone cells and their groups, and phloem fibers, 10 – 20 μm in diameter, and fragments of their bundles. Sometimes, polygonal and elongated epidermal cells, originated from the petiole, having characteristically thickened cell walls. Starch grains are single grains 1 – 7 μm in diameter.

Identification **(1)** To 0.5 g of Powdered Coptis Rhizome add 10 mL of water, allow to stand for 10 minutes with occasional shaking, and filter. To 2 to 3 drops of the filtrate add 1 mL of hydrochloric acid and 1 to 2 drops of hydrogen peroxide TS, and shake: a red-purple color develops.

(2) To 0.5 g of Powdered Coptis Rhizome add 20 mL of methanol, shake for 2 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberine chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the spot among the several spots from the sample solution and a yellow to yellow-green fluorescence spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of Powdered Coptis Rhizome according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm). When the decision is difficult by this method, perform the test as directed under Atomic Absorption Spectrophotometry <*2.23*>. Put 5.0 g of Powdered Coptis Rhizome in a platinum, quartz or porcelain crucible, heat gently, and then incinerate by ignition between 450°C and 550°C. After cooling, add a small amount of 2 mol/L nitric acid TS, filter if necessary, and wash the crucible and filter several times with small portions of 2 mol/L nitric acid TS. Combine the filtrate and the washings, add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the sample solution. Separately, to 2.5 mL of Standard Lead Solution add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the standard solution. Perform the test with the sample solution and the standard solution according to the following conditions: the absorbance of the sample solution is not more than that

of the standard solution (not more than 5 ppm).

Gas: Combustible gas—Acetylene or hydrogen.
Supporting gas—Air.

Lamp: A lead hollow-cathode lamp.

Wavelength: 283.3 nm.

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Coptis Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Phellodendron bark—Under a microscope <*5.01*>, crystal cell rows or mucilage masses are not observable. Stir 0.5 g of Powdered Coptis Rhizome with 2 mL of water: the solution does not become gelatinous.

(4) Curcuma—Place Powdered Coptis Rhizome on a filter paper, drop diethyl ether on it, and allow to stand. Remove the powder from the filter paper, and drop 1 drop of potassium hydroxide TS: no red-purple color develops. Under a microscope <*5.01*>, Powdered Coptis Rhizome does not contain gelatinized starch or secretory cells containing yellow-red resin.

Loss on drying <*5.01*> Not more than 11.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Assay Weigh accurately about 0.5 g of Powdered Coptis Rhizome, add 30 mL of a mixture of methanol and dilute hydrochloric acid (100:1), heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure twice with the residue, using 30-mL and 20-mL portions of a mixture of methanol and dilute hydrochloric acid (100:1). To the last residue add 10 mL of methanol, shake well, and filter. Combine the whole filtrates, add methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (previously determine the water <*2.48*> in the same manner as Berberine Chloride Hydrate), dissolve in methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$)]
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogenphosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: Adjust so that the retention time of berberine is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in methanol to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 5 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Corn Oil

Oleum Maydis

トウモロコシ油

Corn Oil is the fixed oil obtained from the embryo of *Zea mays* Linné (*Gramineae*).

Description Corn Oil is a clear, light yellow oil. It is odorless or has a slight odor, and a mild taste.

It is miscible with diethyl ether and with petroleum ether.

It is slightly soluble in ethanol (95), and practically insoluble in water.

At −7°C, it congeals to an unguentary mass.

Specific gravity d_{25}^{25}: 0.915 – 0.921

Acid value <*1.13*> Not more than 0.2.

Saponification value <*1.13*> 187 – 195

Unsaponifiable matter <*1.13*> Not more than 1.5%.

Iodine value <*1.13*> 103 – 130

Containers and storage Containers—Tight containers.

Cornus Fruit

Corni Fructus

サンシュユ

Cornus Fruit is the pulp of the pseudocarp of *Cornus officinalis* Siebold et Zuccarini (*Cornaceae*).

It contains not less than 0.4% of loganin, calculated on the basis of dried material.

Description Flattened oblong, 1.5 – 2 cm in length, about 1 cm in width; externally dark red-purple to dark purple, lustrous, and with coarse wrinkles; a crack-like scar formed by removal of true fruits; a scar of calyx at one end, and a scar of peduncle at the other; soft in texture.

Odor, slight; taste, acid and occasionally slightly sweet.

Identification To 1 g of coarse cuttings of Cornus Fruit add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of loganin for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (6:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots

obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution. Further, a spot, slightly different in color tone from the above-mentioned spot, is found immediately below of the spot.

Purity **(1)** Foreign matter <*5.01*>—The amount of its peduncles and other foreign matter contained in Cornus Fruit does no exceed 2.0%.

(2) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 5.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 35.0%.

Assay Weigh accurately about 1 g of fine cuttings of Cornus Fruit (separately determine the loss on drying <*5.01*>), put in a glass-stoppered centrifuge tube, add 30 mL of diluted methanol (1 in 2), shake for 20 minutes, centrifuge, and take the supernatant liquid. To the residue add 30 mL of diluted methanol (1 in 2), and repeat the above process twice more. Combine all the extacts, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of loganin for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of loganin in each solution.

$$\text{Amount (mg) of loganin} = M_S \times A_T/A_S$$

M_S: Amount (mg) of loganin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 238 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and methanol (55:4:1).

Flow rate: Adjust so that the retention time of loganin is about 25 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of loganin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of loganin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Corydalis Tuber

Corydalis Tuber

エンゴサク

Corydalis Tuber is the tuber of *Corydalis turtschaninovii* Basser forma *yanhusuo* Y. H. Chou et C. C. Hsu (*Papaveraceae*), usually after being passed through hot water.

It contains not less than 0.08% of dehydrocorydaline [as dehydrocorydaline nitrate ($C_{22}H_{24}N_2O_7$)], calculated on the basis of dried material.

Description Nearly flattened spherical, 1 – 2 cm in diameter, and with stem scar at one end; externally grayish yellow to grayish brown; hard in texture; fractured surface is yellow and smooth or grayish yellow-green in color and granular.

Almost odorless; taste, bitter.

Identification To 2 g of pulverized Corydalis Tuber add 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of dehydrocorydaline nitrate for thin-layer chromatography in 20 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of methanol, ammonium acetate solution (3 in 10) and acetic acid (100) (20:1:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution, and a yellow fluorescent spot appears at the lower side of the spot. Separately, spray evenly Dragendorff's TS for spraying on the plate, air-dry, and then spray sodium nitrite TS: a brown spot appears at an *R*f value of about 0.6.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Corydalis Tuber according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Corydalis Tuber according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 15.0%.

Total ash <*5.01*> Not more than 3.0%.

Assay Weigh accurately about 1 g of pulverized Corydalis Tuber, add 30 mL of a mixture of methanol and dilute hydrochloric acid (3:1), heat under a reflux condenser for 30 minutes, and filter after cooling. To the residue add 15 mL of a mixture of methanol and dilute hydrochloric acid (3:1), and repeat the above procedure. Combine all the filtrates, add a mixture of methanol and dilute hydrochloric acid (3:1) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of dehydrocorydaline nitrate for assay, previously dried in a desiccator (silica gel) for not less than 1 hour, dissolve in a mixture of methanol and dilute hydrochloric acid (3:1) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and

determine the peak areas, A_T and A_S, of dehydrocorydaline in each solution.

Amount (mg) of dehydrocorydaline [as dehydrocorydaline nitrate ($C_{22}H_{24}N_2O_7$)]
$= M_S \times A_T/A_S \times 1/4$

M_S: Amount (mg) of dehydrocorydaline nitrate for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 17.91 g of disodium hydrogen phosphate dodecahydrate in 970 mL of water, and adjust to pH 2.2 with phosphoric acid. To this solution add 14.05 g of sodium perchlorate, dissolve, and add water to make exactly 1000 mL. To this solution add 450 mL of acetonitrile, then dissolve 0.20 g of sodium lauryl sulfate.

Flow rate: Adjust so that the retention time of dehydrocorydaline is about 24 minutes.

System suitability—

System performance: Dissolve 1 mg each of dehydrocorydaline nitrate for assay and berberine chloride hydrate in 20 mL of a mixture of water and acetonitrile (20:9). When the procedure is run with 5 μL of this solution under the above operating conditions, berberine and dehydrocorydaline are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 5 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of dehydrocorydaline is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Corydalis Tuber

Corydalis Tuber Pulveratum

エンゴサク末

Powdered Corydalis Tuber is the powder of Corydalis Tuber.

It contains not less than 0.08% of dehydrocorydaline [as dehydrocorydaline nitrate ($C_{22}H_{24}N_2O_7$)], calculated on the basis of dried material.

Description Powdered Corydalis Tuber occurs as a greenish yellow to grayish yellow powder. Almost odorless; taste, bitter.

Under a microscope <*5.01*>, Powdered Corydalis Tuber reveals mainly, masses of gelatinized starch or light yellow to colorless parenchymatous cells containing starch grains, fragments of cork layers, light yellow stone cells, sclerenchymatous cells, reticulate vessels, spiral vessels and ring vessels; starch grains observed simple grains and 2- to 3-compound grains.

Identification To 2 g of Powdered Corydalis Tuber add 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of dehydrocorydaline nitrate for thin-layer chromatography in 20 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of methanol, ammonium acetate solution (3 in 10) and acetic acid (100) (20:1:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution, and a yellow fluorescent spot appears at the lower side of the spot. Separately, spray evenly Dragendorff's TS for spraying on the plate, air-dry, and then spray sodium nitrite TS: a brown spot appears at an *R*f value of about 0.6.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Corydalis Tuber according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Corydalis Tuber according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 15.0%.

Total ash <*5.01*> Not more than 3.0%.

Assay Weigh accurately about 1 g of Powdered Corydalis Tuber, add 30 mL of a mixture of methanol and dilute hydrochloric acid (3:1), heat under a reflux condenser for 30 minutes, and filter after cooling. To the residue add 15 mL of the mixture of methanol and dilute hydrochloric acid (3:1), and proceed in the same way as above. Combine all the filtrates, add the mixture of methanol and dilute hydrochloric acid (3:1) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of dehydrocorydaline nitrate for assay, previously dried in a desiccator (silica gel) for not less than 1 hour, dissolve in the mixture of methanol and dilute hydrochloric acid (3:1) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of dehydrocorydaline in each solution.

Amount (mg) of dehydrocorydaline [as dehydrocorydaline nitrate ($C_{22}H_{24}N_2O_7$)]
$= M_S \times A_T/A_S \times 1/4$

M_S: Amount (mg) of dehydrocorydaline nitrate for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 17.91 g of disodium hydrogen phosphate dodecahydrate in 970 mL of water, and adjust to pH 2.2 with phosphoric acid. To this solution add 14.05 g of sodium perchlorate, dissolve, and add water to make exactly 1000 mL. Add 450 mL of acetonitrile, and dissolve 0.20 g of sodium lauryl sulfate in this solution.

Flow rate: Adjust so that the retention time of dehydro-

corydaline is about 24 minutes.

System suitability—

System performance: Dissolve 1 mg of dehydrocorydaline nitrate for assay and 1 mg of berberine chloride hydrate in 20 mL of a mixture of water and acetonitrile (20:9). When the procedure is run with 5 μL of this solution under the above operating conditions, berberine and dehydrocorydaline are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 5 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of dehydrocorydaline is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Crataegus Fruit

Crataegi Fructus

サンザシ

Crataegus Fruit is the pseudocarp of **1)** *Crataegus cuneata* Siebold et Zuccarini or **2)** *Crataegus pinnatifida* Bunge var. *major* N. E. Brown (*Rosaceae*) without any treatment or cut crosswise or lengthwise.

Description

1) *Crataegus cuneata* origin—Nearly spherical fruits, 8 – 14 mm in diameter; externally yellow-brown to grayish brown, with fine reticulated wrinkles, remained dent of 4 – 6 mm in diameter at one end, often the base of calyx around the dent, short peduncle or scar at the other end. True fruits, usually five loculus, often split five, mericarp, 5 – 8 mm in length, light brown, usually, containing one seed into each mericarp.

Almost odorless; taste, slightly acid.

Under a microscope <*5.01*>, a transverse section of central parts reveals in the outermost layer composed of epidermis to be covered with comparatively thick cuticle layer, cuticle intrude into lateral cell walls of epidermis, and reveal wedge-like. Cell of the epidermis or 2- to 3-cellular layer of parenchyma cells beneath these observed contents of yellow-brown to red-brown in color followed these appeared parenchyma. Vascular bundles and numerous stone cells appear single or gathered 2 to several cells scattered on the parenchyma, and observed solitary crystals and clustera crystals of calcium oxalate. Pericarp of true fruits composed of mainly sclerenchyma cells, seed covered with seed coats, perisperm, endosperm, cotyledon observed inside seed coats; sclerenchyma cells of true fruits and cells of seed coats containing solitary crystals of calcium oxalate.

2) *Crataegus pinnatifida* var. *major* origin—Approximate to 1), but it is large in size, 17 – 23 mm in diameter, the outer surface red-brown and lustrous, spot-like scars of hairs are distinct. At one end remained dent, 7 – 9 mm in diameter, mericarp, 10 – 12 mm in length, yellow-brown in color, usually ripe seeds are absent.

Odor, characteristic; taste, acid.

Under a microscope <*5.01*>, a transverse section of the central parts approximate to 1), but it contains a few stone cells in parenchyma.

Identification

1) *Crataegus cuneata* origin—To 1.0 g of pulverized Crataegus Fruit add 5 mL of methanol, shake for 30 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of rutin for thin-layer chromatography in 20 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, 2-butanone, water and formic acid (5:3:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the green fluorescent spot from the standard solution, and one or two similar green fluorescent spots are found at an *R*f value of about 0.5. These spots disappear gradually by allowing to cool, and appear again by heating.

2) *Crataegus pinnatifida* var. *major* origin—To 1 g of pulverized Crataegus Fruit add 5 mL of methanol, shake for 30 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of hyperoside for thin-layer chromatography in 20 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, 2-butanone, water and formic acid (5:3:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the green fluorescent spot from the standard solution, and a similar fluorescent spot is found just above the spot. These spots disappear gradually by allowing to cool, and appear again by heating.

Loss on drying <*5.01*> Not more than 17.0%.

Total ash <*5.01*> Not more than 4.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Curcuma Rhizome

Curcumae Rhizoma

ガジュツ

Curcuma Rhizome is the rhizome of 1) *Curcuma zedoaria* Roscoe, 2) *Curcuma phaeocaulis* Valeton or 3) *Curcuma kwangsiensis* S. G. Lee et C. F. Liang (*Zingiberaceae*), usually after being passed through hot water.

Description Nearly ovoid to oblong-ovoid or conical rhizome, 2 – 8 cm in length, 1.5 – 4 cm in diameter; externally grayish yellow-brown to grayish brown; nodes protruded as rings; internode of 0.3 – 0.8 cm, with scars of roots, and small protrusions consisting of scars of branched rhizomes; hard in texture; a transverse section reveals cortex and stele distinctly; cortex 2 – 5 mm in thickness; a transverse section,

grayish brown in rhizome of 1) *Curcuma zedoria* origin, light yellow to grayish yellow or light yellow-green to grayish yellow-green in 2) *Curcuma phaeocaulis* origin and purplish brown to dark purple-brown in 3) *Curcuma kwangsiensis* origin, and sometimes lustrous.

Odor, characteristic; taste, pungent, bitter and cool feeling on chewing.

Under a microscope <*5.01*>, a transverse section of central part reveals the outermost layer usually consisting of a cork layer 4 – 10 cells thick; cortex and stele divided by endodermis, composed of parenchyma cells, vascular bundles scattered; small sized vascular bundles line up beneath the endodermis; oil cells contain yellow-brown to dark brown oily substances, scattered in parenchyma; parenchyma contains gelatinized starch and rarely crystals of calcium oxalate.

Identification To 2.0 g of pulverized Curcuma Rhizome add 5 mL of water, shake, then add 5 mL of hexane, shake for 10 minutes, centrifuge, and use the hexane layer as the sample solution. Perform the test with this solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybezaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: a deep blue to dark brown spot and a red-brown to brown spot appear at *R*f values of about 0.3 and about 0.2, respectively.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Curcuma Rhizome according to Method 3, and perform the test. Prepare the control solution with 1.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Curcuma Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 7.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Curcuma Rhizome, provided that 1 mL of silicon resin is previously added on the sample in the flask: the volume of essential oil is not less than 0.5 mL.

Containers and storage Containers—Well-closed containers.

Cyperus Rhizome

Cyperi Rhizoma

コウブシ

Cyperus Rhizome is the rhizome of *Cyperus rotundus* Linné (*Cyperaceae*).

Description Fusiform rhizome, 1.5 – 2.5 cm in length, 0.5 – 1 cm in diameter; externally grayish brown to grayish blackish brown, with 5 to 8 irregular ring nodes, and with hair-like fiber bundles on each node; hard in texture. The transverse section red-brown to light yellow in color, with waxy luster; thickness of cortex approximately equal to or slightly smaller than the diameter of stele. Under a magnifying glass, a cut surface reveals fiber bundles as brown spots lined in rings along circumference; here and there in the cortex, vascular bundles appear as red-brown spots, and numerous secretory cells scattered as minute yellow-brown spots; in the stele, numerous vascular bundles scattered as spots or lines.

Characteristic odor and taste.

Identification To 2.0 g of pulverized Cyperus Rhizome add 10 mL of diethyl ether, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of diethyl ether, cyclohexane and formic acid (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.35.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Cyperus Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Cyperus Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 3.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Cyperus Rhizome, provided that 1 mL of silicon resin is previously added on the sample in the flask: the volume of essential oil is not less than 0.3 mL.

Containers and storage Containers—Well-closed containers.

Powdered Cyperus Rhizome

Cyperi Rhizoma Pulveratum

コウブシ末

Powdered Cyperus Rhizome is the powder of Cyperus Rhizome.

Description Powdered Cyperus Rhizome occurs as a light red-brown powder, and has a characteristic odor and taste.

Under a microscope <*5.01*>, Powdered Cyperus Rhizome reveals fragments of polygonal parenchyma cells, scalariform vessels, and seta-like fibers; a large quantity of starch, mostly gelatinized; an extremely small number of stone cells.

Identification To 2.0 g of Powdered Cyperus Rhizome add 10 mL of diethyl ether, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of diethyl ether, cyclohexane and formic acid (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.35.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Cyperus Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Cyperus Rhizome according to Method 4, and

perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Cyperus Rhizome does not show extremely lignified cells, except stone cells, and crystals.

Total ash <*5.01*> Not more than 3.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Essential oil content <*5.01*> Perform the test with 50.0 g of Powdered Cyperus Rhizome provided that 1 mL of silicon resin is previously added on the sample in the flask: the volume of essential oil is not less than 0.2 mL.

Containers and storage Containers—Tight containers.

Daiokanzoto Extract

大黄甘草湯エキス

Daiokanzoto Extract contains not less than 3.5 mg of sennoside A ($C_{42}H_{38}O_{20}$: 862.74), and not less than 7 mg and not more than 21 mg (for preparation prescribed 1 g of Glycyrrhiza) or not less than 14 mg and not more than 42 mg (for preparation prescribed 2 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Rhubarb	4 g	4 g
Glycyrrhiza	1 g	2 g

Prepare a dry extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Daiokanzoto Extract occurs as a brown powder. It has a characteristic odor and an astringent first then slightly sweet taste.

Identification (1) To 1.0 g of Daiokanzoto Extract add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Rhubarb).

(2) To 0.5 g of Daiokanzoto Extract add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity (1) Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Daiokanzoto Extract as directed under Extract (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of Daiokanzoto Extract according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> Not more than 7.0% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%.

Assay (1) Sennoside A—Weigh accurately about 0.2 g of Daiokanzoto Extract, add 20 mL of ethyl acetate and 10 mL of water, shake for 10 minutes, centrifuge, and remove the ethyl acetate layer. To the aqueous layer add 20 mL of ethyl acetate, shake for 10 minutes, centrifuge, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of Sennoside A RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of sennoside A in each solution.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (2460:540:1).

Flow rate: 1.0 mL per minute (the retention time of sennoside A is about 14 minutes.)

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of sennoside A are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.2 g of

Daiokanzoto Extract, add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Daisaikoto Extract

大柴胡湯エキス

Daisaikoto Extract contains not less than 1.8 mg and not more than 7.2 mg of saikosaponin b_2, not less than 80 mg and not more than 240 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 26 mg and not more than 78 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)	5)
Bupleurum Root	6 g	6 g	6 g	6 g	6 g
Pinellia Tuber	4 g	4 g	4 g	3 g	4 g
Scutellaria Root	3 g	3 g	3 g	3 g	3 g
Peony Root	3 g	3 g	3 g	3 g	3 g
Jujube	3 g	3 g	3 g	3 g	3 g
Immature Orange	2 g	2 g	2 g	2 g	2 g
Ginger	1 g	1 g	2 g	1 g	1.5 g
Rhubarb	1 g	2 g	1 g	1 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 5), using the crude drugs shown above.

Description Daisaikoto Extract occurs as light yellow-brown to brown powder or black-brown viscous extract, having a slightly order, and a hot first, then a bitter taste.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown to grayish brown spot from the standard solution (Scutellaria Root).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2)

to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzoaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple to purple spot from the standard solution (Peony Root).

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 1.0 g of pulverized immature orange add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in an ammonia gas: two consecutive spots at *R*f values of about 0.7 obtained from the sample solution have respectively the same color tone and *R*f value with the blue-green spot and blue spot underneath from the standard solution (Immature Orange).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Rhubarb).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 11.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, separate the supernatant liquid, combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly

10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, flow through in a column packed with 2 g of polyamide for column chromatography, elute with 20 mL of water, add 1 mL of acetic acid (100), to the effluent, then add water to make exactly 25 mL, and use this as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 5/8$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Digenea

Digenea

マクリ

Digenea is the whole algae of *Digenea simplex* C. Agardh (*Rhodomelaceae*).

Description Rounded, string-like algae, 2 – 3 mm in diameter; externally, dark red-purple to dark grayish red or grayish brown; a few branched rods irregularly forked, covered with short hairy twigs; calcified weeds and other small algae often attached.

Odor, seaweed-like; taste, disagreeable and slightly salty.

Identification To 2 g of pulverized Digenea add 10 mL of dilute ethanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 5 mg of kainic acid in 10 mL of dilute ethanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, water and formic acid (5:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity Foreign matter <5.01>—The amount of other algae in Digenea does not exceed 20.0%.

Loss on drying <5.01> Not more than 22.0%.

Acid-insoluble ash <5.01> Not more than 8.0%.

Containers and storage Containers—Well-closed containers.

Dioscorea Rhizome

Dioscoreae Rhizoma

サンヤク

Dioscorea Rhizome is the rhizome (rhizophore) of *Dioscorea japonica* Thunberg or *Dioscorea batatas* Decaisne (*Dioscoreaceae*), from which the periderm has been removed.

Description Cylindrical or irregular cylindrical rhizome, 5 – 15 cm in length, 1 – 4 cm in diameter, occasionally longitudinally split or transversely cut; externally whitish to yellowish white; fractured surface, whitish, smooth and powdery; hard in texture but breakable.

Practically odorless and tasteless.

Identification **(1)** To the cut surface of Dioscorea Rhizome add diluted iodine TS (1 in 50) dropwise: a dark purple to dark blue color develops.

(2) To 0.2 g of pulverized Dioscorea Rhizome add 2 mL of acetic anhydride, warm on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add 0.5 mL of sulfuric acid carefully to make two layers: a red-brown to purple-brown color appears at the zone of contact.

(3) To 1 g of pulverized Dioscorea Rhizome add 4 mL of a mixture of methanol and water (4:1), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of allantoin for thin-layer chromatography in 2 mL of a mixture of methanol and water (4:1), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (7:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly a solution of 0.2 g of 4-dimethylaminocinnamaldehyde in 10 mL of 6 mol/L hydrochloric acid TS and 10 mL of ethanol (99.5) on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Dioscorea Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Dioscorea Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Powdered Dioscorea Rhizome

Dioscoreae Rhizoma Pulveratum

サンヤク末

Powdered Dioscorea Rhizome is the powder of Dioscorea Rhizome.

Description Powdered Dioscorea Rhizome occurs as nearly yellowish white to white; odorless and tasteless.

Under a microscope <*5.01*>, Dioscorea rhizome powder reveals starch grains; fragments of parenchyma cells containing starch grains; raphides of calcium oxalate, 100 to 200 μm in length and its containing mucilage cells; ring and scalariform vessels, 15 to 35 μm in diameter; starch grain isosceles deltoid or oblong, solitary, 18 to 35 μm, hilum and striation being distinct.

Identification **(1)** To 0.2 g of Powdered Dioscorea Rhizome add 2 mL of acetic anhydride, warm on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add carefully 0.5 mL of sulfuric acid to make two layers: a red-brown to purple-brown color develops at the zone of contact.

(2) To 1 g of Powdered Dioscorea Rhizome add 4 mL of a mixture of methanol and water (4:1), shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of allantoin for thin-layer chromatography in 2 mL of a mixture of methanol and water (4:1), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (7:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly a solution of 0.2 g of 4-dimethylaminocinnamaldehyde in 10 mL of 6 mol/L hydrochloric acid TS and 10 mL of ethanol (99.5) on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Dioscorea Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Dioscorea Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Tight containers.

Dolichos Seed

Dolichi Semen

ヘンズ

Dolichos Seed is the seed of *Dolichos lablab* Linné (*Leguminosae*).

Description Flattened ellipsoidal to flattened orbicular-ovate seed, 9 – 14 mm in length, 6 – 10 mm in width, 4 – 7 mm in thickness; externally light yellowish white to light yellow, smooth and somewhat lustrous; caruncle white, like a half-moon, protrudent at one side; hard in texture.

Almost odorless; taste, slightly sweet and acid.

Under a microscope <*5.01*>, a transverse section reveals the outermost layer of seed coat composed of a single layer of palisade like epidermal cells coated with cuticle; beneath epidermis a single layer of sclerenchymatous and sandglass like cells; inside of the layer mentioned above parenchyma lie, the innermost portion of the parenchyma decayed; cotyledons occur inside of the seed coat; the outermost layer of cotyledon composed of a single layer of epidermal cells, inner part of cotyledon mainly parenchyma, containing aleurone grains and oil drops, and occasionally starch grains.

Identification To 3 g of pulverized Dolichos Seed add 30 mL of methanol, shake for 10 minutes, centrifuge, and take the supernatant liquid. Evaporate the solvent of the supernatant liquid, add 30 mL of water and 50 mL of ethyl acetate to the residue, shake, and take the ethyl acetate layer. To the ethyl acetate add 10 g of anhydrous sodium sulfate, shake, and filter. Evaporate the solvent of the filtrate, add 1 mL of ethyl acetate to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate and acetic acid (100) (100:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a blue-white fluorescent spot appears at an *R*f value of about 0.4.

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 4.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 9.0%.

Containers and storage Containers—Well-closed containers.

Eleutherococcus Senticosus Rhizome

Eleutherococci senticosi Rhizoma

シゴカ

Eleutherococcus Senticosus Rhizome is the rhizome of *Eleutherococcus senticosus* Maximowicz (*Acanthopanax senticosus* Harms) (*Araliaceae*), often with root.

Description Slightly curved subcolumnar rhizome, 15 – 30 cm in length, 1 – 2.5 cm in diameter; externally grayish brown and slightly rough; transversely cut surface light brown, cortex thin, xylem thick with a pith in center; extremely hard in texture.

Odor, slightly characteristics; tasteless or slightly sweet, astringency.

Under a microscope <*5.01*>, a transverse section reveals the outermost layer consisting of a cork layer 3 – 7 cells thick; oil canals scattered in parenchyma; fiber bundles lined stepwise in phloem; phloem and xylem separated clearly by cambium; xylem composed of vessels, xylem fibers and xylem parenchyma; ray composed of 2 – 6 rows of cells; pith composed of parenchyma; parenchyma of cortex and ray contain aggregate crystals of calcium oxalate; occasionally starch grains in ray, parenchyma of cortex and xylem.

Identification To 0.5 g of pulverized Eleutherococcus Senticosus Rhizome add 20 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of eleutheroside B for liquid chromatography in diluted methanol (1 in 2) to make 20 mL. To 2 mL of this solution add diluted methanol (1 in 2) to make 20 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions: the peak corresponding to eleutheroside B in the chromatogram obtained from the sample solution shows the same retention time with the peak of eleutheroside B in the chromatogram from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 265 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water and acetonitrile (9:1).

Flow rate: Adjust so that the retention time of eleutheroside B is about 10 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of eleutheroside B are not less than 5000 and not more than 1.5, respectively.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Eleutherococcus Senticosus Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Eleutherococcus Senticosus Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 2.5%.

Containers and storage Containers—Well-closed containers.

Ephedra Herb

Ephedrae Herba

マオウ

Ephedra Herb is the terrestrial stem of *Ephedra sinica* Stapf, *Ephedra intermedia* Schrenk et C.A. Meyer or *Ephedra equisetina* Bunge (*Ephedraceae*).

Ephedra Herb contains not less than 0.7% of total alkaloids (as ephedrine and pseudoephedrine), calculated on the basis of dried material.

Description Thin cylindrical or ellipsoidal cylinder, 0.1 – 0.2 cm in diameter; 3 – 5 cm in length of internode; light green to yellow-green; numerous parallel vertical furrows on the surface; scaly leaves at the node portion; leaves, 0.2 – 0.4 cm in length, light brown to brown in color, usually being opposite at every node, adhering at the base to form a tubular sheath around the stem. Under a magnifying glass, the transverse section of the stem appears as circle and ellipse, the outer portion grayish green to yellow-green in color, and the center filled with a red-purple substance or hollow. When fractured at internode, the outer part is fibrous and easily split vertically.

Odor, slight; taste, astringent and slightly bitter, giving a slight sensation of numbness on the tongue.

Identification To 0.5 g of pulverized Ephedra Herb add 10 mL of methanol, shake for 2 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying, and heat the plate at 105°C for 5 minutes: a red-purple spot appears at an *R*f value of about 0.35.

Purity **(1)** Woody stem—When perform the test of foreign matter <*5.01*>, the amount of the woody stems contained in Ephedra Herb does not exceed 5.0%.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than woody stems contained in Ephedra Herb does not exceed 1.0%.

Loss on drying <*5.01*> Not more than 12.5% (6 hours).

Total ash <*5.01*> Not more than 11.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Assay Weigh accurately about 0.5 g of moderately fine powder of Ephedra Herb, place in a glass-stoppered centrifuge tube, add 20 mL of diluted methanol (1 in 2), shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), proceed in the same manner, and repeat this procedure twice. Combine all the extracts, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 50 mg of ephedrine hydrochloride for assay of crude drugs, previously dried at 105°C for 3 hours, and dissolve in diluted methanol (1 in 2) to make exactly 20 mL. Pipet 2 mL of the solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine (the relative retention time to ephedrine is about 0.9) obtained from the sample solution, and the peak area, A_S, of ephedrine obtained from the standard solution.

Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)

$$= M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drugs taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, and add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: Adjust so that the retention time of ephedrine is about 27 minutes.

System suitability—

System performance: Dissolve 1 mg of ephedrine hydrochloride for assay of crude drugs and 1 mg of pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Epimedium Herb

Epimedii Herba

インヨウカク

Epimedium Herb is the terrestrial part of *Epimedium koreanum* Nakai, *Epimedium grandiflorum* Morren var. *thunbergianum* Nakai, *Epimedium pubescens* Maximowicz, *Epimedium brevicornu* Maximowicz, *Epimedium wushanense* T. S. Ying, *Epimedium sagittatum* Maximowicz or *Epimedium sempervirens* Nakai (*Berberidaceae*).

Description Epimedium Herb is composed of a stem and a ternate to triternate compound leaf; leaflet ovate to broadly ovate or ovate-lanceolate, 3 – 20 cm in length, 2 – 8 cm in width, petiolule 1.5 – 7 cm in length, apex of leaflet acuminate, needle hair on margin 0.1 – 0.2 cm in length, base of leaflet cordate to deeply cordate, lateral leaflet asymmetry; upper surface green to green-brown, sometimes lustrous, lower surface light green to grayish green-brown, often pilose, especially on vein densely pilose, papery or coriaceous; petiole and stem cylindrical, light yellowish brown to slightly purplish and light green-brown, easily broken.

Odor, slight; taste, slightly bitter.

Under a microscope ⟨*5.01*⟩, a transverse section of the leaf reveals 3 – 6 vascular bundles in midvein; mesophyll composed of upper epidermis, single-layered palisade, spongy tissue and lower epidermis; leaf margins orbicular or oblong, sclerenchymatous; multi-cellular hairs on epidermis; 8 – 20 vascular bundles in petiole and 6 – 15 vascular bundles in petiolule. Under a microscope ⟨*5.01*⟩, a transverse section of the stem reveals a single to several-layered hypodermis, cortex of 4 – 10 cellular layers of sclerenchyma, vascular bundle 13 – 30 in number, oblong to obovate.

Identification To 2 g of pulverized Epimedium Herb add 20 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of icariin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying ⟨*5.01*⟩ Not more than 12.5% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 17.0%.

Containers and storage Containers—Well-closed containers.

Eucalyptus Oil

Oleum Eucalypti

ユーカリ油

Eucalyptus Oil is the essential oil distilled with steam from the leaves of *Eucalyptus globulus* Labillardière or allied plants (*Myrtaceae*).

It contains not less than 70.0% of cineol.

Description Eucalyptus Oil is a clear, colorless or pale yellow liquid. It has a characteristic, aromatic odor and a pungent taste.

It is neutral.

Identification Shake 1 mL of Eucalyptus Oil vigorously with 1 mL of phosphoric acid, and allow to stand: the solution congeals within 30 minutes.

Refractive index ⟨*2.45*⟩ n_D^{20}: 1.458 – 1.470

Specific gravity ⟨*1.13*⟩ d_{20}^{20}: 0.907 – 0.927

Purity **(1)** Clarity of solution—Mix 1.0 mL of Eucalyptus Oil with 5 mL of diluted ethanol (7 in 10): the solution is clear.

(2) Heavy metals ⟨*1.07*⟩—Proceed with 1.0 mL of Eucalyptus Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

Assay Weigh accurately about 0.1 g each of Eucalyptus Oil and cineol for assay, and dissolve each in hexane to make exactly 25 mL. Pipet 5 mL each of these solutions, add exactly 5 mL of the internal standard solution to each, then add hexane to make 100 mL, and use these solutions as the sample solution and the standard solution, respectively. Perform the test with 2 μL each of the sample solution and standard solution as directed under Gas Chromatography ⟨*2.02*⟩ according to the following conditions. Calculate the ratios, Q_T and Q_S, of the peak area of cineol to that of the internal standard.

$$\text{Amount (mg) of cineol} = M_S \times Q_T/Q_S$$

M_S: Amount (mg) of cineol for assay taken

Internal standard solution—A solution of anisole in hexane (1 in 250).

Operating conditions—

Detector: A hydrogen flame-ionization detector.

Column: A glass column 3 mm in inside diameter and 5 m in length, packed with silanized porous silica gel for gas chromatography coated in 5% with alkylene glycol phthalate ester for gas chromatography (150 to 180 μm in particle diameter).

Column temperature: A constant temperature of about 120°C.

Carrier gas: Nitrogen.

Flow rate: Adjust so that the retention time of cineol is about 11 minutes.

System suitability—

System performance: Dissolve 0.1 g each of cineol for assay and limonene in 25 mL of hexane. To 1 mL of this solution add hexane to make 20 mL. When the procedure is run with 2 μL of this solution under the above operating conditions, limonene and cineol are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 2 μL of the standard solution under the above operating conditions, the relative standard deviation of the ratio of the peak area of cineol to that of the internal standard is not more than 1.0%.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Eucommia Bark

Eucommiae Cortex

トチュウ

Eucommia Bark is the bark of *Eucommia ulmoides* Oliver (*Eucommiaceae*).

Description Eucommia Bark is a semi-tubular or plate-like bark, 2 – 6 mm in thickness; externally pale grayish brown to grayish brown, and rough in texture, sometimes red-brown due to the cork layer falling off; internally dark violet, smooth and covered with a linear pattern that runs longitudinally, silk-like threads of gutta-percha (a thermoplastic rubber-like substance) appearing when broken.

It has a faint but characteristic odor and taste.

Under a microscope ⟨*5.01*⟩, transverse section reveals parenchymatous cells containing gutta-percha; phloem with stone-cell and fiber layers; rays in rows of 2 – 3 cells; calcium oxalate crystals absent.

Identification Put 1 g of pulverized Eucommia Bark in a glass-stoppered centrifuge tube, add 10 mL of water and 20 mL of diethyl ether, shake for 15 minutes, and centrifuge. Take the diethyl ether layer so obtained, evaporate the solvent on a water bath, and add 1 mL of ethanol (99.5) to the residue: colloidal substances appear.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 5.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 7.0%.

Containers and storage Containers—Well-closed containers.

Euodia Fruit

Euodiae Fructus

ゴシュユ

Euodia Fruit is the fruit of *Euodia officinalis* Dode (*Evodia officinalis* Dode), *Euodia bodinieri* Dode (*Evodia bodinieri* Dode) or *Euodia ruticarpa* Hooker filius et Thomson (*Evodia rutaecarpa* Bentham) (Rutaceae).

Description Flattened spheroidal or globular fruit, 2 – 5 mm in diameter; externally dark brown to grayish brown, with many oil sacs appearing as hollow pits, and often with peduncle, 2 – 5 mm in length, covered densely with hairs; pericarp in matured split to five loculi, and each loculus containing obovoid or globular seeds of a lustrous brown to black-brown or bluish black color.

Odor, characteristic; taste, acrid, followed by a lasting bitterness.

Identification To 1.0 g of pulverized Euodia Fruit add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, 2-propanol, water and formic acid (7:7:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a blue-white fluorescent spot is observed at an *R*f value of about 0.6. The spot shows a yellow-red color after being sprayed evenly Dragendorff's TS for spraying.

Purity **(1)** Peduncle—The amount of peduncles contained in Euodia Fruit does not exceed 5.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than peduncles contained in Euodia Fruit does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Containers and storage Containers—Well-closed containers.

Fennel

Foeniculi Fructus

ウイキョウ

Fennel is the fruit of *Foeniculum vulgare* Miller (*Umbelliferae*).

Description Cylindrical cremocarp, 3.5 – 8 mm in length, 1 – 2.5 mm in width; externally grayish yellow-green to grayish yellow; two mericarps closely attached with each other, and with five longitudinal ridges; cremocarp often with pedicel 2 – 10 mm in length.

Characteristic odor and taste.

Under a microscope ⟨*5.01*⟩, ridges near the ventral side are far protruded than those on the dorsal side; one large oil canal between each ridge, and two oil canals on the ventral side.

Identification To 0.5 g of pulverized Fennel add 10 mL of hexane, allow to stand for 5 minutes with occasional shaking, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (20:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a spot with a dark purple color appears at an *R*f value of about 0.4.

Purity **(1)** Peduncle—When perform the test of foreign matter ⟨*5.01*⟩, the amount of peduncles contained in Fennel does not exceed 3.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than the peduncle contained in Fennel does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 10.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 50.0 g of pulverized Fennel: the volume of essential oil is not less than 0.7 mL.

Containers and storage Containers—Well-closed containers.

Powdered Fennel

Foeniculi Fructus Pulveratus

ウイキョウ末

Powdered Fennel is the powder of Fennel.

Description Powdered Fennel occurs as a greenish light brown to greenish brown, and is a characteristic odor and taste.

Under a microscope ⟨*5.01*⟩, Powdered Fennel reveals fragments of parenchyma cells of perisperm containing aleurone grain, fragments of parenchyma cells of endosperm containing fatty oil, fragments of sclerenchyma with characteristic simple pits, fragments of oil canal within yellow-brown material, fragments of endocarp shown scalariform, spiral vessels, fragments of epidermis or epidermis with stomata.

Identification To 0.5 g of Powdered Fennel add 10 mL of hexane, allow to stand for 5 minutes with occasional shaking, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate prepared with silica gel with fluorescent indicator for thin-layer chromatography. Then develop the plate with a mixture of hexane and ethyl acetate (20:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a spot with dark purple color appears at an *R*f value of about 0.4.

Total ash ⟨*5.01*⟩ Not more than 10.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 50.0 g of Powdered Fennel: the volume of essential oil is not less than 0.45 mL.

Containers and storage Containers—Tight containers.

Fennel Oil

Oleum Foeniculi

ウイキョウ油

Fennel Oil is the essential oil distilled with steam from the fruit of *Foeniculum vulgare* Miller (*Umbelliferae*) or of *Illicium verum* Hooker filius (*Illiciaceae*).

Description Fennel Oil is a colorless to pale yellow liquid. It has a characteristic, aromatic odor and a sweet taste with a slight, bitter aftertaste.

It is miscible with ethanol (95) and with diethyl ether.

It is practically insoluble in water.

When cold, white crystals or crystalline masses may often separate from the oil.

Identification Dissolve 0.30 g of Fennel Oil in 20 mL of hexane, pipet 1 mL of this solution, add hexane to make exactly 10 mL, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (20:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a spot with a dark purple color appears at the *R*f value of about 0.4.

Refractive index ⟨*2.45*⟩ n_D^{20}: 1.528 – 1.560

Specific gravity ⟨*1.13*⟩ d_{20}^{20}: 0.955 – 0.995

Purity **(1)** Clarity of solution—To 1.0 mL of Fennel Oil add 3 mL of ethanol (95): the solution is clear. To this solution add 7 mL of ethanol (95): the solution remains clear.

(2) Heavy metals ⟨*1.07*⟩—Proceed with 1.0 mL of Fennel Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Foeniculated Ammonia Spirit

アンモニア·ウイキョウ精

Method of preparation

Ammonia Water	170 mL
Fennel Oil	30 mL
Ethanol	a sufficient quantity
	To make 1000 mL

Prepare as directed under Spirits, with the above ingredients. A sufficient quantity of ammonia solution (28) and Purified Water or Purified Water in Containers may be used in place of Ammonia Water.

Description Foeniculated Ammonia Spirit is a colorless to yellow liquid, having a characteristic odor. It has a slightly sweet, pungent taste.

Specific gravity d_{20}^{20}: about 0.85

Alcohol number ⟨*1.01*⟩ Not less than 7.8 (Method 2).

Containers and storage Containers—Tight containers.

Forsythia Fruit

Forsythiae Fructus

レンギョウ

Forsythia Fruit is the fruit of *Forsythia suspensa* Vahl (*Oleaceae*).

Description Ovoid to long ovoid capsule, 1.5 – 2.5 cm in length, 0.5 – 1 cm in width, with acute apex, and sometimes with a peduncle at the base; externally light gray to dark brown, scattered with light gray and small ridged dots, and with two longitudinal furrows; a capsule dehiscing along the longitudinal furrows has the apexes bent backward; the inner surface of dehisced pericarp is yellow-brown in color, with a longitudinal partition-wall in the middle; seeds, slender and oblong, 0.5 – 0.7 cm in length, and usually with a wing.

Odor, slight; taste, slightly bitter.

Identification To 1.0 g of pulverized Forsythia Fruit add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: a red-purple to red-brown spot is observed at an *R*f value of about 0.3.

Purity **(1)** Branchlet—When perform the test of foreign matter ⟨*5.01*⟩, the amount of branchlets contained in Forsythia Fruit does not exceed 5.0%.

(2) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than branchlets contained in Forsythia Fruit does not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not

less than 10.0%.

Containers and storage Containers—Well-closed containers.

Fritillaria Bulb

Fritillariae Bulbus

バイモ

Fritillaria Bulb is the bulb of *Fritillaria verticillata* Willdenow var. *thunbergii* Baker (*Liliaceae*).

Description Fritillaria Bulb is a depressed spherical bulb, 2 – 3 cm in diameter, 1 – 2 cm in height, consisting of 2 thickened scaly leaves often separated; externally and internally white to light yellow-brown in color; inside base is in a slightly dark color; the bulb sprinkled with lime before drying is dusted with white powder; fractured surface, white in color and powdery.

Odor, slight and characteristic; taste, bitter.

Under a microscope <*5.01*>, a transverse section reveals the outermost layer to be composed of an epidermis; numerous vascular bundles scattered throughout the parenchyma inside of the epidermis; parenchyma filled with starch grains; starch grains are mainly simple (rarely 2- to 3-compound), 5 – 60 μm in diameter, narrowly ovate to ovate or triangular to obovate, stratiform figure obvious; epidermal cells and parenchyma cells near the vessels contain solitary crystals of calcium oxalate.

Identification Put 2 g of pulverized Fritillaria Bulb in a glass-stoppered centrifuge tube, add 10 mL of ammonia TS and 20 mL of a mixture of ethyl acetate and diethyl ether (1:1), shake for 20 minutes, and centrifuge. Take the upper layer, add 20 g of anhydrous sodium sulfate to the layer, shake, and filter. Evaporate the solvent of the filtrate, dissolve the residue in 1 mL of ethanol (99.5), and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (17:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate: spots of a yellow-red color appear at *R*f values of about 0.4 and about 0.6.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Fritillaria Bulb according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Fritillaria Bulb according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 16.0% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Gambir

Gambir

アセンヤク

Gambir is the dried aqueous extract prepared from the leaves and young twigs of *Uncaria gambir* Roxburgh (*Rubiaceae*).

Description Brown to dark brown, brittle mass; inside light brown.

Odor, slight; taste, extremely astringent and bitter.

Identification **(1)** To 0.2 g of pulverized Gambir add 10 mL of water, warm in a water bath for 5 minutes with occasional shaking, and filter. Cool the filtrate, and add 2 to 3 drops of gelatin TS: a white turbidity or precipitate is produced.

(2) Shake 0.1 g of pulverized Gambir with 20 mL of dilute ethanol for 2 minutes, and filter. Mix 1 mL of the filtrate with 9 mL of dilute ethanol, and to the solution add 1 mL of vanillin-hydrochloric acid TS: a light red to red-brown color develops.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 70.0%.

Containers and storage Containers—Well-closed containers.

Powdered Gambir

Gambir Pulveratum

アセンヤク末

Powdered Gambir is the powder of Gambir.

Description Powdered Gambir occurs as a red-brown to dark brown powder. It has a slight odor, and an extremely astringent and bitter taste.

Under a microscope <*5.01*>, Powdered Gambir, immersed in olive oil or liquid paraffin, consists of masses of needle crystals or yellow-brown to red-brown angular fragments, and reveals epidermal tissue and thick-walled hairs.

Identification **(1)** To 0.2 g of Powdered Gambir add 10 mL of water, warm in a water bath for 5 minutes with occasional shaking, and filter. Cool the filtrate, and add 2 to 3 drops of gelatin TS: a white turbidity or precipitate is produced.

(2) Shake 0.1 g of Powdered Gambir with 20 mL of dilute ethanol for 2 minutes, and filter. Mix 1 mL of the filtrate with 9 mL of dilute ethanol, and to the solution add 1 mL of vanillin-hydrochloric acid TS: a light red to red-brown color develops.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 70.0%.

Containers and storage Containers—Well-closed contain-

ers.

Gardenia Fruit

Gardeniae Fructus

サンシシ

Gardenia Fruit is the fruit of *Gardenia jasminoides* Ellis (*Rubiaceae*), sometimes after being passed through hot water or steamed.

It contains not less than 2.7% of geniposide, calculated on the basis of dried material.

Description Nearly long ovoid to ovoid fruit, 1 – 5 cm in length, 1 – 1.5 cm in width; externally yellow-brown to yellow-red, usually having 6, rarely 5 or 7, markedly raised ridges; calyx or its scar at one end, and sometimes peduncle at the other end; inner surface of pericarp yellow-brown, smooth and lustrous; internally divided into two loculi, containing a mass of seeds in yellow-red to dark red placenta; seed nearly circular, flat, about 0.5 cm in major axis, black-brown or yellow-red.

Odor, slight; taste, bitter.

Identification **(1)** To 1.0 g of pulverized Gardenia Fruit, previously dried in a desiccator (silica gel) for 24 hours, add 100 mL of hot water, warm the mixture between 60°C and 70°C for 30 minutes with frequent shaking, and filter after cooling. To 1.0 mL of the filtrate add water to make 10 mL: the color of the resulting solution is yellow and is not lighter than that of the following control solution.

Control solution: Dissolve 9.8 mg of carbazochrome sodium sulfonate trihydrate in water to make exactly 10 mL. Pipet 1 mL of this solution, and add water to make exactly 50 mL.

(2) To 1.0 g of pulverized Gardenia Fruit add 20 mL of methanol, warm for 3 minutes on a water bath, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and methanol (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying <*5.01*> Not more than 13.0%.

Total ash <*5.01*> Not more than 6.0%.

Assay Weigh accurately about 0.5 g of pulverized Gardenia Fruit, transfer into a glass-stoppered centrifuge tube, add 40 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and take the supernatant liquid. To the residue add 40 mL of diluted methanol (1 in 2), and repeat the same procedure as above. Combine the extracts so obtained, and add diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 20 mL, use this solution as the sample solution. Separately, weigh accurately about 10 mg of geniposide for assay, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of geniposide in each solution.

$$\text{Amount (mg) of geniposide} = M_S \times A_T/A_S \times 2$$

M_S: Amount (mg) of geniposide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 240 nm).

Column: A stainless steel column 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water and acetonitrile (22:3).

Flow rate: Adjust so that the retention time of geniposide is about 15 minutes.

System suitability—

System performance: Dissolve 1 mg each of geniposide for assay and caffeine in methanol to make 15 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, caffeine and geniposide are eluted in this order with the resolution between these peaks being not less than 3.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of geniposide is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Gardenia Fruit

Gardeniae Fructus Pulveratus

サンシシ末

Powdered Gardenia Fruit is the powder of Gardenia Fruit.

It contains not less than 2.7% of geniposide, calculated on the basis of dried material.

Description Powdered Gardenia Fruit occurs as a yellow-brown powder, and has a slight odor and a bitter taste.

Under a microscope <*5.01*>, Powdered Gardenia Fruit reveals fragments of yellow-brown epidermis consisting of polygonal epidermal cells in surface view; unicellular hairs, spiral and ring vessels, stone cells often containing crystals of calcium oxalate; fragments of thin-walled parenchyma containing yellow pigments, oil drops and rosette aggregates of calcium oxalate (the above elements from fruit receptacle and pericarp); fragments of large and thick-walled epidermis of seed coat, containing a red-brown substance; fragments of endosperm filled with aleuron grains (the above elements from seed).

Identification **(1)** To 1.0 g of Powdered Gardenia Fruit, previously dried in a desiccator (silica gel) for 24 hours, add 100 mL of hot water, warm the mixture between 60°C and 70°C for 30 minutes with frequent shaking, and filter after cooling. To 1.0 mL of the filtrate add water to make 10 mL: the color of the resulting solution is yellow and is not lighter

than that of the following control solution.

Control solution: Dissolve 9.8 mg of carbazochrome sodium sulfonate trihydrate in water to make exactly 10 mL. Pipet 1 mL of this solution, and add water to make exactly 50 mL.

(2) To 1.0 g of Powdered Gardenia Fruit add 20 mL of methanol, warm for 3 minutes on a water bath, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and methanol (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying <*5.01*> Not more than 13.0%.

Total ash <*5.01*> Not more than 6.0%.

Assay Weigh accurately about 0.5 g of Powdered Gardenia Fruit, transfer into a glass-stoppered centrifuge tube, add 40 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and take the supernatant liquid. To the residue add 40 mL of diluted methanol (1 in 2), and repeat the same procedure as above. Combine the extracts so obtained, and add diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 20 mL, use this solution as the sample solution. Separately, weigh accurately about 10 mg of geniposide for assay, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of geniposide in each solution.

$$\text{Amount (mg) of geniposide} = M_S \times A_T/A_S \times 2$$

M_S: Amount (mg) of geniposide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 240 nm).

Column: A stainless steel column 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water and acetonitrile (22:3).

Flow rate: Adjust so that the retention time of geniposide is about 15 minutes.

System suitability—

System performance: Dissolve 1 mg each of geniposide for assay and caffeine in methanol to make 15 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, caffeine and geniposide are eluted in this order with the resolution between these peaks being not less than 3.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of geniposide is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Gastrodia Tuber

Gastrodiae Tuber

テンマ

Gastrodia Tuber is the tuber of *Gastrodia elata* Blume (*Orchidaceae*), after being passed through hot water or steamed.

Description Gastrodia Tuber is an irregularly curved and flattened cylindrical to flattened fusiform tuber, 5 – 15 cm in length, 2 – 5 cm in diameter, 1 – 2 cm in thickness; externally light yellow-brown to light yellow-white; with ring nodes, and irregular longitudinal wrinkles; hard in texture; fractured surface, dark brown to yellow-brown in color, with luster, horny and gluey.

Odor, characteristic; practically tasteless.

Under a microscope <*5.01*>, a transverse section reveals parenchyma cells containing raphides of calcium oxalate; starch grain absent.

Identification To 1 g of pulverized Gastrodia Tuber add 5 mL of methanol, shake for 15 minutes, and filter. Evaporate the filtrate to dryness, dissolve the residue in 1 mL of methanol, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, methanol and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: a red-purple to light brown spot appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Gastrodia Tuber according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Gastrodia Tuber according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 16.0% (6 hours).

Total ash <*5.01*> Not more than 4.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 16.0%.

Containers and storage Containers—Well-closed containers.

Gentian

Gentianae Radix

ゲンチアナ

Gentian is the root and rhizome of *Gentiana lutea* Linné (*Gentianaceae*).

Description Nearly cylindrical pieces, 10 – 50 cm in length, 2 – 4 cm in diameter; externally dark brown; the rhizome short, with fine, transverse wrinkles, and sometimes with

buds and remains of leaves at the upper edge. The root longitudinally and deeply wrinkled, and more or less twisted; fractured surface yellow-brown and not fibrous, and a cambium and its neighborhood tinged dark brown.

Odor, characteristic; taste, sweet at first, later persistently bitter.

Under a microscope <*5.01*>, a transverse section of the root reveals several cellular layers of collenchyma adjoined internally to 4 to 6 cellular layers of thin-walled cork; secondary cortex with irregularly distributed phloem; xylem consisting chiefly of parenchyma, with individual or clustered vessels and tracheids, and exhibiting some sieve tubes of xylem; parenchyma of the xylem and the cortex containing oil droplets, minute needle crystals of calcium oxalate and very rarely starch grains 10 – 20 μm in diameter.

Identification **(1)** Place 0.1 g of pulverized Gentian, previously dried in a desiccator (silica gel) for 48 hours, on a slide glass, put a glass ring 10 mm in both inside diameter and in height on it, then cover with another slide, and heat gently and gradually: pale yellow crystals are sublimed on the upper slide. The crystals are insoluble in water and in ethanol (95), and soluble in potassium hydroxide TS.

(2) To 0.5 g of pulverized Gentian add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of gentiopicroside for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Gentian according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Gentian according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Powdered Gentian

Gentianae Radix Pulverata

ゲンチアナ末

Powdered Gentian is the powder of Gentian.

Description Powdered Gentian occurs as a yellow-brown powder, and has a characteristic odor. It has a sweet taste at first, which later becomes persistently bitter.

Under a microscope <*5.01*>, Powdered Gentian reveals parenchyma cells containing oil droplets and minute needle crystals, vessels, tracheids, cork tissues, and crystals of calcium oxalate. Vessels are chiefly reticulate vessels and scalariform vessels, 20 – 80 μm in diameter. Starch grains are observed very rarely, in simple grains about 10 – 20 μm in diameter.

Identification **(1)** Place 0.1 g of Powdered Gentian, previously dried in a desiccator (silica gel) for 48 hours, on a slide glass, put a glass ring 10 mm in both inside diameter and in height on it, then cover with another slide glass, and heat gently and gradually: light yellow crystals are sublimed on the upper glass. The crystals are insoluble in water and in ethanol (95), and soluble in potassium hydroxide TS.

(2) To 0.5 g of Powdered Gentian add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of gentiopicroside for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of Powdered Gentian according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Gentian according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, stone cell and fiber are not observed.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Containers and storage Containers—Tight containers.

Gentian and Sodium Bicarbonate Powder

ゲンチアナ·重曹散

Method of preparation

Powdered Gentian	300 g
Sodium Bicarbonate	700 g
To make	1000 g

Prepare as directed under Powders, with the above ingredients.

Description Gentian and Sodium Bicarbonate Powder occurs as a light yellow-brown powder, and has a bitter taste.

Identification **(1)** To 2 g of Gentian and Sodium Bicarbonate Powder add 10 mL of water, stir, and filter: the filtrate responds to the Qualitative Tests <*1.09*> (1) for bicarbonate.

(2) To 1.5 g of Gentian and Sodium Bicarbonate Powder add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of gentiopicroside for thin-layer chromatography in 1 mL of

methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Containers and storage Containers—Well-closed containers.

Geranium Herb

Geranii Herba

ゲンノショウコ

Geranium Herb is the terrestrial part of *Geranium thunbergii* Siebold et Zuccarini (*Geraniaceae*).

Description Stem with leaves opposite; stem, slender and long, green-brown; stem and leaf covered with soft hairs; leaf divided palmately into 3 to 5 lobes, and 2 – 4 cm in length, grayish yellow-green to grayish brown; each lobe oblong to obovate, and its upper margin crenate.

Odor, slight; taste, astringent.

Identification Boil 0.1 g of Geranium Herb with 10 mL of water, filter, and to the filtrate add 1 drop of iron (III) chloride TS: a blackish blue color develops.

Purity Foreign matter <5.01>—The amount of the root and other foreign matter contained in Geranium Herb does not exceed 2.0%.

Total ash <5.01> Not more than 10.0%.

Acid-insoluble ash <5.01> Not more than 1.5%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Powdered Geranium Herb

Geranii Herba Pulverata

ゲンノショウコ末

Powdered Geranium Herb is the powder of Geranium Herb.

Description Powdered Geranium Herb occurs as a grayish green to light yellow-brown powder. It has a slight odor and an astringent taste.

Under a microscope <5.01>, Powdered Geranium Herb reveals mainly fibers, spiral vessels, pitted vessels, and unicellular hairs; furthermore, multicellular glandular hairs, epidermis with stomata, fragments of palisade tissue, rosette aggregates of calcium oxalate, and starch grains. Fiber is thick-walled, with somewhat distinct pits; unicellular hair shows small point-like protrusions on the surface; palisade tissue consisting of circular parenchyma cells in surface view, each cell containing one rosette aggregate of calcium oxalate which is about 20 μm in diameter. Starch grains consisting of simple grains but rarely of 2-compound grains, ovoid to spherical, 5 – 30 μm in diameter, with distinct hilum.

Identification Boil 0.1 g of Powdered Geranium Herb with 10 mL of water, filter, and to the filtrate add 1 drop of iron (III) chloride TS: a dark blue color develops.

Purity Foreign matter—Under a microscope <5.01>, Powdered Geranium Herb reveals no stone cells.

Total ash <5.01> Not more than 10.0%.

Acid-insoluble ash <5.01> Not more than 1.5%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 15.0%.

Containers and storage Containers—Well-closed containers.

Ginger

Zingiberis Rhizoma

ショウキョウ

Ginger is the rhizome, with (unpeeled) or without (peeled) the periderm, of *Zingiber officinale* Roscoe (*Zingiberaceae*).

It contains not less than 0.3% of [6]-gingerol, calculated on the basis of dried material.

Description Irregularly compressed and often branched massive rhizome or a part of it; the branched parts are slightly curved ovoid or oblong-ovoid, 2 – 4 cm in length, and 1 – 2 cm in diameter; external surface grayish white to light grayish brown, and often with white powder; fractured surface is somewhat fibrous, powdery, light yellowish brown; under a magnifying glass, a transverse section reveals cortex and stele distinctly divided; vascular bundles and secretes scattered all over the surface as small dark brown dots.

Odor, characteristic; taste, extremely pungent.

Under a microscope <5.01>, a transverse section reveals cork layer, cortex, endodermis and stele in this order from the outside, cork layer often peeled off; cortex and stele, divided by an endodermis, composed of parenchyma; vascular bundles surrounded by fibers scattered in cortex and stele; oil cells contain yellow oily substances, scattered in parenchyma; parenchyma cells contain solitary crystals of calcium oxalate; starch grains in parenchyma cells mainly simple, ovoid, triangular ovoid, ellipsoidal or spherical, with abaxial hilum, usually 10 – 30 μm in long axis.

Identification To 2 g of pulverized Ginger add 5 mL of diethyl ether, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C

for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Ginger according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Ginger according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 8.0%.

Assay Weigh accurately about 1 g of pulverized Ginger (separately determine the loss on drying <*5.01*>, at 105°C for 5 hours), place in a centrifuge tube, add 30 mL of a mixture of methanol and water (3:1), shake for 20 minutes, centrifuge, and separate the supernatant liquid. To the residue add 30 mL of a mixture of methanol and water (3:1), and repeat the extraction twice more. To the combined all extracts add a mixture of methanol and water (3:1) to make exactly 100 mL, use this solution as the sample solution. Separately, weigh accurately about 5 mg of [6]-gingerol for assay, dissolve in a mixture of methanol and water (3:1) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-gingerol in each solution.

$$\text{Amount (mg) of [6]-gingerol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of [6]-gingerol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 205 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile and phosphoric acid (3800:2200:1).

Flow rate: Adjust so that the retention time of [6]-gingerol is about 19 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-gingerol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-gingerol is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Ginger

Zingiberis Rhizoma Pulveratum

ショウキョウ末

Powdered Ginger is the powder of Ginger.

It contains not less than 0.20% of [6]-gingerol, calculated on the basis of dried material.

Description Powdered Ginger occurs as a light grayish brown to light grayish yellow powder. It has a characteristic odor and an extremely pungent taste.

Under a microscope <*5.01*>, Powdered Ginger reveals mainly starch grains and parenchyma cells containing them; also, parenchyma cells containing yellow-brown to dark brown oily substances or single crystals of calcium oxalate; fragments of fibers with distinct pits; fragments of spiral, ring and reticulate vessels, and rarely fragments of cork tissue; starch grains composed of simple, compound or half-compound grains, ovoid, triangular ovoid, ellipsoidal or spherical, with abaxial hilum, usually 10 – 30 μm in long axis.

Identification To 2 g of Powdered Ginger add 5 mL of diethyl ether, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Ginger according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Ginger according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Ginger does not show stone cells, lignified parenchyma cells and other foreign matter.

Total ash <*5.01*> Not more than 8.0%.

Assay Weigh accurately about 1 g of Powdered Ginger (separately determine the loss on drying <*5.01*>, at 105°C for 5 hours), place in a centrifuge tube, add 30 mL of a mixture of methanol and water (3:1), shake for 20 minutes, centrifuge, and separate the supernatant liquid. To the residue add 30 mL of a mixture of methanol and water (3:1), and repeat the extraction twice more. To the combined all extracts add a mixture of methanol and water (3:1) to make exactly 100 mL, use this solution as the sample solution. Separately, weigh accurately about 5 mg of [6]-gingerol for assay, dissolve in a mixture of methanol and water (3:1) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and

determine the peak areas, A_T and A_S, of [6]-gingerol in each solution.

$$\text{Amount (mg) of [6]-gingerol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of [6]-gingerol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 205 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile and phosphoric acid (3800:2200:1).

Flow rate: Adjust so that the retention time of [6]-gingerol is about 19 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-gingerol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-gingerol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Ginseng

Ginseng Radix

ニンジン

Ginseng is the root of *Panax ginseng* C. A. Meyer (*Panax schinseng* Nees) (*Araliaceae*), from which rootlets have been removed, or the root that has been quickly passed through hot water.

It contains not less than 0.10% of ginsenoside Rg_1 ($C_{42}H_{72}O_{14}$: 801.01) and not less than 0.20% of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), calculated on the basis of dried material.

Description Thin and long cylindrical to fusiform root, often branching 2 to 5 lateral roots from the middle; 5 – 20 cm in length, main root 0.5 – 3 cm in diameter; externally light yellow-brown to light grayish brown, with longitudinal wrinkles and scars of rootlets; sometimes crown somewhat constricted and with short remains of rhizome; fractured surface practically flat, light yellow-brown in color, and brown in the neighborhood of the cambium.

Odor, characteristic; taste, at first slightly sweet, followed by a slight bitterness.

Identification (1) On a section of Ginseng add dilute iodine TS dropwise: a dark blue color is produced on the surface.

(2) To 2.0 g of pulverized Ginseng add 10 mL of water and 10 mL of 1-butanol, shake for 15 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of ginsenoside Rg_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (14:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity (1) Heavy metals ⟨*1.07*⟩—Proceed with 1.0 g of pulverized Ginseng according to Method 4, and perform the test. Prepare the control solution with 1.5 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 1.0 g of pulverized Ginseng according to Method 4, and perform the test (not more than 2 ppm).

(3) Foreign matter ⟨*5.01*⟩—The amount of stems and other foreign matter contained in Ginseng does not exceed 2.0%.

(4) Total BHC's and total DDT's ⟨*5.01*⟩—Not more than 0.2 ppm, respectively.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 4.2%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 14.0%.

Assay (1) Ginsenoside Rg_1—Weigh accurately about 1.0 g of pulverized Ginseng, put in a glass-stoppered centrifuge tube, add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. Repeat the procedure with the residue using 15 mL of diluted methanol (3 in 5), combine the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of dilute sodium hydroxide TS, allow to stand for 30 minutes, add 3 mL of 0.1 mol/L hydrochloric acid TS and diluted methanol (3 in 5) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rg_1 RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rg_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rg_1\ (C_{42}H_{72}O_{14}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Ginsenoside Rg_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of ginsenoside Rg_1 is about 25 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rg_1 RS and ginsenoside Re in diluted methanol (3 in 5) to

make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rg_1 and ginsenoside Re are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rg_1 is not more than 1.5%.

(2) Ginsenoside Rb_1—Use the sample solution obtained in (1) as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile (7:3).

Flow rate: Adjust so that the retention time of ginsenoside Rb_1 is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rb_1 RS and ginsenoside Rc in diluted methanol (3 in 5) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rb_1 and ginsenoside Rc are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Ginseng

Ginseng Radix Pulverata

ニンジン末

Powdered Ginseng is the powder of Ginseng.

It contains not less than 0.10% of ginsenoside Rg_1 ($C_{42}H_{72}O_{14}$: 801.01) and not less than 0.20% of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), calculated on the basis of dried material.

Description Powdered Ginseng occurs as a light yellow-white to light yellow-brown powder. It has characteristic odor and is a slight sweet taste followed by a slight bitterness.

Under a microscope <5.01>, Powdered Ginseng reveals round to rectangular parenchyma cells containing starch grains, occasionally gelatinized starch, vessels, secretory cell, sclerenchyma cell, big and thin-walled cork cell; crystals of calcium oxalate and starch. Vessels are reticulate vessel fragments, scalariform vessel and spiral vessel, 15 – 40 μm in diameter. Secretory cell containing a mass of yellow glistened contents; rosette aggregate of calcium oxalate, 20 – 60 μm in diameter, and 1 – 5 μm in diameter, rarely up to 30 μm in diameter of its single crystal; sclerenchymatous cells and thin-walled cork cells. Starch grains are observed in simple grain and 2 to 6-compound grain, simple grain, 3 – 20 μm in diameter.

Identification To 2.0 g of Powdered Ginseng add 10 mL of water and 10 mL of 1-butanol, shake for 15 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of ginsenoside Rg_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (14:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <1.07>—Proceed with 1.0 g of Powdered Ginseng according to Method 4, and perform the test. Prepare the control solution with 1.5 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 1.0 g of Powdered Ginseng according to Method 4, and perform the test (not more than 2 ppm).

(3) Total BHC's and total DDT's <5.01>—Not more than 0.2 ppm, respectively.

Loss on drying <5.01> Not more than 13.0% (6 hours).

Total ash <5.01> Not more than 4.2%.

Acid-insoluble ash <5.01> Not more than 0.5%.

Extract content <5.01> Dilute ethanol-soluble extract; not less than 14.0%.

Assay **(1)** Ginsenoside Rg_1—Weigh accurately about 1.0 g of Powdered Ginseng, put in a glass-stoppered centrifuge tube, add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. Repeat the procedure with the residue using 15 mL of diluted methanol (3 in 5), combine the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of dilute sodium hydroxide TS, allow to stand for 30 minutes, add 3 mL of 0.1 mol/L hydrochloric acid TS and diluted methanol (3 in 5) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rg_1 RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rg_1 in each solution.

Amount (mg) of ginsenoside Rg_1 ($C_{42}H_{72}O_{14}$)
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Ginsenoside Rg_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of ginsenoside Rg_1 is about 25 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rg_1 RS and ginsenoside Re in diluted methanol (3 in 5) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rg_1 and ginsenoside Re are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rg_1 is not more than 1.5%.

(2) Ginsenoside Rb_1—Use the sample solution obtained in (1) as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determined the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

Amount (mg) of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$)
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile (7:3).

Flow rate: Adjust so that the retention time of ginsenoside Rb_1 is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rb_1 RS and ginsenoside Rc in diluted methanol (3 in 5) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rb_1 and ginsenoside Rc are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

Containers and storage Containers—Tight containers.

Glehnia Root and Rhizome

Glehniae Radix cum Rhizoma

ハマボウフウ

Glehnia Root and Rhizome is the root and rhizome of *Glehnia littoralis* Fr. Schmidt ex Miquel (*Umbelliferae*).

Description Cylindrical to long conical root or rhizome, 10 – 20 cm in length, 0.5 – 1.5 cm in diameter; externally light yellow-brown to red-brown. Rhizome short, with fine ring nodes; roots having longitudinal wrinkles and numerous, dark red-brown, warty protrusions or transversely elongated protuberances. Brittle and easily breakable. A transverse section white and powdery, and under a magnifying glass, oil canals scattered as brown dots.

Odor, slight; taste, slightly sweet.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Glehnia Root and Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Glehnia Root and Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Glycyrrhiza

Glycyrrhizae Radix

カンゾウ

Glycyrrhiza is the root and stolon, with (unpeeled) or without (peeled) the periderm, of *Glycyrrhiza uralensis* Fisher or *Glycyrrhiza glabra* Linné (*Leguminosae*).

It contains not less than 2.0% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), calculated on the basis of dried material.

Description Nearly cylindrical pieces, 0.5 – 3 cm in diameter, over 1 m in length; externally dark brown to red-brown, longitudinally wrinkled, and often having lenticels, small buds and scaly leaves; peeled Glycyrrhiza is externally light yellow and fibrous. The transverse section reveals a rather clear border between cortex and xylem, and a radial structure which often has radiating splits. Glycyrrhiza originated from stolon has a pith, but those from root has no pith.

Odor, slight; taste, sweet.

Under a microscope <*5.01*>, a transverse section reveals multicellular layers of yellow-brown cork layers, and 1- to 3-cellular layer of cork cortex inside the cork layer; the secondary cortex exhibiting medullary rays and phloems radiated alternately; the phloem exhibiting fiber bundles with thick but incompletely lignified cell walls, surrounded by crystal cells; peeled Glycyrrhiza sometimes lacks a part of secondary cortex; the xylem exhibiting large yellow vessels and medullary rays in 3 to 10 rows radiated alternately; the vessels

accompanied with xylem fibers surrounded by crystal cells, and with xylem parenchyma cells; the parenchymatous pith only in Glycyrrhiza originated from stolon. The parenchyma cells contain starch grains and often solitary crystals of calcium oxalate. Under a microscope <*5.01*>, a vertical section reveals crystal cells row observed along with phloem fibers or xylem fibers.

Identification To 2 g of pulverized Glycyrrhiza add 10 mL of a mixture of ethanol (95) and water (7:3), heat by shaking on a water bath for 5 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 5 mg of Glycyrrhizic Acid RS or glycyrrhizic acid for thin-layer chromatography in 1 mL of a mixture of ethanol (95) and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Glycyrrhiza according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Glycyrrhiza according to Method 4, and perform the test (not more than 5 ppm).

(3) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 25.0%.

Assay Weigh accurately about 0.5 g of pulverized Glycyrrhiza in a glass-stoppered centrifuge tube, add 70 mL of dilute ethanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 25 mL of dilute ethanol, and proceed in the same manner. Combine all the extracts, add dilute ethanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 25 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in dilute ethanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: Adjust so that the retention time of glycyrrhizic acid is about 15 minutes.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak with the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Glycyrrhiza

Glycyrrhizae Radix Pulverata

カンゾウ末

Powdered Glycyrrhiza is the powder of Glycyrrhiza. It contains not less than 2.0% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), calculated on the basis of dried material.

Description Powdered Glycyrrhiza is light yellow-brown or light yellow to grayish yellow (powder of peeled Glycyrrhiza) in color. It has a slight odor and a sweet taste.

Under a microscope <*5.01*>, Powdered Glycyrrhiza reveals mainly yellow sclerenchymatous fiber bundles accompanied with crystal cell rows; vessels, 80 – 200 μm in diameter, with pitted, reticulate and scalariform pits, and with round perforations; parenchyma cells, containing starch grains and solitary crystals of calcium oxalate, their fragments, and cork tissues; but powder of peeled Glycyrrhiza shows no cork tissue; if any, a very few. Starch grains are simple grains, 2 – 20 μm in diameter; solitary crystals of calcium oxalate, 10 – 30 μm in a diameter.

Identification To 2 g of Powdered Glycyrrhiza add 10 mL of a mixture of ethanol (95) and water (7:3), heat by shaking on a water bath for 5 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 5 mg of Glycyrrhizic Acid RS or glycyrrhizic acid for thin-layer chromatography in 1 mL of a mixture of ethanol (95) and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Glycyrrhiza according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Glycyrrhiza according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Glycyrrhiza shows no stone cells.

(4) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 25.0%.

Assay Weigh accurately about 0.5 g of Powdered Glycyrrhiza in a glass-stoppered centrifuge tube, add 70 mL of dilute ethanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 25 mL of dilute ethanol, and proceed in the same manner. Combine all the extracts, add dilute ethanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 25 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in dilute ethanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: Adjust so that the retention time of glycyrrhizic acid is about 15 minutes.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak with the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Glycyrrhiza Extract

カンゾウエキス

Glycyrrhiza Extract contains not less than 3.6% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93).

Method of preparation **1)** To 1 kg of fine cuttings of Glycyrrhiza or the root and stolon of *Glycyrrhiza glabra* Linné (*Leguminosae*) which meets the requirement of Glycyrrhiza add 5 L of Water, Purified Water or Purified Water in Containers, and macerate for 2 days. Filter the macerated solution through a cloth filter. Add 3 L of Water, Purified Water or Purified Water in Containers to the residue, macerate again for 12 hours, and filter through a cloth filter. Evaporate the combined filtrates until the whole volume becomes 3 L. After cooling, add 1 L of Ethanol, and allow to stand in a cold place for 2 days. Filter, and evaporate the filtrate to a viscous extract.

2) Take Glycyrrhiza or the root and stolon of *Glycyrrhiza glabra* Linné (*Leguminosae*) which meets the requirement of Glycyrrhiza, pulverized to suitable sizes, and prepare the viscous extract as directed under Extracts using Water, Purified Water or Purified Water in Containers as the solvent. Immediately before making a millet jelly-like consistency for the viscous extract, add Ethanol, Anhydrous Ethanol or ethanol (99.5) to the extract, allow it to stand in a cold place, filter, and concentrate the filtrate to prepare.

Description Glycyrrhiza Extract is a brown to blackish brown, viscous extract, and has a characteristic odor and a sweet taste.

It dissolves in water, forming a clear solution, or with a slight turbidity.

Identification To 0.8 g of Glycyrrhiza Extract add 10 mL of a mixture of ethanol (95) and water (7:3), shake for 2 minutes, centrifuge, and use the supernatant liquid as the sample solution. Proceed as directed in the Identification under Glycyrrhiza.

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Glycyrrhiza Extract as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Insoluble matter—Dissolve 2.0 g of Glycyrrhiza Extract in 18 mL of water, and filter. To 10 mL of the filtrate add 5 mL of ethanol (95): a clear solution results.

Assay Weigh accurately about 0.15 g of Glycyrrhiza Extract, place in a glass-stoppered centrifuge tube, add 25 mL of dilute ethanol, and heat at 50°C for 30 minutes with occasional shaking. Cool, centrifuge, and take the supernatant liquid. To the residue add 20 mL of dilute ethanol, and proceed in the same manner. Combine all the extracts, add dilute ethanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 20 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in dilute ethanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: Adjust so that the retention time of glycyrrhizic acid is about 15 minutes.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak with the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Crude Glycyrrhiza Extract

カンゾウ粗エキス

Crude Glycyrrhiza Extract contains not less than 4.8% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93).

Method of preparation Take Glycyrrhiza or the root and stolon of *Glycyrrhiza glabra* Linné (*Leguminosae*) which meets the requirement of Glycyrrhiza, pulverized to suitable sizes, and prepare the dry extracts as directed under Extracts using Water, Purified Water or Purified Water in Containers as the solvent.

Description Crude Glycyrrhiza Extract occurs as lustrous, dark yellow-red to black-brown plates, rods or masses. It is comparatively brittle when cold, and the fractured surface is dark yellow-red, shell-like, and lustrous. It softens when warmed.

It has a characteristic odor and a sweet taste.

It dissolves in water with turbidity.

Identification To 0.6 g of Crude Glycyrrhiza Extract add 10 mL of a mixture of ethanol (95) and water (7:3), dissolve by warming if necessary, cool, centrifuge, and use the supernatant liquid as the sample solution. Proceed as directed in the Identification under Glycyrrhiza.

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Crude Glycyrrhiza Extract as directed in the Extracts (4) under General Rules for Preparations, and perform the test (not more than 30 ppm).

(2) Water-insoluble substances—Boil 5.0 g of pulverized Crude Glycyrrhiza Extract with 100 mL of water. After cooling, filter the mixture through tared filter paper, wash with water, and dry the residue at 105°C for 5 hours: the mass of the residue is not more than 1.25 g.

(3) Foreign matter—The filtrate obtained in (2) does not have a strong bitter taste.

(4) Starch—To about 1 g of pulverized Crude Glycyrrhiza Extract add water to make 20 mL, shake the mixture thoroughly, and filter. Examine the insoluble substance on the filter paper under a microscope: the residue contains no starch grains.

Total ash <*5.01*> Not more than 12.0% (1 g).

Assay Weigh accurately about 0.15 g of Crude Glycyrrhiza Extract, place in a glass-stoppered centrifuge tube, add 25 mL of dilute ethanol, and heat at 50°C for 30 minutes with occasional shaking. Cool, centrifuge, and take the supernatant liquid. To the residue add 20 mL of dilute ethanol, and proceed in the same manner. Combine all the extracts, add dilute ethanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 20 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in dilute ethanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: Adjust so that the retention time of glycyrrhizic acid is about 15 minutes.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak with the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Goreisan Extract

五苓散エキス

Goreisan Extract contains not less than 0.3 mg and not more than 1.2 mg (for preparation prescribed 1.5 g of Cinnamon Bark) or not less than 0.4 mg and not more than 1.6 mg (for preparation prescribed 2 g of Cinnamon Bark) or not less than 0.5 mg and not more than 2.0 mg (for preparation prescribed 2.5 g of Cinnamon Bark) or not less than 0.6 mg and not more than 2.4 mg (for preparation prescribed 3 g of Cinnamon Bark) of (*E*)-cinnamic acid, per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)	5)
Alisma Tuber	5 g	6 g	6 g	4 g	6 g
Polyporus Sclerotium	3 g	4.5 g	4.5 g	3 g	4.5 g
Poria Sclerotium	3 g	4.5 g	4.5 g	3 g	4.5 g
Atractylodes Rhizome	3 g	4.5 g	4.5 g	—	—
Atractylodes Lancea Rhizome	—	—	—	3 g	4.5 g
Cinnamon Bark	2 g	2.5 g	3 g	1.5 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 5), using the crude drugs shown above.

Description Goreisan Extract occurs as a light red-brown to light brown powder, or a black-brown viscous extract. It has a characteristic odor, and a slightly sweet first, bitter, then acrid taste.

Identification **(1)** Weigh exactly 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 20 mL of water and 2 mL of ammonia solution (28), and shake. Add 20 mL of a mixture of hexane and ethyl acetate (20:1), shake, centrifuge, and separate the upper layer. Add 20 mL of a mixture of hexane and ethyl acetate (20:1) to the aqueous layer, shake, centrifuge, and separate the upper layer. Combine these extracts, evaporate the solvent under low pressure (in vacuo), add exactly 2 mL of methanol to the residue, and use this solution as the sample solution. Separately, weigh exactly 10 mg of alisol A for thin-layer chromatography, and dissolve in exactly 10 mL of methanol. Pipet 1 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, water and formic acid (30:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution, and it is larger and more intense than the spot from the standard solution (Alisma Tuber).

(2) For preparation prescribed Atractylodes Rhizome—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red to red-purple spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 25 mL of hexane, and shake. Separate the hexane layer, and evaporate the solvent under low pressure (in vacuo), add 0.5 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a green-brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 50 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydradine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 5 mL of hexane, and shake. Centrifuge this solution, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance

of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay Conduct this procedure using light-resistant vessels. Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-cinnamic acid for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of (*E*)-cinnamic acid in each solution.

$$\text{Amount (mg) of (}E\text{)-cinnamic acid} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of (*E*)-cinnamic acid for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 273 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (750:250:1).

Flow rate: 1.0 mL per minute (the retention time of (*E*)-cinnamic acid is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of (*E*)-cinnamic acid are not less than 5000 and not more than1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of (*E*)-cinnamic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Goshajinkigan Extract

牛車腎気丸エキス

Goshajinkigan Extract contains not less than 4 mg and not more than 16 mg of loganin, not less than 6 mg and not more than 18 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 0.2 mg (for preparation prescribed Powdered Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride, or as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride) or not less than 0.1 mg (for preparation prescribed Powdered Processed Aconite Root 2) of total alkaloids (as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Rehmannia Root	5 g	5 g
Cornus Fruit	3 g	3 g
Dioscorea Rhizome	3 g	3 g
Alisma Tuber	3 g	3 g
Poria Sclerotium	3 g	3 g
Moutan Bark	3 g	3 g
Cinnamon Bark	1 g	1 g
Powdered Processed Aconite Root (Powdered Processed Aconite Root 1)	1 g	—
Powdered Processed Aconite Root (Powdered Processed Aconite Root 2)	—	1 g
Achyranthes Root	3 g	3 g
Plantago Seed	3 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Goshajinkigan Extract occurs as a brown to dark brown powder or black-brown viscous extract. It has slightly a characteristic odor and an acid taste.

Identification **(1)** To 1.0 g of the dry extract (or 3.0 g of the viscous extract), add 10 mL of water, shake, then add 30 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of water, methanol and 1-butanol (1:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool; a dark-green spot is observed at an *R*f value of about 0.6 (Rehmannia Root).

(2) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of loganin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer

chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (6:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybezaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Cornus Fruit).

(3) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of sodium carbonate TS, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of alisol A for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Alisma Tuber).

(4) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and diethyl ether (5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot from the standard solution (Moutan Bark).

(5) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate 1 mL of the hexane layer, add 0.5 mL of sodium hydroxide TS, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 50 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(6) To 3.0 g of the dry extract (or 9.0 g of the viscous extract), add 20 mL of diethyl ether and 2 mL of ammonia TS, shake for 10 minutes, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of acetonitrile to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of benzoylmesaconine hydrochloride for thin-layer chromatography in 10 mL of ethanol (99.5), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Powdered Processed Aconite Root).

(7) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 0.3 g of pulverized plantago seed for thin-layer chromatography, add 1 mL of methanol, warm on a water bath for 3 minutes, centrifuge after cooling, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate, water and acetic acid (100) (10:10:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the deep blue spot (*R*f value: about 0.3) from the standard solution (Plantago Seed).

(8) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 2 g of achyranthes root for thin-layer chromatography, add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate and water (4:4:3) to a distance of about 10 cm, and air-dry the plate. Spray evenly diluted sul-

furic acid on the plate and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value (around 0.4) with the dark red spot from the standard solution (Achyranthes Root).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

(3) Aconitum diester alkaloids (aconitine, jesaconitine, hypaconitine and mesaconitine)—Weigh accurately 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, and evaporate to dryness under low pressure (in vacuo). Dissolve the residue with exactly 10 mL of a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1). Centrifuge this solution, and use the supernatant liquid as the sample solution. Separately, pipet 1 mL of aconitum diester alkaloids standard solution for purity, add a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 40 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions: the heights of the peaks corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the sample solution are not higher than the respective heights corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for aconitine, hypaconitine and mesaconitine; 254 nm for jesaconitine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of mesaconitine is about 31 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of aconitum diester alkaloids standard solution for purity under the above operating conditions, using 254 nm, mesaconitine, hypaconitine, aconitine and jesaconitine are eluted in this order, and each resolution between their peaks is not less than 1.5 respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, using 231 nm, the relative standard deviation of the peak height of mesaconitine is not more than 1.5%.

Loss on drying <*2.41*> The dry extract: Not more than 9.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Loganin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of loganin for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of loganin in each solution.

$$\text{Amount (mg) of loganin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of loganin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 238 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and methanol (55:4:1).

Flow rate: 1.2 mL per minute (the retention time of loganin is about 25 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and symmetry factor of the peak of loganin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of loganin is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Total alkaloids—Weigh accurately about 1 g of the dry extract (or an amount of the viscous extract, equivalent to about 1 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, and evaporate to dryness under low pressure (in vacuo). Dissolve the residue with a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL. Centrifuge this solution, and use the supernatant liquid as the sample solution. Perform the test with exactly 20 μL each of the sample solution and the aconitum monoester alkaloids standard solution TS for assay as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine, A_{TM} and A_{SM}, A_{TH} and A_{SH}, as well as A_{TA} and A_{SA}, in each solution, respectively.

Amount (mg) of benzoylmesaconine hydrochloride
$= C_{SM} \times A_{TM}/A_{SM} \times 10$

Amount (mg) of benzoylhypaconine hydrochloride
$= C_{SH} \times A_{TH}/A_{SH} \times 10$

Amount (mg) of 14-anisoylaconine hydrochloride
$= C_{SA} \times A_{TA}/A_{SA} \times 10$

C_{SM}: Concentration (mg/mL) of benzoylmesaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SH}: Concentration (mg/mL) of benzoylhypaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SA}: Concentration (mg/mL) of 14-anisoylaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for benzoylmesaconine and benzoylhypaconine; 254 nm for 14-anisoylaconine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer solution for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of benzoylmesaconine is about 15 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of benzoylmesaconine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions, the relative standard deviation of the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Goshuyuto Extract

呉茱萸湯エキス

Goshuyuto Extract contains not less than 0.3 mg (for preparation prescribed 3 g of Euodia Fruit) or not less than 0.4 mg (for preparation prescribed 4 g of Euodia Fruit) of evodiamine, not less than 0.5 mg and not more than 2.0 mg (for preparation prescribed 1 g of Ginger) or not less than 0.7 mg and not more than 2.8 mg (for preparation prescribed 1.5 g of Ginger) of [6]-gingerol, and not less than 1.2 mg (for preparation prescribed 2 g of Ginseng) or not less than 1.8 mg (for preparation prescribed 3 g of Ginseng) of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Euodia Fruit	3 g	4 g	3 g
Ginger	1 g	1.5 g	1.5 g
Ginseng	2 g	3 g	2 g
Jujube	4 g	3 g	4 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 3), using the crude drugs shown above.

Description Goshuyuto Extract occurs as a light brown to light red-yellow powder, or a black-brown viscous extract. It has a slight odor and a hot and bitter taste.

Identification (1) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, to 1 g of pulverized euodia fruit add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under

Thin-layer Chromatography ⟨*2.03*⟩. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, 2-propanol, water and formic acid (7:7:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot (*R*f value: about 0.5) from the standard solution (Euodia Fruit).

(2) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(3) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

Purity **(1)** Heavy metals ⟨*1.07*⟩—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying ⟨*2.41*⟩ The dry extract: Not more than 11.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Evodiamine—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 30 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of evodiamine for assay, and dissolve in methanol to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of evodiamine in each solution.

$$\text{Amount (mg) of evodiamine} = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of evodiamine for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 282 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (620:380:1).

Flow rate: 1.0 mL per minute (the retention time of evodiamine is about 18 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of evodiamine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of evodiamine is not more than 1.5%.

(2) [6]-Gingerol—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 30 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of [6]-gingerol for assay, dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-gingerol in each solution.

$$\text{Amount (mg) of [6]-gingerol} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of [6]-gingerol for assay taken

Operating conditions—

Detector, column, column temperature and mobile phase: Proceed as directed in the operating conditions in (1).

Flow rate: 1.0 mL per minute (the retention time of [6]-gingerol is about 14 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-gingerol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operat-

ing conditions, the relative standard deviation of the peak area of [6]-gingerol is not more than 1.5%.

(3) Ginsenoside Rb_1—Weigh accurately about 2 g of the dry extract (or an amount of the viscous extract, equivalent to about 2 g of the dried substance), add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 15 mL of diluted methanol (3 in 5), and repeat the same procedure. Combine all the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, then add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 5 mL of this solution to a column [about 10 mm in inside diameter, packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), and washed just before using with methanol and then diluted methanol (3 in 10)], and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S \times 1/5$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl group bound silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (400:100:1).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

Containers and storage Containers—Tight containers.

Gypsum

Gypsum Fibrosum

セッコウ

Gypsum is natural hydrous calcium sulfate. It possibly corresponds to the formula $CaSO_4.2H_2O$.

Description Gypsum occurs as lustrous, white, heavy, fibrous, crystalline masses, which easily split into needles or very fine crystalline powder.

It is odorless and tasteless.

It is slightly soluble in water.

Identification To 1 g of pulverized Gypsum add 20 mL of water, allow to stand with occasional shaking for 30 minutes, and filter: the filtrate responds to the Qualitative Tests <*1.09*> (2) and (3) for calcium salt and to the Qualitative Tests <*1.09*> for sulfate.

Purity **(1)** Heavy metals <*1.07*>—Boil 4.0 g of pulverized Gypsum with 4 mL of acetic acid (100) and 96 mL of water for 10 minutes, cool, add water to make exactly 100 mL, and filter. Perform the test using 50 mL of the filtrate as the test solution. Prepare the control solution as follows: to 4.0 mL of Standard Lead Solution add 2 mL of dilute acetic acid and water to make 50 mL (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Gypsum according to Method 2, and perform the test (not more than 5 ppm).

Containers and storage Containers—Well-closed containers.

Exsiccated Gypsum

Gypsum Exsiccatum

焼セッコウ

Exsiccated Gypsum possibly corresponds to the formula $CaSO_4.\frac{1}{2}H_2O$.

Description Exsiccated Gypsum occurs as a white to grayish white powder. It is odorless and tasteless.

It is slightly soluble in water, and practically insoluble in ethanol (95).

It absorbs moisture slowly on standing in air to lose its solidifying property.

When it is heated to yield an anhydrous compound at a temperature above 200°C, it loses its solidifying property.

Identification Shake 1 g of Exsiccated Gypsum with 20 mL of water for 5 minutes, and filter: the filtrate responds to the Qualitative Tests <*1.09*> (2) and (3) for calcium salt and to the Qualitative Tests <*1.09*> for sulfate.

Purity Alkalinity—Take 3.0 g of Exsiccated Gypsum in a glass-stoppered test tube, add 10 mL of water and 1 drop of phenolphthalein TS, and shake vigorously: no red color develops.

Solidification To 10.0 g of Exsiccated Gypsum add 10 mL of water, stir immediately for 3 minutes, and allow to stand: the period until water no longer separates, when the material is pressed with a finger, is not more than 10 minutes from the time when the water was added.

Containers and storage Containers—Tight containers.

Hachimijiogan Extract

八味地黄丸エキス

Hachimijiogan Extract contains not less than 4 mg and not more than 16 mg of loganin, not less than 6 mg and not more than 18 mg (for preparation prescribed 3 g of Moutan Bark) or not less than 5 mg and not more than 15 mg (for preparation prescribed 2.5 g of Moutan Bark) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 0.7 mg (for preparation prescribed 1 g of Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride), or not less than 0.2 mg (for preparation prescribed 1 g of Powdered Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride, or as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride), or not less than 0.1 mg (for preparation prescribed 1 g of Powdered Processed Aconite Root 2) of total alkaloids (as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride), or not less than 0.1 mg (for preparation prescribed 0.5 g of Powdered Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride, or as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Rehmannia Root	5 g	5 g	5 g	6 g
Cornus Fruit	3 g	3 g	3 g	3 g
Dioscorea Rhizome	3 g	3 g	3 g	3 g
Alisma Tuber	3 g	3 g	3 g	3 g
Poria Sclerotium	3 g	3 g	3 g	3 g
Moutan Bark	3 g	3 g	3 g	2.5 g
Cinnamon Bark	1 g	1 g	1 g	1 g
Processed Aconite Root (Processed Aconite Root 1)	1 g	—	—	—
Powdered Processed Aconite Root (Powdered Processed Aconite Root 1)	—	1 g	—	0.5 g
Powdered Processed Aconite Root (Powdered Processed Aconite Root 2)	—	—	1 g	—

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Hachimijiogan Extract occurs as a grayish brown to brown powder or black-brown viscous extract. It has a characteristic odor and a slightly bitter and acid taste.

Identification **(1)** To 1.0 g of the dry extract (or 3.0 g of the viscous extract), add 10 mL of water, shake, then add 30 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of water, methanol and 1-butanol (1:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool; a dark green spot is observed at an *R*f value of about 0.6 (Rehmannia Root).

(2) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of loganin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (6:1:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybezaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Cornus Fruit).

(3) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of sodium carbonate TS, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of alisol A for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Alisma Tuber).

(4) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and diethyl ether (5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot from the standard solution (Moutan Bark).

(5) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour,

separate 1 mL of the hexane layer, add 0.5 mL of sodium hydroxide TS, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 50 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(6) To 3.0 g of the dry extract (or 9.0 g of the viscous extract), add 20 mL of diethyl ether and 2 mL of ammonia TS, shake for 10 minutes and centrifuge. Separate the diethyl ether layer, evaporate the diethyl ether layer under low pressure (in vacuo), add 1 mL of acetonitrile to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of benzoylmesaconine hydrochloride for thin-layer chromatography in 10 mL of ethanol (99.5), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Processed Aconite Root or Powdered Processed Aconite Root).

Purity (1) Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

(3) Aconitum diester alkaloids (aconitine, jesaconitine, hypaconitine and mesaconitine)—Weigh accurately 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue with exactly 10 mL of a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1), centrifuge this solution, and use the supernatant liquid as the sample solution. Separately, pipet exactly 1 mL of aconitum diester alkaloids standard solution for purity, add a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 40 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions: the heights of the peaks corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the sample solution are not higher than the respective heights corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for aconitine, hypaconitine and mesaconitine; 254 nm for jesaconitine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of mesaconitine is about 31 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of aconitum diester alkaloids standard solution for purity under the above operating conditions, using 254 nm, mesaconitine, hypaconitine, aconitine and jesaconitine are eluted in this order, and each resolution between their peaks is not less than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, using 231 nm, the relative standard deviation of the peak height of mesaconitine is not more than 1.5 %.

Loss on drying <*2.41*> The dry extract: Not more than 8.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay (1) Loganin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of loganin for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> ac-

cording to the following conditions, and determine the peak areas, A_T and A_S, of loganin in each solution.

$$\text{Amount (mg) of loganin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of loganin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 238 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and methanol (55:4:1).

Flow rate: 1.2 mL per minute (the retention time of loganin is about 25 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and symmetry factor of the peak of loganin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of loganin is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Total alkaloids—Weigh accurately about 1 g of the dry extract (or an amount of the viscous extract, equivalent to about 1 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue with a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL. Centrifuge this solution, and use the supernatant liquid as the sample solution. Perform the test with exactly 20 μL each of the sample solution and the aconitum monoester alkaloids standard solution TS for assay as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine, A_{TM} and A_{SM}, A_{TH} and A_{SH}, as well as A_{TA} and A_{SA}, in each solution, respectively.

$$\text{Amount (mg) of benzoylmesaconine hydrochloride} = C_{SM} \times A_{TM}/A_{SM} \times 10$$

$$\text{Amount (mg) of benzoylhypaconine hydrochloride} = C_{SH} \times A_{TH}/A_{SH} \times 10$$

$$\text{Amount (mg) of 14-anisoylaconine hydrochloride} = C_{SA} \times A_{TA}/A_{SA} \times 10$$

C_{SM}: Concentration (mg/mL) of benzoylmesaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SH}: Concentration (mg/mL) of benzoylhypaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SA}: Concentration (mg/mL) of 14-anisoylaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for benzoylmesaconine and benzoylhypaconine; 254 nm for 14-anisoylaconine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer solution for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of benzoylmesaconine is about 15 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of benzoylmesaconine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions,

the relative standard deviation of the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Hangekobokuto Extract

半夏厚朴湯エキス

Hangekobokuto Extract contains not less than 2 mg and not more than 6 mg of magnolol, not less than 4 mg (for preparation prescribed 2 g of Perilla Herb) or not less than 6 mg (for preparation prescribed 3 g of Perilla Herb) of rosmarinic acid, and not less than 0.6 mg and not more than 2.4 mg (for preparation prescribed 1 g of Ginger) or not less than 0.8 mg and not more than 3.2 mg (for preparation prescribed 1.3 g of Ginger) or not less than 0.9 mg and not more than 3.6 mg (for preparation prescribed 1.5 g of Ginger) of [6]-gingerol, per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Pinellia Tuber	6 g	6 g	6 g	6 g
Poria Sclerotium	5 g	5 g	5 g	5 g
Magnolia bark	3 g	3 g	3 g	3 g
Perilla Herb	2 g	3 g	2 g	2 g
Ginger	1 g	1 g	1.3 g	1.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Hangekobokuto Extract is a light brown to dark brown powder or black-brown viscous extract. It has a characteristic odor and has a bitter and astringent taste first then pungent later.

Identification (1) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of magnolol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Magnolia Bark).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of 0.1 mol/L hydrochloric acid TS, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of rosmarinic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (60:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Perilla Herb).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

Purity (1) Heavy metals ⟨*1.07*⟩—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying ⟨*2.41*⟩ The dry extract: Not more than 11.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 14.0%, calculated on the dried basis.

Assay (1) Magnolol—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of magnolol for assay, and dissolve in diluted methanol (7 in 10) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of magnolol in each solution.

$$\text{Amount (mg) of magnolol} = M_S \times A_T/A_S \times 1/8$$

M_S: Amount (mg) of magnolol for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wave-

length: 289 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (50:50:1).

Flow rate: 1.0 mL per minute (the retention time of magnolol is about 15 minutes).

System suitability—

System performance: Dissolve 1 mg each of magnolol for assay and honokiol in diluted methanol (7 in 10) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, honokiol and magnolol are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of magnolol is not more than 1.5%.

(2) Rosmarinic acid—Conduct this procedure using light-resistant vessels. Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of rosmarinic acid for assay, dissolve in diluted methanol (7 in 10) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of rosmarinic acid in each solution.

$$\text{Amount (mg) of rosmarinic acid} = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of rosmarinic acid for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 330 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (800:200:1).

Flow rate: 1.0 mL per minute (the retention time of rosmarinic acid is about 11 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of rosmarinic acid are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of rosmarinic acid is not more than 1.5%.

(3) [6]-Gingerol—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of [6]-gingerol for assay, dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-gingerol in each solution.

$$\text{Amount (mg) of [6]-gingerol} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of [6]-gingerol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 282 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (620:380:1).

Flow rate: 1.0 mL per minute (the retention time of [6]-gingerol is about 15 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-gingerol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-gingerol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Hangeshashinto Extract

半夏瀉心湯エキス

Hangeshashinto Extract contains not less than 70 mg and not more than 210 mg (for preparation prescribed 2.5 g of Scutellaria Root) or not less than 80 mg and not more than 240 mg (for preparation prescribed 3 g of Scutellaria Root) of baicalin ($C_{21}H_{18}O_{11}$: 446.36), not less than 18 mg and not more than 54 mg (for preparation prescribed 2.5 g of Glycyrrhiza) or not less than 20 mg and not more than 60 mg (for preparation prescribed 3 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), and not less than 7 mg and not more than 21 mg of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Pinellia Tuber	5 g	6 g	5 g
Scutellaria Root	2.5 g	3 g	2.5 g
Processed Ginger	2.5 g	3 g	—
Ginger	—	—	2.5 g
Ginseng	2.5 g	3 g	2.5 g
Glycyrrhiza	2.5 g	3 g	2.5 g
Jujube	2.5 g	3 g	2.5 g
Coptis Rhizome	1 g	1 g	1 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1), 2) or 3), using the crude drugs shown above.

Description Hangeshashinto Extract is a light yellow to yellow-brown powder or black-brown viscous extract. It has a slightly odor and a hotter, bitter and slightly sweet taste.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Take the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(2) For preparation prescribed Processed Ginger—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of cyclohexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Processed Ginger).

(3) For preparation prescribed Ginger—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(4) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rg_1 RS or ginsenoside Rg_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Ginseng).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 10 mL of methanol, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of coptisine chloride for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ammonia solution (28) and methanol (15:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Coptis Rhizome).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract—Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract—Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Baicalin—Weigh accurately about 0.1 g of the

dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(2) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of baicalein for resolution check in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and baicalein is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

(3) Berberine—Weigh accurately about 0.2 g of the dry

extract (or an amount of the viscous extract, equivalent to about 0.2 g of the dried substance), add exactly 50 mL of the mobile phase, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (separately determine the water <2.48> in the same manner as Berberine Chloride Hydrate), dissolve in the mobile phase to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine chloride ($C_{20}H_{18}ClNO_4$) $= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogen phosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: 1.0 mL per minute (the retention time of berberine is about 8 minutes).

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in the mobile phase to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Hedysarum Root

Hedysari Radix

シンギ

Hedysarum Root is the root of *Hedysarum polybotrys* Handel-Mazzetti (*Leguminosae*).

Description Hedysarum Root is nearly cylindrical, 20 – 100 cm in length, 0.5 – 2.5 cm in diameter; outer surface yellow-brown to red-brown, with irregular longitudinal wrinkles; often horizontal lenticels and scars of lateral roots; periderm peeled easily, internally light yellow-brown to light red-brown; soft in texture, flexible and difficult to break; fractured surface fibrous, powdery; in transverse section nearly white in cortex, brownish around cambium, light yellow-brown in xylem; ray obvious.

Odor, slightly characteristic; taste, slightly sweet.

Under a microscope <5.01>, a transverse section reveals cork layer 6 – 8 cells layered, 2 – 4 cells layered parenchyma cells with sparingly thick wall inside the cork layer; ray obvious in secondary cortex and often appearing cracked tissue in outer portion of secondary cortex; phloem fiber bungles arranged stepwise in phloem; ray obvious in xylem, reticulate, scalariform, pitted, and spiral vessels; xylem tissues around vessels; thin walled cells containing solitary crystals of calcium oxalate in peripheral region of phloem fibers and xylem fibers and appearing as crystal cell rows in a longitudinal section; solitary crystals of calcium oxalate 7 – 20 μm in diameter, starch grains simple or 2- to 8-compound grains in parenchyma.

Identification To 1.0 g of pulverized Hedysarum Root add 10 mL of methanol, shake for 10 minutes, and filter. Evaporate the solvent of the filtrate under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, 2-butanone and formic acid (60:40:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a blue-white fluorescent spot at an *R*f value of about 0.4 is observed.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of pulverized Hedysarum Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.40 g of pulverized Hedysarum Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <5.01> Not more than 16.0% (6 hours).

Total ash <5.01> Not more than 5.5%.

Acid-insoluble ash <5.01> Not more than 1.0%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 25.0%.

Containers and storage Containers—Well-closed containers.

Hemp Fruit

Cannabis Fructus

マシニン

Hemp Fruit is the fruit of *Cannabis sativa* Linné (*Moraceae*).

Description Hemp Fruit is a slightly compressed void fruit, 4 – 5 mm in length, 3 – 4 mm in diameter; externally grayish green to grayish brown; pointed at one end, a scar of gynophore at the other end, and ridge on both sides; outer surface lustrous with white mesh-like pattern; slightly hard pericarp; seed, slightly green in color and internally has grayish white albumen; 100 fruits weigh 1.6 – 2.7 g.

Practically odorless, aromatic on chewing; taste, mild and oily.

Under a microscope <5.01>, a transverse section reveals the exocarp composed of an epidermis; mesocarp composed of parenchyma, a pigment cell layer and rows of short, small cells; endocarp made up of a single cellular layer of radially elongated stone cells; seed coat comprises a tubular cellular layer and spongy tissue. Inside of the seed; exosperm con-

sists of one cellular layer of parenchymatous cells, endosperm of one to several cellular layers of parenchymatous cells; most of the embryo composed of parenchyma, vascular bundles occurring in the center of hypocotyls and cotyledons; embryo parenchyma contains aleurone grains and oil drops.

Identification To 0.3 g of pulverized Hemp Fruit add 3 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and ethyl acetate (9:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a dark blue-purple spot appears at an *R*f value of about 0.6.

Purity Bract—When perform the test of foreign matter <*5.01*>, Hemp Fruit does not contain bract.

Loss on drying <*5.01*> Not more than 9.0% (6 hours).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Containers and storage Containers—Well-closed containers.

Hochuekkito Extract

補中益気湯エキス

Hochuekkito Extract contains not less than 16 mg and not more than 64 mg of hesperidin, not less than 0.3 mg and not more than 1.2 mg (for preparation prescribed 1 g of Bupleurum Root) or not less than 0.6 mg and not more than 2.4 mg (for preparation prescribed 2 g of Bupleurum Root) of saikosaponin b_2, and not less than 10 mg and not more than 30 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)	5)	6)
Ginseng	4 g	4 g	4 g	4 g	4 g	4 g
Atractylodes Rhizome	4 g	—	4 g	—	4 g	4 g
Atractylodes Lancea Rhizom	—	4 g	—	4 g	—	—
Astragalus Root	4 g	4 g	4 g	4 g	3 g	4 g
Japanese Angelica Root	3 g	3 g	3 g	3 g	3 g	3 g
Citrus Unshiu Peel	2 g	2 g	2 g	2 g	2 g	2 g
Jujube	2 g	2 g	2 g	2 g	2 g	2 g
Bupleurum Root	2 g	2 g	1 g	1 g	2 g	1 g
Glycyrrhiza	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g
Ginger	0.5 g	0.5 g	0.5 g	0.5 g	0.5 g	—
Processed Ginger	—	—	—	—	—	0.5 g
Cimicifuga Rhizome	1 g	1 g	0.5 g	0.5 g	1 g	0.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 6), using the crude drugs shown above.

Description Hochuekkito Extract occurs as a light brown to brown powder or black-brown viscous extract. It has a slight odor, and a sweet and bitter taste.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, and shake. Centrifuge this solution, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(2) For preparation prescribed Atractylodes Rhizome—To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, shake, and separate the hexane layer. Evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated the plate at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 40 mL of a solution of potassium hydroxide in methanol (1 in 50), shake for 15 minutes, centrifuge, and evaporate the solvent under low pressure (in vacuo). Add 30 mL of water and 20 mL of diethyl ether to the residue, shake, remove the diethyl ether layer, and separate the aqueous layer. To the aqueous layer add 20 mL of 1-butanol, shake, and separate the 1-butanol layer. To the 1-butanol

layer add 20 mL of water, shake, separate the 1-butanol layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of astragaloside IV for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of octadecylsilanized silica gel for thin-layer chromatography. Develop the plate with a mixture of methanol, water, 1-butanol and acetic acid (100) (60:30:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-brown spot from the standard solution (Astragalus Root).

(5) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root).

(6) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of 1-butanol, shake, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), add 3 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of hesperidin for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL of the sample solution and 20 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone, water and acetic acid (100) (10:6:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and expose to ammonia vapor: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (Citrus Unshiu Peel).

(7) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, and shake. Centrifuge this solution, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(8) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of 1-butanol, shake, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), add 3 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(9) For preparation prescribed Ginger—To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(10) For preparation prescribed Processed Ginger—Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 60 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of cyclohexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Processed Ginger).

(11) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 30 mL of water, shake, then add 50 mL of 1-butanol, shake, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), add 3 mL of methanol to the residue, and use this solution as the sample solution. Use (*E*)-isoferulic acid-(*E*)-ferulic acid TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone and water (20:12:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the light yellow-white fluorescent spot from the standard solution (Cimicifuga Rhizome).

Purity **(1)** Heavy metals ⟨*1.07*⟩—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed in the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying ⟨*2.41*⟩ The dry extract: Not more than 11.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Hesperidin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted tetrahydrofuran (1 in 4), shake for 30 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of hesperidin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add diluted tetrahydrofuran (1 in 4) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of hesperidin in each solution.

$$\text{Amount (mg) of hesperidin} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of hesperidin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (82:18:1).

Flow rate: 1.0 mL per minute (the retention time of hesperidin is about 15 minutes).

System suitability—

System performance: Dissolve 1 mg each of hesperidin for assay and naringin for thin-layer chromatography in diluted methanol (1 in 2) to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, naringin and hesperidin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of hesperidin is not more than 1.5%.

(2) Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the

standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Honey

Mel

ハチミツ

Honey is the saccharine substances obtained from the honeycomb of *Apis mellifera* Linné or *Apis cerana* Fabricius (*Apidae*).

Description Honey is a light yellow to light yellow-brown, syrupy liquid. Usually it is transparent, but often opaque with separated crystals.

It has a characteristic odor and a sweet taste.

Specific gravity <2.56> Mix 50.0 g of Honey with 100 mL of water: the specific gravity of the solution is not less than d_{20}^{20}: 1.111.

Purity **(1)** Acidity—Mix 10 g of Honey with 50 mL of water, and titrate <2.50> with 1 mol/L potassium hydroxide VS (indicator: 2 drops of phenolphthalein TS): not more than 0.5 mL is required.

(2) Sulfate—Mix 1.0 g of Honey with 2.0 mL of water, and filter. To the filtrate add 2 drops of barium chloride TS: the solution does not show any change immediately.

(3) Ammonia-coloring substances—Mix 1.0 g of Honey with 2.0 mL of water, and filter. To the filtrate add 2 mL of ammonia TS: the solution does not show any change immediately.

(4) Resorcinol-coloring substances—Mix well 5 g of Honey with 15 mL of diethyl ether, filter, and evaporate the diethyl ether solution at ordinary temperature. To the residue add 1 to 2 drops of resorcinol TS: a yellow-red color may develop in the solution of resorcinol and in the residue, and a red to red-purple color which does not persist more than 1 hour.

(5) Starch or dextrin—(i) Shake 7.5 g of Honey with 15 mL of water, warm the mixture on a water bath, and add 0.5 mL of tannic acid TS. After cooling, filter, and to 1.0 mL of the filtrate add 1.0 mL of ethanol (99.5) containing 2 drops of hydrochloric acid: no turbidity is produced.

(ii) To 2.0 g of Honey add 10 mL of water, warm in a water bath, mix, and allow to cool. Shake 1.0 mL of the mixture with 1 drop of iodine TS: no blue, green or red-brown color develops.

(6) Foreign matter—Mix 1.0 g of Honey with 2.0 mL of water, centrifuge the mixture, and examine the precipitate microscopically <5.01>: no foreign substance except pollen grains is observable.

Total ash <5.01> Not more than 0.4%.

Containers and storage Containers—Tight containers.

Houttuynia Herb

Houttuyniae Herba

ジュウヤク

Houttuynia Herb is the terrestrial part of *Houttuynia cordata* Thunberg (*Saururaceae*), collected during the flowering season.

Description Stem with alternate leaves and spikes; stem light brown, with longitudinal furrows and protruded nodes; when soaked in water and smoothed out, leaves wide ovate and cordate, 3 – 8 cm in length, 3 – 6 cm in width; light green-brown; margin entire, apex acuminate; petiole long, and membranous stipule at the base; spike, 1 – 3 cm in length, with numerous light yellow-brown achlamydeous florets, and the base enclosed by 4 long ovate, light yellow to light yellow-brown involucres.

Odor, slight; taste, slight.

Identification Heat 2 g of pulverized Houttuynia Herb with 20 mL of ethyl acetate under a reflux condenser for 15 minutes, and filter. Evaporate the filtrate to dryness, add 10 mL of water to the residue, warm the mixture on a water bath for 2 minutes, and, after cooling, filter. Shake well the filtrate with 20 mL of ethyl acetate in a separator, take 15 mL of ethyl acetate solution, and evaporate the solution on a water bath to dryness. Dissolve the residue in 5 mL of methanol, add 0.1 g of magnesium ribbon and 1 mL of hydrochloric acid, and allow the mixture to stand: a light red to red color develops.

Purity Foreign matter <5.01>—The amount of the rhizome, roots and other foreign matter contained in Houttuynia Herb is not more than 2.0%.

Total ash <5.01> Not more than 14.0%.

Acid-insoluble ash <5.01> Not more than 3.0%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 10.0%.

Containers and storage Containers—Well-closed containers.

Immature Orange

Aurantii Fructus Immaturus

キジツ

Immature Orange is the immature fruit or the fruit cut crosswise of *Citrus aurantium* Linné var. *daidai* Makino, *Citrus aurantium* Linné or *Citrus natsudaidai* Hayata (*Rutaceae*).

Description Nearly spherical fruit, 1 – 2 cm in diameter, or semispherical, 1.5 – 4.5 cm in diameter; external surface, deep green-brown to brown, and without luster, with numerous small dents associated with oil sacs; the outer portion of transverse section exhibits pericarp and mesocarp about 0.4 cm in thickness, yellow-brown in color in the region contacting epidermis, and light grayish brown color in the other parts; the central portion is radially divided into 8 to 16 small loculi; each loculus is brown and indented, often containing immature seeds.

Odor, characteristic; taste, bitter.

Identification To 0.5 g of pulverized Immature Orange add 10 mL of methanol, boil gently for 2 minutes, and filter. To 5 mL of the filtrate add 0.1 g of magnesium ribbon and 1 mL of hydrochloric acid, and allow to stand: a red-purple color develops.

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Containers and storage Containers—Well-closed containers.

Imperata Rhizome

Imperatae Rhizoma

ボウコン

Imperata Rhizome is the rhizome of *Imperata cylindrica* Beauvois (*Gramineae*), from which rootlets and scale leaves have been removed.

Description Long and thin cylindrical rhizome, 0.3 – 0.5 cm in diameter; sometimes branched; externally yellow-white, with slight longitudinal wrinkles, and with nodes at 2 – 3 cm intervals; difficult to break; fractured surface fibrous. Cross section irregularly round; thickness of cortex is slightly smaller than the diameter of the stele; pith often forms a hollow. Under a magnifying glass, a transverse section reveals cortex, yellow-white, and with scattered brown spots; stele, yellow-brown in color.

Almost odorless, and tasteless at first, but later slightly sweet.

Identification To 1 g of pulverized Imperata Rhizome add 20 mL of hexane, allow the mixture to stand for 30 minutes with occasional shaking, and filter. Evaporate the solvent of the filtrate under low pressure (in vacuo), dissolve the residue in 5 mL of acetic anhydride, place 0.5 mL of this solution in a test tube, and add carefully 0.5 mL of sulfuric acid to make two layers: a red-brown color develops at the zone of contact, and the upper layer acquires a blue-green to blue-purple color.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Imperata Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Imperata Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Rootlet and scaly leaf—When perform the test of foreign matter ⟨*5.01*⟩, the amount of the rootlets and scaly leaves contained in Imperata Rhizome is not more than 3.0 %.

(4) Foreign matter ⟨*5.01*⟩—The amount of foreign matter other than rootlets and scaly leaves is not more than 1.0%.

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Ipecac

Ipecacuanhae Radix

トコン

Ipecac is the root and rhizome of *Cephaelis ipecacuanha* A. Richard or *Cephaelis acuminata* Karsten (*Rubiaceae*).

It contains not less than 2.0% of the total alkaloids (emetine and cephaeline), calculated on the basis of dried material.

Description Slender, curved, cylindrical root, 3 – 15 cm in length, 0.3 – 0.9 cm in diameter; mostly twisted, and sometimes branched; outer surface gray, dark grayish brown, red-brown in color and irregularly annulated; when root fractured, cortex easily separable from the xylem; the cortex on the fractured surface is grayish brown, and the xylem is light brown in color: thickness of cortex up to about two-thirds of radius in thickened portion. Scales in rhizome opposite.

Odor, slight; powder irritates the mucous membrane of the nose; taste, slightly bitter and unpleasant.

Under a microscope ⟨*5.01*⟩, the transverse section of Ipecac reveals a cork layer, consisting of brown thin-walled cork cells; in the cortex, sclerenchyma cells are absent; in the xylem, vessels and tracheids arranged alternately; parenchyma cells filled with starch grains and sometimes with raphides of calcium oxalate.

Identification To 0.5 g of pulverized Ipecac add 2.5 mL of hydrochloric acid, allow to stand for 1 hour with occasional shaking, and filter. Collect the filtrate into an evaporating dish, and add a small pieces of chlorinated lime: circumference of it turns red.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Ipecac according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Ipecac according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Assay Weigh accurately about 0.5 g of pulverized Ipecac, in a glass-stoppered centrifuge tube, add 30 mL of 0.01 mol/L hydrochloric acid TS, shake for 15 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure twice with the residue using 30-mL portions of 0.01 mol/L hydrochloric acid TS. Combine all the extracts, add 0.01 mol/L hydrochloric acid TS to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of emetine hydrochloride for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 50°C) for 5 hours, dissolve in 0.01 mol/L hydrochloric acid TS to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas, A_{TE} and A_{TC}, of emetine and cephaeline obtained with the sample solution, and the peak area, A_{SE}, of emetine obtained with the standard solution.

$$\text{Amount (mg) of total alkaloids (emetine and cephaeline)} = M_S \times \{A_{TE} + (A_{TC} \times 0.971)\}/A_{SE} \times 0.868$$

M_S: Amount (mg) of emetine hydrochloride for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 283 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 2.0 g of sodium 1-heptane sulfonate in 500 mL of water, adjust the pH 4.0 with acetic acid (100), and add 500 mL of methanol.

Flow rate: Adjust so that the retention time of emetine is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg each of emetine hydrochloride for assay and cephaeline hydrobromide in 0.01 mol/L hydrochloric acid TS to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, cephaeline and emetine are eluted in this order with the resolution between these peaks being not less than 5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of emetine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Ipecac

Ipecacuanhae Radix Pulverata

トコン末

Powdered Ipecac is the powder of Ipecac or its powder diluted with Potato Starch.

It contains not less than 2.0% and not more than 2.6% of the total alkaloids (emetine and cephaeline), calculated on the basis of dried material.

Description Powdered Ipecac occurs as a light grayish yellow to light brown powder. It has a slight odor, which is irritating to the nasal mucosa, and has a somewhat bitter and unpleasant taste.

Under a microscope ⟨*5.01*⟩, Powdered Ipecac reveals starch grains and needle crystals of calcium oxalate; fragments of parenchyma cells containing starch grains or the needle crystals; substitute fibers,thin-walled cork tissue; vessels and tracheids with simple or bordered pits; a few wood fibers and wood parenchyma. Starch grains inherent in Ipecac, mainly 2 – 8-compound grains, rarely simple grains 4 – 22 μm in diameter; and needle crystals of calcium oxalate 25 – 60 μm in length.

Identification To 0.5 g of Powdered Ipecac add 2.5 mL of hydrochloric acid, allow to stand for 1 hour with occasional shaking, and filter. Collect the filtrate into an evaporating dish, and add a small pieces of chlorinated lime: circumference of it turns red.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of Powdered Ipecac according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of Powdered Ipecac according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope ⟨*5.01*⟩, groups of stone cells and sclerenchymatous fibers are not observed.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Assay Weigh accurately about 0.5 g of Powdered Ipecac, transfer into a glass-stoppered centrifuge tube, add 30 mL of 0.01 mol/L hydrochloric acid TS, shake for 15 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure twice with the residue using 30-mL portions of 0.01 mol/L hydrochloric acid TS. Combine all the extracts, add 0.01 mol/L hydrochloric acid TS to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of emetine hydrochloride for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 50°C) for 5 hours, dissolve in 0.01 mol/L hydrochloric acid TS to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas, A_{TE} and A_{TC}, of emetine and cephaeline obtained with the sample solution, and the peak area, A_{SE}, of emetine obtained with the standard solution.

Amount (mg) of total alkaloids (emetine and cephaeline)
$= M_S \times \{A_{TE} + (A_{TC} \times 0.971)\}/A_{SE} \times 0.868$

M_S: Amount (mg) of emetine hydrochloride for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 283 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 2.0 g of sodium 1-heptane sulfonate in 500 mL of water, adjust the pH 4.0 with acetic acid (100), and add 500 mL of methanol.

Flow rate: Adjust so that the retention time of emetine is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg each of emetine hydrochloride for assay and cephaeline hydrobromide in 10 mL of 0.01 mol/L hydrochloric acid TS. When the procedure is run with 10 μL of this solution under the above operating conditions, cephaeline and emetine are eluted in this order with the resolution between these peaks being not less than 5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of emetine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Ipecac Syrup

トコンシロップ

Ipecac Syrup is a syrup containing not less than 0.12 g and not more than 0.15 g of the total alkaloids (emetine and cephaeline) per 100 mL.

Method of preparation Take coarse powder of Ipecac, prepare the fluidextract as directed under Fluidextracts using a mixture of Ethanol and Purified Water or Purified Water in Containers (3:1), and evaporate the mixture under low pressure (in vacuo) or add a suitable amount of Ethanol or Purified Water or Purified Water in Containers if necessary to get a solution containing 1.7 to 2.1 g of the total alkaloids (emetine and cephaeline) per 100 mL. To 70 mL of this solution add 100 mL of Glycerin and Simple Syrup to make 1000 mL, as directed under Syrups.

Description Ipecac Syrup is a yellow-brown, viscous liquid. It has a sweet taste and a bitter aftertaste.

Identification Take 2 mL of Ipecac Syrup into an evaporating dish, mix with 1 mL of hydrochloric acid, and add small pieces of chlorinated lime: circumference of it turns orange.

Purity Ethanol—Take exactly 5 mL of Ipecac Syrup, add exactly 5 mL of the internal standard solution and water to make 50 mL, and use this solution as the sample solution. Separately, pipet 5 mL of ethanol (99.5), and add water to make exactly 100 mL. To exactly 5 mL of this solution add exactly 5 mL of the internal standard solution and water to make 50 mL, and use this solution as the standard solution. Perform the test with 2 μL each of the sample solution and standard solution as directed under Gas Chromatography ⟨*2.02*⟩ according to the following conditions, and calculate the rate of peak height of ethanol to that of the internal standard, Q_T and Q_S: Q_T is not larger than Q_S.

Internal standard solution—A solution of acetonitrile (1 in 20).

Operating conditions—

Detector: A hydrogen flame-ionization detector.

Column: A glass-column about 3 mm in inside diameter and about 1.5 m in length, packed with ethylvinylbenzene-divinylbenzene porous co-polymer for gas chromatography (150 to 180 μm in particle diameter).

Column temperature: A constant temperature of between 105°C and 115°C.

Carrier gas: Nitrogen.

Flow rate: Adjust so that the retention time of ethanol is 5 to 10 minutes.

System suitability—

System performance: When the procedure is run with 2 μL of the standard solution under the above operating conditions, ethanol and the internal standard are eluted in this order with the resolution between these peaks being not less than 1.5.

Microbial limit ⟨*4.05*⟩ The acceptance criteria of TAMC and TYMC are 10^3 CFU/mL and 10^2 CFU/mL, respectively. *Escherichia coli*, *Salmonella*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* are not observed.

Assay Take exactly 5 mL of Ipecac Syrup, add 0.01 mol/L hydrochloric acid TS to make exactly 50 mL, and use the solution as the sample solution. Separately, weigh accurately about 10 mg of emetine hydrochloride for assay, previously dried in a desiccator (in vacuum, phosphorus (V) oxide, 50°C) for 5 hours, dissolve in 0.01 mol/L hydrochloric acid TS to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas, A_{TE} and A_{TC}, of emetine and cephaeline with the sample solution, and the peak area, A_{SE}, of emetine with the standard solution.

Amount (mg) of total alkaloids (emetine and cephaeline)
$= M_S \times \{A_{TE} + (A_{TC} \times 0.971)\}/A_{SE} \times 1/2 \times 0.868$

M_S: Amount (mg) of emetine hydrochloride for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 283 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 2.0 g of sodium l-heptane sulfonate in 500 mL of water, adjust the pH to 4.0 with acetic acid (100), and add 500 mL of methanol.

Flow rate: Adjust so that the retention time of emetine is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg each of emetine hydrochloride for assay and cephaeline hydrobromide in 10 mL of 0.01 mol/L hydrochloric acid TS. When the procedure is run with 10 μL of this solution under the above operating conditions, cephaeline and emetine are eluted in this order with the resolution between these peaks being not less than 5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operat-

ing conditions, the relative standard deviation of the peak area of emetine is not more than 1.5%.

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Japanese Angelica Root

Angelicae Acutilobae Radix

トウキ

Japanese Angelica Root is the root of *Angelica acutiloba* Kitagawa or *Angelica acutiloba* Kitagawa var. *sugiyamae* Hikino (*Umbelliferae*), usually after being passed through hot water.

Description Thick and short main root, with numerous branched roots, nearly fusiform; 10 – 25 cm in length; externally dark brown to red-brown, with longitudinal wrinkles and horizontal protrusions composed of numerous scars of fine rootlets; fractured surface is dark brown to yellow-brown in color, and smooth; and with a little remains of leaf sheath at the crown.

Odor, characteristic; taste, slightly sweet, followed by slight pungency.

Under a microscope <*5.01*>, a transverse section reveals 4 to 10 cellular layers of cork, with several cellular layers of collenchyma inside of the layer; the cortex exhibits many oil canals surrounded by secretory cells and often large hollows appear; boundary of cortex and xylem is distinct; in the xylem, numerous vessels radiate alternately with medullary rays; vessels in the outer part of the xylem are singly or in several groups, and disposed rather densely in a cuneiform pattern, but vessels in the region of the center are scattered very sparsely; starch grains are simple grains, not more than 20 μm in diameter, and rarely 2- to 5-compound grains, some times up to 25 μm in diameter; starch grains often gelatinized.

Identification To 1.0 g of pulverized Japanese Angelica Root add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution (1). Dissolve 1 mg of scopoletin for thin-layer chromatography in 10 mL of methanol, and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL each of the standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (30:25:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): two of the several spots obtained from the sample solution have the same color tones and *R*f values with the corresponding blue-white fluorescent spots from the standard solutions (1) and (2).

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Japanese Angelica Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Japanese Angelica Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Leaf sheath—When perform the test of foreign matter <*5.01*>, the amount of leaf sheath contained in Japanese Angelica Root does not exceed 3.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than leaf sheath contained in Japanese Angelica Root does not exceed 1.0%.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 35.0%.

Containers and storage Containers—Well-closed containers.

Powdered Japanese Angelica Root

Angelicae Acutilobae Radix Pulverata

トウキ末

Powdered Japanese Angelica Root is the powder of Japanese Angelica Root.

Description Powdered Japanese Angelica Root occurs as a light grayish brown powder. It has a characteristic odor and a slight, sweet taste with a slightly pungent aftertaste.

Under a microscope <*5.01*>, Powdered Japanese Angelica Root reveals starch grains or masses of gelatinized starch, and fragments of parenchyma containing them; fragments of light yellow-brown cork tissue; fragments of rather thick-walled collenchyma and phloem tissue; fragments of oil canal surrounded by secretory cells; fragments, 20 – 60 μm in diameter, of scalariform and reticulate vessels with simple perforation; starch grains composed of simple grains not more than 20 μm in diameter, and rarely 2- to 5-compound grains, sometimes comes up to 25 μm.

Identification To 1.0 g of Powdered Japanese Angelica Root add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution (1). Dissolve 1 mg of scopoletin for thin-layer chromatography in 10 mL of methanol, and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL each of the standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, acetone and acetic acid (100) (30:25:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): two of the several spots obtained from the sample solution have the same color tones and *R*f values with the corresponding blue-white fluorescent spots from the standard solutions (1) and (2).

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Japanese Angelica Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Japanese Angelica Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Japanese Angelica Root does not show remarkably lignified sclerenchymatous cells.

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 35.0%.

Containers and storage Containers—Tight containers. Storage—Light-resistant.

Japanese Gentian

Gentianae Scabrae Radix

リュウタン

Japanese Gentian is the root and rhizome of *Gentiana scabra* Bunge, *Gentiana manshurica* Kitagawa or *Gentiana triflora* Pallas (*Gentianaceae*).

Description Irregular, cylindrical, short rhizome with numerous, slender roots around, and externally yellow-brown to grayish yellow-brown. The root is 10 – 15 cm in length, about 0.3 cm in diameter, and has longitudinal, coarse wrinkles on the outer surface; flexible; fractured surface, smooth and yellow-brown in color. The rhizome is about 2 cm in length, about 0.7 cm in diameter, and has buds or short remains of stems at the top.

Odor, slight; taste, extremely bitter and lasting.

Under a microscope ⟨*5.01*⟩, a transverse section of the young root reveals epidermis, exodermis and a few cellular layers of primary cortex; usually, the outermost layer is endodermis consisting of characteristic cells divided into a few daughter cells, often with collenchyma of 1 to 2 cellular layers contacting the inner side; secondary cortex having rents here and there, and irregularly scattered sieve tubes; vessels arranged rather radially in xylem, sieve tubes existing in xylem; the rhizome has a large pith, rarely with sieve tubes; parenchyma cells contain small needle, plate or sand crystals of calcium oxalate and oil drops; starch grains usually absent.

Identification To 0.5 g of pulverized Japanese Gentian add 10 mL of methanol, shake for 20 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of gentiopicroside for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and the same *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Japanese Gentian according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Japanese Gentian according to Method 4, and perform the test (not more than 5 ppm).

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Powdered Japanese Gentian

Gentianae Scabrae Radix Pulverata

リュウタン末

Powdered Japanese Gentian is the powder of Japanese Gentian.

Description Powdered Japanese Gentian occurs as a grayish yellow-brown powder. It has a slight odor and a lasting, extremely bitter taste.

Under a microscope ⟨*5.01*⟩, Powdered Japanese Gentian reveals fragments of parenchyma cells containing oil droplets and fine crystals, fragments of endodermis divided into daughter cells with suberized cell wall, exodermis, and vessels; vessels mainly reticulate vessels and scalariform vessels, 20 – 30 μm in diameter.

Identification To 0.5 g of Powdered Japanese Gentian add 10 mL of methanol, shake for 20 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of gentiopicroside for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of Powdered Japanese Gentian according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of Powdered Japanese Gentian according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope ⟨*5.01*⟩, Powdered Japanese Gentian usually reveals no stone cells and fibers. No starch grains; if any, very few.

Total ash ⟨*5.01*⟩ Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Japanese Valerian

Valerianae Fauriei Radix

カノコソウ

Japanese Valerian is the root and rhizome of *Valeriana fauriei* Briquet (*Valerianaceae*).

Description Obovoid, short rhizome with numerous, fine and long roots; externally dark brown to grayish brown. The

root, 10 – 15 cm in length, 0.1 – 0.3 cm in diameter; externally, with fine longitudinal wrinkles; brittle. The rhizome, 1 – 2 cm in length, 1 – 2 cm in diameter, with buds and remains of stem at the crown; hard in texture and difficult to break; flank of rhizome sometimes accompanied with stolons having thick and short or thin, long and extremely small, scaly leaves. Under a magnifying glass, the transverse section reveals a thick, light grayish brown cortex, and a grayish brown stele.

Odor, strong and characteristic; taste, slightly bitter.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Japanese Valerian according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Japanese Valerian according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 5.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Japanese Valerian provided that 1 mL of silicon resin is previously added to the sample in the flask: the volume of essential oil is not less than 0.3 mL.

Containers and storage Containers—Tight containers.

Powdered Japanese Valerian

Valerianae Fauriei Radix Pulverata

カノコソウ末

Powdered Japanese Valerian is the powder of Japanese Valerian.

Description Powdered Japanese Valerian occurs as a dark grayish brown powder. It is somewhat moist to the touch. It has a strong, characteristic odor and a slightly bitter taste.

Under a microscope <*5.01*>, Powdered Japanese Valerian reveals starch grains and fragments of parenchyma cells containing them; fragments of pitted vessels, reticulate vessels, ring vessels, and spiral vessels; fragments of exodermis containing oil droplets and composed of cells suberized and divided into daughter cells; fragments of yellow stone cells from the rhizome and the stolon; and very rarely, some fragments of epidermis and phloem fibers. Starch grains, simple grains 10 – 20 μm in diameter and 2- to 4-compound grains; oil droplets stained red with sudan III TS.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Japanese Valerian according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Japanese Valerian according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 5.0%.

Essential oil content <*5.01*> Perform the test with 50.0 g of Powdered Japanese Valerian provided that 1 mL of silicon resin is previously added to the sample in the flask: the volume of essential oil is not less than 0.2 mL.

Containers and storage Containers—Tight containers.

Japanese Zanthoxylum Peel

Zanthoxyli Piperiti Pericarpium

サンショウ

Japanese Zanthoxylum Peel is the pericarps of the ripe fruit of *Zanthoxylum piperitum* De Candolle (*Rutaceae*), from which the seeds separated from the pericarps have been mostly removed.

Description Pericarps of capsules of 2 or 3 flattened spheroidal mericarps, which are dehiscent in 2 pieces about 5 mm in diameter; the outer surface of pericarp, dark yellow-red to dark red-brown, with numerous dented spots originated from oil sacs; the inner surface, light yellow-white.

Odor, characteristically aromatic; taste, acrid, which gives numbing sensation to the tongue.

Under a microscope <*5.01*>, a transverse section of Japanese Zanthoxylum Peel reveals the external epidermis and the adjoined unicellular layer containing red-brown tannin; the mesocarp holds oil sacs being up to approximately 500 μm in diameter and sporadically vascular bundles consisting mainly of spiral vessels; the endocarp consists of stone cell layers; inner epidermal cells very small.

Identification To 2 g of pulverized Japanese Zanthoxylum Peel add 10 mL of water, shake for 5 minutes, add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane, methanol and acetic acid (100) (20:20:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a spot with an *R*f value of about 0.3 is observed.

Purity **(1)** Seed—When perform the test of foreign matter <*5.01*>, the amount of the seeds contained in Japanese Zanthoxylum Peel does not exceed 20.0%.

(2) Peduncle and twig—The amount of the peduncles and twigs contained in Japanese Zanthoxylum Peel does not exceed 5.0%.

(3) Foreign matter <*5.01*>—The amount of foreign matter other than peduncles and twigs contained in Japanese Zanthoxylum Peel does not exceed 1.0%.

Total ash <*5.01*> Not more than 8.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Essential oil content <*5.01*> Perform the test with 30.0 g of pulverized Japanese Zanthoxylum Peel: the volume of essential oil is not less than 1.0 mL.

Containers and storage Containers—Well-closed containers.

Powdered Japanese Zanthoxylum Peel

Zanthoxyli Piperiti Pericarpium Pulveratum

サンショウ末

Powdered Japanese Zanthoxylum Peel is the powder of Japanese Zanthoxylum Peel.

Description Powdered Japanese Zanthoxylum Peel occurs as a dark yellow-brown powder. It has a strong, characteristic aroma and an acrid taste, leaving a sensation of numbness on the tongue.

Under a microscope ⟨*5.01*⟩, Powdered Japanese Zanthoxylum Peel reveals fragments of inner tissue of endocarp consisting of stone cells with cell walls about 2.5 μm in thickness; fragments of spiral and ring vessels 10 – 15 μm in diameter; fragments of oil sacs containing essential oil or resin; fragments of epidermal cells, polygonal in surface view, containing tannin; numerous oil drops; masses of tannin, colored red by adding vanillin-hydrochloric acid TS.

Identification To 2 g of Powdered Japanese Zanthoxylum Peel add 10 mL of water, shake for 5 minutes, add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane, methanol and acetic acid (100) (20:20:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a spot with an *R*f value of about 0.3 is observed.

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Essential oil content ⟨*5.01*⟩ Perform the test with 30.0 g of Powdered Japanese Zanthoxylum Peel: the volume of essential oil is not less than 0.8 mL.

Containers and storage Containers—Tight containers.

Jujube

Ziziphi Fructus

タイソウ

Jujube is the fruit of *Ziziphus jujuba* Miller var. *inermis* Rehder (*Rhamnaceae*).

Description Ellipsoidal or broad ovoid fruit, 2 – 3 cm in length, 1 – 2 cm in diameter; externally red-brown with coarse wrinkles, or dark grayish red with fine wrinkles, and both lustrous; both ends slightly dented, with a scar of style on one end and a scar of peduncle on the other; epicarp thin and leather; mesocarp thick, dark grayish brown in color, spongy, soft and adhesive; endocarp extremely hard, fusiform, and divided into two loculi; seeds flat and ovoid.

Odor, slight and characteristic; taste, sweet.

Purity **(1)** Rancidity—Jujube has no unpleasant, rancid odor and taste.

(2) Total BHC's and total DDT's ⟨*5.01*⟩ Not more than 0.2 ppm, respectively.

Total ash ⟨*5.01*⟩ Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Jujube Seed

Ziziphi Semen

サンソウニン

Jujube Seed is the seed of *Ziziphus jujuba* Miller var. *spinosa* Hu ex H. F. Chow (*Rhamnaceae*).

Description Jujube Seed is a compressed ovate to orbicular, lenticular seed, 5 – 9 mm in length, 4 – 6 mm in width, 2 – 3 mm in thickness, externally brown to dark red-brown, glossy; hilum at one end, chalaza at the other end; seed coat sightly flexible, covering, milky white endosperm and light yellow embryo. 100 seeds weigh 3.0 – 4.5 g.

Odor, slightly oily; taste, mild and slightly oily.

Under a microscope ⟨*5.01*⟩, transverse section reveals seed coat composed of an upper epidermis, parenchyma and lower epidermis; upper epidermal cells sclerified and elongated in radial direction; lower epidermis covered with cuticle; endosperm composed of parenchyma, containing aggregated crystals of calcium oxalate, aleurone grains and starch grains; cotyledons composed of parenchyma that contains aleurone grains, starch grains and oil drops.

Identification To 2 g of pulverized Jujube Seed add 10 mL of methanol, and heat under a reflux condenser for 10 minutes. After cooling, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography, develop the plate with a mixture of acetone, ethyl acetate, water and acetic acid (100) (10:10:3:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two spots appear at the *R*f value of about 0.3 and about 0.4, and these spots exhibit a blue-white fluorescence when examined under ultraviolet light (main wavelength: 365 nm) after spraying evenly dilute sulfuric acid on the plate and heating at 105°C for 5 minutes.

Purity Foreign matter ⟨*5.01*⟩—Jujube Seed contains not more than 1.0% of the endocarp and other foreign matters.

Loss on drying ⟨*5.01*⟩ Not more than 11.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 8.5%.

Containers and storage Containers—Well-closed containers.

Juzentaihoto Extract

十全大補湯エキス

Juzentaihoto Extract contains not less than 1.5 mg (for preparation prescribed 2.5 g of Ginseng) or not less than 1.8 mg (for preparation prescribed 3 g of Ginseng) of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), not less than 26 mg and not more than 78 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 6 mg and not more than 18 mg (for preparation prescribed 1 g of Glycyrrhiza) or not less than 10 mg and not more than 30 mg (for preparation prescribed 1.5 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Ginseng	3 g	3 g	2.5 g	3 g
Astragalus Root	3 g	3 g	2.5 g	3 g
Atractylodes Rhizome	3 g	—	3.5 g	3 g
Atractylodes Lancea Rhizome	—	3 g	—	—
Poria Sclerotium	3 g	3 g	3.5 g	3 g
Japanese Angelica Root	3 g	3 g	3.5 g	3 g
Peony Root	3 g	3 g	3 g	3 g
Rehmannia Root	3 g	3 g	3.5 g	3 g
Cnidium Rhizome	3 g	3 g	3 g	3 g
Cinnamon Bark	3 g	3 g	3 g	3 g
Glycyrrhiza	1.5 g	1.5 g	1 g	1g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Juzentaihoto Extract is a light brown to brown powder or black-brown viscous extract. It has a slight odor and a sweet and bitter taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 15 mL of sodium hydroxide TS, centrifuge, and separate the supernatant liquid. To the liquid add 10 mL of 1-butanol, shake, centrifuge, and separate the 1-butanol layer. To the 1-butanol layer add 10 mL of water, shake, centrifuge, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark brown spot from the standard solution (Ginseng).

(2) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 15 mL of sodium hydroxide TS, centrifuge, and separate the supernatant liquid. To the liquid add 10 mL of 1-butanol, shake, centrifuge, and separate the 1-butanol layer. To the 1-butanol layer add 10 mL of water, shake, centrifuge, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of astragaloside IV for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-brown spot from the standard solution (Astragalus Root).

(3) For preparation prescribed Atractylodes Rhizome—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 5 mL of diethyl ether, shake, and centrifuge. Use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red spot from the standard solution (Atractylodes Rhizome).

(4) For preparation prescribed Atractylodes Lancea Rhizome—Shake 5.0 g of the dry extract (or 15.0 g of the viscous extract) with 10 mL of water, add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 15 mL of water and 5 mL of 0.1 mol/L hydrochloric acid TS, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365

nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Cnidium Rhizome; Japanese Angelica Root).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(7) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 30 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of water, methanol and 1-butanol (1:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: a dark green spot is observed at an *R*f value of about 0.6 (Rehmannia Root).

(8) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 50 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(9) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Ginsenoside Rb_1—Weigh accurately about 2 g of the dry extract (or an amount of the viscous extract, equivalent to about 2 g of the dried substance), add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 15 mL of diluted methanol (3 in 5), and repeat the same procedure. Combine the supernatant liquids, add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, then add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 5 mL of this solution to a column (about 10 mm in inside diameter and packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), washed just before use with methanol and then with diluted methanol (3 in 10)), and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

Amount (mg) of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$)
$= M_S \times A_T/A_S \times 1/5$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl groups bound silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (400:100:1).

Flow rate: 1.0 mL per minute (the retention time of ginsenoside Rb_1 is about 16 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

Amount (mg) of paeoniflorin ($C_{23}H_{28}O_{11}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this

solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Kakkonto Extract

葛根湯エキス

Kakkonto Extract contains not less than 7 mg and not more than 21 mg (for preparation prescribed 3 g of Ephedra Herb) or not less than 10 mg and not more than 30 mg (for preparation prescribed 4 g of Ephedra Herb) of total alkaloids (ephedrine and pseudoephedrine), not less than 14 mg and not more than 56 mg (for preparation prescribed 2 g of Peony Root) or not less than 21 mg and not more than 84 mg (for preparation prescribed 3 g of Peony Root) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 15 mg and not more than 45 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Pueraria Root	8 g	4 g	4 g	4 g
Ephedra Herb	4 g	4 g	3 g	3 g
Jujube	4 g	3 g	3 g	3 g
Cinnamon Bark	3 g	2 g	2 g	2 g
Peony Root	3 g	2 g	2 g	2 g
Glycyrrhiza	2 g	2 g	2 g	2 g
Ginger	1 g	1 g	1 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Kakkonto Extract occurs as a light brown to brown powder or black-brown viscous extract. It has a characteristic odor, and a sweet first, then hot, and slightly bitter taste.

Identification **(1)** To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Puerarin RS or puerarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Pueraria Root).

(2) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <2.03>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (4:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot is observed at an *R*f value of about 0.5 (Ephedra Herb).

(3) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) To 2.0 g of the dry extract (or 6.0 g of the viscous extract), add 10 mL of water, shake, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solu-

tion.

(4) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatog-raphy in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(5) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed in the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Total alkaloids (ephedrine and pseudoephedrine)—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. In addition, repeat twice in the same manner for the aqueous layer using 1.0 mL of ammonia TS and 20 mL of diethyl ether. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL. Centrifuge this solution, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of ephedrine hydrochloride for assay of crude drugs, previously dried at 105°C for 3 hours, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine obtained with the sample solution, and the peak area, A_S, of ephedrine with the standard solution.

Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)

$$= M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drugs taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, and add 650 mL of water and 1 mL of phosphoric acid to dissolve sodium lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of ephedrine is about 27 minutes).

System suitability—

System performance: Dissolve 1 mg each of ephedrine hydrochloride for assay of crude drugs and pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent

to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, flow through in a column packed with 2 g of polyamide for column chromatography, elute with 20 mL of water, add 1 mL of acetic acid (100) and water to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 5/8$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about

0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Kakkontokasenkyushin'i Extract

葛根湯加川芎辛夷エキス

Kakkontokasenkyushin'i Extract contains not less than 9.5 mg and not more than 28.5 mg (for preparation prescribed 3 g of Ephedra Herb) or not less than 13 mg and not more than 39 mg (for preparation prescribed 4 g of Ephedra Herb) of total alkaloids (ephedrine and pseudoephedrine), not less than 17 mg and not more than 51 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), and not less than 1.5 mg and not more than 6 mg (for preparation prescribed 2 g of Magnolia Flower) or not less than 2 mg and not more than 8 mg (for preparation prescribed 3 g of Magnolia Flower) of magnoflorine [as magnoflorine iodide ($C_{20}H_{24}INO_4$: 469.31)], per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Pueraria Root	4 g	4 g
Ephedra Herb	4 g	3 g
Jujube	3 g	3 g
Cinnamon Bark	2 g	2 g
Peony Root	2 g	2 g
Glycyrrhiza	2 g	2 g
Ginger	1 g	1 g
Cnidium Rhizome	3 g	2 g
Magnolia Flower	3 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Kakkontokasenkyushin'i Extract occurs as a light brown to brown powder or black-brown viscous extract, having a characteristic order, and a sweet first, then a bitter and hot taste.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Puerarin RS or puerarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Pueraria Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (4:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot is observed at an *R*f value of about 0.5 (Ephedra Herb).

(3) Perform the test according to the following (i) or (ii) (Cinnamon Bark).

(i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect the apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

(ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzoaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple to purple spot from the standard solution (Peony Root).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample

solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(7) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 15 mL of water and 5 mL of 0.1 mol/L hydrochloric acid TS, and then shake with 25 mL of diethyl ether. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Cnidium Rhizome).

(8) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, to 1 g of powdered magnolia flower add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the brown spot (*R*f value: about 0.4) from the standard solution (Magnolia Flower).

Purity **(1)** Heavy metals ⟨*1.07*⟩—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed in Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying ⟨*2.41*⟩ The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Total alkaloids (ephedrine and pseudoephedrine)—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. In addition, repeat twice in the same manner for the aqueous layer using 1.0 mL of ammonia TS and 20 mL of diethyl ether. Combine the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of ephedrine hydrochloride for assay of crude drugs, previously dried at 105°C for 3 hours, dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine with the sample solution, and peak area, A_S, of ephedrine with standard solution.

Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)

$$= M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drugs taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, then add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of ephedrine is about 27 minutes).

System suitability—

System performance: Dissolve 1 mg each of ephedrine hydrochloride for assay of crude drugs and pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, flow through in a column packed with 2 g of polyamide for column chromatography, elute with 20 mL of water, add 1 mL of acetic acid (100) to the effluent, then add water to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 5/8$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

(4) Magnoflorine—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, add 3.0 mL of diluted sodium hydroxide TS (1 in 10), shake for 10 minutes, centrifuge, and remove the diethyl ether layer. Add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 3.0 mL of 0.1 mol/L hydrochloric acid TS and 20 mL of diluted methanol (1 in 2), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2) shake for 15 minutes, centrifuge, and separate the supernatant liquid. Combine the previous supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of magnoflorine iodide for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of magnoflorine in each solution.

Amount (mg) of magnoflorine [as magnoflorine iodide ($C_{20}H_{24}INO_4$)]
$= M_S \times A_T/A_S \times 1/20$

M_S: Amount (mg) of magnoflorine iodide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 303 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, then add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of magnoflorine is about 20 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of magnoflorine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of magnoflorine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Kamikihito Extract

加味帰脾湯エキス

Kamikihito Extract contains not less than 0.8 mg and not more than 3.2 mg of saikosaponin b_2, not less than 27 mg and not more than 81 mg of geniposide, and not less than 6 mg and not more than 18 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Ginseng	3 g	3 g	3 g	3 g
Atractylodes Rhizome	3 g			3 g
Atractylodes Lancea Rhizome		3 g	3 g	
Poria Sclerotium	3 g	3 g	3 g	3 g
Jujube Seed	3 g	3 g	3 g	3 g
Longan Aril	3 g	3 g	3 g	3 g
Astragalus Root	2 g	3 g	2 g	3 g
Japanese Angelica Root	2 g	2 g	2 g	2 g
Polygala Root	1.5 g	2 g	1 g	2 g
Bupleurum Root	3 g	3 g	3 g	3 g
Gardenia Fruit	2 g	2 g	2 g	2 g
Glycyrrhiza	1 g	1 g	1 g	1 g
Saussurea Root	1 g	1 g	1 g	1 g
Jujube	1.5 g	2 g	1 g	2 g
Ginger	0.5 g	1 g	1 g	0.5g
Moutan Bark				2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the preparation 1) to 4), using the crude drugs shown above. Or, prepare a dry extract by adding Light Anhydrous Silicic Acid to an extractive prepared as directed under Extracts, according to the preparation 2, using the crude drugs shown above.

Description Kamikihito Extract is a light yellow-brown to brown powder or black-brown viscous extract. It has a slightly odor, and a slightly sweet, hot and bitter taste.

Identification (1) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of sodium hydroxide TS, shake, centrifuge, and separate the supernatant liquid. To the liquid add 10 mL of 1-butanol, shake, centrifuge, and separate the 1-butanol layer. To this layer add 10 mL of water, shake, centrifuge, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of methanol, and use the solution as the sample solution. Separately, dissolve 1 mg of ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100)

(7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(2) For preparation prescribed Atractylodes Rhizome—To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of diethyl ether, and use the solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red to red-purple spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), then dissolve the residue in 2 mL of hexane, and use the solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark violet spot is observed at an *R*f value of about 0.5, and this spot exhibits greenish brown when the plate is sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated the plate at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of sodium hydroxide TS, shake, centrifuge, and separate the supernatant liquid. To the liquid add 10 mL of 1-butanol, shake, centrifuge, and separate 1-butanol layer. To the 1-butanol layer add 10 mL of water, shake, centrifuge, separate the 1-butanol layer, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue in 2 mL of methanol, and use this solution as the sample solution. Separately, dissolve 1 mg of astragaloside IV for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 2-propanol, water and ammonia solution (28) (9:2:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to blue-purple spot from the standard solution (Astragalus Root).

(5) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then add 2 mL of diethyl ether to the residue, and use the solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of butyl acetate and hexane (2:1) to a distance of about 7 cm, and air-dry the plate. Examine the plate under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root).

(6) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 30 mL of 1 mol/L hydrochloric acid TS, and heat for 10 minutes. After cooling, to 10 mL of this solution add 10 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the sample solution. Separately, to 2.0 g of a powder of polygala root add 30 mL of 1 mol/L hydrochloric acid TS, and heat for 10 minutes. After cooling, to 10 mL of this solution add 10 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol and acetic acid (100) (7:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfric acid TS on the plate, heat the plate at 105°C for 1 minute, and observe while hot: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purplish red spot (at an *R*f value of about 0.5) from the standard solution (Polygala Root).

(7) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of sodium hydroxide TS, shake, centrifuge, and separate the supernatant liquid. To the liquid add 10 mL of 1-butanol, shake, centrifuge, and separate the 1-butanol layer. To this layer add 10 mL of water, shake, centrifuge, and separate the 1-butanol layer. Evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of methanol, and use this solution as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(8) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography

<2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, ammonia solution (28) (6:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minute: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Gardenia Fruit).

(9) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(10) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of water, shake, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, to 1.0 g of a powder of saussurea root add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 2 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (Saussurea Root).

(11) To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of water, shake, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(12) For preparation prescribed Moutan Bark—To 3.0 g of the dry extract (or 9.0 g of the viscous extract) add 15 mL of water, shake, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and diethyl ether (5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot from the standard solution (Moutan Bark).

Purity **(1)** Heavy metals <1.07>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <2.41> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <5.01> Not less than 8.0%, calculated on the dried basis. However, for the dry extract prepared by adding Light Anhydrous Silicic Acid, between 9.0% and 18.0%.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. Centrifuge, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and separate the supernatant liquid. Combine all the supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Geniposide—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of geniposide for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of geniposide in each solution.

$$\text{Amount (mg) of geniposide} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of geniposide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 240 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (900:100:1).

Flow rate: 1.0 mL per minute (retention time of geniposide is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of geniposide are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of geniposide is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Kamishoyosan Extract

加味逍遙散エキス

Kamishoyosan Extract contains not less than 28 mg and not more than 84 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), not less than 25 mg and not more than 75 mg of geniposide, and not less than 10 mg and not more than 30 mg (for preparation prescribed 1.5 g of Glycyrrhiza) or not less than 13 mg and not more than 39 mg (for preparation prescribed 2 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)	5)	6)
Japanese Angelica Root	3 g	3 g	3 g	3 g	3 g	3 g
Peony Root	3 g	3 g	3 g	3 g	3 g	3 g
Atractylodes Rhizome	3 g	—	3 g	—	3 g	3 g
Atractylodes Lancea Rhizome	—	3 g	—	3 g	—	—
Poria Sclerotium	3 g	3 g	3 g	3 g	3 g	3 g
Bupleurum Root	3 g	3 g	3 g	3 g	3 g	3 g
Moutan Bark	2 g	2 g	2 g	2 g	2 g	2 g
Gardenia Fruit	2 g	2 g	2 g	2 g	2 g	2 g
Glycyrrhiza	2 g	2 g	1.5 g	1.5 g	1.5 g	1.5 g
Ginger	1 g	1 g	1 g	1 g	1.5 g	0.5 g
Mentha Herb	1 g	1 g	1 g	1 g	1 g	1 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 6), using the crude drugs shown above.

Description Kamishoyosan Extract occurs as a yellow-brown to brown powder or black-brown viscous extract. It has slightly a characteristic odor, and a sweet, slightly hot, then bitter taste.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root).

(2) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of albiflorin in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool for more than 30 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Peony Root).

(3) For preparation prescribed Atractylodes Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red spot from the standard solution (Atractylodes Rhizome).

(4) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heat the plate at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(5) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(6) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 15 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and diethyl ether (5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot from the standard solution (Moutan Bark).

(7) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the

sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Gardenia Fruit).

(8) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(9) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(10) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of diluted phosphoric acid (1 in 30), shake, then add 15 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the sample solution. Separately, shake 0.2 g of pulverized mentha herb with 10 mL of diluted phosphoric acid (1 in 30), add 15 mL of ethyl acetate, shake, centrifuge, and use the ethyl acetate layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate, water and acetic acid (100) (10:10:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-brown spot (around *R*f value 0.4) from the standard solution (Mentha Herb).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(2) Geniposide—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of geniposide for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solu-

tion as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of geniposide in each solution.

$$\text{Amount (mg) of geniposide} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of geniposide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 240 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (900:100:1).

Flow rate: 1.0 mL per minute (the retention time of geniposide is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of geniposide are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of geniposide is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Keishibukuryogan Extract

桂枝茯苓丸エキス

Keishibukuryogan Extract contains not less than 0.6 mg and not more than 2.4 mg (for preparation prescribed 3 g of Cinnamon Bark) or not less than 0.8 mg and not more than 3.2 mg (for preparation prescribed 4 g of Cinnamon Bark) of (*E*)-cinnamic acid, not less than 30 mg and not more than 90 mg (for preparation prescribed 3 g each of Moutan Bark and Peony Root) or not less than 40 mg and not more than 120 mg (for preparation prescribed 4 g each of Moutan Bark and Peony Root) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 21 mg and not more than 63 mg (for preparation prescribed 3 g of Peach Kernel) or not less than 28 mg and not more than 84 mg (for preparation prescribed 4 g of Peach Kernel) of amygdalin, per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Cinnamon Bark	4 g	3 g
Poria Sclerotium	4 g	3 g
Moutan Bark	4 g	3 g
Peach Kernel	4 g	3 g
Peony Root	4 g	3 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) using the crude drugs shown above, or prepare a dry extract by adding Light Anhydrous Silicic Acid to an extractive, prepared as directed under Extracts, according to the prescription 2), using the crude drugs shown above.

Description Keishibukuryogan Extract is a light brown to brown powder or black-brown viscous extract. It has a characteristic odor and has a taste slightly sweet first then bitter later.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of (*E*)-

cinnamic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate, formic acid and water (60:40:4:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Cinnamon Bark).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and diethyl ether (5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot from the standard solution (Moutan Bark).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of methanol, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate and water (4:4:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the green-brown spot from the standard solution (Peach Kernel).

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of albiflorin in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (6:3:2) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool for more than 30 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Peony Root).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis. However, for the dry extract prepared by adding Light Anhydrous Silicic Acid, between 9.0% and 18.0%.

Assay **(1)** (*E*)-Cinnamic acid—Conduct this procedure using light-resistant vessels. Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-cinnamic acid for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of (*E*)-cinnamic acid in each solution.

$$\text{Amount (mg) of (}E\text{)-cinnamic acid} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of (*E*)-cinnamic acid for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 273 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (750:250:1).

Flow rate: 1.0 mL per minute (the retention time of (*E*)-cinnamic acid is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of (*E*)-cinnamic acid are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of (*E*)-cinnamic acid is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg),

dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Amygdalin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

$$\text{Amount (mg) of amygdalin} = M_S \times A_T/A_S$$

M_S: Amount (mg) of amygdalin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Koi

Koi

コウイ

Koi is a saccharized substance obtained by hydrolysis of the starch of *Zea mays* Linné (*Gramineae*), *Manihot esculenta* Crantz (*Euphorbiaceae*), *Solanum tuberosum* Linné (*Solanaceae*), *Ipomoea batatas* Poiret (*Convolvulaceae*) or *Oryza sativa* Linné (*Gramineae*), or the seed of *Oryza sativa* Linné from which the seed coat is removed.

Koi is prepared by the following processes 1 or 2, and contains mainly maltose, sometimes glucose and maltotriose also.

Process 1. Saccharize starch with hydrochloric acid, oxalic acid, amylase or wort, then concentrate to dryness, and powder.

Process 2. To starch or a paste of starch prepared by adding water and heating, add hydrochloric acid, oxalic acid, amylase or wort to saccharize, and dry or concentrate.

Koi prepared by Process 1 is termed ''Koi 1'' and by Process 2 is termed ''Koi 2''. The label states the process.

Description

Koi 1: A white crystalline powder. It is odorless and has a sweet taste.

Koi 2: Colorless or brown, clear or semi-translucent, masses or viscous liquid. It is odorless and has a sweet taste.

Identification Dissolve exactly 0.50 g of Koi in a mixture of water and methanol (1:1) to make exactly 50 mL, and use this solution as the sample solution. Separately, dissolve exactly 20.0 mg of maltose hydrate in a mixture of water and methanol (1:1) to make exactly 5 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography in equal size of circular spot each other. Develop the plate with a mixture of 2-butanone, water and acetic acid (100) (3:1:1) to a distance of about 7 cm, and dry at 105°C for 10 minutes the plate. Spray evenly 2,3,5-triphenyl-2*H*-tetrazolium chloride-methanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange spot obtained from the standard solution, and it is larger and more intense than the spot from the standard solution.

Purity **(1)** Clarity of solution—A solution obtained by dissolving 2.0 g of Koi in 20 mL of hot water is practically clear.

(2) Heavy metals ⟨*1.07*⟩

Koi 1: Proceed with 1.0 g of Koi 1 according to Method

1, and perform the test. Prepare the control solution with 1.0 mL of Standard Lead Solution (not more than 10 ppm).

Koi 2: Proceed with 1.0 g of Koi 2 according to Method 2, and perform the test. Prepare the control solution with 1.0 mL of Standard Lead Solution (not more than 10 ppm).

(3) Arsenic <*1.11*>—Prepare the test solution with 1.0 g of Koi according to Method 3, and perform the test (not more than 2 ppm).

Loss on drying <*5.01*>

Koi 1: Not more than 3.0% (1 g, 80°C, 4 hours).

Koi 2: Not more than 15.0% (1 g, 80°C, 4 hours). In the case where the sample is in masses, crush the masses, weigh accurately the mass, and put in a desiccator. In the case in viscous liquid, put in a weighing bottle to spread about 1 mm thick, weigh accurately the mass, and put the bottle in a desiccator.

Total ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Hydrous Lanolin

加水ラノリン

Hydrous Lanolin is Purified Lanolin to which water is added. It contains not less than 70% and not more than 75% of Purified Lanolin (as determined by the test for Residue on evaporation).

Description Hydrous Lanolin is a yellow-white, ointment-like substance, and has a slight, characteristic odor, which is not rancid.

It is soluble in diethyl ether and in cyclohexane, with the separation of water.

When melted by heating on a water bath, it separates into a clear oily layer and a clear aqueous layer.

Melting point: about 39°C.

Identification Dissolve 1 g of Hydrous Lanolin in 50 mL of cyclohexane, and remove the separated water. Superimpose carefully 1 mL of the cyclohexane solution on 2 mL of sulfuric acid: a red-brown color develops at the zone of contact, and sulfuric acid layer shows a green fluorescence.

Acid value <*1.13*> Not more than 1.0.

Iodine value 18 – 36 Heat a suitable amount of Hydrous Lanolin on a water bath to remove its almost moisture, then weigh accurately about 0.8 g of the treated Hydrous Lanolin in a glass-stoppered 500-mL flask, and add 10 mL of cyclohexane to dissolve, and add exactly 25 mL of Hanus's TS, and mix well. If a clear solution is not obtained, add more cyclohexane to make clear, and allow the mixture to stand for 1 hour between 20°C and 30°C in a light-resistant, well-closed container while occasional shaking. Add 20 mL of a solution of potassium iodide (1 in 10) and 100 mL of water, shake, and titrate <*2.50*> the liberated iodine with 0.1 mol/L sodium thiosulfate VS (indicator: 1 mL of starch TS). Perform a blank determination in the same manner.

$$\text{Iodine value} = (a - b) \times 1.269/M$$

M: amount (g) of Hydrous Lanolin taken

a: Volume (mL) of 0.1 mol/L sodium thiosulfate VS consumed in the blank determination

b: Volume (mL) of 0.1 mol/L sodium thiosulfate VS consumed in the titration

Purity **(1)** Acidity or alkalinity—To 5 g of Hydrous Lanolin add 25 mL of water, boil for 10 minutes, and cool. Add water to restore the previous mass, and separate the aqueous layer: the aqueous layer is neutral.

(2) Chloride <*1.03*>—To 2.0 g of Hydrous Lanolin add 40 mL of water, boil for 10 minutes, and cool. Add water to restore the previous mass, and filter. To 20 mL of the filtrate add 6 mL of dilute nitric acid and water to make 50 mL. Use this solution as the test solution, and perform the test. Prepare the control solution with 1.0 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.036%).

(3) Ammonia—To 10 mL of the aqueous layer obtained in (1) add 1 mL of sodium hydroxide TS, and boil: the gas evolved does not turn moistened red litmus paper to blue.

(4) Water-soluble organic substances—To 5 mL of the aqueous layer obtained in (1) add 0.25 mL of 0.002 mol/L potassium permanganate VS, and allow to stand for 5 minutes: the red color of the solution does not disappear.

(5) Petrolatum—Dissolve 1.0 g of the dried residue obtained in the Residue on evaporation in 10 mL of a mixture of tetrahydrofuran and isooctane (1:1), and use this solution as the sample solution. Add dissolve 20 mg of vaseline in 10 mL of a mixture of tetrahydrofuran and isooctane (1:1), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 25 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with isooctane to a distance of about 10 cm, and air-dry the plate. Spray evenly diluted sulfuric acid (1 in 2) on the plate, heat the plate at 80°C for 5 minutes, cool, and examine under ultraviolet light (main wavelength: 365 nm): no fluorescent spot is observed in the same level with the spot of standard solution. For this test use a thin-layer plate previously developed with isooctane to the upper end, dried in air, and heated at 110°C for 60 minutes.

Residue on evaporation Weigh accurately about 12.5 g of Hydrous Lanolin, dissolve in 50 mL of diethyl ether, put it in a separator, transfer the separated aqueous layer to another separator, add 10 mL of diethyl ether, shake, and combine the diethyl ether layer and diethyl ether in the first separator. Shake the diethyl ether layer with 3 g of anhydrous sodium sulfate, and filter through dry filter paper. Wash the separator and the filter paper with two 20-mL portions of diethyl ether, combine the washings with the filtrate, evaporate on a water bath until the odor of diethyl ether is no longer perceptible, and dry in a desiccator (in vacuum, silica gel) for 24 hours: the content is not less than 70% and not more than 75%.

Containers and storage Containers—Well-closed containers.

Storage—Not exceeding 30°C.

Purified Lanolin

Adeps Lanae Purificatus

精製ラノリン

Purified Lanolin is the purified product of the fat-like substance obtained from the wool of *Ovis aries* Linné (*Bovidae*).

Description Purified Lanolin is a light yellow to yellowish brown, viscous, ointment-like substance, and has a faint,

characteristic but not rancid odor.

It is very soluble in diethyl ether and in cyclohexane, freely soluble in tetrahydrofuran and in toluene, and very slightly soluble in ethanol (95). It is practically insoluble in water, but miscible without separation with about twice its mass of water, retaining ointment-like viscosity.

Melting point: 37 – 43°C

Identification Superimpose carefully 1 mL of a solution of Purified Lanolin in cyclohexane (1 in 50) on 2 mL of sulfuric acid: a red-brown color develops at the zone of contact, and the sulfuric acid layer shows a green fluorescence.

Acid value ⟨*1.13*⟩ Not more than 1.0.

Iodine value 18 – 36 Weigh accurately about 0.8 g of Purified Lanolin in a glass-stoppered 500-mL flask, add 20 mL of cyclohexane to dissolve, and add exactly 25 mL of Hanus' TS, and mix well. If a clear solution is not obtained, add more cyclohexane to make clear, and allow the mixture to stand for 1 hour between 20°C and 30°C in light-resistant, well-closed containers, with occasional shaking. Add 20 mL of a solution of potassium iodide (1 in 10) and 100 mL of water, shake, and titrate the liberated iodine with 0.1 mol/L sodium thiosulfate VS (indicator: 1 mL of starch TS). Perform a blank determination in the same manner.

$$\text{Iodine value} = (a - b) \times 1.269/M$$

M: amount (g) of Purified Lanolin taken
a: Volume (mL) of 0.1 mol/L sodium thiosulfate VS used in the blank determination
b: Volume (mL) of 0.1 mol/L sodium thiosulfate VS used in the titration of the sample

Purity **(1)** Acid or alkali—To 5 g of Purified Lanolin add 25 mL of water, boil for 10 minutes, and cool. Add water to restore the previous mass, and separate the aqueous layer: the aqueous layer is neutral.

(2) Chloride ⟨*1.03*⟩—To 2.0 g of Purified Lanolin add 40 mL of water, boil for 10 minutes, and cool. Add water to restore the previous mass, and filter. To 20 mL of the filtrate add 6 mL of dilute nitric acid and water to make 50 mL. Use this solution as the test solution, and perform the test. Prepare the control solution with 1.0 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.036%).

(3) Ammonia—To 10 mL of the aqueous layer obtained in (1) add 1 mL of sodium hydroxide TS, and boil: the gas evolved does not turn moistened red litmus paper to blue.

(4) Water-soluble organic substances—To 5 mL of the aqueous layer obtained in (1) add 0.25 mL of 0.002 mol/L potassium permanganate VS, and allow to stand for 5 minutes: the red color of the solution does not disappear.

(5) Petrolatum—Dissolve 1.0 g of Purified Lanolin in 10 mL of a mixture of tetrahydrofuran and isooctane (1:1), and use this solution as the sample solution. And dissolve 20 mg of vaseline in 10 mL of a mixture of tetrahydrofuran and isooctane (1:1), and use this solution as the standard solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 25 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with isooctane to a distance of about 10 cm, and air-dry the plate. Spray evenly diluted sulfuric acid (1 in 2) on the plate, heat the plate at 80°C for 5 minutes, cool, and examine under ultraviolet light (main wavelength: 365 nm): no fluorescent spot is observable same level of the spot of standard solution. Use a thin-layer plate previously developed with isooctane to the upper end, dried in air, and heated at 110°C for 60 minutes.

Loss on drying ⟨*2.41*⟩ Not more than 0.5% (1 g, 105°C, 2 hours).

Total ash ⟨*5.01*⟩ Not more than 0.1%.

Containers and storage Containers—Well-closed containers.

Storage—Not exceeding 30°C.

Lard

Adeps Suillus

豚脂

Lard is the fat obtained from *Sus scrofa* Linné var. *domesticus* Gray (*Suidae*).

Description Lard occurs as a white, soft, unctuous mass, and has a faint, characteristic odor and a bland taste.

It is freely soluble in diethyl ether and in petroleum ether, very slightly soluble in ethanol (95), and practically insoluble in water.

Melting point: 36 – 42°C

Congealing point of the fatty acids: 36 – 42°C

Acid value ⟨*1.13*⟩ Not more than 2.0.

Saponification value ⟨*1.13*⟩ 195 – 203

Iodine value ⟨*1.13*⟩ 46 – 70

Purity **(1)** Moisture and coloration—Melt 5 g of Lard by heating on a water bath: it forms a clear liquid, from which no water separates. Observe the liquid in a layer 10 mm thick: the liquid is colorless to slightly yellow.

(2) Alkalinity—To 2.0 g of Lard add 10 mL of water, melt by warming on a water bath, and shake vigorously. After cooling, add 1 drop of phenolphthalein TS to the separated aqueous layer: the layer is colorless.

(3) Chloride ⟨*1.03*⟩—To 1.5 g of Lard add 30 mL of ethanol (95), heat under a reflux condenser for 10 minutes, and filter after cooling. To 20 mL of the filtrate add 5 drops of a solution of silver nitrate in ethanol (95) (1 in 50): the opalescence of the mixture does not exceed that of the following control solution.

Control solution: To 1.0 mL of 0.01 mol/L hydrochloric acid VS add ethanol (95) to make 20 mL, and add 5 drops of a solution of silver nitrate in ethanol (95) (1 in 50).

(4) Beef tallow—Dissolve 5 g of Lard in 20 mL of diethyl ether, stopper lightly with absorbent cotton, and allow to stand at 20°C for 18 hours. Collect the separated crystals, moisten them with ethanol (95), and examine under a microscope of 200 magnifications: the crystals are in the form of rhomboidal plates grouped irregularly, and do not contain prisms or needles grouped in fan-shaped clusters.

Containers and storage Containers—Well-closed containers.

Storage—Not exceeding 30°C.

Leonurus Herb

Leonuri Herba

ヤクモソウ

Leonurus Herb is the aerial part of *Leonurus japonicus* Houttuyn or *Leonurus sibiricus* Linné (*Labiatae*), collected during the flowering season.

Description Stem, leaves, and flowers usually cross sectioned, stems square, 0.2 – 3 cm in diameter, yellow-green to green-brown in color, covered densely with white short hairs; the pith white, a great parts of central of cut surface. Light in texture. Leaves opposite, petiolated, 3-dissected to 3-incised, each lobe splits pinnately, and end lobes reveals linear-lanceolate, acute or acuminate, the upper surface light green, the lower surface bristle with white short hairs, grayish green. Flower, verticillate; sepal, tubular, and the upper end acerate with five lobes; light green to light green-brown in color, corolla labiate, light red-purple to light brown.

Odor, slightly; taste, slightly bitter, astringent.

Under a microscope <*5.01*>, a transverse section of stem reveals four ridges, a part of the ridge of *Leonurus sibiricus* Linné protruding knobby. Epidermis, observed non-glandular hairs from 1 to 3 cells, glandular hairs with head of 1 to 4 celled or glandular scale with 8 cells. Each ridge parts, beneath epidermis, collenchyma developed, development of xylem fibers remarkably. Cortex composed of several cellular layers parenchymatous cells. Collateral vascular bundle arranged in a circle. Phloem fibers observed at the outer portion of phloem. Parenchymatous cells of cortex and pith observe needle crystals or plate-like crystals of calcium oxalate.

Identification To 1 g of pulverized Leonurus Herb add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of water and methanol (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS followed by immediate spraying of sodium nitrite TS on the plate: a grayish brown spot appears at an *R*f value of about 0.5, which color fades soon and then disappears after air-drying the plate.

Loss on drying <*5.01*> Not more than 12.0%.

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 12.0%.

Containers and storage Containers—Well-closed containers.

Lilium Bulb

Lilii Bulbus

ビャクゴウ

Lilium Bulb is the scaly leaves of *Lilium lancifolium* Thunberg, *Lilium brownii* F.E.Brown var. *colchesteri* Wilson, *Lilium brownii* F.E.Brown or *Lilium pumilum* De Candolle (*Liliaceae*), usually with the application of steaming.

Description Lilium Bulb reveals oblong with narrowed apex, lanceolate, or narrowly triangular boat-shaped, translucent, 1.3 – 6 cm in length, 0.5 – 2.0 cm in diameter, externally milky white to light yellow-brown occasionally purplish in color, nearly smooth; central portion somewhat thickened, circumferential portion thin, slightly waved, occasionally rolled inside; usually several lines of vascular bundles longitudinally in parallel are seen through parenchyma; hard in texture, easy to break; fractured surface horny and flat.

Almost odorless; taste, slightly acid and bitter.

Under a microscope <*5.01*>, the surface reveals epidermal cells rectangular to almost square, stomata nearly circular, the cells adjacent to stomata mostly 4 in number. Under a microscope <*5.01*>, a transverse section reveals the outermost layer composed of epidermal cells covered with smooth cuticle; epidermis circular to quadrangular parenchymatous cells distributed evenly, palisade tissue not observed; in parenchyma of mesophyll collateral vascular bundles extended from adaxial side to abaxial side of scaly leaves are arranged almost in a transverse line; starch grains contained in parenchymatous cells, usually gelatinized.

Identification To 3 g of pulverized Lilium Bulb add 10 mL of 1-butanol, shake, add 10 mL of water, shake for 30 minutes, and centrifuge. Evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, shake gently, and use the supernatant liquid so obtained as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (12:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two spots appear at an *R*f value of about 0.3. When examine these spots under ultraviolet light (main wavelength: 365 nm) after spraying with sodium carbonate TS, they appear as blue-purple fluorescent spots.

Loss on drying <*5.01*> Not more than 16.0%.

Total ash <*5.01*> Not more than 4.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Lindera Root

Linderae Radix

ウヤク

Lindera Root is the root of *Lindera strychnifolia* Fernandez-Villar (*Lauraceae*).

Description Fusiform or rosary-like root, 10 – 15 cm in length, 10 – 25 mm in diameter; externally yellow-brown to brown, with a few scars of rootlets; a transversely cut surface reveals cortex brown, xylem light yellow-brown, concentric circles and radially arranged lines brown; dense and hard in texture.

Odor, camphor-like; taste, bitter.

Under a microscope <*5.01*>, a transverse section of the root with periderm reveals a cork layer several cells thick, partially consisting of cork stone cells; cortex parenchyma sometimes contains oil cells and fibers; in xylem, vessels-xylem fibers and rays are arranged alternately; parenchyatous cells of cortex and xylem contain sandy and columnar crystals of calcium oxalate, simple starch grains 1 – 15 μm in diameter, and 2- to 4- compound starch grains.

Identification To 3 g of pulverized Lindera Root add 40 mL of hexane, and heat under a reflux condenser for 30 minutes. After cooling, filter, to the residue add 10 mL of ammonia TS and 30 mL of a mixture of ethyl acetate and diethyl ether (1:1), shake vigorously for 20 minutes, and centrifuge. Separate the upper layer, add 10 g of anhydrous sodium sulfate, shake, and filter. Evaporate the solvent of the filtrate, dissolve the residue with 0.5 mL of ethanol (99.5), and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, methanol and ammonia water (28) (10:2:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate: a yellow-brown spot appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Lindera Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Lindera Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 2.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 6.0%.

Containers and storage Containers—Well-closed containers.

Lithospermum Root

Lithospermi Radix

シコン

Lithospermum Root is the root of *Lithospermum erythrorhizon* Siebold et Zuccarini (*Boraginaceae*).

Description Rather slender conical root, often branched, 6 – 10 cm in length, 0.5 – 1.5 cm in diameter; externally dark purple, coarse in texture, thin and easily peeled; mostly with twisted and deep longitudinal furrows, which sometimes reach to xylem; sometimes remains of stem at the crown; easily broken; fractured surface granular and with many clefts. Under a magnifying glass, a transverse section reveals a dark purple color at the outer portion of cortex, and light brown inner portion making irregular wave; xylem yellowish in color; the center of the crown is often cracked, and the surrounding part red-purple.

Odor, slight; taste, slightly sweet.

Identification **(1)** Heat 0.5 g of pulverized Lithospermum Root in a test tube: red vapor evolves, which condenses on the wall of the upper part of the tube into red-brown oil drops.

(2) Shake 0.5 g of pieces or powder of Lithospermum Root with 1 mL of ethanol (95), and to the red solution thereby obtained add 1 drop of sodium hydroxide TS: the red color changes to blue-purple. To this solution add 1 to 2 drops of dilute hydrochloric acid: the color turns red again.

(3) To 0.5 g of pulverized Lithospermum Root add 5 mL of ethanol (95), shake for 30 minutes, filter, and evaporate the solvent of the filtrate at a temperature not higher than 40°C under low pressure (in vacuo). Add 1 mL of ethanol (95) to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and ethanol (95) (3:1) to a distance of about 7 cm, and air-dry the plate: a red-purple spot appears at an *R*f value of about 0.75.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 2.0 g of pulverized Lithospermum Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Lithospermum Root according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 11.0%.

Acid-insoluble ash <*5.01*> Not more than 3.5%.

Containers and storage Containers—Well-closed containers.

Longan Aril

Longan Arillus

リュウガンニク

Longan Aril is the aril of *Euphoria longana* Lamarck (*Sapindaceae*).

Description Depressed ellipsoidal aril, 1 – 2 cm in length, about 1 cm in width; yellow-red-brown to black-brown; soft in texture and mucous; when immersed in water, bell-shaped, with the tip split in several parts.

Odor, characteristic; taste, sweet.

Under a microscope ⟨*5.01*⟩, a transverse section reveals the outmost layer composed of an epidermis, beneath this observed parenchyma consisting of depressed parenchyma cells; the innermost layer composed of slightly thick-walled epidermis; parenchyma contains red-brown to brown contents as well as solitary crystals, amorphous crystals and sand crystals of calcium oxalate.

Identification To 1 g of coarse cuttings of Longan Aril, add 10 mL of water, shake thoroughly, and filter. To 3 mL of the filtrate, add 3 mL of Fehling solution, and heat on a water bath: a red precipitate is produced.

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: Not less than 75.0%.

Containers and storage Containers—Well-closed containers.

Longgu

Fossilia Ossis Mastodi

リュウコツ

Longgu is the ossified bone of large mammal, and is mainly composed of calcium carbonate.

For Longgu used only for extracts, infusions and decoctions, the label states the restricted utilization forms.

Description Irregular masses or fragments, occasionally cylindrical masses; externally light grayish white, sometimes with grayish black or yellow-brown spots here and there; the outer part consists of a layer 2 – 10 mm in thickness, and is minute in texture, surrounding the light brown, porous portion; heavy and hard, but somewhat fragile in texture; when crushed, it changes into pieces and powder.

Odorless, tasteless, and strongly adhesive to the tongue on licking.

Identification **(1)** Dissolve 0.5 g of pulverized Longgu in 10 mL of dilute hydrochloric acid: it evolves a gas, and forms a slightly brownish and turbid solution. Pass the gas evolved through calcium hydroxide TS: a white precipitate is produced.

(2) The turbid solution obtained in (1) has a characteristic odor. Filter this solution and neutralize filtrate with ammonia TS: this solution responds to Qualitative Tests ⟨*1.09*⟩ (1), (2) and (3) for calcium salt.

(3) Dissolve 0.1 g of pulverized Longgu in 5 mL of nitric acid by warming, and add hexaammonium heptamolybdate TS: a yellow precipitate is produced.

Purity **(1)** Heavy metals ⟨*1.07*⟩—To 2.0 g of pulverized Longgu, add 5 mL of water, shake, add gradually 6 mL of hydrochloric acid, evaporate on a water bath to dryness, dissolve the residue in 50 mL of water, and filter. To 25 mL of the filtrate, add 2 mL of dilute acetic acid, 1 drop of ammonia TS and water to make 50 mL. Perform the test with this solution as the test solution. Prepare the control solution as follows: evaporate 3 mL of hydrochloric acid on a water bath to dryness, add 2 mL of dilute acetic acid, 2.0 mL of Standard Lead Solution and water to make 50 mL, and use this solution as the control solution (not more than 20 ppm).

When being shown as extracts, infusions and decoctions on the label, the procedure and the limit are as follows.

To 20.0 g of pulverized Longgu, add 80 mL of water, shake occasionally in a water bath, heat to make about 40 mL, allow to cool, and filter. Proceed with this solution according to Method 3, and perform the test. To the control solution, add 1.0 mL of Standard Lead Solution (not more than 0.5 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.20 g of pulverized Longgu according to Method 2, and perform the test (not more than 10 ppm).

When being shown the restricted utilization forms as ''extracts, infusions and decoctions only'', the procedure and the limit are as follows.

Put 4.0 g of pulverized Longgu in a centrifuge tube, add 30 mL of water, and heat in a water bath with occasional shaking to make about 15 mL. After cooling, centrifuge, and perform the test using the supernatant liquid as the test solution (not more than 0.5 ppm).

Containers and storage Containers—Well-closed containers.

Powdered Longgu

Fossilia Ossis Mastodi Pulveratum

リュウコツ末

Powdered Longgu is the powder of Longgu.

Description Powdered Longgu occurs as a light grayish white to light grayish brown. It is odorless and tasteless.

Identification **(1)** Dissolve 0.5 g of Powdered Longgu in 10 mL of dilute hydrochloric acid: it evolves a gas, and forms a slightly brownish and turbid solution. Pass the gas evolved through calcium hydroxide TS: a white precipitate is produced.

(2) The turbid solution obtained in (1) has a characteristic odor. Filter this solution, and neutralize filtrate with ammonia TS: the solution responds to Qualitative test ⟨*1.09*⟩ (1), (2) and (3) for calcium salt.

(3) Dissolve 0.1 g of Powdered Longgu in 5 mL of nitric acid by warming, and add hexaammonium heptamolybdate TS: a yellow precipitate is produced.

Purity **(1)** Heavy metals ⟨*1.07*⟩—To 2.0 g of Powdered Longgu add 5 mL of water, shake to mix, add gradually 6 mL of hydrochloric acid, and evaporate on a water bath to dryness. Dissolve the residue in 50 mL of water, and filter. To 25 mL of the filtrate add 2 mL of dilute acetic acid, 1 drop of ammonia TS and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the con-

trol solution as follows: evaporate 3 mL of hydrochloric acid on a water bath to dryness, add 2 mL of dilute acetic acid and 2.0 mL of Standard Lead Solution, and add water to make 50 mL (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.20 g of Powdered Longgu according to Method 2, and perform the test (not more than 10 ppm).

Containers and storage Containers—Well-closed containers.

Lonicera Leaf and Stem

Lonicerae Folium Cum Caulis

ニンドウ

Lonicera Leaf and Stem is the leaves and stems of *Lonicera japonica* Thunberg (*Caprifoliaceae*).

Description Leaves and opposite leaves on short stem; leaf, ovate and entire, with short petiole, 3 – 7 cm in length, 1 – 3 cm in width; upper surface green-brown, lower surface light grayish green; under a magnifying glass, both surfaces pubescent. Stem, 1 – 4 mm in diameter; externally grayish yellow-brown to purplish brown, a transversely cut surface of stem, round and hollow.

Almost odorless; taste, slightly astringent, followed by a litter bitterness.

Under a microscope <*5.01*>, a transverse section of leaf reveals the outermost layer of upper and lower surfaces to be composed of a single-layered epidermis, uni-cellular non-glandular hairs and multi-cellular glandular hairs on epidermis; in midvein, several-cellular-layered collenchyma present beneath the epidermis and vascular bundles in the center; in mesophyll, palisade layer adjacent to upper epidermis, spongy tissue adjacent to lower epidermis; glandular hairs contain brown secretion, parenchymatous cells contain aggregate crystals of calcium oxalate, and occasionally starch grains.

Identification To 1 g of pulverized Lonicera Leaf and Stem add 5 mL of methanol, shake for 5 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of chlorogenic acid for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution (1). Separately, dissolve 1 mg of loganin for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (6:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (1). Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution (2).

Purity Stem—Lonicera Leaf and Stem does not contains the stems larger than 5 mm in diameter.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 9.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 12.0%.

Containers and storage Containers—Well-closed containers.

Loquat Leaf

Eriobotryae Folium

ビワヨウ

Loquat Leaf is the leaf of *Eriobotrya japonica* Lindley (*Rosaceae*).

Description Loquat Leaf is an oblong to wide lanceolate leaf, 12 – 30 cm in length, 4 – 9 cm in width; pointed at the apex and wedge-shaped at the base; roughly serrate leaf with short petiole; occasionally being cut into strips 5 – 10 mm in shorter diameter and several cm in longer diameter; upper surface green to green-brown in color, lower surface light green-brown with light brown woolly hairs; vein, light yellow-brown in color, raised out on the lower surface of the leaf.

Odor, slight; practically tasteless.

Under a microscope <*5.01*>, a transverse section of Loquat Leaf reveals thick cuticle on both surfaces; palisade tissue, mostly 4 to 5 cellular layers with several large cells without chloroplast; at main vein, ring of collateral bundle partly cut by intruding fundamental tissue at xylem side, and group of fiber attaching to phloem; solitary and clustered crystals of calcium oxalate in mesophyll; woolly hair, unicellular and curved, about 25 μm in thickness, and up to 1.5 mm in length.

Identification To 0.3 g of pulverized Loquat Leaf add 10 mL of methanol, warm on a water bath for 5 minutes with occasional shaking, cool, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of water and acetonitrile (3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 10 minutes: a red-purple principal spot appears at an *R*f value of about 0.5.

Purity Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 10.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 16.0%.

Containers and storage Containers—Well-closed containers.

Lycium Bark

Lycii Cortex

ジコッピ

Lycium Bark is the root bark of *Lycium chinense* Miller or *Lycium barbarum* Linné (*Solanaceae*).

Description Tubular to semitubular bark, 1 – 6 mm in thickness; externally light brown to light yellow-brown, periderm peeled easily as scale; internally grayish brown, longitudinally striate; brittle in texture; fractured surface, grayish white, not fibrous.

Odor, weak and characteristic; taste, slightly sweet at first.

Under a microscope ⟨*5.01*⟩, a transverse section reveals periderm composed of a cork layer of several cellular layers of thin-walled cork cells; in cortex parenchyma cells containing sandy crystals of calcium oxalate sparsely distributed, occasionally a few fibers observed; parenchyma cells contain starch grains, 1 – 10 μm in diameter; stone cells very rare.

Identification To 1.0 g of pulverized Lycium Bark add 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, ammonium acetate solution (1 in 20) and acetic acid (100) (2:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, heat the plate at 105°C for 2 minutes, then spray evenly sodium nitrite TS, and allow to stand for 5 minutes: a dark brown principal spot appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 2.0 g of pulverized Lycium Bark according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Lycium Bark according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 11.5% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 20.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 3.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 10.0%.

Containers and storage Containers—Well-closed containers.

Lycium Fruit

Lycii Fructus

クコシ

Lycium Fruit is the fruit of *Lycium chinense* Miller or *Lycium barbarum* Linné (*Solanaceae*).

Description Fusiform fruit with acute apex, 6 – 20 mm in length, 3 – 8 mm in diameter, pericarp red to dark red, externally roughly wrinkled; under a magnifying glass, a transverse section of fruit reveals two locules containing numerous seeds; seed light brown to light yellow-brown, about 2 mm in a diameter, compressed reniform.

Odor, characteristic; taste, sweet, occasionally slightly bitter.

Identification To 1.0 g of pulverized Lycium Fruit add 5 mL of ethyl acetate, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and ethyl acetate (10:1) to a distance of about 7 cm, and air-dry the plate: a yellow principal spot appears at an *R*f value of about 0.6.

Purity Foreign matter ⟨*5.01*⟩—It contains not more than 2.0% of foreign matter such as peduncle or others.

Total ash ⟨*5.01*⟩ Not more than 8.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 35.0%.

Containers and storage Containers—Well-closed containers.

Magnolia Bark

Magnoliae Cortex

コウボク

Magnolia Bark is the bark of the trunk of *Magnolia obovata* Thunberg (*Magnolia hypoleuca* Siebold et Zuccarini), *Magnolia officinalis* Rehder et Wilson or *Magnolia officinalis* Rehder et Wilson var. *biloba* Rehder et Wilson (*Magnoliaceae*).

It contains not less than 0.8% of magnolol.

Description Plate-like or semi-tubular bark, 2 – 7 mm in thickness; externally grayish white to grayish brown, and rough, sometimes cork layer removed, and externally red-brown; internally light brown to dark purple-brown; cut surface extremely fibrous, and light red-brown to purple-brown.

Odor, slight; taste, bitter.

Under a microscope ⟨*5.01*⟩, a transverse section reveals a thick cork layer or several thin cork layers, and internally adjoining the circular tissue of stone cells of approximately equal in diameter; primary cortex thin; fiber groups scattered in the pericycle; groups of phloem fibers lined alternately with the other tissue of phloem between medullary rays in the secondary cortex, and then these tissues show a latticework; oil cells scattered in the primary and secondary cortex, but sometimes observed in the narrow medullary rays.

Identification To 1.0 g of pulverized Magnolia Bark add 10 mL of methanol, stir for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS on the plate: a yellow

spot appears at an *R*f value of about 0.3.

Total ash <*5.01*> Not more than 6.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 11.0%.

Assay Weigh accurately about 0.5 g of pulverized Magnolia Bark, add 40 mL of diluted methanol (7 in 10), heat under a reflux condenser for 20 minutes, cool, and filter. Repeat the above procedure with the residue, using 40 mL of diluted methanol (7 in 10). Combine the whole filtrates, add diluted methanol (7 in 10) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of magnolol for assay, dissolve in diluted methanol (7 in 10) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of magnolol in each solution.

$$\text{Amount (mg) of magnolol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of magnolol for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 289 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (50:50:1).

Flow rate: Adjust so that the retention time of magnolol is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg each of magnolol for assay and honokiol in diluted methanol (7 in 10) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, honokiol and magnolol are eluted in this order with the resolution between these peaks being not less than 5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of magnolol is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Magnolia Bark

Magnoliae Cortex Pulveratus

コウボク末

Powdered Magnolia Bark is the powder of Magnolia Bark.

It contains not less than 0.8% of magnolol.

Description Powdered Magnolia Bark occurs as a yellow-brown powder, and has a slight odor and a bitter taste.

Under a microscope <*5.01*>, Powdered Magnolia Bark reveals starch grains and parenchyma cells containing them; stone cells of various sizes or its groups; fibers 12 to 25 μm in diameter; yellow-red-brown cork tissue; oil cells containing a yellow-brown to red-brown substance. Simple starch grains about 10 μm in diameter and 2- to 4-compound starch grains.

Identification To 1.0 g of Powdered Magnolia Bark add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS on the plate: a yellow spot appears at an *R*f value of about 0.3.

Total ash <*5.01*> Not more than 6.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 11.0%.

Assay Weigh accurately about 0.5 g of Powdered Magnolia Bark, add 40 mL of diluted methanol (7 in 10), heat under a reflux condenser for 20 minutes, cool, and filter. Repeat the above procedure with the residue, using 40 mL of diluted methanol (7 in 10). Combine the whole filtrates, add diluted methanol (7 in 10) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of magnolol for assay, dissolve in diluted methanol (7 in 10) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of magnolol in each solution.

$$\text{Amount (mg) of magnolol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of magnolol for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 289 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (50:50:1).

Flow rate: Adjust so that the retention time of magnolol is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg each of magnolol for assay and honokiol in diluted methanol (7 in 10) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, honokiol and magnolol are eluted in this order with the resolution between these peaks being not less than 5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of magnolol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Magnolia Flower

Magnoliae Flos

シンイ

Magnolia Flower is the flower bud of *Magnolia biondii* Pampanini, *Magnolia heptapeta* Dandy (*Magnolia denudata* Desrousseaux), *Magnolia sprengeri* Pampanini, *Magnolia salicifolia* Maximowicz, or *Magnolia kobus* De Candolle (*Magnoliaceae*).

Description Magnolia Flower is a fusiform flower bud, 15 – 45 mm in length, 6 – 20 mm in diameter at central part; often having ligneous peduncles on base; usually 3 bracts, externally with sparse hairs, brown to dark brown, or with dense hairs, grayish white to light yellow-brown, and the inner surface of 3 bracts smooth and dark brown in color; interior perianth of 9 pieces or 12 pieces, same size or outer three pieces are smaller; 50 – 100 stamens and numerous pistils. Brittle in texture.

Odor, characteristic; taste, acrid and slightly bitter.

Identification To 1 g of pulverized Magnolia Flower add 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, acetone, water and formic acid (5:3:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate: a yellow-red spot appears at an *R*f value of about 0.3.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Extract content ⟨*5.01*⟩ Dilute ethanol-extract: not less than 13.0%.

Essential oil content ⟨*5.01*⟩ Perform the test with 50.0 g of pulverized Magnolia Flower: the volume of essential oil is not less than 0.5 mL.

Containers and storage Containers—Well-closed containers.

Mallotus Bark

Malloti Cortex

アカメガシワ

Mallotus Bark is the bark of *Mallotus japonica* Müeller Argoviensis (*Euphorbiaceae*).

Description Mallotus Bark is flat or semitubular pieces of bark, 1 – 3 mm in thickness; externally greenish gray to brownish gray brow in color, with a vertically striped shape gathering numerous lenticels; internal surface light yellow-brown to grayish brown in color, and smooth with numerous fine striped lines; easy to break; slightly fibrous at fractured surface.

Mallotus Bark has a slight odor, a bitter taste and slightly astringent.

Identification To 0.5 g pulverized Mallotus Bark add 10 mL of methanol, warm on a water bath for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of bergenin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (100:17:13) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying ⟨*5.01*⟩ Not more than 13.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 12.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.5%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 11.0%.

Containers and storage Containers—Well-closed containers.

Malt

Fructus Hordei Germinatus

バクガ

Malt is the dried ripe caryopsis of *Hordeum vulgare* Linné (*Gramineae*), after being germinated.

Description Oval caryopsis, 10 mm in length, 3 – 4 mm in width, furrowed on one surface; externally light yellow, sometimes with plumule at one end, with hairs and sometimes with roots at the other end; cut surface of caryopsis white and powdery; easily broken and light in texture.

Odor, slight; taste, slightly sweet.

Under a microscope ⟨*5.01*⟩, a transverse section of the caryopsis reveals glume, pericarp, seed coat and endosperm in this order from the outside; 2 – 4 cellular layered aleurone layers on the circumference of endosperm; endosperm filled with starch grains; starch grains as spheroidal or ellipsoidal, large grains about 20 μm and small grains about 2 μm in diameter mixed together.

Identification To 3.0 g of pulverized Malt add 5 mL of methanol, shake for 15 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Liquid Chromatography ⟨*2.03*⟩. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of methanol, water and acetic acid (100) (8:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly a solution of 0.1 g of 2,3-indolinedione in 50 mL of acetone on the plate, and heat the plate at 105°C for 5 minutes: a blue-purple spot appears at an *R*f value of about 0.4.

Loss on drying ⟨*5.01*⟩ Not more than 11.0%.

Total ash ⟨*5.01*⟩ Not more than 2.6%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.8%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: Not

less than 15.0%.

Containers and storage Containers—Well-closed containers.

Maoto Extract

麻黄湯エキス

Maoto Extract contains not less than 15 mg and not more than 45 mg of total alkaloids (ephedrine and pseudoephedrine), not less than 48 mg and not more than 192 mg of amygdalin, and not less than 11 mg and not more than 33 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)
Ephedra Herb	5 g
Apricot Kernel	5 g
Cinnamon Bark	4 g
Glycyrrhiza	1.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1), using the crude drugs shown above, or prepare a dry extract by adding Light Anhydrous Silicic Acid to an extractive prepared as directed under Extracts, according to the prescription 1), using the crude drugs shown above.

Description Maoto Extract occurs as a light brown powder or black-brown viscous extract, having a slightly order, and a sweet and bitter, then a slightly astringent taste.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (4:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot is observed at an *R*f value of about 0.5 (Ephedra Herb).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate and water (4:4:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the green-brown spot from the standard solution (Apricot Kernel).

(3) Perform the test according to the following (i) or (ii) (Cinnamon Bark).

(i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect the apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

(ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 13.0%, calculated on the dried basis. However, for the dry extract prepared by adding Light Anhydrous Silicic Acid, between 10.0% and 22.0%.

Assay (1) Total alkaloids (ephedrine and pseudoephedrine)—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. In addition, repeat twice in the same manner for the aqueous layer using 1.0 mL of ammonia TS and 20 mL of diethyl ether. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of ephedrine hydrochloride for assay of crude drugs, previously dried at 105°C for 3 hours, dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine obtained from the sample solution, and peak area, A_S, of ephedrine from the standard solution.

Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)
$= M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drugs taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, then add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of ephedrine is about 27 minutes).

System suitability—

System performance: Dissolve 1 mg each of ephedrine hydrochloride for assay of crude drugs and pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

(2) Amygdalin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, flow through in a column packed with 2 g of polyamide for column chromatography, then elute with water to make exactly 20 mL, and use this effluent as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for 24 hours or more, and dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

Amount (mg) of amygdalin $= M_S \times A_T/A_S \times 4$

M_S: Amount (mg) of amygdalin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about

40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Mentha Herb

Menthae Herba

ハッカ

Mentha Herb is the terrestrial part of *Mentha arvensis* Linné var. *piperascens* Malinvaud (*Labiatae*).

Description Stem with opposite leaves; stem, square, light brown to red-purple in color, and with fine hairs; when smoothed by immersing in water, leaf, ovate to oblong, with acute apex and base, 2 – 8 cm in length, 1 – 2.5 cm in width, margin irregularly serrated; the upper surface, light brown-yellow to light green-yellow, and the lower surface, light green to light green-yellow in color; petiole 0.3 – 1 cm in length. Under a magnifying glass, leaf reveals hairs, glandular hairs and scales.

It has a characteristic aroma and gives a cool feeling on keeping in the mouth.

Identification To 1.0 g of pulverized Mentha Herb add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of menthol in 1 mL of diethyl ether, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity Foreign matter <*5.01*>—The amount of roots and other foreign matter contained in Mentha Herb does not exceed 2.0%.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 12.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Essential oil content <*5.01*> Perform the test with 50.0 g of pulverized Mentha Herb after adding 1 mL of silicone resin to the sample in the flask: the volume of essential oil is not less than 0.4 mL.

Containers and storage Containers—Well-closed containers.

Mentha Oil

Oleum Menthae Japonicae

ハッカ油

Mentha Oil is the essential oil which is distilled with steam from the terrestrial parts of *Mentha arvensis* Linné var. *piperascens* Malinvaud (*Labiatae*), and from which solids are removed after cooling.

It contains not less than 30.0% of menthol.

Description Mentha Oil is a colorless or pale yellow, clear

liquid. It has a characteristic, pleasant aroma and has a pungent taste, followed by a cool aftertaste.

It is miscible with ethanol (95), with ethanol (99.5), with warm ethanol (95), and with diethyl ether.

It is practically insoluble in water.

Refractive index <*2.45*> n_D^{20}: 1.455 – 1.467

Optical rotation <*2.49*> α_D^{20}: −17.0 – −36.0° (100 mm).

Specific gravity <*1.13*> d_{25}^{25}: 0.885 – 0.910

Acid value <*1.13*> Not more than 1.0.

Purity **(1)** Clarity and color of solution—To 1.0 mL of Mentha Oil add 3.5 mL of diluted ethanol (7 in 10), and shake: Mentha Oil dissolves clearly. To the solution add 10 mL of ethanol (95): the solution is clear or has no more turbidity, if any, than the following control solution.

Control solution: To 0.70 mL of 0.01 mol/L hydrochloric acid VS add 6 mL of dilute nitric acid and water to make 50 mL, add 1 mL of silver nitrate TS, and allow to stand for 5 minutes.

(2) Heavy metals <*1.07*>—Proceed with 1.0 mL of Mentha Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

Assay Weigh accurately about 5 g of Mentha Oil, and dissolve in ethanol (95) to make exactly 20 mL. Pipet 10 mL of this solution, add exactly 10 mL of the internal standard solution, and use this solution as the sample solution. Separately, weigh accurately about 10 g of *l*-menthol for assay, and dissolve in ethanol (95) to make exactly 100 mL. Pipet 10 mL of this solution, add exactly 10 mL of the internal standard solution, and use this solution as the standard solution. Perform the test with 1 μL each of the sample solution and standard solution as directed under Gas Chromatography <*2.02*> according to the following conditions. Calculate the ratios, Q_T and Q_S, of the peak area of menthol to that of the internal standard.

$$\text{Amount (mg) of menthol} = M_S \times Q_T/Q_S \times 1/5$$

M_S: Amount (mg) of *l*-menthol for assay taken

Internal standard solution—A solution of *n*-ethyl caprylate in ethanol (95) (1 in 25).

Operating conditions—

Detector: A hydrogen flame-ionization detector.

Column: A glass column about 3 mm in inside diameter and about 2 m in length, packed with 25% of polyethylene glycol 6000 for gas chromatography supported on acid-washed 180 – 250 μm siliceous earth for gas chromatography.

Column temperature: A constant temperature of about 150°C.

Carrier gas: Nitrogen.

Flow rate: Adjust so that the retention time of the internal standard is about 10 minutes.

System suitability—

System performance: When the procedure is run with 1 μL of the standard solution under the above operating conditions, the internal standard and *l*-menthol are eluted in this order with the resolution between these peaks being not less than 5.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Mentha Water

ハッカ水

Method of preparation

Mentha Oil	2 mL
Purified Water or Purified Water in Containers	a sufficient quantity
	To make 1000 mL

Prepare as directed under Aromatic Waters, with the above ingredients.

Description Mentha Water is a clear, colorless liquid, having the odor of mentha oil.

Containers and storage Containers—Tight containers.

Moutan Bark

Moutan Cortex

ボタンピ

Moutan Bark is the root bark of *Paeonia suffruticosa* Andrews (*Paeonia moutan* Sims) (*Paeoniaceae*).

It contains not less than 0.9% of paeonol.

Description Tubular to semi-tubular bark, about 0.5 cm in thickness, 5 – 8 cm in length, 0.8 – 1.5 cm in diameter; externally dark brown to purple-brown, with small and transversely elongated ellipsoidal scars of lateral roots, and with longitudinal wrinkles; internally, light grayish brown to purplish brown and smooth; fractured surface coarse; white crystals often attached on the internal and fractured surfaces.

Odor, characteristic; taste, slightly pungent and bitter.

Identification To 2.0 g of pulverized Moutan Bark add 10 mL of hexane, shake for 3 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of hexane, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Moutan Bark according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Moutan Bark according to Method 4, and perform the test (not more than 5 ppm).

(3) Xylem—When perform the test of foreign matter <*5.01*>, the amount of the xylem contained in Moutan Bark is not more than 5.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than xylem contained in Moutan Bark is not exceed 1.0%.

(5) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Assay Weigh accurately about 0.3 g of pulverized Moutan Bark, add 40 mL of methanol, heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure with the residue, using 40 mL of methanol. Combine all the filtrates, add methanol to make exactly 100 mL, then pipet 10 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of paeonol for assay, dissolve in methanol to make exactly 100 mL, then pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeonol in each solution.

$$\text{Amount (mg) of paeonol} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of paeonol for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 274 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile, and acetic acid (100) (65:35:2).

Flow rate: Adjust so that the retention time of paeonol is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg of paeonol for assay and 5 mg of butyl parahydroxybenzoate for resolution check in methanol to make 25 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, paeonol and butyl parahydroxybenzoate are eluted in this order with the resolution between these peaks being not less than 2.0.

System repeatability: When the test is repeated 6 times with the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeonol is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Moutan Bark

Moutan Cortex Pulveratus

ボタンピ末

Powdered Moutan Bark is the powder of Moutan Bark.

It contains not less than 0.6% of paeonol.

Description Powdered Moutan Bark occurs as a light grayish yellow-brown powder. It has a characteristic odor and a slight, pungent and bitter taste.

Under a microscope <*5.01*>, Powdered Moutan Bark reveals starch grains and fragments of parenchyma containing them; fragments of cork tissue containing tannin; fragments of somewhat thick-walled collenchyma, medullary rays, and phloem parenchyma; rosette aggregates of calcium oxalate and also fragments of parenchyma cells containing them. Starch grains are simple or 2- to 10-compound grains, 10 – 25 μm in diameter; rosette aggregates are 20 – 30 μm in diameter.

Identification **(1)** To 2.0 g of Powdered Moutan Bark add 10 mL of hexane, shake for 3 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of paeonol for thin-layer chromatography in 1 mL of hexane, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

(2) Evaporate the solvent to dryness 1 mL of the sample solution obtained in (1), dissolve the residue in 50 mL of ethanol (95), and determine the absorption spectrum of this solution as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits maxima at around 228 nm, 274 nm and 313 nm.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Moutan Bark according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Moutan Bark according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, usually vessels and other sclerenchymatous cells are not observable.

(4) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Assay Weigh accurately about 0.5 g of Powdered Moutan Bark, add 40 mL of methanol, heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure with the residue, using 40 mL of methanol. Combine all the filtrates, add methanol to make exactly 100 mL, then pipet 10 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of paeonol for assay, dissolve in methanol to make exactly 100 mL, then pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeonol in each solution.

$$\text{Amount (mg) of paeonol} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of paeonol for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 274 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile, and acetic acid (100) (65:35:2).

Flow rate: Adjust so that the retention time of paeonol is about 14 minutes.

System suitability—

System performance: Dissolve 1 mg of paeonol for assay and 5 mg of butyl parahydroxybenzoate for resolution check in methanol to make 25 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, paeonol and butyl parahydroxybenzoate are eluted in this order with the resolution between these peaks being not less than 2.0.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeonol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Mukoi-Daikenchuto Extract

無コウイ大建中湯エキス

Mukoi-Daikenchuto Extract contains not less than 1.8 mg of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), and not less than 1.4 mg and not more than 4.2 mg of [6]-shogaol, per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)
Japanese Zanthoxylum Peel	2 g
Ginseng	3 g
Processed Ginger	5 g

Prepare a dry extract as directed under Extracts, according to the prescription 1), using crude drugs shown above.

Description Mukoi-Daikenchuto Extract is a light brown powder. It has a slight odor, and has a pungent taste.

Identification (1) Shake 2.0 g of Mukoi-Daikenchuto Extract with 10 mL of water, add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, shake 2.0 g of pulverized japanese zanthoxylum peel with 10 mL of water, add 5 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane, methanol and acetic acid (100) (20:20:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot (*R*f value: about 0.3) from the standard solution (Japanese Zanthoxylum Peel).

(2) Shake 2.0 g of Mukoi-Daikenchuto Extract with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(3) Shake 2.0 g of Mukoi-Daikenchuto Extract with 10 mL of water, add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Processed ginger).

Purity (1) Heavy metals <*1.07*>—Prepare the test solution with 2.0 g of Mukoi-Daikenchuto Extract as directed under Extracts (4), and perform the test (not more than 15 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 2.0 g of Mukoi-Daikenchuto Extract according to Method 3, and perform the test (not more than 1 ppm).

Loss on drying <*2.41*> Not more than 5.9% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%.

Assay (1) Ginsenoside Rb_1—Weigh accurately about 2 g of Mukoi-Daikenchuto Extract, add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 15 mL of diluted methanol (3 in 5), and repeat the same procedure. Combine the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 5 mL of this solution to a column [10 mm in inside diameter, packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), and washed just before using with methanol and then diluted methanol (3 in 10)], and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and

standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S \times 1/5$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl group bound silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (400:100:1).

Flow rate: 1.0 mL per minute (the retention time of ginsenoside Rb_1 is about 16 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

(2) [6]-Shogaol—Weigh accurately about 0.5 g of Mukoi-Daikenchuto Extract, add exactly 50 mL of diluted methanol (3 in 4), shake for 15 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of [6]-shogaol for assay, dissolve in diluted methanol (3 in 4) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (3 in 4) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-shogaol in each solution.

$$\text{Amount (mg) of [6]-shogaol} = M_S \times A_T/A_S \times 1/10$$

M_S: Amount (mg) of [6]-shogaol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 225 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 0.1 g of oxalic acid dihydrate in 600 mL of water, and add 400 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of [6]-shogaol is about 30 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-shogaol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-shogaol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Mulberry Bark

Mori Cortex

ソウハクヒ

Mulberry Bark is the root bark of *Morus alba* Linné (*Moraceae*).

Description Tubular, semi-tubular or cord-like bark, 1 – 6 mm thick, often in fine lateral cuttings; externally, white to yellow-brown; in the case of the bark with periderm, its periderm is yellow-brown in color, easy to peel, with numerous longitudinal, fine wrinkles and numerous red-purple lenticels laterally elongated; inner surface, dark yellow-brown in color and flat; cross section, white to light brown in color, and fibrous.

Odor, slight; taste, slight.

Under a microscope <5.01>, a transverse section of bark with periderm reveals 5 to 12 cellular layers of cork cells in the outer portion; phloem fibers or their bundles scattered in the cortex, arranged alternately and stepwise with phloem parenchyma; lactiferous tubes; solitary crystals of calcium oxalate; starch grains as spheroidal or ellipsoidal, simple or compound grains, simple grain 1 – 7 μm in diameter.

Identification Heat 1 g of pulverized Mulberry Bark with 20 mL of hexane under a reflux condenser for 15 minutes, and filter. Evaporate the solvent of the filtrate under low pressure (in vacuo), dissolve the residue in 10 mL of acetic anhydride, place 0.5 mL of the solution in a test tube, and add carefully 0.5 mL of sulfuric acid to make two layers: a red-brown color develops at the zone of contact.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of pulverized Mulberry Bark according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.40 g of pulverized Mulberry Bark according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter <5.01>—The amount of the root xylem and other foreign matter is not more than 1.0%.

Total ash <5.01> Not more than 11.0%.

Acid-insoluble ash <5.01> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Nelumbo Seed

Nelumbinis Semen

レンニク

Nelumbo Seed is the seed of *Nelumbo nucifera* Gaertner (*Nymphaeaceae*), usually with the endocarp, sometime being removed the embryo.

Description Ovoid to ellipsoidal seed, at the base a papillate protuberance surrounded with shallow depression, 1.0 – 1.7 cm in length, 0.5 – 1.2 cm in width; externally light red-brown to light yellow-brown; projection part dark reddish brown; endocarp not lustrous and hardly peeled off; endosperm yellow-white, a green embryo in the center.

Almost odorless; taste, slightly sweet and oily, embryo is extremely bitter.

Under a microscope ⟨*5.01*⟩, a transverse section of the seed at central portion reveals endocarp composed of parenchyma or endocarp occasionally left out; seed coat composed of epidermis and parenchyma of compressed cells; vascular bundles scattered in parenchyma; endosperm composed of epidermis and parenchyma; aggregate crystals of calcium oxalate and tannin-like substances contained in endocarp remained; parenchymatous cells of seed coat contain tannin-like substances; parenchyma of endosperm contain starch grains.

Identification To 0.5 g of pulverized Nelumbo Seed add 5 mL of water, shake for 5 minutes, and centrifuge. To 0.5 mL of the supernatant liquid add 1 drop of a solution of 1-naphthol in ethanol (99.5) (1 in 5), mix, then add gently 1 mL of sulfuric acid: the solution shows a purple color.

Loss on drying ⟨*5.01*⟩ Not more than 14.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 5.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 14.5%.

Containers and storage Containers—Well-closed containers.

Notopterygium

Notopterygii Rhizoma

キョウカツ

Notopterygium is the rhizome and root of *Notopterygium incisum* Ting ex H. T. Chang or *Notopterygium forbesii* Boissieu (*Umbelliferae*).

Description Notopterygium is slightly curved, cylindrical to conical, 3 – 10 cm in length, 5 – 20 mm in diameter; rhizome occasionally branched; externally yellow-brown to dark brown. The rhizome with nearly orbicular, hollowed stem scars at the apex, sometimes having short residue of stem; externally node rising, internode short; root scars in warty processes on the node; externally root has coarse longitudinal wrinkles and lateral root scars in warty processes; light and slightly brittle in texture, easy to break. The transverse section of the rhizome reveals numerous radial cracks; cortex yellow-brown to brown; xylem light yellow to light grayish yellow; pith grayish white to light brown. Under a magnifying glass, the rhizome reveals brown, fine points of resin canals in the cortex and pith.

Odor, characteristic; taste, a slight characteristic taste at first, followed by a slightly pungent and slightly numbing aftertaste.

Under a microscope ⟨*5.01*⟩, transverse section shows the outermost layer to be composed of a cork layer several to a dozen or so cells thick; several-layered collenchyma just inside of the cork layer; oil canals scattered in cortex, large ones more than 300 μm in diameter; intercellular space occurring in radial direction in cortex; oil canals scattered in pith, large ones more than 500 μm in diameter; parenchymatous cells contain simple and 2- to 3-compound starch grains.

Identification To 0.3 g of pulverized Notopterygium add 3 mL of hexane in a glass-stoppered centrifuge tube, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of octadecylsilanized silica gel with fluorescent indicator for thin-layer chromatography, develop the plate with a mixture of methanol and water (9:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a blue-white fluorescent spot appears at an *R*f value of about 0.5, which shows a dark purple color under ultraviolet light (main wavelength: 254 nm).

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Notopterygium according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Notopterygium according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 13.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 6.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.5%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 20.0%.

Containers and storage Containers—Well-closed containers.

Nuphar Rhizome

Nupharis Rhizoma

センコツ

Nuphar Rhizome is the longitudinally split rhizome of *Nuphar japonica* De Candolle, *Nuphar pumila* De Candolle, or their interspecific hybrids (*Nymphaeaceae*).

Description Usually, longitudinally split irregular column, twisted, bent or somewhat pressed, 20 – 30 cm in length, about 2 cm in width; the outer surface, dark brown, and the cut surface, white to grayish white in color; one side shows nearly round to blunt triangular scars of petiole about 1 cm in diameter, and the other side numerous scars of roots less than 0.3 cm in diameter; light, spongy in texture, and easily broken; fractured surface flat and powdery. Under a magnifying glass, a transverse section reveals a black outer por-

tion, and porous tissue with scattered vascular bundles in the inner portion.

Odor, slight; taste, slightly bitter and unpleasant.

Identification To 1.0 g of pulverized Nuphar Rhizome add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, methanol and ammonia solution (28) (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate: a yellow-brown spot appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Nuphar Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Nuphar Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Petiole—When perform the test of foreign matter <*5.01*>, the amount of the petioles contained in Nuphar Rhizome does not exceed 3.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than petioles is not more than 1.0%.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 10.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Nutmeg

Myristicae Semen

ニクズク

Nutmeg is the seed of *Myristica fragrans* Houttuyn (*Myristicaceae*), usually from which the seed coat is removed.

Description Ovoid-globose to ellipsoidal seeds, 1.5 – 3.0 cm in length, 1.3 – 2.0 cm in diameter; externally grayish brown, with wide and shallow longitudinal furrows and fine wrinkles; usually, reveals a hilum at one end, the hilum grayish white to grayish yellow and slightly protruding, and a chalaza at the other end, the chalaza grayish brown to dark brown and slightly concave; dark brown thin perisperm extending irregularly into the light yellow-white to light brown endosperm, by which cut surface exhibiting a marble-like appearance.

Odor, characteristic and strong; taste, acrid and slightly bitter.

Under a microscope <*5.01*>, a transverse section reveals perisperm composed of outer and inner layers; the outer layer composed of parenchyma containing red-brown to dark red-brown contents; the inner layer, composed of parenchyma filled with red-brown to dark red-brown contents, often extends into endosperm, where numerous oil cells are scattered: several vascular bundles present in the inner layer, the vessels spiral; in parenchyma cells of endosperm, simple or compound starch grains and aleurone grains observed; and in the parenchyma cells in the outer layer of perisperm and the marginal part of endosperm, numerous crystals of calcium oxalate observed.

Identification To 1 g of pulverized Nutmeg add 5 mL of methanol, allow to stand for 10 minutes with occasional shaking, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of myristicin for thin-layer chromatography in 1 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (9:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly diluted sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying <*5.01*> Not more than 16.0% (6 hours).

Total ash <*5.01*> Not more than 2.5%.

Essential oil content <*5.01*> When the test is performed with 10.0 g of pulverized Nutmeg, the essential oil content is not less than 0.5 mL.

Containers and storage Containers—Well-closed containers.

Nux Vomica

Strychni Semen

ホミカ

Nux Vomica is the seed of *Strychnos nux-vomica* Linné (*Loganiaceae*).

When dried, it contains not less than 1.07% of strychnine.

Description Disk, often slightly bent, 1 – 3 cm in diameter, 0.3 – 0.5 cm in thickness; externally light grayish yellow-green to light grayish brown, covered densely with lustrous appressed hairs radiating from the center to the circumference; on both sides, the margin and the central part bulged a little; the dot-like micropyle situated at one point on the margin, and from which usually a raised line runs to the center on one side; extremely hard in texture; when cracked upon soaking in water, the seed coat thin, the interior consisting of two horny, light grayish yellow endosperms, and leaving a central narrow cavity at the center; a white embryo, about 0.7 cm in length, situated at one end between the inner surfaces of the endosperms.

Odorless.

Identification **(1)** To 3 g of pulverized Nux Vomica add 3 mL of ammonia TS and 20 mL of chloroform, macerate for 30 minutes with occasional shaking, and filter. Remove most of the chloroform from the filtrate by warming on a water bath, add 5 mL of diluted sulfuric acid (1 in 10), and warm on a water bath while shaking well until the odor of chloroform is no longer perceptible. After cooling, filter through a pledget of absorbent cotton, and add 2 mL of nitric acid to 1 mL of the filtrate: a red color develops.

(2) To the remaining filtrate obtained in (1) add 1 mL of potassium dichromate TS, and allow to stand for 1 hour: a yellow-red precipitate is produced. Collect the precipitate by

filtration, and wash with 1 mL of water. Transfer a part of the precipitate to a small test tube, add 1 mL of water, dissolve by warming, cool, and add 5 drops of sulfuric acid dropwise carefully along the wall of the test tube: the layer of sulfuric acid shows a purple color which turns immediately red to red-brown.

Total ash ⟨*5.01*⟩ Not more than 3.0%.

Assay Weigh accurately about 1 g of pulverized Nux Vomica, previously dried at 60°C for 8 hours, place in a glass-stoppered centrifuge tube, and moisten with 1 mL of ammonia solution (28). To this solution add 20 mL of diethyl ether, stopper the centrifuge tube tightly, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diethyl ether, proceed in the same manner, and repeat this procedure three times. Combine all the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 10 mL of the mobile phase, add exactly 10 mL of the internal standard solution, and add the mobile phase to make 100 mL. Filter this solution through a membrane filter with a porosity not more than 0.8 μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 75 mg of strychnine nitrate for assay (separately determine the loss on drying), and dissolve in the mobile phase to make exactly 50 mL. Pipet 10 mL of this solution, add exactly 10 mL of the internal standard solution, then add the mobile phase to make 100 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Calculate the ratio, Q_T and Q_S, of the peak area of strychnine to that of the internal standard.

$$\text{Amount (mg) of strychnine} = M_S \times Q_T/Q_S \times 1/5 \times 0.841$$

M_S: Amount (mg) of strychnine nitrate for assay taken, calculated on the dried basis

Internal standard solution—A solution of barbital sodium in the mobile phase (1 in 500).

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: Room temperature.

Mobile phase: Dissolve 6.8 g of potassium dihydrogenphosphate in water to make 1000 mL, and mix with acetonitrile and triethylamine (45:5:1), and adjust the mixture with phosphoric acid to pH 3.0.

Flow rate: Adjust so that the retention time of Strychnine is about 17 minutes.

System suitability—

System performance: When the procedure is run with 5 μL of the standard solution under the above operating conditions, the internal standard and strychnine are eluted in this order with the resolution between these peaks being not less than 1.5.

Containers and storage Containers—Well-closed containers.

Nux Vomica Extract

ホミカエキス

Nux Vomica Extract contains not less than 6.15% and not more than 6.81% of strychnine ($C_{21}H_{22}N_2O_2$: 334.41).

Method of preparation After defatting 1000 g of coarse powder of Nux Vomica with hexane, extract with the percolation method, using a mixture of 750 mL of Ethanol, 10 mL of Acetic Acid and 240 mL of Purified Water or Purified Water in Containers as the first solvent, and 70 vol% ethanol as the second solvent. Combine the extracts, and prepare the dry extract as directed under Extracts. Where, an appropriate quantity of Ethanol and Purified Water or Purified Water in Containers may be used instead of 70 vol% ethanol.

Description Nux Vomica Extract occurs as yellow-brown to brown powder. It has a slight characteristic odor, and an extremely bitter taste.

Identification Extract 0.5 g of Nux Vomica Extract with 0.5 mL of ammonia TS and 10 mL of chloroform with occasional shaking. Filter the chloroform extract, evaporate the filtrate on a water bath until most of the chloroform is removed, and proceed as directed in the Identification under Nux Vomica.

Purity Heavy metals ⟨*1.07*⟩—Prepare the test solution with 1.0 g of Nux Vomica Extract as directed in the Extracts (4), and perform the test (not more than 30 ppm).

Assay Weigh accurately about 0.2 g of Nux Vomica Extract, place in a glass-stoppered centrifuge tube, add 15 mL of ammonia TS, and shake. Add 20 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge to disperse the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, proceed in the same manner, and repeat this procedure three times. Combine the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 10 mL of the mobile phase, add exactly 10 mL of the internal standard solution, and add the mobile phase to make 100 mL. Then, proceed as directed in the Assay under Nux Vomica.

$$\text{Amount (mg) of strychnine} = M_S \times Q_T/Q_S \times 1/5 \times 0.841$$

M_S: Amount (mg) of strychnine nitrate for assay taken, calculated on the dried basis

Internal standard solution—A solution of barbital sodium in the mobile phase (1 in 500).

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Nux Vomica Extract Powder

ホミカエキス散

Nux Vomica Extract Powder contains not less than 0.61% and not more than 0.68% of strychnine.

Method of preparation

Nux Vomica Extract	100 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
To make	1000 g

To Nux Vomica Extract add 100 mL of Purified Water or Purified Water in Containers, then warm, and soften with stirring. Cool, add 800 g of Starch, Lactose Hydrate or their mixture little by little, and mix well. Dry, preferably at a low temperature, and dilute with a sufficient additional quantity of Starch, Lactose or their mixture to make 1000 g of the homogeneous powder.

Description Nux Vomica Extract Powder occurs as a yellow-brown to grayish brown powder. It has a slight, characteristic odor and a bitter taste.

Identification **(1)** To 3 g of Nux Vomica Extract Powder add 3 mL of ammonia TS and 20 mL of chloroform, macerate for 30 minutes with occasional shaking, and filter. Remove most of the chloroform from the filtrate by warming on a water bath, add 5 mL of diluted sulfuric acid (1 in 10), and warm on a water bath while shaking well until the odor of chloroform is no longer perceptible. After cooling, filter through a pledget of absorbent cotton, and add 2 mL of nitric acid to 1 mL of the filtrate: a red color develops.

(2) To the remaining filtrate obtained in (1) add 1 mL of potassium dichromate TS, and allow to stand for 1 hour: a yellow-red precipitate is produced. Collect the precipitate by filtration, and wash with 1 mL of water. Transfer a part of the precipitate to a small test tube, add 1 mL of water, dissolve by warming, cool, and add 5 drops of sulfuric acid dropwise carefully along the wall of the test tube: the layer of sulfuric acid shows a purple color which turns immediately red to red-brown.

Assay Weigh accurately about 2.0 g of Nux Vomica Extract Powder, place in a glass-stoppered centrifuge tube, add 15 mL of ammonia TS, and shake. Add 20 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge to separate the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, proceed in the same manner, and repeat this procedure three times. Combine the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 10 mL of the mobile phase, add exactly 10 mL of the internal standard solution, and add the mobile phase to make 100 mL. Filter this solution through a membrane filter with a porosity not more than 0.8 μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 75 mg of strychnine nitrate for assay (separately determine the loss on drying), and dissolve in the mobile phase to make exactly 50 mL. Pipet 10 mL of this solution, add exactly 10 mL of the internal standard solution, then add the mobile phase to make 100 mL, and use this solution as the standard solution. Perform the test with exactly 5 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Calculate the ratio, Q_T and Q_S, of the peak area of strychnine to that of the internal standard.

Amount (mg) of strychnine = $M_S \times Q_T/Q_S \times 1/5 \times 0.841$

M_S: Amount (mg) of strychnine nitrate for assay taken, calculated on the dried basis

Internal standard solution—A solution of barbital sodium in the mobile phase (1 in 500).

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: Room temperature.

Mobile phase: A mixture of a solution of potassium dihydrogenphosphate (6.8 in 1000), acetonitrile and triethylamine (45:5:1), adjusted the pH to 3.0 with phosphoric acid.

Flow rate: Adjust so that the retention time of strychnine is about 17 minutes.

System suitability—

System performance: When the procedure is run with 5 μL of the standard solution under the above operating conditions, the internal standard and strychnine are eluted in this order with the resolution between these peaks being not less than 1.5.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Nux Vomica Tincture

ホミカチンキ

Nux Vomica Tincture contains not less than 0.097 w/v% and not more than 0.116 w/v% of strychnine.

Method of preparation

Nux Vomica, in coarse powder	100 g
70 vol% Ethanol	a sufficient quantity
To make	1000 mL

Prepare as directed under Tinctures, with the above ingredients. May be prepared with an appropriate quantity of Ethanol and Purified Water or Purified Water in Containers.

Description Nux Vomica Tincture is a yellow-brown liquid. It has an extremely bitter taste.

Specific gravity d_{20}^{20}: about 0.90

Identification Heat 20 mL of Nux Vomica Tincture on a water bath to remove ethanol, cool, transfer to a separator, add 2 mL of ammonia TS and 20 mL of chloroform, and shake well for 2 to 3 minutes. Filter the chloroform layer through a pledget of absorbent cotton, warm the filtrate on a water bath to remove most of chloroform, and proceed as directed in the Identification under Nux Vomica.

Alcohol number <*1.01*> Not less than 6.7 (Method 2).

Assay Pipet 3 mL of Nux Vomica Tincture into a glass-stoppered centrifuge tube, add 10 mL of ammonia TS and 20 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge to separate the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, proceed in the same manner, and repeat this procedure twice. Combine the extracts,

and evaporate the solvent on a water bath. Dissolve the residue with 10 mL of the mobile phase, add exactly 5 mL of the internal standard solution, and add the mobile phase to make 50 mL. Filter the solution through a membrane filter with a pore size not exceeding 0.8-μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 75 mg of strychnine nitrate for assay (separately determine the loss on drying), and dissolve in the mobile phase to make exactly 100 mL. Pipet 5 mL of this solution, add exactly 5 mL of the internal standard solution, add the mobile phase to make 50 mL, and use this solution as the standard solution. Proceed with the sample solution and the standard solution as directed in the Assay under Nux Vomica.

Amount (mg) of strychnine = $M_S \times Q_T/Q_S \times 1/20 \times 0.841$

M_S: Amount (mg) of strychnine nitrate for assay taken, calculated on the dried basis

Internal standard solution—A solution of barbital sodium in the mobile phase (1 in 500).

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Olive Oil

Oleum Olivae

オリブ油

Olive Oil is the fixed oil obtained by expression from the ripe fruit of *Olea europaea* Linné (*Oleaceae*).

Description Olive Oil is a light yellow oil. It has a faint odor, which is not rancid, and has a bland taste.
It is miscible with diethyl ether, with petroleum ether.
It is slightly soluble in ethanol (95).
The whole or a part of it congeals between 0°C and 6°C.
Congealing point of the fatty acids: 17 – 26°C

Specific gravity ⟨*1.13*⟩ d_{25}^{25}: 0.908 – 0.914

Acid value ⟨*1.13*⟩ Not more than 1.0.

Saponification value ⟨*1.13*⟩ 186 – 194

Unsaponifiable matters ⟨*1.13*⟩ Not more than 1.5%.

Iodine value ⟨*1.13*⟩ 79 – 88

Purity **(1)** Drying oil—Mix 2 mL of Olive Oil with 10 mL of diluted nitric acid (1 in 4), add 1 g of powdered sodium nitrite little by little with thorough shaking, and allow to stand in a cold place for 4 to 10 hours: the mixture congeals to a white solid.

(2) Peanut oil—Weigh exactly 1.0 g of Olive Oil, dissolve in 60 mL of sulfuric acid-hexane-methanol TS, heat for 2.5 hours under a reflux condenser, cool, transfer to a separator, and add 100 mL of water. Wash the vessel for the reflux extraction with 50 mL of petroleum ether, add the washing to the separator, shake, allow to stand, and separate the petroleum ether layer. Extract the aqueous layer with another 50 mL of petroleum ether, and combine the petroleum ether layer with the former petroleum ether solution. Wash the petroleum ether solution repeatedly with 20-mL portions of water until the washings show no more acidity to methyl orange TS. Then add 5 g of anhydrous sodium sulfate, shake, filter, wash anhydrous sodium sulfate with two 10-mL portions of petroleum ether, and filter the washings using the former separator. Combine all the filtrates, distil the solvent on a water bath, passing nitrogen. Dissolve the residue in acetone to make exactly 20 mL, and use this solution as the sample solution. Separately, dissolve 0.067 g of methyl behenate in acetone to make exactly 50 mL. Pipet 2 mL of this solution, add acetone to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 2 μL each of the sample solution and standard solution as directed under Gas Chromatography ⟨*2.02*⟩ according to the following conditions. Measure the peak heights, H_T and H_S, of methyl behenate of respective solutions: H_T is not higher than H_S.

Operating conditions—
Detector: A hydrogen flame-ionization detector.
Column: A glass column about 3 mm in inside diameter and about 2 m in length, packed with silanized siliceous earth for gas chromatography (150 to 180 μm in particle diameter), coated with polyethylene glycol 20 mol/L in a ratio of 5%.
Column temperature: A constant temperature of about 220°C.
Carrier gas: Nitrogen.
Flow rate: Adjust so that the retention time of methyl behenate is about 18 minutes.

System suitability—
Test for required detectability: Adjust so that the peak height of methyl behenate obtained with 2 μL of the standard solution is 5 to 10 mm.

Containers and storage Containers—Tight containers.

Ophiopogon Root

Ophiopogonis Radix

バクモンドウ

Ophiopogon Root is the enlarged part of the root of *Ophiopogon japonicus* Ker-Gawler (*Liliaceae*).

Description Fusiform root, 1 – 2.5 cm in length, 0.3 – 0.5 cm in diameter, somewhat sharp at one end, and somewhat rounded at the other; externally light yellow to light yellow-brown, with longitudinal wrinkles of various sizes; when fractured, cortex flexible and friable, stele strong; fractured surface of cortex light yellow-brown in color, slightly translucent and viscous.
Odor, slight; taste, slightly sweet and mucous.
Under a microscope ⟨*5.01*⟩, a transverse section reveals brown, 4- to 5-cellular layer velamen internally adjoining the epidermis; a single-layer exodermis inside the velamen, and cortex of parenchyma cells inside the exodermis; endodermis is distinct; about 20 protoxylems in actionstele; cortex parenchyma contains columnar crystals and raphides of calcium oxalate; oil drops in the exodermis.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Ophiopogon Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Ophiopogon Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Rootlets—When perform the test of foreign matter ⟨*5.01*⟩, the amount of the rootlets contained in Ophiopogon Root is not exceed 1.0%.

Total ash ⟨*5.01*⟩ Not more than 3.0%.

Containers and storage Containers—Well-closed containers.

Powdered Opium

Opium Pulveratum

アヘン末

Powdered Opium is a homogeneous powder of opium obtained from *Papaver somniferum* Linné (*Papaveraceae*). Starch or Lactose Hydrate may be added.

Powdered Opium contains not less than 9.5% and not more than 10.5% of morphine ($C_{17}H_{19}NO_3$: 285.34).

Description Powdered Opium occurs as a yellow-brown to dark brown powder.

Identification **(1)** To 0.1 g of Powdered Opium add 5 mL of diluted ethanol (7 in 10), dissolve by sonicating for 10 minutes, and add diluted ethanol (7 in 10) to make 10 mL. Filter this solution, and use the filtrate as the sample solution. Separately, dissolve 25 mg of Morphine Hydrochloride Hydrate, 12 mg of Codeine Phosphate Hydrate, 2 mg of Papaverine Hydrochloride, and 12 mg of Noscapine Hydrochloride Hydrate separately in 25 mL of diluted ethanol (7 in 10), and use these solutions as the standard solution (1), the standard solution (2), the standard solution (3) and the standard solution (4), respectively. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solutions on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, toluene, ethanol (99.5) and ammonia water (28) (20:20:3:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate: each spot obtained from the sample solution shows the same color tone and *R*f value of each spot from the standard solution (1), the standard solution (2), the standard solution (3), and the standard solution (4) (morphine, codeine, papaverine and noscapine), respectively.

(2) To 0.1 g of Powdered Opium add 5 mL of water, and shake the mixture for 5 minutes. Filter, to the filtrate add 1 mL of a solution of hydroxylammonium chloride (3 in 10) and 1 drop of iron (III) chloride TS, and shake: a red-brown color is produced. To this solution add immediately 5 mL of diethyl ether, and shake: the diethyl ether layer has no red-purple color (meconic acid).

Loss on drying ⟨*2.41*⟩ Not more than 8.0% (1 g, 105°C, 5 hours).

Assay Place about 5 g of Powdered Opium, accurately weighed, in a mortar, and triturate it with exactly 10 mL of water. Add 2 g of calcium hydroxide and exactly 40 mL of water, and stir the mixture for 20 minutes. Filter, and shake 30 mL of the filtrate with 0.1 g of magnesium sulfate heptahydrate for 1 minute. To the mixture add 0.3 g of calcium hydroxide, shake for 1 minute, and allow to stand for 1 hour. Filter, place 20 mL of the filtrate, exactly measured, in a glass-stoppered flask, and add 10 mL of diethyl ether and 0.3 g of ammonium chloride. Shake vigorously with caution. When crystals begin to separate out, shake for 30 minutes with a mechanical shaker, and set aside overnight at a temperature of 5°C to 10°C. Decant the diethyl ether layer and filter first, and then the aqueous layer through filter paper 7 cm in diameter. Wash the adhering crystals in the flask with three 5-mL portions of water saturated with diethyl ether, and wash the crystals on the filter paper with each of these washings. Wash the top of the glass-stoppered flask and the upper part of the filter paper with final 5 mL of water saturated with diethyl ether. Transfer the crystals and the filter paper to a beaker. Dissolve the crystals remaining in the glass-stoppered flask with the aid of 15 mL of 0.05 mol/L sulfuric acid VS, accurately measured, and pour the solution into the beaker. Wash the glass-stoppered flask with four 5-mL portions of water, and add the washings to the solution in the beaker. Titrate ⟨*2.50*⟩ the excess sulfuric acid with 0.1 mol/L sodium hydroxide VS (indicator: 4 drops of methyl red-methylene blue TS).

Each mL of 0.05 mol/L sulfuric acid VS
= 28.53 mg of $C_{17}H_{19}NO_3$

Containers and storage Containers—Tight containers.

Diluted Opium Powder

アヘン散

Diluted Opium Powder contains not less than 0.90% and not more than 1.10% of morphine ($C_{17}H_{19}NO_3$: 285.34).

Method of preparation

Powdered Opium	100 g
Starch or a suitable diluent	a sufficient quantity
To make	1000 g

Prepare as directed under Powders, with the above ingredients. Lactose Hydrate should not be used.

Description Diluted Opium Powder occurs as a light brown powder.

Identification **(1)** Proceed with 1 g of Diluted Opium Powder as directed in the Identification (1) under Powdered Opium.

(2) Proceed with 1 g of Diluted Opium Powder as directed in the Identification (2) under Powdered Opium.

Assay Place about 50 g of Diluted Opium Powder, accurately weighed, in a glass-stoppered flask, and stir with 250 mL of dilute ethanol in a water bath at 40°C for 1 hour. Filter the mixture through a glass filter (G3). Transfer the residue on the filter to the first glass-stoppered flask, and add 50 mL of dilute ethanol. Stir the mixture in a water bath at 40°C for 10 minutes, and filter through the same glass filter. Repeat the extraction with three 50-mL portions of dilute ethanol. Evaporate the combined filtrate in a mortar to dryness on a water bath. Add 10 mL of ethanol (99.5) to the residue, evaporate to dryness again, and, after cooling, triturate it with exactly 10 mL of water. Proceed with this solution as directed in Assay under Powdered Opium.

Each mL of 0.05 mol/L sulfuric acid VS
= 28.53 mg of $C_{17}H_{19}NO_3$

Containers and storage Containers—Tight containers.

Opium Tincture

アヘンチンキ

Opium Tincture contains not less than 0.93 w/v% and not more than 1.07 w/v% of morphine ($C_{17}H_{19}NO_3$: 285.34).

Method of preparation

Powdered Opium	100 g
35 vol% Ethanol	a sufficient quantity
To make	1000 mL

Prepare as directed under Tinctures, with the above ingredients. May be prepared with an appropriate quantity of Ethanol and Purified Water or Purified Water in Containers in place of 35 vol% Ethanol.

Description Opium Tincture is a dark red-brown liquid. It is affected by light.

Identification **(1)** To 1 mL of Opium Tincture add diluted ethanol (7 in 10) to make 10 mL, filter, and use the filtrate as the sample solution. Proceed as directed in the Identification (1) under Powdered Opium.

(2) Evaporate 1 mL of Opium Tincture to dryness on a water bath, and proceed with the residue as directed in the Identification (2) under Powdered Opium.

Alcohol number <*1.01*> Not less than 3.5 (Method 1).

Assay Evaporate 50 mL of Opium Tincture, accurately measured, on a water bath to dryness. Add 10 mL of ethanol (99.5) to the residue, evaporate to dryness again, cool, and triturate with exactly 10 mL of water. Proceed with this solution as directed in the Assay under Powdered Opium.

Each mL of 0.05 mol/L sulfuric acid VS
= 28.53 mg of $C_{17}H_{19}NO_3$

Containers and storage Containers—Tight containers. Storage—Light-resistant.

Opium Ipecac Powder

アヘン·トコン散

Opium Ipecac Powder contains not less than 0.90% and not more than 1.10% of morphine ($C_{17}H_{19}NO_3$: 285.34).

Method of preparation

Powdered Opium	100 g
Powdered Ipecac	100 g
Starch or a suitable ingredient	a sufficient quantity
To make	1000 g

Prepare as directed under Powders, with the above ingredients. Lactose Hydrate should not be used.

Description Opium Ipecac Powder occurs as a light brown powder.

Identification **(1)** Proceed with 1 g of Opium Ipecac Powder as directed in the Identification (1) under Powdered Opium.

(2) Proceed with 1 g of Opium Ipecac Powder as directed in the Identification (2) under Powdered Opium.

(3) Shake frequently a mixture of 3 g of Opium Ipecac Powder and 5 mL of hydrochloric acid, and allow to stand for 1 hour. Filter the solution into an evaporating dish. Add 5 mg of chlorinated lime to the filtrate: an orange color is produced at the circumference of the chlorinated lime (emetine).

Assay Weigh accurately about 50 g of Opium Ipecac Powder in a glass stoppered flask, add 250 mL of dilute ethanol, warm in a water bath at 40°C for 1 hour with stirring, and filter through a glass filter (G3). Transfer the residue on the filter to the first glass-stoppered flask, add 50 mL of dilute ethanol, warm in a water bath at 40°C for 10 minutes with stirring, and filter through the glass filter. Repeat the extraction with three 50-mL portions of dilute ethanol. Combine all the filtrates in a mortar, evaporate on a water bath to dryness, add 10 mL of ethanol (99.5) to the residue, and evaporate again. After cooling, triturate the residue with an exactly measured 10 mL of water, add 2 g of calcium hydroxide and an exactly measured 40 mL of water, stir the mixture for 20 minutes, and filter. To 30 mL of the filtrate add 0.1 g of magnesium sulfate heptahydrate, shake for 1 minute, then add 0.3 g of calcium hydroxide, shake for 1 minute, allow to stand for 1 hour, and filter. To an exactly measured 20 mL of the filtrate add 5 mL of sodium hydroxide TS, and adjust the pH to between 9.0 and 9.2 with ammonium chloride. Extract the solution successively with 60 mL, 40 mL and 30 mL of a mixture of chloroform and ethanol (95) (3:1). Combine all the extracts, distil, then evaporate the solvent on a water bath. Dissolve the residue in 20 mL of dilute sodium hydroxide TS and 10 mL of diethyl ether with shaking, add 0.5 g of ammonium chloride, shake vigorously with caution, and proceed as directed in the Assay under Powdered Opium.

Each mL of 0.05 mol/L sulfuric acid VS
= 28.53 mg of $C_{17}H_{19}NO_3$

Containers and storage Containers—Tight containers.

Orange Oil

Oleum Aurantii

オレンジ油

Orange Oil is the essential oil obtained by expression from the peel of the edible fruit of *Citrus* species (*Rutaceae*).

Description Orange Oil is a yellow to yellow-brown liquid. It has a characteristic, aromatic odor, and a slightly bitter taste.

It is miscible with an equal volume of ethanol (95) with turbidity.

Refractive index <*2.45*> n_D^{20}: 1.472 – 1.474

Optical rotation <*2.49*> α_D^{20}: +43 – +50° (50 mm).

Specific gravity <*1.13*> d_{20}^{20}: 0.842 – 0.848

Purity Heavy metals <*1.07*>—Proceed with 1.0 mL of Orange Oil according to Method 2, and perform the test. Prepare the control solution with 4.0 mL of Standard Lead Solution (not more than 40 ppm).

Containers and storage Containers—Tight containers. Storage—Light-resistant.

Orange Peel Syrup

トウヒシロップ

Method of preparation

Orange Peel Tincture	200 mL
Simple Syrup	a sufficient quantity
To make	1000 mL

Prepare as directed under Syrups, with the above ingredients. An appropriate quantity of Sucrose and Purified Water or Purified Water in Containers may be used in place of Simple Syrup.

Description Orange Peel Syrup is a brownish yellow to reddish brown liquid. It has a characteristic odor, a sweet taste and a bitter aftertaste.

Specific gravity d_{20}^{20}: about 1.25

Identification To 25 mL of Orange Peel Syrup add 50 mL of ethyl acetate, shake for 5 minutes, allow to stand until clear ethyl acetate layer separate, and take the ethyl acetate layer, and evaporate on a water bath to dryness. Dissolve the residue in 10 mL of ethanol (95), filter if necessary, and use this solution as the sample solution. Separately, dissolve 10 mg of naringin for thin-layer chromatography in 10 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly dilute 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in ammonia gas: one of the several spots obtained from the sample solution has the same color tone and the same *R*f value with the spot from the standard solution.

Containers and storage Containers—Tight containers.

Orange Peel Tincture

トウヒチンキ

Method of preparation

Bitter Orange Peel, in coarse powder	200 g
70 vol% Ethanol	a sufficient quantity
To make	1000 mL

Prepare as directed under Tinctures, with the above ingredients. An appropriate quantity of Ethanol and Purified Water or Purified Water in Containers may be used in place of 70 vol% Ethanol.

Description Orange Peel Tincture is a yellowish brown liquid. It has a characteristic odor, and a bitter taste.

Specific gravity d_{20}^{20}: about 0.90

Identification To 5.0 mL of Orange Peel Tincture add 5 mL of ethanol (95), filter if necessary, and use the filtrate as the sample solution. Proceed as directed in the Identification under Bitter Orange Peel.

Alcohol number <*1.01*> Not less than 6.6 (Method 2).

Containers and storage Containers—Tight containers.

Orengedokuto Extract

黄連解毒湯エキス

Orengedokuto Extract contains not less than 20 mg and not more than 80 mg of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], not less than 80 mg and not more than 240 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 30 mg and not more than 90 mg (for preparation prescribed 2 g of Gardenia Fruit) or not less than 45 mg and not more than 135 mg (for preparation prescribed 3 g of Gardenia Fruit) of geniposide, per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Coptis Rhizome	1.5 g	1.5 g	2 g	2 g
Phellodendron Bark	1.5 g	3 g	2 g	1.5 g
Scutellaria Root	3 g	3 g	3 g	3 g
Gardenia Fruit	2 g	3 g	2 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Orengedokuto Extract occurs as a yellow-brown to red-brown powder or blackish brown viscous extract. It has a characteristic odor and a very bitter taste.

Identification **(1)** Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 10 mL of methanol, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of coptisine chloride for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ammonia solution (28) and methanol (15:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Coptis Rhizome).

(2) Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 5 mL of water, then add 25 mL of ethyl acetate, and shake. Separate the ethyl acetate layer, evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of limonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the pur-

ple spot from the standard solution (Phellodendron Bark).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(4) Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 10 mL of methanol, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Gardenia Fruit).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Lead—Take 5.0 g of the dry extract (or an amount of the viscous extract, equivalent to 5.0 g of the dried substance) in a platinum, quartz or porcelain crucible, heat gently, and then incinerate by ignition at 450 to 550°C. After cooling, add a small amount of 2 mol/L nitric acid TS, filter if necessary, and wash the crucible and filter several times with small portions of 2 mol/L nitric acid TS. Combine the washings and the filtrate, add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the sample solution. Separately, to 2.5 mL of Standard Lead Solution add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the standard solution. Perform the test with the sample solution and the standard solution as directed under Atomic Absorption Spectrophotometry <*2.23*> according to the following conditions: the absorbance of the sample solution is not more than that of the standard solution (not more than 5 ppm).

Gas: Combustible gas—Acetylene or hydrogen.
Supporting gas—Air.
Lamp: A lead hollow-cathode lamp.
Wavelength: 283.3 nm.

(3) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 7.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 12.0%, calculated on the dried basis.

Assay **(1)** Berberine—Weigh accurately about 0.2 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.2 g of dried substance), add exactly 50 mL of the mobile phase, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (separately determine the water <*2.48*> in the same manner as Berberine Chloride Hydrate), dissolve in the mobile phase to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine chloride ($C_{20}H_{18}ClNO_4$) $= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogen phosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: 1.0 mL per minute (the retention time of berberine is about 8 minutes).

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in the mobile phase to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, add diluted methanol (7 in 10) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

Amount (mg) of baicalin ($C_{21}H_{18}O_{11}$) $= M_S \times A_T/A_S$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Geniposide—Weigh accurately about 0.2 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.2 g of dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of geniposide for assay, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of geniposide in each solution.

$$\text{Amount (mg) of geniposide} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of geniposide for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 240 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (900:100:1).

Flow rate: 1.0 mL per minute (the retention time of geniposide is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of geniposide are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of geniposide is not more than 1.5%.

Containers and storage Containers—Tight containers.

Oriental Bezoar

Bezoar Bovis

ゴオウ

Oriental Bezoar is a stone formed in the gall sac of *Bos taurus* Linné var. *domesticus* Gmelin (*Bovidae*).

It contains not less than 10.0% of bilirubin.

Description Spherical or massive stone, 1 – 4 cm in diameter; externally yellow-brown to red-brown; light, fragile and easily broken. Fractured surface shows yellow-brown to red-brown annular rings, often containing white granular substances or thin layers in these annular rings.

Odor, slight; taste, slightly bitter, followed by slight sweetness.

Identification To 25 mg of pulverized Oriental Bezoar add 10 mL of methanol, shake for 5 minutes, and centrifuge. Take the supernatant liquid, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 0.5 mL of methanol, and use this solution as the sample solution. Separately, dissolve 5 mg each of cholic acid for thin-layer chromatography and deoxycholic acid for thin-layer chromatography in 5 mL of methanol, respectively, and use these solutions as the standard solution (1) and the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution, standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, formic acid and methanol (30:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 10 minutes: two of the several spots obtained from the sample solution have the same color tone and *R*f value with each spot from the standard solutions (1) and (2).

Purity **(1)** Synthetic dye—To 2 mg of pulverized Oriental Bezoar add 1 mL dilute hydrochloric acid: no violet color develops.

(2) Starch—To 5 mg of pulverized Oriental Bezoar add 2 mL of water, and heat on a water bath for 5 minutes. Cool and add 2 to 3 drops of iodine TS: no blue-purple color develops.

(3) Sucrose—To 0.02 g of pulverized Oriental Bezoar add 10 mL of water, shake for 15 minutes, and filter. To 1 mL of the filtrate add 2 mL of anthrone TS, and shake: no deep blue-green to dark green color develops.

Total ash <*5.01*> Not more than 10.0%.

Assay Conduct this procedure without exposure to light using light-resistant vessels. The following sample solution and standard solution should be prepared before use. Weigh accurately about 10 mg of pulverized Oriental Bezoar, add 10 mL of a mixture of dimethyl sulfoxide and acetic acid (100) (9:1), warm at 60°C for 20 minutes, and add a mixture of dimethyl sulfoxide and acetic acid (100) (9:1) to make exactly 50 mL. Pipet 5 mL of this solution, add the mobile phase to make exactly 50 mL, filter through a membrane filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of bilirubin for assay, add about 350 mg of L-ascorbic acid, and dissolve in a mixture of dimethyl sulfoxide and acetic acid (100) (9:1) to make exactly 100 mL. Pipet 5 mL of this solution, add the mobile phase to make exactly 50 mL, and use this solution as the standard solution.

Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of bilirubin in each solution.

$$\text{Amount (mg) of bilirubin} = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of bilirubin for assay taken

Operating conditions—

Detector: A visible absorption photometer (wavelength: 450 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of acetonitrile and diluted acetic acid (100) (1 in 100) (19:1).

Flow rate: Adjust so that the retention time of bilirubin is about 10 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of bilirubin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of bilirubin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Otsujito Extract

乙字湯エキス

Otsujito Extract contains not less than 1.2 mg and not more than 4.8 mg of saikosaponin b_2, not less than 80 mg and not more than 240 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), not less than 14 mg and not more than 42 mg (for preparation prescribed 2 g of Glycyrrhiza) or not less than 20 mg and not more than 60 mg (for preparation prescribed 3 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), and not less than 0.5 mg of sennoside A ($C_{42}H_{38}O_{20}$: 862.74) or not less than 1.5 mg of rhein (for preparation prescribed 0.5 g of Rhubarb) or not less than 1 mg of sennoside A ($C_{42}H_{38}O_{20}$: 862.74) or not less than 3 mg of rhein (for preparation prescribed 1 g of Rhubarb), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Japanese Angelica Root	6 g	6 g	6 g
Bupleurum Root	5 g	5 g	5 g
Scutellaria Root	3 g	3 g	3 g
Glycyrrhiza	2 g	2 g	3 g
Cimicifuga Rhizome	1.5 g	1 g	1 g
Rhubarb	1 g	0.5 g	1 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 3), using the crude drugs shown above.

Description Otsujito Extract occurs as light brown to brown powder or black-brown viscous extract, having a slightly order, and a hot and slight sweet taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 10 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, add 10 mL of sodium hydroxide TS, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of butyl acetate and hexane (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, and heat the plate at 105°C for 5 minutes. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown to grayish brown spot from the standard solution (Scutellaria Root).

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>.

Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Use (*E*)-isoferulic acid-(*E*)-ferulic acid TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone and water (20:12:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the light yellow-white fluorescent spot from the standard solution (Cimicifuga Rhizome).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Rhubarb).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.5%, calculated on the dried basis.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, shake for 10 minutes, and centrifuge. After removing the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, separate the supernatant liquid, combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, shake for 10 minutes, and centrifuge. After removing the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

(4) Sennoside A—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and centrifuge. Pipet 10 mL of the supernatant liquid, pour it into a column about 10 mm in inside diameter (previously prepared by packing 0.36 g of strongly basic ion-exchange resin for column chromatography, and washing with 10 mL of methanol and 10 mL of diluted methanol (1 in 2)) to flow out, wash out the column with 10 mL of diluted methanol (1 in 2), then flow out with a mixture of water, methanol and formic acid (25:25:1) to obtain exactly 5 mL of the outflow liquid, and use this liquid as the sample solution. Separately, weigh accurately about 5 mg of Sennoside A RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of sennoside A in each solution.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_S \times A_T/A_S \times 1/8$$

M_S: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (2460:540:1).

Flow rate: 1.0 mL per minute (the retention time of sennoside A is about 14 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of sennoside A are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

(5) Rhein—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 80 mL of water, shake, and add water to make exactly 100 mL. Pipet 5 mL of this solution, add 20 mL of iron (III) chloride TS, heat under a reflux condenser for 30 minutes, add 3 mL of hydrochloric acid, and heat in addition under a reflux condenser for 30 minutes. After cooling, extract three times with 25 mL each of diethyl ether, combine all the diethyl ether layers, evaporate the solvent under low pressure (in vacuo), dissolve the residue in methanol to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of rhein for assay, and dissolve in acetone to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the

standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of rhein in each solution.

$$\text{Amount (mg) of rhein} = M_S \times A_T/A_S \times 2/5$$

M_S: Amount (mg) of rhein for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 278 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (650:350:1).

Flow rate: 1.0 mL per minute (the retention time of rhein is about 17 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of rhein are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of rhein is not more than 1.5%.

Containers and storage Containers—Tight containers.

Oyster Shell

Ostreae Testa

ボレイ

Oyster Shell is the shell of *Ostrea gigas* Thunberg (*Ostreidae*).

Description Irregularly curved, foliaceous or lamellated broken pieces. The unbroken oyster shell forms a bivalve 6 – 10 cm in length and 2 – 5 cm in width. The upper valve is flat and the lower one is somewhat hollow. Both the upper and lower edges of the valve are irregularly curved and bite with each other. The surface of the valve is externally light green-gray-brown and internally milky in color.

Almost odorless and tasteless.

Identification **(1)** Dissolve 1 g of sample pieces of Oyster Shell in 10 mL of dilute hydrochloric acid by heating: it evolves a gas, and forms a very slightly red, turbid solution in which a transparent, thin suspended matter remains. Pass the evolved gas through calcium hydroxide TS: a white precipitate is produced.

(2) The solution obtained in (1) has a slight, characteristic odor. Filter this solution and neutralize with ammonia TS: the solution responds to Qualitative Tests <*1.09*> for calcium salt.

(3) Ignite 1 g of pulverized Oyster Shell: it turns black-brown in color at first, and evolves a characteristic odor. Ignite it further: it becomes almost white.

Purity Barium—Dissolve 1 g of pulverized Oyster Shell in 10 mL of dilute hydrochloric acid: the solution does not respond to Qualitative Tests (1) <*1.09*> for barium salt.

Containers and storage Containers—Well-closed containers.

Powdered Oyster Shell

Ostreae Testa Pulverata

ボレイ末

Powdered Oyster Shell is the powder of Oyster Shell.

Description Powdered Oyster Shell occurs as a grayish white powder. It is almost odorless and tasteless.

Identification **(1)** Dissolve 1 g of Powdered Oyster Shell in 10 mL of dilute hydrochloric acid by heating: it evolves a gas, and forms a very slightly red, turbid solution. Pass the gas evolved through calcium hydroxide TS: a white precipitate is produced.

(2) The solution obtained in (1) has a slight, characteristic odor. Filter this solution, and neutralize with ammonia TS: the solution responds to Qualitative Tests <*1.09*> for calcium salt.

(3) Ignite 1 g of Powdered Oyster Shell: it turns black-brown in color at first evolving a characteristic odor. Ignite it further: it becomes almost white.

Purity **(1)** Water-soluble substances—Shake 3.0 g of Powdered Oyster Shell with 50 mL of freshly boiled and cooled water for 5 minutes, filter, and evaporate 25 mL of the filtrate to dryness. Dry the residue at 105°C for 1 hour, cool, and weigh: the mass of the residue does not exceed 15 mg.

(2) Acid-insoluble substances—To 5.0 g of Powdered Oyster Shell add 100 mL of water, and add hydrochloric acid in small portions with stirring until the solution becomes acid. Boil the acidic mixture with additional 1 mL of hydrochloric acid. After cooling, collect the insoluble substance by filtration, and wash it with hot water until the last washing no longer gives any reaction in Qualitative Tests <*1.09*> (2) for chloride. Ignite the residue and weigh: the mass of the residue does not exceed 25 mg.

(3) Barium—Dissolve 1 g of Powdered Oyster Shell in 10 mL of dilute hydrochloric acid: the solution does not respond to Qualitative Tests <*1.09*> (1) for barium salt.

Loss on drying <*2.41*> Not more than 4.0% (1 g, 180°C, 4 hours).

Containers and storage Containers—Tight containers.

Panax Japonicus Rhizome

Panacis Japonici Rhizoma

チクセツニンジン

Panax Japonicus Rhizome is the rhizome of *Panax japonicus* C. A. Meyer (*Araliaceae*), usually after being treated with hot water.

Description Irregularly cylidrical rhizome with distinct nodes, 3 – 20 cm in length, 1 – 1.5 cm in diameter, internode 1 – 2 cm; externally light yellow-brown, with fine longitudi-

nal wrinkles; stem scars, hollowed at the center, protruding on the upper surface, and root scars protruding as knobs on internodes; easily broken; fractured surface approximately flat, and light yellow-brown in color; horny in texture.

Odor, slight; taste, slightly bitter.

Identification Shake 0.5 g of pulverized Panax Japonicus Rhizome with 10 mL of methanol for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of chikusetsusaponin IV for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (5:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 110°C for 5 minutes: one of the several spots obtained from the sample solution shows the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Panax Japonicus Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Panax Japonicus Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Containers and storage Containers—Well-closed containers.

Powdered Panax Japonicus Rhizome

Panacis Japonici Rhizoma Pulveratum

チクセツニンジン末

Powdered Panax Japonicus Rhizome is the powder of Panax Japonicus Rhizome.

Description Powdered Panax Japonicus Rhizome occurs as a light grayish yellow-brown powder, and has a slight odor and a slightly bitter taste.

Under a microscope <*5.01*>, Powdered Panax Japonicus Rhizome reveals mainly starch grains or gelatinized starch masses, and fragments of parenchyma cells containing them; also fragments of cork tissue, somewhat thick-walled collenchyma, phloem tissue, and reticulate vessels; rarely fragments of scalariform vessels with a simple perforation, fibers and fiber bundles, rosette aggregates of calcium oxalate, and parenchyma cells containing them; yellow to orange-yellow resin; starch grains consisting of simple grains or 2- to 4-compound grains, simple grains, 3 – 18 μm in diameter; rosette aggregates of calcium oxalate are 20 – 60 μm in diameter.

Identification Shake 0.5 g of Powdered Panax Japonicus Rhizome with 10 mL of methanol for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of chikusetsusaponin IV for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (5:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 110°C for 5 minutes: one of the several spots obtained from the sample solution shows the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Panax Japonicus Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Panax Japonicus Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Containers and storage Containers—Well-closed containers.

Peach Kernel

Persicae Semen

トウニン

Peach Kernel is the seed of *Prunus persica* Batsch or *Prunus persica* Batsch var. *davidiana* Maximowicz (*Rosaceae*).

It contains not less than 1.2% of amygdalin, calculated on the basis of dried material.

Description Flattened, asymmetric ovoid seed, 1.2 – 2.0 cm in length, 0.6 – 1.2 cm in width, and 0.3 – 0.7 cm in thickness; somewhat sharp at one end, and round at the other end with chalaza; seed coat red-brown to light brown; externally, its surface being powdery by easily detachable stone cells of epidermis; numerous vascular bundles running and rarely branching from chalaza through the seed coat, and, appearing as dented longitudinal wrinkles; when soaked in boiling water and softened, the seed coat and thin, translucent, white albumen easily separated from the cotyledone; cotyledone white in color.

Almost odorless; taste, slightly bitter and oily.

Under a microscope <*5.01*>, the outer surface of seed coat reveals polygonal, long polygonal, or obtuse triangular stone cells on the protrusion from vascular bundles, shape of which considerably different according to the position, and their cell walls almost equally thickened; in lateral view, appearing as a square, rectangle or obtuse triangle.

Identification To 1.0 g of ground Peach Kernel add 10 mL of methanol, immediately heat under a reflux condenser for 10 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography.

Develop the plate with a mixture of ethyl acetate, methanol and water (20:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly thymol-sulfuric acid-methanol TS for spraying upon the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Rancidity—Grind Peach Kernel with boiling water: no odor of rancid oil is perceptible.

(2) Foreign matter <*5.01*>—When perform the test with not less than 250 g of Peach Kernel, it contains not more than 0.10% of broken pieces of endocarp.

Loss on drying <*5.01*> Not more than 8.0% (6 hours).

Assay Weigh accurately 0.5 g of ground Peach Kernel, add 40 mL of diluted methanol (9 in 10), heat immediately under a reflux condenser for 30 minutes, and cool. Filter the mixture, add diluted methanol (9 in 10) to make exactly 50 mL. Pipet 5 mL of this solution, add water to make exactly 10 mL, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

$$\text{Amount (mg) of amygdalin} = M_S \times A_T/A_S \times 2$$

M_S: Amount (mg) of amygdalin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilianized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Peach Kernel

Persicae Semen Pulveratum

トウニン末

Powdered Peach Kernel is the powder of the Peach Kernel.

It contains not less than 1.2% of amygdalin, calculated on the basis of dried material.

Description Powdered Peach Kernel occurs as a reddish-light brown to light brown powder. It is almost odorless and is oily and has slightly a bitter taste.

Under a microscope <*5.01*>, Powdered Peach Kernel fragments of outer seed coat epidermis; elliptical to ovoid, containing yellow-brown compound 50 to 80 μm in diameter and stone cell; cap-like shape to ovoid, yellow-brown in color. The stone cell is element of epidermis, 50 to 80 μm in diameter and 70 to 80 μm in height, cell wall of the top, 12 to 25 μm thickness, the base 4 μm in thickness, with obvious and numerous pits. Inner seed coat, yellow-brown, irregular and somewhat long polygon, 15 to 30 μm in diameter; and fragments of cotyledon and albumen containing aleurone grains and fatted oil, Aleurone grains are almost spherical grains, 5 to 10 μm in diameter.

Identification To 1.0 g of Powdered Peach Kernel add 10 mL of methanol, and immediately heat under a reflux condenser for 10 minutes. After cooling, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly thymol-sulfuric acid-methanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Loss on drying <*5.01*> Not more than 8.5% (6 hours).

Total ash <*5.01*> Not more than 3.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Assay Weigh accurately 0.5 g of Powdered Peach Kernel, add 40 mL of diluted methanol (9 in 10), heat immediately under a reflux condenser for 30 minutes, and cool. Filter the mixture, add diluted methanol (9 in 10) to make exactly 50 mL. Pipet 5 mL of this solution, add water to make exactly 10 mL, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for not less than 24 hours, dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test exactly with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

$$\text{Amount (mg) of amygdalin} = M_S \times A_T/A_S \times 2$$

M_S: Amount (mg) of amygdalin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilianized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Peanut Oil

Oleum Arachidis

ラッカセイ油

Peanut Oil is the fixed oil obtained from the seeds of *Arachis hypogaea* Linné (*Leguminosae*).

Description Peanut Oil is a pale yellow, clear oil. It is odorless or has a slight odor. It has a mild taste.

It is miscible with diethyl ether and with petroleum ether.

It is slightly soluble in ethanol (95).

Specific gravity d_{25}^{25}: 0.909 – 0.916

Congealing point of the fatty acids: 22 – 33°C

Identification Saponify 5 g of Peanut Oil by boiling with 2.5 mL of sodium hydroxide solution (3 in 10) and 12.5 mL of ethanol (95). Evaporate the ethanol, dissolve the residue in 50 mL of hot water, and add dilute hydrochloric acid in excess until the free fatty acids separate as an oily layer. Cool the mixture, remove the separated fatty acids, and dissolve them in 75 mL of diethyl ether. To the diethyl ether solution add a solution of 4 g of lead (II) acetate trihydrate in 40 mL of ethanol (95), and allow the mixture to stand for 18 hours. Filter the supernatant liquid, transfer the precipitate to the filter with the aid of diethyl ether, and filter by suction. Place the precipitate in a beaker, heat it with 40 mL of dilute hydrochloric acid and 20 mL of water until the oily layer is entirely clear, cool, and decant the aqueous layer. Boil the fatty acids with 50 mL of diluted hydrochloric acid (1 in 100). When the solution prepared by dissolving 0.1 g of the fatty acids in 10 mL of ethanol (95) is not darkened by the addition of 2 drops of sodium sulfide TS, allow the fatty acids to solidify, and press them between dry filter papers to exclude moisture. Dissolve the solid fatty acid in 25 mL of diluted ethanol (9 in 10) with the aid of gentle heat, and then cool to 15°C to crystallize the fatty acids. Recrystallize them from diluted ethanol (9 in 10) and dry in a desiccator (phosphorus (V) oxide, in vacuum) for 4 hours: the melting point <*1.13*> of the dried crystals is between 73°C and 76°C.

Acid value <*1.13*> Not more than 0.2.

Saponification value <*1.13*> 188 – 196

Unsaponifiable matters <*1.13*> Not more than 1.5%.

Iodine value <*1.13*> 84 – 103

Containers and storage Containers—Tight containers.

Peony Root

Paeoniae Radix

シャクヤク

Peony Root is the root of *Paeonia lactiflora* Pallas (*Paeoniaceae*).

It contains not less than 2.0% of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), calculated on the basis of dried material.

Description Cylindrical root, 7 – 20 cm in length, 1 – 2.5 cm in diameter; externally brown to light grayish brown, with distinct longitudinal wrinkles, with warty scars of lateral roots, and with laterally elongated lenticels; fractured surface dense in texture, light grayish brown, and with light brown radial lines in xylem.

Odor, characteristic; taste, slightly sweet at first, followed by an astringency and a slight bitterness.

Identification **(1)** Shake 0.5 g of pulverized Peony Root with 30 mL of ethanol (95) for 15 minutes, and filter. Shake 3 mL of the filtrate with 1 drop of iron (III) chloride TS: a blue-purple to blue-green color is produced, and it changes to dark blue-purple to dark green.

(2) To 2 g of pulverized Peony Root add 10 mL of methanol, warm on a water bath for 5 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Peony Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Peony Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Assay Weigh accurately about 0.5 g of pulverized Peony Root, add 50 mL of diluted methanol (1 in 2), heat under a reflux condenser for 30 minutes, cool, and filter. To the residue add 50 mL of diluted methanol (1 in 2), and proceed

in the same manner. Combine the filtrates, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions. Determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: Adjust so that the retention time of paeoniflorin is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Peony Root

Paeoniae Radix Pulverata

シャクヤク末

Powdered Peony Root is the powder of Peony Root.

It contains not less than 2.0% of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), calculated on the basis of dried material.

Description Powdered Peony Root occurs as a light grayish brown powder, and has a characteristic odor and a slightly sweet taste at first, followed by an astringency and a slight bitterness.

Under a microscope <5.01>, Powdered Peony Root reveals starch grains and fragments of parenchyma cells containing them; fragments of cork cells, vessels, tracheids and xylem fibers; rosette aggregates of calcium oxalate, and fragments of rows of crystal cells containing them. Starch grains consist of simple grains, 5 – 25 μm in diameter, occasionally 2- to 3-compound grains.

Identification **(1)** Shake 0.5 g of Powdered Peony Root with 30 mL of ethanol (95) for 15 minutes, and filter. To 3 mL of the filtrate add 1 drop of iron (III) chloride TS, and mix: a blue-purple to blue-green color is produced, and thereafter it changes to dark blue-purple to dark green.

(2) To 2 g of Powdered Peony Root add 10 mL of methanol, warm on a water bath for 5 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of Powdered Peony Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.40 g of Powdered Peony Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <5.01>, Powdered Peony Root does not show groups of light yellow stone cells and fibers.

Loss on drying <5.01> Not less than 14.0% (6 hours).

Total ash <5.01> Not more than 6.5%.

Acid-insoluble ash <5.01> Not more than 0.5%.

Assay Weigh accurately about 0.5 g of Powdered Peony Root, add 50 mL of diluted methanol (1 in 2), heat under a reflux condenser for 30 minutes, cool, and filter. To the residue add 50 mL of diluted methanol (1 in 2), and proceed in the same manner. Combine the filtrates, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions. Determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: Adjust so that the retention time of paeoniflorin is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Perilla Herb

Perillae Herba

ソヨウ

Perilla Herb is the leaves and the tips of branches of *Perilla frutescens* Britton var. *crispa* W. Deane (*Labiatae*).

It contains not less than 0.07% of perillaldehyde, calculated on the basis of dried material.

Description Usually, contracted and wrinkled leaves, often with thin stems. Both surfaces of the leaf are brownish purple, or the upper surface is grayish green to brownish green, and the lower surface is brownish purple in color. When smoothed by immersion in water, the lamina is ovate to obcordate, 5 – 12 cm in length, 5 – 8 cm in width; the apex, acuminate; the margin, serrate; the base, broadly cuneate; petiole, 3 – 5 cm in length; cross sections of stem and petiole, square. Under a magnifying glass, hairs are observed on both surfaces of the leaf, but abundantly on the vein and sparsely on other parts; small glandular hairs are observed on the lower surface.

Odor, characteristic; taste slightly bitter.

Identification To 0.6 g of pulverized Perilla Herb, add 10 mL of diethyl ether, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of perillaldehyde for thin-layer chromatography in 10 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid-ethanol TS for spray on the plate, and heat the plate at 105°C for 2 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Stem—When perform the test of foreign matter <*5.01*>, Perilla Herb does not contain its stems equal to or greater than 3 mm in diameter.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than the stems contained in Perilla Herb does not exceed 1.0%.

(3) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 16.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Assay Weigh accurately about 0.2 g of freshly prepared pulverized Perilla Herb, put in a glass-stoppered centrifuge tube, add 20 mL of methanol, shake for 10 minutes, centrifuge, and separate the supernatant liquid. To the residue, add 20 mL of methanol, and proceed in the same manner. Combine all the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of diphenyl sulfone for assay, and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T of perillaldehyde and A_S of diphenyl sulfone, in each solution.

$$\text{Amount (mg) of perillaldehyde} = M_S \times A_T/A_S \times 1/20 \times 0.700$$

M_S: Amount (mg) of diphenyl sulfone for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 234 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile (13:7).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: Dissolve 1 mg of (*E*)-asarone and 1 mg of perillaldehyde for thin-layer chromatography in the standard solution to make 50 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, diphenyl sulfone, perillaldehyde, and (*E*)-asarone are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of diphenyl sulfone is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Peucedanum Root

Peucedani Radix

ゼンコ

Peucedanum Root is the root of **1)** *Peucedanum praeruptorum* Dunn (Peucedanum Praeruptorum Root) or **2)** *Angelica decursiva* Franchet et Savatier (*Peucedanum decursivum* Maximowicz) (*Umbelliferae*) (Angelica Decursiva Root).

Description **1)** Peucedanum Praeruptorum Root—Slender obconical to cylindrical root, occasionally dichotomized at the lower part, 3 – 15 cm in length, 0.8 – 1.8 cm in diame-

ter at the crown; externally light brown to dark brown; ring-node-like wrinkles numerous at the crown, sometimes with hair-like remains of leaf sheath; the root having somewhat deep longitudinal wrinkles and scars of cutting off of lateral roots; transverse section surface light brown to whitish in color; brittle in texture.

Odor, characteristic; taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section reveals the outermost layer composed of a cork layer, inner tangential walls of some cork cells thickened; collenchyma just inside of the cork layer; in cortex numerous oil canals scattered and intercellular air spaces observed; occasionally phloem fibers observed at the terminal portion of phloem; vessels and scattered oil canals in xylem; starch grains in parenchyma, 2 to 10 several-compound grains.

2) Angelica Decursiva Root—Similar to 1), but without hair-like remains of leaf sheath at the crown.

Odor, characteristic; taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section reveals, similar to 1), but cell wall of cork cells not thickened, phloem fibers not observed at the terminal portion of phloem, nor oil canals observed in xylem.

Identification **1)** Peucedanum Praeruptorum Root—To 1 g of pulverized Peucedanum Root add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of (±)-praeruptorin A for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of diethyl ether and hexane (3:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white to blue-purple fluorescent spot from the standard solution.

2) Angelica Decursiva Root—To 1 g of pulverized Peucedanum Root add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of nodakenin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (12:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white to blue-purple fluorescent spot from the standard solution.

Purity Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Peucedanum Root according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

Loss on drying <*5.01*> Not more than 13.0%.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 20.0%.

Containers and storage Containers—Well-closed containers.

Pharbitis Seed

Pharbitidis Semen

ケンゴシ

Pharbitis Seed is the seed of *Pharbitis nil* Choisy (*Convolvulaceae*).

Description Longitudinally quartered or sexpartite globe, 4 – 6 mm in length, 3 – 5 mm in width; externally black to grayish red-brown or grayish white, smooth, but slightly shrunken and coarsely wrinkled. The transverse section almost fan-shaped, light yellow-brown to light grayish brown, and dense in texture. Under a magnifying glass, the surface of the seed coat reveals dense, short hairs; dented hilum at the bottom of the raphe. Seed coat thin, the outer layer dark gray, and the inner layer light gray; two irregularly folded cotyledons in the transverse section at one end; two septa from the center of the dorsal side to the ridge separating cotyledons but unrecognizable in the transverse section of the other end having hilum; dark gray secretory pits in the section of the cotyledon. 100 seeds weighing about 3.5 g.

When cracked, odor, slight; taste, oily and slightly pungent.

Total ash <*5.01*> Not more than 6.0%.

Containers and storage Containers—Well-closed containers.

Phellodendron Bark

Phellodendri Cortex

オウバク

Phellodendron Bark is the bark of *Phellodendron amurense* Ruprecht or *Phellodendron chinense* Schneider (*Rutaceae*), from which the periderm has been removed.

It contains not less than 1.2% of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], calculated on the basis of dried material.

Description Flat or rolled semi-tubular pieces of bark, 2 – 4 mm in thickness; externally grayish yellow-brown to grayish brown, with numerous traces of lenticels; the internal surface yellow to dark yellow-brown in color, with fine vertical lines, and smooth; fractured surface fibrous and bright yellow.

Odor, slight; taste, extremely bitter; mucilaginous; it colors the saliva yellow on chewing.

Under a microscope <*5.01*>, a transverse section reveals primary ray expands outward and looks fan shaped in secondary cortex, and sometimes ray differentiated later converges outward; groups of stone cells yellow and scattered in primary ray; groups of phloem fibers light yellow to yellow, lined alternately with the other tissue of phloem between rays, and then these tissues show obviously latticework; solitary crystals of calcium oxalate, single and compound starch grains observed in parenchyma.

Identification (1) To 1 g of pulverized Phellodendron Bark add 10 mL of diethyl ether, allow to stand for 10 minutes with occasional shaking, and filter to remove the diethyl ether. Collect the powder on the filter paper, add 10 mL of ethanol (95), allow to stand for 10 minutes with occasional shaking, and filter. To 2 to 3 drops of the filtrate add 1 mL of hydrochloric acid, add 1 to 2 drops of hydrogen peroxide TS, and shake: a red-purple color develops.

(2) Use the filtrate obtained in (1) as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberine chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution and a spot with yellow to yellow-green fluorescence from the standard solution show the same color tone and the same *R*f value.

(3) Stir up pulverized Phellodendron Bark with water: the solution becomes gelatinous owing to mucilage.

Loss on drying <*5.01*> Not more than 11.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Assay Weigh accurately about 0.5 g of pulverized Phellodendron Bark, add 30 mL of a mixture of methanol and dilute hydrochloric acid (100:1), heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure twice with the residue, using 30-mL and 20-mL portions of a mixture of methanol and dilute hydrochloric acid (100:1). To the last residue add 10 mL of methanol, shake well, and filter. Combine the whole filtrates, add methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (separately determine the water <*2.48*> in the same manner as Berberine Chloride Hydrate), dissolve in methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$)]
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 mm in particle diameter).

Column temperature: A constant temperature of about 40°C

Mobile phase: Dissolve 3.4 g of potassium dihydrogenphosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: Adjust so that the retention time of berberine is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in methanol to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 5 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Phellodendron Bark

Phellodendri Cortex Pulveratus

オウバク末

Powdered Phellodendron Bark is the powder of Phellodendron Bark.

It contains not less than 1.2% of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], calculated on the basis of dried material.

Description Powdered Phellodendron Bark occurs as a bright yellow to yellow powder. It has a slight odor and an extremely bitter taste, is mucilaginous, and colors the saliva yellow on chewing.

Under a microscope <*5.01*>, Powdered Phellodendron Bark reveals fragments of yellow, thick-walled fiber bundles or fibers, and fibers often accompanied by crystal cell rows; fewer groups of stone cells together with idioblasts; fragments of parenchyma cells containing starch grains and oil droplets; fragments of medullary ray and phloem; mucilage cells and mucilage masses. Numerous solitary crystals of calcium oxalate, 7 – 20 μm in diameter; starch grains, simple grains and 2- to 4-compound grains, simple grain, 2 – 6 μm in diameter; oil droplets, stained red with sudan III TS.

Identification (1) To 1 g of Powdered Phellodendron Bark add 10 mL of diethyl ether, allow to stand for 10 minutes with occasional shaking, and filter to remove the diethyl ether. Collect the powder on the filter paper, add 10 mL of ethanol (95), allow to stand for 10 minutes with occasional shaking, and filter. To 2 to 3 drops of the filtrate add 1 mL of hydrochloric acid, add 1 to 2 drops of hydrogen peroxide TS, and shake: a red-purple color develops.

(2) Use the filtrate obtained in (1) as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberine chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution and a spot with yellow to yellow-green fluorescence from the standard solution show the same color tone and the same *R*f value.

(3) Stir up Powdered Phellodendron Bark with water: the solution becomes gelatinous owing to mucilage.

Purity Curcuma—Place Powdered Phellodendron Bark on filter paper, drop diethyl ether on it, and allow to stand. Take the powder off the filter paper, and drip 1 drop of potassium hydroxide TS: no red-purple color develops. Under a microscope ⟨*5.01*⟩, Powdered Phellodendron Bark does not contain gelatinized starch or secretory cells containing yellow-red resin.

Loss on drying ⟨*5.01*⟩ Not more than 11.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 7.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%.

Assay Weigh accurately about 0.5 g of Powdered Phellodendron Bark, add 30 mL of a mixture of methanol and dilute hydrochloric acid (100:1), heat under a reflux condenser for 30 minutes, cool, and filter. Repeat the above procedure twice with the residue, using 30-mL and 20-mL portions of a mixture of methanol and dilute hydrochloric acid (100:1). To obtained residue add 10 mL of methanol, shake well, and filter. Combine the whole filtrates, add methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (separately determine the water ⟨*2.48*⟩ in the same manner as Berberine Chloride Hydrate), dissolve in methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$)]
$= M_S \times A_T/A_S$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogenphosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: Adjust so that the retention time of berberine is about 10 minutes.

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in methanol to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 5 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Compound Phellodendron Powder for Cataplasm

パップ用複方オウバク散

Method of preparation

Powdered Phellodendron Bark	660 g
Powdered Gardenia Fruit	325 g
d- or *dl*-Camphor	10 g
dl- or *l*-Menthol	5 g
To make	1000 g

Prepare as directed under Powders, with the above ingredients.

Description Compound Phellodendron Powder for Cataplasm occurs as a yellow-brown powder, having a characteristic odor.

Identification Shake thoroughly 0.2 g of Compound Phellodendron Powder for Cataplasm with 5 mL of methanol, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberine chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use the solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 10 cm, air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow to yellow-green fluorescent spot from the standard solution (phellodendron bark).

Containers and storage Containers—Tight containers.

Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder

オウバク·タンナルビン·ビスマス散

Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder contains not less than 12.9% and not more than 16.3% of bismuth (Bi: 208.98).

Method of preparation

Powdered Phellodendron Bark	300 g
Albumin Tannate	300 g
Bismuth Subnitrate	200 g
Scopolia Extract	10 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
To make	1000 g

Prepare as directed under Powders, with the above ingredients. Scopolia Extract Powder may be used in place of Scopolia Extract.

Description Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder is brownish yellow in color, and has a bitter taste.

Identification **(1)** Shake thoroughly 0.1 g of Phelloden-

dron, Albumin Tannate and Bismuth Subnitrate Powder with 5 mL of methanol, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Berberine Chloride RS or berberine chloride hydrate for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 10 cm, air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one spot of the several spots obtained from the sample solution and a spot with yellow to yellow-green fluorescence from the standard solution show the same color tone and the same *R*f value (phellodendron bark).

(2) To 0.3 g of Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder add 20 mL of ethanol (95), heat in a water bath for 3 minutes with shaking, cool, and filter. To 10 mL of the filtrate add 1 drop of iron (III) chloride TS: a blue-green color is produced. Allow to stand: a bluish black precipitate is produced (albumin tannate).

(3) To 0.3 g of Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder add 10 mL of diluted pyridine (1 in 5), warm in a water bath for 3 minutes with shaking, cool, and filter. Add 1 mL of ninhydrin-ascorbic acid TS to the filtrate, and heat in a water bath: a blue color is produced (albumin tannate).

(4) To 0.5 g of Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder add 5 mL of dilute hydrochloric acid and 10 mL of water, warm, shake thoroughly, and filter. The filtrate responds to the Qualitative Tests <*1.09*> for bismuth salt.

Assay Weigh accurately about 0.7 g of Phellodendron, Albumin Tannate and Bismuth Subnitrate Powder, shake well with 10 mL of water and 20 mL of diluted nitric acid (1 in 3), add water to make exactly 100 mL, and filter. Discard the first 20 mL of the filtrate, pipet the subsequent 10 mL of the filtrate, and add water to make exactly 100 mL. Pipet 25 mL of this solution, add diluted nitric acid (1 in 100) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.23 g of bismuth nitrate pentahydrate, add 20 mL of diluted nitric acid (1 in 3) and water to make exactly 100 mL. Pipet 10 mL of this solution, and add water to make exactly 100 mL. Pipet 25 mL of this solution, add diluted nitric acid (1 in 100) to make exactly 100 mL, and use this solution as the standard solution. Determine the absorbances, A_T and A_S, of the sample solution and standard solution as directed under Atomic Absorption Spectrophotometry <*2.23*> according to the following conditions. On the other hand, determine the absorbance A_0 of the solution prepared in the same manner using 20 mL of diluted nitric acid (1 in 3) instead of the standard solution.

Gas: Combustible gas—Acetylene.
Supporting gas—Air.
Lamp: A bismuth hollow-cathode lamp.
Wavelength: 223.1 nm.

$$\text{Amount (mg) of bismuth (Bi)} = M \times (A_T - A_0)/(A_S - A_0) \times 0.431$$

M: Amount (mg) of bismuth nitrate pentahydrate taken

Containers and storage Containers—Well-closed containers.

Picrasma Wood

Picrasmae Lignum

ニガキ

Picrasma Wood is the wood of *Picrasma quassioides* Bennet (*Simaroubaceae*).

Description Light yellow chips, slices or short pieces of wood; a transverse section reveals distinct annual rings and thin medullary rays; tissue dense in texture.

Odorless; taste, extremely bitter and lasting.

Under a microscope <*5.01*>, it reveals medullary rays consisting of 1 – 5 cells wide for transverse section, and 5 – 50 cells high for longitudinal cut surface; vessels of spring wood up to about 150 μm in diameter, but those of autumn wood only one-fifth as wide; vessels, single or in groups, scattered in the xylem parenchyma; wall of wood fibers extremely thickened; medullary rays and xylem parenchyma cells contain rosette aggregates of calcium oxalate and starch grains. Vivid yellow or red-brown, resinous substance often present in the vessels.

Purity Foreign matter <*5.01*>—The amount of foreign matter contained in Picrasma Wood does not exceed 1.0%.

Total ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Well-closed containers.

Powdered Picrasma Wood

Picrasmae Lignum Pulveratum

ニガキ末

Powdered Picrasma Wood is the powder of Picrasma Wood.

Description Powdered Picrasma occurs as a grayish white to light yellow powder. It is odorless, and has an extremely bitter and lasting taste.

Under a microscope <*5.01*>, Powdered Picrasma Wood reveals fragments of vessels of various sizes, xylem fibers and xylem parenchyma cells; fragments of medullary rays containing starch grains; all tissues lignified; a few crystals of calcium oxalate observed. Starch grains are 5 to 15 μm in diameter.

Total ash <*5.01*> Not more than 4.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Pinellia Tuber

Pinelliae Tuber

ハンゲ

Pinellia Tuber is the tuber of *Pinellia ternata* Breitenbach (*Araceae*), from which the cork layer has been removed.

Description Slightly flattened spherical to irregular-shaped tuber; 0.7 – 2.5 cm in diameter and 0.7 – 1.5 cm in height; externally white to grayish white-yellow; the upper end dented, where the stem has been removed, with root scars dented as numerous small spots on the circumference; dense in texture; cross section white and powdery.

Almost odorless; tasteless at first, slightly mucous, but leaving a strong acrid taste.

Under a microscope <*5.01*>, a transverse section reveals mainly tissue of parenchyma filled with starch grains, and scattered with a few mucilage cells containing raphides of calcium oxalate. Starch grains mostly 2- to 3-compound grains, usually 10 – 15 μm in diameter, and simple grains, usually 3 – 7 μm in diameter; raphides of calcium oxalate 25 – 150 μm in length.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Pinellia Tuber according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Pinellia Tuber according to Method 4, and perform the test (not more than 5 ppm).

(3) Rhizome of Arisaema species and others—Under a microscope <*5.01*>, no mucilage canal is revealed on the outer layer of cortex.

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 3.5%.

Containers and storage Containers—Well-closed containers.

Plantago Herb

Plantaginis Herba

シャゼンソウ

Plantago Herb is the entire plant of *Plantago asiatica* Linné (*Plantaginaceae*), collected during the flowering season.

Description Usually wrinkled and contracted leaf and spike, grayish green to dark yellow-green in color; when soaked in water and smoothed out, the lamina is ovate to orbicular-ovate, 4 – 15 cm in length, 3 – 8 cm in width; apex acute, and base sharply narrowed; margin slightly wavy, with distinct parallel veins; glabrous or nearly glabrous; petiole is rather longer than the lamina, and its base is slightly expanded with thin-walled leaf-sheath; scape is 10 – 50 cm in length, one-third to one-half of the upper part forming the spike, with dense florets; the lower part of inflorescence often shows pyxidia; roots usually removed, but, if any, fine roots are closely packed.

Odor, slight; practically tasteless.

Identification To 2.0 g of pulverized Plantago Herb add 10 mL of methanol, warm on a water bath for 3 minutes, cool, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride TS on the plate: a dark blue spot appears at an *R*f value of about 0.55.

Total ash <*5.01*> Not more than 15.0%.

Acid-insoluble ash <*5.01*> Not more than 4.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 14.0%.

Containers and storage Containers—Well-closed containers.

Plantago Seed

Plantaginis Semen

シャゼンシ

Plantago Seed is the seed of *Plantago asiatica* Linné (*Plantaginaceae*).

Description Flattened ellipsoidal seed, 2 – 2.5 mm in length, 0.7 – 1 mm in width, 0.3 – 0.5 mm in thickness; externally brown to yellow-brown and lustrous. Under a magnifying glass, the surface of the seed is practically smooth, with the dorsal side protruding like a bow, and with the ventral side somewhat dented; micropyle and raphe not observable. 100 seeds weigh about 0.05 g.

Almost odorless; taste, slightly bitter and mucous.

Under a microscope <*5.01*>, a transverse section reveals a seed coat consisting of three layers of epidermis composed of cells containing mucilage, a vegetative layer, and a pigment layer of approximately equidiameter cells; in the interior, endosperm thicker than seed coat, enclosing two cotyledons.

Identification **(1)** To 1 g of Plantago Seed add 2 mL of warm water, and allow the mixture to stand for 10 minutes: the seed coat swells to discharge mucilage.

(2) To 1.0 g of pulverized Plantago Seed add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, to 0.2 g of powdered plantago seed for thin-layer chromatography add 1 mL of methanol, and warm on a water bath for 3 minutes. After cooling, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate, water and acetic acid (100) (10:10:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: the spot appeared at an *R*f value of about 0.25 obtained from the sample solution has the same color tone with the dark blue spot appeared at an *R*f value of about 0.25 from the standard solution.

Purity Foreign matter <*5.01*>—The amount of foreign matter contained in Plantago Seed does not exceed 2.0%.

Total ash ⟨*5.01*⟩ Not more than 5.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Containers and storage Containers—Well-closed containers.

Platycodon Root

Platycodi Radix

キキョウ

Platycodon Root is the root of *Platycodon grandiflorus* A. De Candollе (*Campanulaceae*).

Description Irregular, somewhat thin and long fusiform to conical root, often branched; externally grayish brown, light brown or white; main root 10 – 15 cm in length, 1 – 3 cm in diameter; the upper end, with dented scars of removed stems; the neighborhood, with fine lateral wrinkles and longitudinal furrows and also slightly constricted; the greater part of the root, except the crown, covered with coarse longitudinal wrinkles, lateral furrows and lenticel-like lateral lines; hard in texture, but brittle; fractured surface not fibrous, often with cracks. Under a magnifying glass, a transverse section reveals cambium and its neighborhood often brown in color; cortex slightly thinner than xylem, almost white and with scattered cracks; xylem white to light brown in color, and the tissue slightly denser than cortex.

Odor, slight; tasteless at first, later acrid and bitter.

Identification **(1)** Warm 0.2 g of pulverized Platycodon Root with 2 mL of acetic anhydride on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add carefully 0.5 mL of sulfuric acid to make two layers: a red to red-brown color develops at the zone of contact, and the upper layer acquires a blue-green to green color.

(2) To 2.0 g of pulverized Platycodon Root add 20 mL of sodium carbonate TS, and shake. Add 5 mL of 1-butanol, shake for 10 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of platycodin D for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (5:3:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Platycodon Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Platycodon Root according to Method 4, and perform the test (not more than 5 ppm).

Total ash ⟨*5.01*⟩ Not more than 4.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 25.0%.

Containers and storage Containers—Well-closed containers.

Powdered Platycodon Root

Platycodi Radix Pulverata

キキョウ末

Powdered Platycodon Root is the powder of Platycodon Root.

Description Powdered Platycodon Root occurs as a light grayish yellow to light grayish brown powder. It has a slight odor, and is tasteless at first, later acrid and bitter.

Under a microscope ⟨*5.01*⟩, Powdered Platycodon Root reveals numerous fragments of colorless parenchyma cells; fragments of reticulate vessels and scalariform vessels; fragments of sieve tubes and lactiferous tubes; fragments of cork layer are sometimes observed. Usually, starch grains are not observed, but very rarely simple grain.

Identification **(1)** Warm 0.2 g of Powdered Platycodon Root with 2 mL of acetic anhydride on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add carefully 0.5 mL of sulfuric acid to make two layers: a red to red-brown color develops at the zone of contact, and the upper layer acquires a blue-green to green color.

(2) To 2.0 g of Powdered Platycodon Root add 20 mL of sodium carbonate TS, and shake. Add 5 mL of 1-butanol, shake for 10 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of platycodin D for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (5:3:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution and a spot from the standard solution show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of Powdered Platycodon Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of Powdered Platycodon Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope ⟨*5.01*⟩, Powdered Platycodon Root does not show fibers, stone cells or other foreign matter.

Total ash ⟨*5.01*⟩ Not more than 4.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 1.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 25.0%.

Containers and storage Containers—Well-closed containers.

Platycodon Fluidextract

キキョウ流エキス

Method of preparation 1) Take coarse powder of Platycodon Root, and prepare the fluidextract as directed under Fluidextracts using 25 vol% ethanol. An appropriate quantity of Ethanol and Purified Water or Purified Water in Containers may be used in place of 25 vol% ethanol.

2) Take Platycodon Root pulverized to suitable sizes, and prepare the fluidextract as directed under Fluidextracts using 25 vol% ethanol or diluted ethanol (1 in 4) as the solvent.

Description Platycodon Fluidextract is a red-brown liquid. It is miscible with water, producing slight turbidity. It has a mild taste at first, followed by an acrid and bitter taste.

Identification To 2 mL of Platycodon Fluidextract add 20 mL of water and 5 mL of 1-butanol, mix, shake for 10 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of platycodin D for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (5:3:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Alcohol number <*1.01*> Apply to Platycodon Fluidextract prepared by the Method of preparation 2). 2.0 – 3.0 (Method 1).

Purity (1) Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Platycodon Fluidextract as directed in the Fluidextracts (4), and perform the test (not more than 30 ppm).

(2) Starch—Mix 1 mL of Platycodon Fluidextract with 4 mL of water, and add 1 drop of dilute iodine TS: no purple or blue color develops.

Content of the active principle Transfer exactly 5 mL of Platycodon Fluidextract to a tared beaker or porcelain dish, evaporate to dryness on a water bath, and dry at 105°C for 5 hours: the mass of the residue is not less than 0.50 g.

Containers and storage Containers—Tight containers.
Storage—Light-resistant.

Pogostemi Herb

Pogostemi Herba

カッコウ

Pogostemon Herb is the terrestrial part of *Pogostemon cablin* Bentham (*Labiatae*).

Description Stems with opposite leaves, leaves wrinkled and shriveled. When smoothed by immersion in water, leaves are obovate to ovate-oblong, 2.5 – 10 cm in length, 2.5 – 7 cm in width, with obtusely serrate margins and petioles at the cuneate bases; the upper surface of leaves dark brown, the lower surface grayish brown, both sides covered densely with hairs. Stems are square, solid, grayish green, covered with grayish white to yellow-white hairs; the pith broad, whitish, spongy. Under a magnifying glass, leaf reveals hairs, glandular hairs and glandular scales.

Odor, distinct; taste, slightly bitter.

Under a microscope <*5.01*>, a transverse section of petiole reveals central portion of the adaxial side protruding remarkably, with collenchyma cells beneath epidermis; vascular bundles at the center divided into two groups. Under a microscope <*5.01*>, a transverse section of the midvein of lamina reveals the adaxial side protruding remarkably, with collenchyma cells beneath epidermis; vascular bundles at the center arranged in fan-shape. Under a microscope <*5.01*>, a transverse section of stem reveals several-cells-layered collenchyma beneath epidermis, occasionally with cork layer developed; beneath cortex, collateral vascular bundles arranged in a circle, phloem fibers in groups observed at the outer portion of phloem; oil droplets observed in parenchymat cells of cortex, needle, solitary or columnar crystals of calcium oxalate in parenchyma cells of pith.

Identification To 0.5 g of pulverized Pogostemon Herb, add 5 mL of methanol, shake for 3 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of hexane and acetone (9:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes; a blue-purple spot appears at an *R*f value of about 0.4.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 13.0%.

Acid-insoluble ash <*5.01*> Not more than 3.0%.

Essential oil content <*5.01*> When the test is performed with 50.0 g of pulverized Pogostemon Herb in a flask with 1 mL of silicon resin added, the essential oil content is not less than 0.3 mL.

Containers and storage Containers—Well-closed containers.

Polygala Root

Polygalae Radix

オンジ

Polygala Root is the root or the root bark of *Polygala tenuifolia* Willdenow (*Polygalaceae*).

Description Thin, long and bent, cylindrical or tubular root; main root, 10 – 20 cm in length, 0.2 – 1 cm in diameter, sometimes with one to several lateral roots; externally light grayish brown, with coarse longitudinal wrinkles, and with deep lateral furrows cracked to some degree here and there; brittle, and fractured surface not fibrous; under a magnifying glass, margin of the transverse section irregularly undulate; cortex, comparatively thick, with large cracks here and there; xylem usually round to elliptical, light brown in color, and often tears in a wedge-like shape.

Odor, slight; taste, slightly acrid.

Identification **(1)** Shake vigorously 0.5 g of pulverized Polygala Root with 10 mL of water: a lasting fine foam is produced.

(2) To 1.0 g of pulverized Polygala Root add 10 mL of a solution of sodium hydroxide (1 in 10), and heat under a reflux condenser for 20 minutes. After cooling, add 10 mL of dilute hydrochloric acid, and shake. After cooling, add 10 mL of 1-butanol, shake for 10 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, water and acetic acid (100) (20:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 10 minutes: a red-brown to light brown spot appears at an *R*f value of about 0.35.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Polygala Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Polygala Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Stem—When perform the test of foreign matter <*5.01*>, the amount of the stems contained in Polygala Root does not exceed 10.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than the stems is not more than 1.0%.

(5) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 6.0%.

Containers and storage Containers—Well-closed containers.

Powdered Polygala Root

Polygalae Radix Pulverata

オンジ末

Powdered Polygala Root is the powder of Polygala Root.

Description Powdered Polygala Root occurs as a light yellow-grayish brown powder. It has a slight odor and a slightly acrid taste.

Under a microscope <*5.01*>, Powdered Polygala Root reveals fragments of cork layers, pitted vessels, reticulate vessels and tracheids; fragments of xylem fibers and xylem parenchyma cells with a small number of simple pits; fragments of parenchyma cells containing substances such as oil droplets, rosette aggregates and solitary crystals of calcium oxalate. Oil drop-like contents stained red with sudan III TS.

Identification **(1)** Shake vigorously 0.5 g of Powdered Polygala Root with 10 mL of water: a lasting fine foam is produced.

(2) To 1.0 g of Powdered Polygala Root add 10 mL of a solution of sodium hydroxide (1 in 10), and heat under a reflux condenser for 20 minutes. After cooling, add 10 mL of dilute hydrochloric acid, and shake. After cooling, add 10 mL of 1-butanol, shake for 10 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, water and acetic acid (100) (20:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 10 minutes: a red-brown to light brown spot appears at an *R*f value of about 0.35.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Polygala Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Polygala Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Polygala Root does not show stone cells or starch grains.

(4) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Total ash <*5.01*> Not more than 6.0%.

Containers and storage Containers—Well-closed containers.

Polygonatum Rhizome

Polygonati Rhizoma

オウセイ

Polygonatum Rhizome is the rhizome of *Polygonatum kingianum* Collett et Hemsley, *Polygonatum sibiricum* Redouté, *Polygonatum cyrtonema* Hua or *Polygonatum falcatum* A. Gray (*Liliaceae*), usually after being steamed.

Description Irregularly cylindrical rhizome, 3 – 10 cm in length, 0.5 – 3 cm in diameter; or irregular massive rhizome, 5 – 10 cm in length, 2 – 6 cm in diameter, occasionally branched; both rhizomes with many cyclic nodes and longitudinally striate; externally yellow-brown to black-brown; stem scars, round, concave at their center, and protuberant on the upper surface; root scars on the lower surface; cut surface flat and horny.

Odor, slight; taste, slightly sweet.

Under a microscope <*5.01*>, a transverse section of the rhizome reveals an epidermis coated with cuticle; inside of epidermis parenchyma lie; numerous vascular bundles and mucilage cells scattered in parenchyma; vascular bundles collateral or amphivasal concentric; mucilage cells contain raphides of calcium oxalate.

Identification **(1)** To 0.5 g of fine cutted Polygonatum Rhizome add 2 mL of acetic anhydride, warm on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add gently 0.5 mL of sulfuric acid: a red-brown color appears at the zone of contact.

(2) To 1.0 g of fine cutted Polygonatum Rhizome add 10 mL of dilute hydrochloric acid, boil gently for 2 minutes, and filter. Neutralize the filtrate with sodium hydroxide TS. To 3 mL of this solution add 1 mL of Fehling's TS, and warm: red precipitates appear.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Polygonutum Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Polygonutum Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Polygonum Root

Polygoni Multiflori Radix

カシュウ

Polygonum Root is the root of *Polygonum multiflorum* Thunberg (*Polygonaceae*), often being cut into round slices.

Description Polygonum Root is nearly fusiform, 10 – 15 cm in length, 2 – 5 cm in diameter; externally red-brown to dark brown; roughly wrinkled; a cross section light red-brown or light grayish brown, with numerous abnormal vascular bundles scattering irregularly around the large vascular bundles near center; heavy and hard in texture.

Odor, slight and characteristic; taste, astringent and slightly bitter.

Under a microscope <*5.01*>, transverse section reveals the outermost layer to be several cells thick and composed of cork; cork cells contain brown substances; cortex composed of parenchyma; abnormal vascular bundles, exhibiting a ring of cambium; xylem lies inside of the cambium, and phloem outside; fibers lie outside the phloem; central portion of root lignified; parenchymatous cells contain aggregated crystals of calcium oxalate, and both simple and 2- to 8-compound starch grains; navel of starch grain obvious.

Identification To 1 g of pulverized Polygonum Root add 10 mL of methanol, shake for 15 minutes, and filter. Evaporate the filtrate to dryness, dissolve the residue in 2 mL of methanol, and use this as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, water, methanol and acetic acid (100) (200:10:10:3) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): a fluorescent blue-white spot appears at an *R*f value of about 0.3.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Polygonum Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Polygonum Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 14.0% (6 hours).

Total ash <*5.01*> Not more than 5.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 17.0%.

Containers and storage Containers—Well-closed containers.

Polyporus Sclerotium

Polyporus

チョレイ

Polyporus Sclerotium is the sclerotium of *Polyporus umbellatus* Fries (*Polyporaceae*).

Description Irregularly shaped mass, usually 5 – 15 cm in length; externally black-brown to grayish brown, with numerous dents and coarse wrinkles; breakable; fractured surface rather soft and cork-like, and almost white to light brown in color, and a white speckled pattern on the inner region; light in texture.

Practically odorless and tasteless.

Identification Warm, while shaking, 0.5 g of pulverized Polyporus Sclerotium with 5 mL of acetone on a water bath for 2 minutes, filter, and evaporate the filtrate to dryness. Dissolve the residue in 5 drops of acetic anhydride, and add 1 drop of sulfuric acid: a red-purple color develops, and immediately changes to dark green.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Polyporus Sclerotium according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Polyporus Sclerotium according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 16.0%.

Acid-insoluble ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Well-closed containers.

Powdered Polyporus Sclerotium

Polyporus Pulveratus

チョレイ末

Powdered Polyporus Sclerotium is the powder of the Polyporus Sclerotium.

Description Powdered Polyporus Sclerotium occurs as a light grayish brown to light brown powder. Almost odorless and tasteless.

Under a microscope <*5.01*>, Powdered Polyporus Sclerotium reveals hypha, 1 to 2 μm, rarely up to 13 μm in diameter, and colorless transparent; granule strongly refracting light; and a few mucilage plates; sometimes fragments of false tissue consisting of them; somewhat brown false tissues; and solitary crystal of calcium oxalate. Solitary crystal is 10 to 40 μm in diameter, sometimes 100 μm in diameter.

Identification Warm, while shaking, 0.5 g of Powdered Polyporus Sclerotium with 5 mL of acetone on a water bath for 2 minutes, filter and evaporate the filtrate to dryness. Dissolve the residue in 5 drops of acetic anhydride, and add 1 drop of sulfuric acid: a red-purple color develops, and immediately changes to dark green.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Polyporus Sclerotium according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Polyporus Sclerotium according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 16.0%.

Acid-insoluble ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Tight containers.

Poria Sclerotium

Poria

ブクリョウ

Poria Sclerotium is the sclerotium of *Wolfiporia cocos* Ryvarden et Gilbertson (*Poria cocos* Wolf) (*Polyporaceae*), from which usually the external layer has been mostly removed.

Description Mass, about 10 – 30 cm in diameter, up to 0.1 – 2 kg in mass; usually it appears as broken or chipped pieces; white or slightly reddish white; sclerotium with remaining outer layer is dark brown to dark red-brown in color, coarse, which fissures; hard in texture, but brittle.

Almost odorless, almost tasteless, and slightly mucous.

Identification **(1)** Warm 1 g of pulverized Poria Sclerotium with 5 mL of acetone on a water bath for 2 minutes with shaking, and filter. Evaporate the filtrate to dryness, dissolve the residue in 0.5 mL of acetic anhydride, and add 1 drop of sulfuric acid: a light red color develops, which changes immediately to dark green.

(2) To a cut surface or powder of Poria Sclerotium add 1 drop of iodine TS: a deep red-brown color is produced.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Poria Sclerotium according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Poria Sclerotium according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Powdered Poria Sclerotium

Poria Pulveratum

ブクリョウ末

Powdered Poria Sclerotium is the powder of Poria Sclerotium.

Description Powdered Poria Sclerotium occurs as a white to grayish white powder. It is almost odorless and almost tasteless, but is slightly mucous.

Under a microscope <*5.01*>, Powdered Poria Sclerotium reveals colorless and transparent hyphae strongly refracting light, and fragments of false tissue consisting of granules and mucilage plates. Thin hyphae, 2 – 4 μm in diameter; thick ones, usually 10 – 20 μm, up to 30 μm.

Identification **(1)** Warm 1 g of Powdered Poria Sclerotium with 5 mL of acetone on a water bath for 2 minutes with shaking, and filter. Evaporate the filtrate to dryness, dissolve the residue in 0.5 mL of acetic anhydride, and add 1 drop of sulfuric acid: a light red color develops, which changes immediately to dark green.

(2) To Powdered Poria Sclerotium add 1 drop of iodine TS: a deep red-brown color is produced.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Poria Sclerotium according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Poria Sclerotium according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Poria Sclerotium does not show starch grains.

Total ash <*5.01*> Not more than 1.0%.

Containers and storage Containers—Well-closed containers.

Prepared Glycyrrhiza

Glycyrrhizae Radix Praeparata

シャカンゾウ

Prepared Glycyrrhiza is prepared by roasting Glycyrrhiza.

It contains not less than 2.0% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), calculated on the basis of dried material.

Description Usually cut; external surface dark brown to dark red-brown and with longitudinal wrinkles; cut surface brown to light yellow-brown; in case periderm fallen off, external surface brown to light yellow-brown and fibrous; on transversely cut surface cortex and xylem almost distinctly defined, and exhibits radial structure; sometimes radial cleft observed.

Odor, fragrant; taste sweet, followed by slight bitterness.

Identification To 2.0 g of pulverized Prepared Glycyrrhiza add 10 mL of ethyl acetate, shake for 15 minutes, centrifuge, and separate the ethyl acetate layer. Shake the residue with 5 mL of ethyl acetate and 5 mL of 0.1 mol/L hydrochloric acid TS for 15 minutes, centrifuge, and use the ethyl acetate layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (7:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, heat the plate at 105°C for 3 minutes, and allow to cool: a red-purple spot is observed at an *R*f value of about 0.6.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Prepared Glycyrrhiza according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g

of pulverized Prepared Glycyrrhiza according to Method 4, and perform the test (not more than 5 ppm).

(3) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 8.0% (6 hours).

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 25.0%.

Assay Weigh accurately about 0.5 g of pulverized Prepared Glycyrrhiza in a glass-stoppered centrifuge tube, add 70 mL of dilute ethanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 25 mL of dilute ethanol, and proceed in the same manner. Combine all the extracts, add dilute ethanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 25 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in dilute ethanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: Adjust so that the retention time of glycyrrhizic acid is about 15 minutes.

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak with the relative retention time of about 0.9 to glycyrrhizic acid, and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Processed Aconite Root

Aconiti Radix Processa

ブシ

Processed Aconite Root is the tuberous root of *Aconitum carmichaeli* Debeaux or *Aconitum japonicum* Thunberg (*Ranunculaceae*) prepared by the following processes.

Process 1: Autoclaving. [Processed Aconite Root 1]

Process 2: Heating or autoclaving after rinsing in salt or rock salt solution. [Processed Aconite Root 2]

Process 3: Treating with calcium hydroxide after rinsing in salt solution. [Processed Aconite Root 3]

Processed Aconite Root 1, Processed Aconite Root 2 and Processed Aconite Root 3 contain the total alkaloid [as benzoyl aconin ($C_{32}H_{45}NO_{10}$: 603.70)] of not less than 0.7% and not more than 1.5%, not less than 0.1% and not more than 0.6%, and not less than 0.5% and not more than 0.9%, calculated on the dried bases, respectively.

The label indicates the treating process.

Description **1)** Processed Aconite Root 1: Cut pieces irregularly polygonal, less than 10 mm in diameter; externally dark grayish brown to black-brown; hard in texture; cut surface flat, light brown to dark brown, usually horny and lustrous.

Odor, weak and characteristic.

Under a microscope <*5.01*>, transverse and longitudinal sections reveal pitted, scaraliform, reticulate and spiral vessels; starch grains in parenchymatous cells usually gelatinized but sometimes not gelatinized; starch grains, simple, spherical or ellipsoid, 2 – 25 μm in diameter, or 2- to a dozen or so- compound, hilum of starch grain distinct.

2) Processed Aconite Root 2: Nearly obconical root, 15 – 30 mm in length, 12 – 16 mm in diameter, slices cut longitudinally or transversely, 20 – 60 mm in length, 15 – 40 mm in width, and 200 – 700 μm in thickness, or cut pieces irregularly polygonal, less than 12 mm in diameter; externally light brown to dark brown or yellow-brown; hard in texture, usually without wrinkles; cut surface flat, light brown to dark brown or yellow-white to light yellow-brown, usually horny, semi-transparent and lustrous.

Odor, weak and characteristic.

Under a microscope <*5.01*>, transverse and longitudinal sections reveal metaderm, primary cortex, endodermis, secondary cortex, cambium, and xylem; primary cortex contains oblong to oblong-square sclerenchymatous cells, 30 – 75 μm in short axis, 60 – 150 μm in long axis; endodermis single layered cell, endodermal cells elongated in tangential direction; cambium, star shaped or irregular polygons to orbicular; a group of vessel in xylem v-shaped; sometimes isolated ring of cambium appears in secondary cortex or in pith; vessels, pitted, scaraliform, reticulate and spiral; starch grains in parenchymatous cells gelatinized.

3) Processed Aconite Root 3: Cut pieces irregularly polygonal, less than 5 mm in diameter; externally grayish brown; hard in texture; cut surface flat, light grayish brown to grayish white, not lustrous.

Odor, weak and characteristic.

Under a microscope <*5.01*>, transverse and longitudinal sections reveal pitted, scaraliform, reticulate and spiral ves-

sels; starch grains, simple, spherical or ellipsoid, 2 – 25 μm in diameter, or 2- to a dozen or so- compound, hilum of starch grain distinct.

Identification To 3 g of pulverized Processed Aconite Root in a glass-stoppered centrifuge tube add 20 mL of diethyl ether and 2 mL of ammonia TS, shake for 10 minutes, centrifuge, and take the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), dissolve the residue in 1 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of benzoylmesaconine hydrochloride for thin-layer chromatography in 5 mL of ethanol (99.5), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, ethanol (99.5) and ammonia water (28) (40:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, air-dry the plate, and spray evenly sodium nitrite TS: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Processed Aconite Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Processed Aconite Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Aconitum diester alkaloids (aconitine, jesaconitine, hypaconitine and mesaconitine)—Weigh accurately about 0.5 g of pulverized Processed Aconite Root, put in a glass-stoppered centrifuge tube, suspend in 3.0 mL of water by shaking, and add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, evaporate under low pressure (in vacuo), and dissolve the residue with exactly 10 mL of a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1). Centrifuge and use the supernatant liquid as the sample solution. Perform the test with exactly 20 μL each of the sample solution and aconitum diester alkaloids standard solution for purity as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the heights of the peaks corresponding to aconitine, H_{TA} and H_{SA}, jesaconitine, H_{TJ} and H_{SJ}, hypaconitine, H_{TH} and H_{SH}, and mesaconitine, H_{TM} and H_{SM}, respectively, and calculate the amounts of them by the following formulae: the amounts of aconitine, jesaconitine, hypaconitine and mesaconitine per g calculated on the dried basis are not more than 60 μg, 60 μg, 280 μg and 140 μg, respectively, and the total amount of them is not more than 450 μg.

$$\text{Amount } (\mu g) \text{ of aconitine} = C_{SA}/M \times H_{TA}/H_{SA} \times 10$$

$$\text{Amount } (\mu g) \text{ of jesaconitine} = C_{SJ}/M \times H_{TJ}/H_{SJ} \times 10$$

$$\text{Amount } (\mu g) \text{ of hypaconitine} = C_{SH}/M \times H_{TH}/H_{SH} \times 10$$

$$\text{Amount } (\mu g) \text{ of mesaconitine} = C_{SM}/M \times H_{TM}/H_{SM} \times 10$$

C_{SA}: Concentration (μg/mL) of aconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SJ}: Concentration (μg/mL) of jesaconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SH}: Concentration (μg/mL) of hypaconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SM}: Concentration (μg/mL) of mesaconitine for purity in aconitum diester alkaloids standard solution for purity

M: Amount (g) of Processed Aconite Root taken, calculated on the dried basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for aconitine, hypaconitine and mesaconitine; 254 nm for jesaconitine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer solution for processed aconite root and tetrahydrofuran (183:17).

Flow rate: Adjust so that the retention time of mesaconitine is about 31 minutes.

System suitability—

System performance: When the procedure is run with 20 μL of aconitum diester alkaloids standard solution for purity under the above operating conditions, using 254 nm, mesaconitine, hypaconitine, aconitine and jesaconitine are eluted in this order, and each resolution between their peaks is not less than 1.5, respectively.

System repeatability: To 1 mL of aconitum diester alkaloids standard solution for purity add a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make 10 mL. When the test is repeated 6 times with 20 μL of this solution under the above operating conditions, using 231 nm, the relative standard deviation of the peak height of mesaconitine is not more than 1.5%.

Loss on drying <*5.01*> Not more than 15.0% (6 hours).

Total ash <*5.01*>

Processed Aconite Root 1: Not more than 4.0%.
Processed Aconite Root 2: Not more than 12.0%.
Processed Aconite Root 3: Not more than 19.0%.

Acid-insoluble ash <*5.01*> Not more than 0.9%.

Assay Weigh accurately about 2 g of pulverized Processed Aconite Root, put in a glass-stoppered centrifuge tube, and add 1.6 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 0.8 mL of ammonia TS and 20 mL of diethyl ether, and proceed as above. Repeat this process more three times. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 5 mL of ethanol (99.5), add 30 mL of freshly boiled and cooled water, and titrate <*2.50*> with 0.01 mol/L hydrochloric acid VS until the color of the solution changes from green to gray-blue through blue-green (indicator: 3 drops of methyl red-methylene blue TS). Perform a blank determination in the same manner, and make any necessary correction.

Each mL of 0.01 mol/L hydrochloric acid VS
= 6.037 mg of total alkaloid [as benzoylaconine ($C_{32}H_{45}NO_{10}$)]

Containers and storage Containers—Well-closed containers.

Powdered Processed Aconite Root

Aconiti Radix Processa et Pulverata

ブシ末

Powdered Processed Aconite Root is the powder of Processed Aconite Root prepared by the process 1 or process 2, the powder of Processed Aconite Root prepared by process 1, or the powder of Processed Aconite Root prepared by the process 1 to which Corn Starch or Lactose Hydrate is added.

Process 1: Autoclaving. [Powdered Processed Aconite Root 1]

Process 2: Heating or autoclaving after rinsing in salt or rock salt solution. [Powdered Processed Aconite Root 2]

Powdered Processed Aconite Root 1 and Powdered Processed Aconite Root 2 contain the total alkaloid [as benzoyl aconin ($C_{32}H_{45}NO_{10}$: 603.70)] of not less than 0.4% and not more than 1.2%, and not less than 0.1% and not more than 0.3%, calculated on the dried bases, respectively.

The label indicates the treating process.

Description 1) Powdered Processed Aconite Root 1: Powdered Processed Aconite Root 1 occurs as a light grayish brown powder. It has a characteristic odor.

Under a microscope ⟨*5.01*⟩, Powered Processed Aconite Root 1 reveals gelatinized starch masses or starch grains and parenchymatous cells containing them, fragments of red-brown metaderm, fragments of pitted, scaraliform, reticulate and spiral vessels; also square to oblong-square sclerenchymatous cells, 30 – 150 μm in diameter, 100 – 250 μm in length, cell wall of sclerenchymatous cells, 6 – 12 μm in thickness; starch grains of *Aconitum carmichaeli Debeaux* or *Aconitum japonicum* Thunberg (*Ranunculaceae*) origin, simple, spherical or ellipsoid, 2 – 25 μm in diameter, or 2- to a dozen or so- compound, hilum of starch grain distinct.

2) Powdered Processed Aconite Root 2: Powdered Processed Aconite Root 2 occurs as a light yellow-white powder. It has a characteristic odor.

Under a microscope ⟨*5.01*⟩, Powered Processed Aconite Root 2 reveals gelatinized starch masses and parenchymatous cells containing them, fragments of red-brown metaderm, fragments of pitted, scaraliform, reticulate and spiral vessels; also square to oblong-square sclerenchymatous cells, 30 – 150 μm in diameter, 100 – 250 μm in length, cell wall of sclerenchymatous cells, 6 – 12 μm in thickness.

Identification To 3 g of Powdered Processed Aconite Root in a glass-stoppered centrifuge tube add 20 mL of diethyl ether and 2 mL of ammonia TS, shake for 10 minutes, centrifuge, and take the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), dissolve the residue in 1 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of benzoylmesaconine hydrochloride for thin-layer chromatography in 5 mL of ethanol (99.5), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, ethanol (99.5) and ammonia water (28) (40:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, air-dry the plate, and spray evenly sodium nitrite TS: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity (1) Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of Powdered Processed Aconite Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of Powdered Processed Aconite Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Aconitum diester alkaloids (aconitine, jesaconitine, hypaconitine and mesaconitine)—Weigh accurately about 0.5 g of Powdered Processed Aconite Root, put in a glass-stoppered centrifuge tube, suspend in 3.0 mL of water by shaking, and add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), and dissolve the residue with exactly 10 mL of a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1). Centrifuge this solution, and use the supernatant liquid as the sample solution. Perform the test with exactly 20 μL each of the sample solution and aconitum diester alkaloids standard solution for purity as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the heights of the peaks corresponding to aconitine, H_{TA} and H_{SA}, jesaconitine, H_{TJ} and H_{SJ}, hypaconitine, H_{TH} and H_{SH}, and mesaconitine, H_{TM} and H_{SM}, respectively, and calculate the amounts of them by the following formulae: the amounts of aconitine, jesaconitine, hypaconitine and mesaconitine per g calculated on the dried basis are not more than 55 μg, 40 μg, 55 μg and 120 μg, respectively, and the total amount of them is not more than 230 μg.

$$\text{Amount } (\mu g) \text{ of aconitine} = C_{SA}/M \times H_{TA}/H_{SA} \times 10$$

$$\text{Amount } (\mu g) \text{ of jesaconitine} = C_{SJ}/M \times H_{TJ}/H_{SJ} \times 10$$

$$\text{Amount } (\mu g) \text{ of hypaconitine} = C_{SH}/M \times H_{TH}/H_{SH} \times 10$$

$$\text{Amount } (\mu g) \text{ of mesaconitine} = C_{SM}/M \times H_{TM}/H_{SM} \times 10$$

C_{SA}: Concentration (μg/mL) of aconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SJ}: Concentration (μg/mL) of jesaconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SH}: Concentration (μg/mL) of hypaconitine for purity in aconitum diester alkaloids standard solution for purity

C_{SM}: Concentration (μg/mL) of mesaconitine for purity in aconitum diester alkaloids standard solution for purity

M: Amount (g) of Powdered Processed Aconitine Root taken, calculated on the dried basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for aconitine, hypaconitine and mesaconitine; 254 nm for jesaconitine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer solution for processed aconite root and tetrahydrofuran (183:17).

Flow rate: Adjust so that the retention time of mesaconitine is about 31 minutes.

System suitability—

System performance: When the procedure is run with 20 μL of aconitum diester alkaloids standard solution for purity under the above operating conditions, using 254 nm, mesaconitine, hypaconitine, aconitine and jesaconitine are eluted in this order, and each resolution between their peaks is not less than 1.5, respectively.

System repeatability: To 1 mL of aconitum diester alkaloids standard solution for purity add a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make 10 mL. When the test is repeated 6 times with 20 μL of this solution under the above operating conditions, using 231 nm, the relative standard deviation of the peak height of mesaconitine is not more than 1.5%.

Loss on drying ⟨*5.01*⟩ Not more than 11.0% (6 hours).

Total ash ⟨*5.01*⟩

Powdered Processed Aconite Root 1: Not more than 4.0%.

Powdered Processed Aconite Root 2: Not more than 7.0%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.7%.

Assay Weigh accurately about 2 g of Powdered Processed Aconite Root, put in a glass-stoppered centrifuge tube, and add 1.6 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 0.8 mL of ammonia TS and 20 mL of diethyl ether, and proceed as above. Repeat this process more three times. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 5 mL of ethanol (99.5), add 30 mL of freshly boiled and cooled water, and titrate ⟨*2.50*⟩ with 0.01 mol/L hydrochloric acid VS until the color of the solution changes from green to gray-blue through blue-green (indicator: 3 drops of methyl red-methylene blue TS). Perform a blank determination in the same manner, and make any necessary correction.

Each mL of 0.01 mol/L hydrochloric acid VS
= 6.037 mg of total alkaloid [as benzoylaconine ($C_{32}H_{45}NO_{10}$)]

Containers and storage Containers—Well-closed containers.

Processed Ginger

Zingiberis Rhizoma Processum

カンキョウ

Processed Ginger is the rhizome of *Zingiber officinale* Roscoe (*Zingiberaceae*), after being passed through hot water or being steamed.

It contains not less than 0.10% of [6]-shogaol, calculated on the basis of dried material.

Description Irregularly compressed and often branched massive rhizome; branched parts slightly curved ovoid or oblong- ovoid, 2 – 4 cm in length, and 1 – 2 cm in diameter; external surface grayish yellow to grayish yellow-brown, with wrinkles and ring node; fractured surface brown to dark brown, transparent and horny; under a magnifying glass, a transverse cut surface reveals cortex and stele distinctly divided; vascular bundles scattered throughout the surface.

Odor, characteristic; taste, extremely pungent.

Under a microscope ⟨*5.01*⟩, a transverse section reveals cork layer, cortex and stele in this order from the outside; cortex and stele, divided by an endodermis, composed of parenchyma; vascular bundles surrounded by fibers scattered in cortex and stele; oil cells contain yellow oily substances, scattered in parenchyma; parenchyma cells contain solitary crystals of calcium oxalate, and gelatinized starch.

Identification To 2 g of pulverized Processed Ginger add 5 mL of diethyl ether, shake for 10 minutes, filter, and use the filtrate as the sample solution (1). To the residue add 5 mL of methanol, proceed in the same manner as above, and use so obtained solution as the sample solution (2). Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution (1). Separately, dissolve 1 mg of sucrose in 2 mL of methanol, and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL each of the sample solution (1) and standard solution (1) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution (1) has the same color tone and *R*f value with the spot from the standard solution (1). Spot 10 μL each of the sample solution (2) and standard solution (2) on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of 1-butanol, water and acetic acid (100) (8:5:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1,3-naphthalenediol TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution (2) has the same color tone and *R*f value with the spot from the standard solution (2).

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 1.0 g of pulverized Processed Ginger according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Processed Ginger according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 15.0% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.0%.

Assay Weigh accurately about 1 g of pulverized Processed Ginger, place in a centrifuge tube, add 30 mL of the mobile phase, shake for 20 minutes, centrifuge, and separate the supernatant liquid. To the residue add 30 mL of the mobile phase, and repeat the extraction twice more. To the combined all extracts add the mobile phase to make exactly 100 mL, use this solution as the sample solution. Separately, weigh accurately about 5 mg of [6]-shogaol for assay, dissolve in the mobile phase to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-shogaol in each solution.

$$\text{Amount (mg) of [6]-shogaol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of [6]-shogaol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 225 nm).

Column: A stainless steel column 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of acetonitrile and water (3:2).

Flow rate: Adjust so that the retention time of [6]-shogaol is about 14 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-shogaol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-shogaol is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Prunella Spike

Prunellae Spica

カゴソウ

Prunella Spike is the spike of *Prunella vulgaris* Linné var. *lilacina* Nakai (*Labiatae*).

Description Spikes in nearly cylindrical and wheat ear-like shape, 3 – 6 cm in length, 1 – 1.5 cm in diameter, externally grayish brown; spikes composed of a floral axis having numerous bracts and calyxes; corollas often remaining on the upper part; a calyx usually enclosing four mericarps; bract, cordate to eccentric, and exhibiting white hairs on the vein, as on the calyx; light in texture.

Almost odorless and tasteless.

Purity **(1)** Stem—When perform the test of foreign matter <*5.01*>, the amount of the stems contained in Prunella Spike does not exceed 5.0%.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than the stems contained in Prunella Spike does not exceed 1.0%.

Total ash <*5.01*> Not more than 13.0%.

Acid-insoluble ash <*5.01*> Not more than 5.0%.

Containers and storage Containers—Well-closed containers.

Pueraria Root

Puerariae Radix

カッコン

Pueraria Root is the root of *Pueraria lobata* Ohwi (*Leguminosae*), from which periderm has been removed.

It contains not less than 2.0% of puerarin ($C_{21}H_{20}O_9$: 416.38), calculated on the basis of dried material.

Description Usually cut into small pieces of irregular hexagons of about 0.5 cm cube, or cut into longitudinally plate-like pieces 20 – 30 cm in length, 5 – 10 cm in width, and about 1 cm in thickness; externally light grayish yellow to grayish white; transverse section showing concentric annulate ring or part of it formed by abnormal growth of cambium. Under a magnifying glass, phloem light grayish yellow in color; in xylem, numerous vessels appearing as small dots; medullary rays slightly dented; vertical section showing longitudinal patterns formed alternately by fibrous xylem and parenchyma; easily breakable lengthwise, and its section extremely fibrous.

Almost odorless; taste, at first slightly sweet, followed by a slight bitterness.

Under a microscope <*5.01*>, a transverse section reveals fiber bundles accompanied by crystal cells in phloem; distinct vessels and xylem fibers in xylem; starch grains numerous in parenchyma, mainly composed of polygonal simple grains, rarely 2- to 3-compound grains, 2 – 18 μm, mostly 8 – 12 μm, in size, with hilum or cleft in the center, and also with striae.

Identification To 2 g of pulverized Pueraria Root add 10 mL of methanol, shake for 3 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Puerarin RS or puerarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (12:2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Pueraria Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g

of pulverized Pueraria Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not less than 13.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 6.0%.

Assay Weigh accurately about 0.3 g of pulverized Pueraria Root, add 50 mL of diluted methanol (1 in 2), and heat under a reflex condenser for 30 minutes, cool, and filter. To the residue add 50 mL of diluted methanol (1 in 2), and perform as the same as above. Combine the filtrates, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Puerarin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of puerarin in each solution.

$$\text{Amount (mg) of puerarin } (C_{21}H_{20}O_9) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Puerarin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 250 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (9:1).

Flow rate: Adjust so that the retention time of puerarin is about 15 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry coefficient of the peak of puerarin are not less than 3000 and not more than 2.0, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of puerarin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Quercus Bark

Quercus Cortex

ボクソク

Quercus Bark is the bark of *Quercus acutissima* Carruthers, *Quercus serrata* Murray, *Quercus mongolica* Fischer ex Ledebour var. *crispula* Ohashi or *Quercus variabilis* Blume (*Fagaceae*).

Description Plate-like or semi-tubular pieces of bark, 5 – 15 mm in thickness; externally grayish brown to dark brown, with thick periderm and longitudinal coarse splits; internally brown to light brown, with longitudinal ridges, the transverse section brown to light brown, white small spots composed of stone cells in groups observed sporadically.

Almost odorless, tasteless.

Under a microscope ⟨*5.01*⟩, a transverse section reveals a cork layer with scattered cork stone cells; in secondary cortex fiber bundles lined almost stepwise, large groups of stone cells arranged irregularly; in parenchyma aggregated crystals of calcium oxalate scattered; adjacent to stone cells and fiber cells, cells containing solitary crystals of calcium oxalate observed, and these cells form crystal cell rows in a longitudinal section.

Identification To 2 g of pulverized Quercus Bark, add 10 mL of ethyl acetate, shake for 10 minutes, and centrifuge to remove the supernatant liquid. Add 10 mL of acetone to the residue, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate, methanol and water (7:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): Two consecutive blue to blue-white fluorescent spots in different color tone are observed at an *R*f value of about 0.4. Then, spray evenly diluted sulfuric acid on the plate, heat the plate at 105°C. Examine under ultraviolet light (main wavelength: 365 nm): one of these spots produces blue to blue-white fluorescence.

Loss on drying ⟨*5.01*⟩ Not more than 11.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 8.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Rape Seed Oil

Oleum Rapae

ナタネ油

Rape Seed Oil is the fixed oil obtained from the seed of *Brassica napus* Linné or *Brassica rapa* Linné var. *oleifera* De Candolle (*Cruciferae*).

Description Rape Seed Oil is a clear, pale yellow, slightly viscous oil. It is odorless or has a slight odor and a mild taste.

It is miscible with diethyl ether and with petroleum diethyl ether. It is slightly soluble in ethanol (95).

Specific gravity d_{25}^{25}: 0.906 – 0.920

Acid value ⟨*1.13*⟩ Not more than 0.2.

Saponification value ⟨*1.13*⟩ 169 – 195

Unsaponifiable matters ⟨*1.13*⟩ Not more than 1.5%.

Iodine value ⟨*1.13*⟩ 95 – 127

Containers and storage Containers—Tight containers.

Red Ginseng

Ginseng Radix Rubra

コウジン

Red Ginseng is the root of *Panax ginseng* C. A. Meyer (*Panax schinseng* Nees) (*Araliaceae*), after being steamed.

It contains not less than 0.10% of ginsenoside Rg_1 ($C_{42}H_{72}O_{14}$: 801.01) and not less than 0.20% of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), calculated on the basis of dried material.

Description Thin and long cylindrical to fusiform root, often branching out into 2 to 5 lateral roots from the middle; 5 – 25 cm in length, main root 0.5 – 3 cm in diameter; externally light yellow-brown to red-brown, and translucent and with longitudinal wrinkles; crown somewhat constricted, and sometimes with short remains of stem; fractured surface flat; horny and hard in texture.

Odor, characteristic; taste, at first slightly sweet, followed by a slight bitterness.

Identification **(1)** To 0.2 g of pulverized Red Ginseng add 2 mL of acetic anhydride, warm on a water bath for 2 minutes, and filter. To 1 mL of the filtrate add gently 0.5 mL of sulfuric acid to make two layers: a red-brown color develops at the zone of contact.

(2) To 2.0 g of pulverized Red Ginseng add 10 mL of water and 10 mL of 1-butanol, shake for 15 minutes, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of ginsenoside Rg_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (14:5:4) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Red Ginseng according to Method 4, and perform the test. Prepare the control solution with 1.5 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 1.0 g of pulverized Red Ginseng according to Method 4, and perform the test (not more than 2 ppm).

(3) Foreign matter <*5.01*>—The amount of stems and other foreign matter contained in Red Ginseng does not exceed 2.0%.

(4) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 15.5% (6 hours).

Total ash <*5.01*> Not more than 4.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 18.0%.

Assay **(1)** Ginsenoside Rg_1—Weigh accurately about 1 g of pulverized Red Ginseng, put in a glass-stoppered centrifuge tube, add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. Repeat the procedure with the residue using 15 mL of diluted methanol (3 in 5), combine the supernatant liquids, and add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of dilute sodium hydroxide TS, allow to stand for 30 minutes, add 3 mL of 0.1 mol/L hydrochloric acid TS and diluted methanol (3 in 5) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rg_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg) dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rg_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rg_1\ (C_{42}H_{72}O_{14}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Ginsenoside Rg_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of ginsenoside Rg_1 is about 25 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rg_1 RS and ginsenoside Re in diluted methanol (3 in 5) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rg_1 and ginsenoside Re are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rg_1 is not more than 1.5%.

(2) Ginsenoside Rb_1—Use the sample solution obtained in (1) as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (3 in 5) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water and acetonitrile (7:3).

Flow rate: Adjust so that the retention time of ginsenoside Rb_1 is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of Ginsenoside Rb_1 RS and ginsenoside Rc in diluted methanol (3 in 5) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, ginsenoside Rb_1 and ginsenoside Rc are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Rehmannia Root

Rehmanniae Radix

ジオウ

Rehmannia Root is the root of *Rehmannia glutinosa* Liboschitz var. *purpurea* Makino or *Rehmannia glutinosa* Liboschitz (*Scrophulariaceae*), with the application of steaming (prepared one: Juku-jio) or without it (non-prepared one: Kan-jio).

Description **1)** Kan-jio—Massive or fusiform root, narrow at one or both ends, 5 – 10 cm in length, 0.5 – 3.0 cm in diameter, sometimes broken or markedly deformed in shape; externally yellow-brown, black-brown or black, with deep, longitudinal wrinkles and constrictions; soft in texture; transversely cut surface yellow-brown, black-brown, or black and peripheral portion darker.

Odor, characteristic; taste, slightly sweet at first, followed by a slight bitterness.

Under a microscope <*5.01*>, a transverse section reveals 7 – 15 cellular layers of cork layer; cortex composed entirely of parenchyma; cells containing brown secretes scattered in cortex; xylem practically filled with parenchyma; vessels radially lined, mainly reticulate vessels.

2) Juku-jio—Irregularly massive root, or massive or fusiform root, narrow at one or both ends, 5 – 10 cm in length, 0.5 – 3.0 cm in diameter; externally black, usually lustrous, with deep, longitudinal wrinkles and constrictions; soft in texture and mucous; transversely cut surface black.

Odor, characteristic; taste, sweet at first, followed by a slight bitterness.

Under a microscope <*5.01*>, a transverse section reveals 7 – 15 cellular layers of cork layer; cortex composed entirely of parenchyma; cells containing brown secretes scattered in cortex; xylem practically filled with parenchyma, often parenchyma partially broken and gaps observed; vessels radially lined, mainly reticulate vessels.

Identification **1)** Kan-jio—Sake 0.5 g of the fine cutting of Rehmannia Root with 5 mL of water, add 20 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 2 mg of stachyose for thin-layer chromatography in 1 mL of a mixture of water and methanol (1:1), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 2-propanol, water and methanol (3:2:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1,3-naphthalenediol TS on the plate, heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution. When further heat for more than 5 minutes, a blue spot is not observed at just lower than the spot mentioned above, or even appears it is only few.

2) Juku-jio—Sake 0.5 g of the fine cutting of Rehmannia Root with 5 mL of water, add 20 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 2 mg of fructose for thin-layer chromatography in 1 mL of a mixture of water and methanol (1:1), and use this solution as the standard solution (1). Separately, dissolve 3 mg of manninotriose for thin-layer chromatography in 1 mL of a mixture of water and methanol (1:1), and use this solution as the standard solution (2). Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and the standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 2-propanol, water and methanol (3:2:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1,3-naphthalenediol TS on the plate, heat the plate at 105°C for 10 minutes: the principal spot obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution (1). Furthermore, one of the several spots from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (2).

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Rehmannia Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Rehmannia Root according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Containers and storage Containers—Well-closed containers.

Rhubarb

Rhei Rhizoma

ダイオウ

Rhubarb is usually the rhizome of *Rheum palmatum* Linné, *Rheum tanguticum* Maximowicz, *Rheum officinale* Baillon, *Rheum coreanum* Nakai or their interspecific hybrids (*Polygonaceae*).

It contains not less than 0.25% of sennosides A ($C_{42}H_{38}O_{20}$: 862.74), calculated on the basis of dried material.

Description Ovoid, oblong-ovoid or cylindrical rhizome, often cut crosswise or longitudinally, 4 – 10 cm in diameter, 5 – 15 cm in length. In the case of Rhubarb without most part of cortex, the outer surface is flat and smooth, yellow-

brown to light brown in color, and sometimes exhibiting white, fine reticulations; thick and hard in texture. In the case of Rhubarb with cork layer, externally dark brown or red-black, and with coarse wrinkles; rough and brittle in texture. The fractured surface of Rhubarb is not fibrous; transverse section grayish brown, light grayish brown or brown in color, having patterns of black-brown tissue complicated with white and light brown tissues; near the cambium, the patterns often radiate, and in pith, consist of whirls of tissues radiated from the center of a small brown circle 1 – 3 mm in diameter and arranged in a ring or scattered irregularly.

Odor, characteristic; taste, slightly astringent and bitter; when chewed, gritty between the teeth, and coloring the saliva yellow.

Under a microscope <*5.01*>, the transverse section reveals mostly parenchyma cells; small abnormal cambium-rings scattered here and there in the pith; the cambium-rings produce phloem inside and xylem outside, accompanied with 2 to 4 cell rows of medullary rays containing brown-colored substances, and the rays run radiately from the center of the ring towards the outside forming whirls of tissues; parenchyma cells contain starch grains, brown-colored substances or crystal druses of calcium oxalate.

Identification To 1.0 g of pulverized Rhubarb add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography<*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution, and the spot develops a red color on spraying sodium carbonate TS.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Rhubarb according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Rhubarb according to Method 4, and perform the test (not more than 5 ppm).

(3) Rhaponticin—To 0.1 g of pulverized Rhubarb add exactly 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of rhaponticin for purity in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, 2-butanon, water and formic acid (10:7:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): the chromatogram obtained from the sample solution shows no spot having the same color tone and *R*f value with the blue fluorescent spot from the standard solution.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 13.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Assay Weigh accurately about 0.5 g of pulverized Rhubarb, add exactly 50 mL of a solution of sodium hydrogen carbonate (1 in 1000), shake for 30 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Sennoside A RS, (separately determine the water <*2.48*> by coulometric titration, using 10 mg) dissolve in a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 50 mL. Pipet 5 mL of this solution, add a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 20 mL and use this solution as the standard solution. Perform the test with exactly 10 μL of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of sennoside A in each solution.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4 – 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted acetic acid (100) (1 in 80) and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of sennoside A is about 15 minutes.

System suitability—

System performance: Dissolve 1 mg each of Sennoside A RS and naringin for thin-layer chromatography in a solution of sodium hydrogen carbonate (1 in 1000) to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, sennoside A and naringin are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Rhubarb

Rhei Rhizoma Pulveratum

ダイオウ末

Powdered Rhubarb is the powder of Rhubarb.

It contains not less than 0.25% of sennoside A ($C_{42}H_{38}O_{20}$: 862.74), calculated on the basis of dried materials.

Description Powdered Rhubarb occurs as a brown powder. It has a characteristic odor and a slightly astringent and bitter taste; is gritty between the teeth and colors the saliva yellow on chewing.

Under a microscope <5.01>, Powdered Rhubarb reveals starch grains, dark brown substances or druses of calcium oxalate, fragments of parenchyma cells containing them, and reticulate vessels. The starch grains are spherical, simple, or 2- to 4-compound grains. Simple grain, 3 – 18 μm in diameter, rarely 30 μm; crystal druses of calcium oxalate, 30 – 60 μm in diameter, sometimes exceeding 100 μm.

Identification To 1.0 g of Powdered Rhubarb add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution, and the spot develops a red color on spraying sodium carbonate TS.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of Powdered Rhubarb according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.40 g of Powdered Rhubarb according to Method 4, and perform the test (not more than 5 ppm).

(3) Rhaponticin—To 0.1 g of Powdered Rhubarb add exactly 10 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of rhaponticin for purity in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, 2-butanon, water and formic acid (10:7:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): the chromatogram obtained from the sample solution shows no spot having the same color tone and *R*f value with the blue fluorescent spot from the standard solution.

Loss on drying <5.01> Not more than 13.0% (6 hours).

Total ash <5.01> Not more than 13.0%.

Acid-insoluble ash <5.01> Not more than 2.0%.

Extract content <5.01> Dilute ethanol-soluble extract: not less than 30.0%.

Assay Weigh accurately about 0.5 g of Powdered Rhubarb, add exactly 50 mL of a solution of sodium hydrogen carbonate (1 in 1000), shake for 30 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Sennoside A RS, (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 50 mL. Pipet 5 mL of this solution, add a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of sennoside A in each solution.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column about 4 – 6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted acetic acid (100) (1 in 80) and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of sennoside A is about 15 minutes.

System suitability—

System performance: Dissolve 1 mg each of Sennoside A RS and naringin for thin-layer chromatography in a solution of sodium hydrogen carbonate (1 in 1000) to make 10 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, sennoside A and naringin are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Compound Rhubarb and Senna Powder

複方ダイオウ·センナ散

Method of preparation

Powdered Senna Leaves		110 g
Powdered Rhubarb		110 g
Sulfur		555 g
Magnesium Oxide		225 g
	To make	1000 g

Prepare as directed under Powders, with the above ingredients.

Description Compound Rhubarb and Senna Powder occurs as a yellow-brown powder, having a characteristic odor and a bitter taste.

Identification To 2 g of Compound Rhubarb and Senna Powder add 50 mL of water, warm on a water bath for 30 minutes, and filter. Add 2 drops of dilute hydrochloric acid to the filtrate, shake with two 20-mL portions of diethyl ether, and remove the diethyl ether layer. Add 5 mL of hydrochloric acid to the aqueous layer, and heat it on a water bath for 30 minutes. Cool, shake with 20 mL of diethyl ether, take the diethyl ether layer, add 10 mL of sodium hydrogen carbonate TS, and shake: the aqueous layer is red in color.

Containers and storage Containers—Well-closed contain-

ers.

Rikkunshito Extract

六君子湯エキス

Rikkunshito Extract contains not less than 2.4 mg of ginsenoside Rb_1 ($C_{54}H_{92}O_{23}$: 1109.29), not less than 16 mg and not more than 48 mg of hesperidin, and not less than 6 mg and not more than 18 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Ginseng	4 g	4 g
Atractylodes Rhizome	4 g	—
Atractylodes Lancea Rhizome	—	4 g
Poria Sclerotium	4 g	4 g
Pinellia Tuber	4 g	4 g
Citrus Unshiu Peel	2 g	2 g
Jujube	2 g	2 g
Glycyrrhiza	1 g	1 g
Ginger	0.5 g	0.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Rikkunshito Extract is a light brown to brown powder or black-brown viscous extract. It has an odor and a sweet and bitter taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(2) For preparation prescribed Atractylodes Rhizome—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 25 mL of hexane, shake, and centrifuge. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of hesperidin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, acetone, water and acetic acid (100) (10:6:3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,6-dibromo-*N*-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in an ammonia gas: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue spot from the standard solution (Citrus Unshiu Peel).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 30 μL of the sample solution and 5 μL of

the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Ginsenoside Rb_1—Weigh accurately about 2 g of the dry extract (or an amount of the viscous extract, equivalent to about 2 g of the dried substance), add 30 mL of diluted methanol (3 in 5), shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 15 mL of diluted methanol (3 in 5), repeat the same procedure. Combine the supernatant liquids, add diluted methanol (3 in 5) to make exactly 50 mL. Pipet 10 mL of this solution, add 3 mL of sodium hydroxide TS, allow to stand for 30 minutes, then add 3 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 20 mL. Apply exactly 5 mL of this solution to a column (about 10 mm in inside diameter and packed with 0.36 g of octadecylsilanized silica gel for pre-treatment (55 – 105 μm in particle size), washed just before use with methanol and then with diluted methanol (3 in 10)), and wash the column in sequence with 2 mL of diluted methanol (3 in 10), 1 mL of sodium carbonate TS and 10 mL of diluted methanol (3 in 10). Finally, elute with methanol to collect exactly 5 mL, and use this as the sample solution. Separately, weigh accurately about 10 mg of Ginsenoside Rb_1 RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of ginsenoside Rb_1 in each solution.

$$\text{Amount (mg) of ginsenoside } Rb_1\ (C_{54}H_{92}O_{23}) = M_S \times A_T/A_S \times 1/5$$

M_S: Amount (mg) of Ginsenoside Rb_1 RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 203 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with carbamoyl groups bound silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 60°C.

Mobile phase: A mixture of acetonitrile, water and phosphoric acid (400:100:1).

Flow rate: 1.0 mL per minute (the retention time of ginsenoside Rb_1 is about 16 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of ginsenoside Rb_1 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ginsenoside Rb_1 is not more than 1.5%.

(2) Hesperidin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted tetrahydrofuran (1 in 4), shake for 30 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of hesperidin for assay, previously dried in a desiccator (silica gel) for more than 24 hours, dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, add diluted tetrahydrofuran (1 in 4) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of hesperidin in each solution.

$$\text{Amount (mg) of hesperidin} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of hesperidin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (82:18:1).

Flow rate: 1.0 mL per minute (the retention time of hesperidin is about 15 minutes).

System suitability—

System performance: Dissolve 1 mg each of hesperidin for assay and naringin for thin-layer chromatography in diluted methanol (1 in 2) to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, naringin and hesperidin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of hesperidin is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10

mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Rose Fruit

Rosae Fructus

エイジツ

Rose Fruit is false or true fruit of *Rosa multiflora* Thunberg (*Rosaceae*).

Description The pseudocarp, spherical, ellipsoidal or spheroidal, 5 – 9.5 mm in length, 3.5 – 8 mm in diameter; the external surface red to dark brown in color, smooth and lustrous; often with peduncle about 10 mm in length at one end, and with pentagonal remains of calyx without sepal at the other end; internal wall of receptacle covered densely with silvery hairs; the interior containing 5 – 10 mature nuts; the nut, irregularly angular ovoid, about 4 mm in length, about 2 mm in diameter; external surface, light yellow-brown; obtuse at one end, and slightly acute at the other.

Odor, slight; taste of fruit receptacle, sweet and acid, and of nut, mucilaginous at first, later astringent, bitter and irritative.

Identification Boil gently 1 g of pulverized Rose Fruit with 20 mL of methanol for 2 minutes, and filter. To 5 mL of the filtrate add 0.1 g of magnesium in ribbon form and 0.5 mL of hydrochloric acid, and allow the mixture to stand: a light red to red color develops.

Purity Foreign matter <*5.01*>—The amount of the peduncle and other foreign matter contained in Rose Fruit is not more than 1.0%.

Total ash <*5.01*> Not more than 6.0%.

Containers and storage Containers—Well-closed containers.

Powdered Rose Fruit

Rosae Fructus Pulveratus

エイジツ末

Powdered Rose Fruit is the powder of Rose Fruit.

Description Powdered Rose Fruit occurs as a grayish yellow-brown powder. It has a slight odor, and has a slightly mucilaginous, astringent, bitter, and slightly acid taste.

Under a microscope <*5.01*>, Powdered Rose Fruit reveals fragments of extremely thick-walled hairs 35 – 70 μm in diameter, fragments of epidermis and hypodermis containing brown tannin masses, fragments of thin-walled fundamental tissue containing grayish brown substances, fragments of fine vessels, and solitary or twin crystals or rosette agregates of calcium oxalate (components of fruit receptacle); fragments of sclerenchyma, fiber groups, fine vessels, and fragments of epidermis containing brown tannin and mucilage (components of pericarp); fragments of endosperm composed of polygonal cells containing aleuron grains and fatty oil, fragments of outer epidermis composed of polygonal cells containing tannin, and fragments of inner epidermis composed of elongated cells having wavy lateral walls (components of seed).

Identification Boil gently 1 g of Powdered Rose Fruit with 20 mL of methanol for 2 minutes, and filter. To 5 mL of the filtrate add 0.1 g of magnesium in ribbon form and 0.5 mL of hydrochloric acid, and allow the mixture of stand: a light red to red color develops.

Total ash <*5.01*> Not more than 6.0%.

Containers and storage Containers—Well-closed containers.

Rosin

Resina Pini

ロジン

Rosin is the resin obtained from the exudation of plants of *Pinus* species (*Pinaceae*) from which essential oil has been removed.

Description Rosin occurs as a light yellow to light brown, glassily transparent, brittle mass, the surfaces of which are often covered with a yellow powder. The fractured surface is shell-like and lustrous.

It has a slight odor.

It melts easily, and burns with a yellow-brown flame.

It is freely soluble in ethanol (95), in acetic acid (100) and in diethyl ether.

A solution of Rosin in ethanol (95) is acidic.

Acid value ⟨*1.13*⟩ 150 – 177

Total ash ⟨*5.01*⟩ Not more than 0.1%.

Containers and storage Containers—Well-closed containers.

Royal Jelly

Apilac

ローヤルゼリー

Royal Jelly is the viscous liquid or its dried substance secreted by the secreting gland on the head of *Apis mellifera* Linné or *Apis cerana* Fabricius (*Apidae*).

It contains not less than 4.0% and not more than 8.0% of 10-hydroxy-2-(*E*)-decenoic acid, calculated on the basis of dried material.

Description Slightly viscous liquid or powder, milky white to light yellow in color. Odor, characteristic; taste, astringent and acid.

Identification To a portion of Royal Jelly, equivalent to 0.2 g of dried substance, add 5 mL of water, 1 mL of dilute hydrochloric acid and 10 mL of diethyl ether, shake for 15 minutes, and centrifuge. Take the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 5 mL of methanol, and use this solution as the sample solution. Separately, dissolve 2 mg of 10-hydroxy-2-(*E*)-decenoic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 20 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography, develop the plate with a mixture of 1-propanol and ammonia solution (28) (7:3) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): the spot obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with a portion of Royal Jelly, equivalent to 1.0 g of the dried substance, according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 30 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with an amount of Royal Jelly, equivalent to 0.40 g of the dried substance according to Method 3, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ The slightly viscous liquid: Not less than 57.0% and not more than 77.0% (6 hours).

The powder: Not less than 7.0% and not more than 13.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 4.0%, calculated on the dried basis.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 0.5%, calculated on the dried basis.

Assay Weigh accurately a portion of Royal Jelly, equivalent to 0.2 g of the dried substance, add 20 mL of methanol, sonicate for 30 minutes, and add methanol to make exactly 50 mL. Centrifuge this solution, pipet 2 mL of the supernatant liquid, add exactly 2 mL of the internal standard solution, then add 25 mL of water and methanol to make 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of 10-hydroxy-2-(*E*)-decenoic acid for assay, dissolve in methanol to make exactly 100 mL. Pipet 3 mL of this solution, add exactly 2 mL of the internal standard solution, then add 25 mL of water and methanol to make 50 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and calculate the ratios, Q_T and Q_S, of the peak area of 10-hydroxy-2-(*E*)-decenoic acid to that of the internal standard.

$$\text{Amount (mg) of 10-hydroxy-2-(}E\text{)-decenoic acid} = M_S \times Q_T/Q_S \times 3/4$$

M_S: Amount (mg) of 10-hydroxy-2-(*E*)-decenoic acid for assay taken

Internal standard solution—A solution of propyl parahydroxybenzoate in methanol (1 in 5000).

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 215 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, methanol for liquid chromatography and phosphoric acid (550:450:1).

Flow rate: Adjust so that the retention time of 10-hydroxy-2-(*E*)-decenoic acid is about 10 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, 10-hydroxy-2-(*E*)-decenoic acid and the internal standard are eluted in this order with the resolution between these peaks being not less than 6.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the ratio of the peak area of 10-hydroxy-2-(*E*)-decenoic acid to that of the internal standard is not more than 1.0%.

Containers and storage Containers—Tight containers.

Storage—At not exceeding 10°C.

Ryokeijutsukanto Extract

苓桂朮甘湯エキス

Ryokeijutsukanto Extract contains not less than 1 mg and not more than 4 mg of (*E*)-cinnamic acid, and not less than 17 mg and not more than 51 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Poria Sclerotium	6 g	6 g
Cinnamon Bark	4 g	4 g
Atractylodes Rhizome	3 g	—
Atractylodes Lancea Rhizome	—	3 g
Glycyrrhiza	2 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Ryokeijutsukanto Extract occurs as a brown powder or black-brown viscous extract. It has an odor, and a sweet first then bitter taste.

Identification **(1)** To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate, formic acid and water (60:40:4:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Cinnamon Bark).

(2) For preparation prescribed Atractylodes Rhizome—To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, and shake. Centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) of Ryokeijutsukanto Extract as directed in the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) of Ryokeijutsukanto Extract according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 8.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g 105°C, 5 hours).

Total ash <*5.01*> Not more than 8.0%, calculated on the dried basis.

Assay **(1)** (*E*)-Cinnamic acid—Conduct this procedure using light-resistant vessels. Weigh accurately about 0.5 g of dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance) of Ryokeijutsukanto Extract, add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-cinnamic acid for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of (*E*)-cinnamic acid in each solution.

$$\text{Amount (mg) of (}E\text{)-cinnamic acid} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of (*E*)-cinnamic acid for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wave-

length: 273 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (750:250:1).

Flow rate: 1.0 mL per minute (the retention time of (*E*)-cinnamic acid is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of (*E*)-cinnamic acid are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of (*E*)-cinnamic acid is not more than 1.5%.

(2) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Safflower

Carthami Flos

コウカ

Safflower is the tubulous flower of *Carthamus tinctorius* Linné (*Compositae*) without any treatment or with most of the yellow pigment removed, and sometimes with pressed into a flat slab.

Description Red to red-brown corolla, yellow style and stamen, rarely mixed with immature ovary; total length about 1 cm; corolla, tubular and with 5 lobes; 5 stamens surrounding long pistil; pollen grains yellow and approximately spherical, about 50 μm in diameter, with fine protrusions on the surface. The pressed slab, about 0.5 cm in thickness, consists of a collection of numerous corollas.

Odor, characteristic; taste, slightly bitter.

Identification To 1.0 g of pulverized Safflower add 10 mL of a mixture of acetone and water (4:1), shake for 10 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water, formic acid and methanol (35:15:10:2) to a distance of about 7 cm, and air-dry the plate: a red spot appears at an *R*f value of about 0.5.

Purity Foreign matter <*5.01*>—The amount of ovaries, stems, leaves and other foreign matter contained in Safflower does not exceed 2.0%.

Total ash <*5.01*> Not more than 18.0%.

Containers and storage Containers—Well-closed containers.

Storage—Light-resistant.

Saffron

Crocus

サフラン

Saffron is the stigma of *Crocus sativus* Linné (*Iridaceae*).

Description Thin cord-like stigma, externally dark yellow-red to red-brown, 1.5 – 3.5 cm in length, tripartite or separate; the end of partite part widened and the other end narrowed gradually.

Odor, strong and characteristic; taste, bitter; colors the saliva yellow on chewing.

Under a microscope <*5.01*>, when softened by immersion in water, the upper end has numerous tubular protrusions about 150 μm in length, with a small number of pollen grains.

Identification Use the sample solution obtained in the Purity (1) as the sample solution. Perform the test with this solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, water and acetic acid (100) (20:5:4:1) to a distance of about 10 cm, and air-dry the plate: three yellow spots appear at the *R*f values of about 0.1, about 0.25 and about 0.4.

Purity **(1)** Synthetic dye—To 0.10 g of pulverized Saffron add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol, water and acetic acid (100) (20:5:4:1) to a distance of about 10 cm, and air-dry the plate: any spot other than yellow spots does not appear upper in position than a yellow spot at an *R*f value of about 0.4 and any yellow spot does not appear between yellow spots at *R*f values of about 0.25 and about 0.4. Spray evenly diluted sulfuric acid on the plate, and heat the plate at 105°C for 10 minutes: any clear yellow spot does not appear at an *R*f value of less than 0.1, and any orange spot does not appear between blue-purple spots at *R*f values of about 0.1 and about 0.25.

(2) Glycerol, sugar or honey—Saffron has no sweet taste. Press it between two pieces of paper: no spot is left on the paper.

(3) Yellow style—When perform the test of foreign matter <*5.01*>, the yellow style in Saffron does not exceed 10.0%.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Content of the active principle Crocin—Dry Saffron in a desiccator (silica gel) for 24 hours, and powder. To exactly 0.100 g of the powder add 150 mL of warm water, warm the mixture between 60°C and 70°C for 30 minutes with frequent shaking, cool, and filter. Pipet 1 mL of the filtrate, add water to make exactly 10 mL, and use this solution as the sample solution. Separately, dissolve exactly 98 mg of carbazochrome sodium sulfonate trihydrate in water to make exactly 100 mL. Pipet 5 mL of this solution, add water to make exactly 100 mL, and use this solution as the standard solution. Determine the absorbances of the sample solution and standard solution at 438 nm as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: the absorbance of the sample solution is larger than that of the standard solution.

Containers and storage Containers—Well-closed containers.

Storage—Light-resistant.

Saibokuto Extract

柴朴湯エキス

Saibokuto Extract contains not less than 2 mg and not more than 8 mg of saikosaponin b_2, not less than 90 mg and not more than 270 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Bupleurum Root	7 g	7 g
Pinellia Tuber	6 g	5 g
Poria Sclerotium	5 g	5 g
Scutellaria Root	3 g	3 g
Magnolia Bark	3 g	3 g
Jujube	3 g	3 g
Ginseng	3 g	3 g
Glycyrrhiza	2 g	2 g
Perilla Herb	2 g	2 g
Ginger	1 g	1 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Saibokuto Extract is a light brown powder or black-brown viscous extract, having a slightly odor and a slight sweet first, then a bitter taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-

butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of magnolol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Magnolia Bark).

(4) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of 0.1 mol/L hydrochloric acid TS, add 25 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of rosmarinic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water and formic acid (60:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Perilla Herb).

(7) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than

9.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of baicalein for resolution check in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and baicalein is not less than 1.5.

System repeatability: When the test is repeated 6 times

with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Saikokeishito Extract

柴胡桂枝湯エキス

Saikokeishito Extract contains not less than 1.5 mg and not more than 6 mg of saikosaponin b_2, not less than 60 mg and not more than 180 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), not less than 17 mg and not more than 51 mg (for preparation prescribed 2 g of Peony Root) or not less than 21 mg and not more than 63 mg (for preparation prescribed 2.5 g of Peony Root) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 10 mg and not more than 30 mg (for preparation prescribed 1.5 g of Glycyrrhiza) or not less than 14 mg and not more than 42 mg (for preparation prescribed 2 g of Glycyrrhiza) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Bupleurum Root	5 g	5 g	5 g	5 g
Pinellia Tuber	4 g	4 g	4 g	4 g
Scutellaria Root	2 g	2 g	2 g	2 g
Peony Root	2 g	2.5 g	2 g	2 g
Jujube	2 g	2 g	2 g	2 g
Ginseng	2 g	2 g	2 g	2 g
Cinnamon Bark	2.5 g	2.5 g	2.5 g	2 g
Glycyrrhiza	1.5 g	1.5 g	1.5 g	2 g
Ginger	0.5 g	1 g	1 g	1 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Saikokeishito Extract is a yellow-brown powder or black-brown viscous extract, having a slightly odor and a slight sweet first, then a bitter and slightly pungent taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for

thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(4) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(5) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect an apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use this solution as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 50 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, diethyl ether and methanol (15:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydradine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 5 mL of hexane, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(6) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(7) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Lead—Take 5.0 g of the dry extract (or an amount of the viscous extract, equivalent to 5.0 g of the dried substance) in a platinum, quartz or porcelain crucible, heat gently, and then incinerate by ignition at 450 to 550°C. After cooling, add a small amount of 2 mol/L nitric acid TS to the residue, filter if necessary, and wash the crucible several times with small portions of 2 mol/L nitric acid TS. Combine the washings and the filtrate, add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the sample solution. Separately, to 2.5 mL of Standard Lead Solution add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the standard solution. Perform the test with the sample solution and standard solution as directed under Atomic Absorption Spectrophotometry <*2.23*> according to the following conditions: the absorbance of the sample solution is not more than that of the standard solution (not more than 5 ppm).

Gas: Combustible gas—Acetylene or hydrogen.
Supporting gas—Air.
Lamp: A lead hollow-cathode lamp.
Wavelength: 283.3 nm.

(3) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash ⟨*5.01*⟩ Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(4) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as

the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography and 1 mg of baicalein for resolution check in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, two peaks other than glycyrrhizic acid are observed with the resolutions between the peak of glycyrrhizic acid and each of the two peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Saireito Extract

柴苓湯エキス

Saireito Extract contains not less than 2 mg and not more than 8 mg of saikosaponin b_2, not less than 80 mg and not more than 240 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Bupleurum Root	7 g	7 g
Pinellia Tuber	5 g	5 g
Ginger	1 g	1 g
Scutellaria Root	3 g	3 g
Jujube	3 g	3 g
Ginseng	3 g	3 g
Glycyrrhiza	2 g	2 g
Alisma Tuber	6 g	5 g
Polyporus Sclerotium	4.5 g	3 g
Poria Sclerotium	4.5 g	3 g
Atractylodes Rhizome	4.5 g	—
Atractylodes Lancea Rhizome	—	3 g
Cinnamon Bark	3 g	2 g

Prepare a dry extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Saireito Extract occurs as a light yellow-brown powder. It has slightly a characteristic odor, and a sweet, then bitter taste.

Identification (1) To 2.0 g of Saireito Extract add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatogra-

phy <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(2) To 1.0 g of Saireito Extract add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 15 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(3) To 1.0 g of Saireito Extract add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(4) To 2.0 g of Saireito Extract add 10 mL of sodium hydroxide TS, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(5) To 2.0 g of Saireito Extract add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) To 2.0 g of Saireito Extract add 10 mL of sodium carbonate TS, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of alisol A for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Alisma Tuber).

(7) For preparation prescribed Atractylodes Rhizome—To 1.0 g of Saireito Extract add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Atractylodes Rhizome).

(8) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of Saireito Extract add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes

Lancea Rhizome).

(9) To 1.0 g of Saireito Extract add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamic acid for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate, formic acid and water (60:40:4:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the dark purple spot from the standard solution (Cinnamon Bark).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Saireito Extract as directed under Extract (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of Saireito Extract according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> Not more than 10.0% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of Saireito Extract, add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of Saireito Extract, add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of Saireito Extract, add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography and 1 mg of baicalein for resolution check in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, two peaks other than glycyrrhizic acid are observed with the resolutions between the peak of glycyrrhizic acid and each of the two peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of Saireito Extract, add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Salvia Miltiorrhiza Root

Salviae Miltiorrhizae Radix

タンジン

Salvia Miltiorrhiza Root is the root of *Salvia miltiorrhiza* Bunge (*Labiatae*).

Description Nearly cylindrical root, 5 – 25 cm in length, 0.3 – 1.5 cm in diameter; slightly curved, often with lateral roots; outer surface red-brown, dark red-brown or black-brown; with irregular rough wrinkles; hard in texture, and easily broken; fracture surface fine or rough with clefts; cortex grayish yellow-white or red-brown, xylem light yellow-white or black-brown.

Odor, slight; taste, sweet at first and followed by slight bitterness and astringency.

Under a microscope <*5.01*>, a transverse section reveals usually cork layer in the outermost part, or rarely parenchyma or endodermis at the outside of the cork layer; several sclerenchyma cells observed or not in secondary cortex; cambium obvious; vessels radially arranged in secondary xylem, sometimes radial lines of vessels unite in the center of root; xylem fibers surrounding vessels; primary xylem divided into 2 – 3; vessels of secondary xylem mainly pitted vessels and reticulate vessels in a longitudinal section.

Identification To 1.0 g of pulverized Salvia Miltiorrhiza Root add 10 mL of diethyl ether, allow to stand for 10 minutes with occasional shaking, and filter. Evaporate the filtrate on a water bath to dryness, dissolve the residue in 1 mL of ethyl acetate, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (3:1) to a distance of about 10 cm, and air-dry the plate: a red-brown spot at an *R*f value of about 0.4 is observed.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Salvia Miltiorrhiza Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Salvia Miltiorrhiza Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 16.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 42.0%.

Containers and storage Containers—Well-closed containers.

Saposhnikovia Root and Rhizome

Saposhnikoviae Radix

ボウフウ

Saposhnikovia Root and Rhizome is the root and rhizome of *Saposhnikovia divaricata* Schischkin (*Umbelliferae*).

Description Long and narrow, conical rhizome and root, 15 – 20 cm in length, 0.7 – 1.5 cm in diameter; externally light brown; rhizome reveals dense crosswise wrinkles like ring nodes, and sometimes reveals brown and hair-like remains of leaf sheath; the root reveals many longitudinal wrinkles and scars of rootlets; in a transverse section, cortex is grayish brown in color and reveals many lacunae, and xylem is yellow in color.

Odor, slight; taste, slightly sweet.

Identification To 1.0 g of pulverized Saposhnikovia Root and Rhizome, add 5 mL of methanol, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of 4′-*O*-glucosyl-5-*O*-methylvisamminol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 4 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, formic acid, 2-butanone, and water (20:5:5:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 2.0 g of pulverized Saposhnikovia Root and Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Saposhnikovia Root and Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter <*5.01*>—The amount of stems and other foreign matter is not more than 2.0%.

(4) Peucedanum ledebourielloides—Place 1.0 g of pulverized Saposhnikovia Root and Rhizome in a glass-stoppered centrifuge tube, add 5 mL of hexane, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, place 1.0 g of peucedanum ledebourielloides for purity in a glass-stoppered centrifuge tube, add 5 mL of hexane, shake for 10 minutes, centrifuge, and use the supernatant liquid as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate and acetic acid (100) (20:10:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): the sample solution shows no spot corresponding to the blue fluorescent spot at an *R*f value of about 0.4 obtained from the standard solution.

Total ash <*5.01*> Not more than 7.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 20.0%.

Containers and storage Containers—Well-closed containers.

Sappan Wood

Sappan Lignum

ソボク

Sappan Wood is the duramen of *Caesalpinia sappan* Linné (*Leguminosae*).

Description Chips, slices or short pieces of wood; yellow-red to grayish yellow-brown, sometimes with light brown to grayish white splint woods; hard in texture; a transverse section shows a pattern like annual ring.

Almost odorless; almost tasteless.

Under a microscope <*5.01*>, a transverse section reveals ray composed of 1 – 2 cell rows of slender and long cells; the area between rays filled with fiber cells, and large and oblong vessels scattered there; solitary crystals of calcium oxalate in parenchymatous cells of the innermost of xylem.

Identification To 1 g of pulverized Sappan Wood add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, water, formic acid and 2-propanol (20:1:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly sodium carbonate TS on the plate, and air-dry the plate: a red-purple spot appears at an *R*f value of about 0.7.

Purity Put a small piece of Sappan Wood in calcium hydroxide TS: no purple-blue color develops.

Loss on drying <*5.01*> Not more than 11.5% (6 hours).

Total ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 7.0%.

Containers and storage Containers—Well-closed containers.

Saussurea Root

Saussureae Radix

モッコウ

Saussurea Root is the root of *Saussurea lappa* Clarke (*Compositae*).

Description Nearly cylindrical roots, 5 – 20 cm in length, 1 – 6 cm in diameter; some of them slightly bent, and sometimes longitudinally cut; scar of stem dented on the top of the root with crown; externally yellow-brown to grayish brown, with coarse longitudinal wrinkles and fine reticulate furrows, and also with remains of lateral roots; sometimes root from which periderm has been removed; hard and dense

in texture, and difficult to break. A transverse section is yellow-brown to dark brown, and cambium part has a dark color. Under a magnifying glass, a transverse section reveal obvious medullary rays, large clefts here and there, and brown oil sacs scattered; in old root, pith existing in the center, and often forming a hollow.

Odor, characteristic; taste, bitter.

Identification To 1.0 g of pulverized Saussurea Root add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and cool: a red-purple spot at an *R*f value of about 0.5 and a grayish blue to grayish brown spot just below it are observed.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 1.0 g of pulverized Saussurea Root according to Method 3, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Saussurea Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Add iodine TS dropwise to a transverse section: no blue-purple color develops.

Total ash <*5.01*> Not more than 4.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 17.0%.

Containers and storage Containers—Well-closed containers.

Schisandra Fruit

Schisandrae Fructus

ゴミシ

Schisandra Fruit is the fruit of *Schisandra chinensis* Baillon (*Schisandraceae*).

Description Sap fruit of irregular sphere or spheroid, about 6 mm in diameter; externally dark red to black-brown in color, with wrinkles, and occasionally with white powder; seeds, kidney-shaped, externally yellow-brown to dark red-brown, lustrous, with distinct raphe on the dorsal side; external seed coat easily peeled but internal seed coat adhering closely to the albumen.

Odor, slight; taste, acid, later astringent and bitter.

Identification To 1.0 g of pulverized Schisandra Fruit add 10 mL of methanol, warm on a water bath for 3 minutes with shaking, cool, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of schisandrin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity Foreign matter <*5.01*>—The amount of fruit receptacle, peduncle and other foreign matter contained in Schisandra Fruit is not more than 1.0%.

Total ash <*5.01*> Not more than 5.0%.

Containers and storage Containers—Well-closed containers.

Schizonepeta Spike

Schizonepetae Spica

ケイガイ

Schizonepeta Spike is the spike of *Schizonepeta tenuifolia* Briquet (*Labiatae*).

Description Oblong spike, 5 – 10 cm in length, 0.5 – 0.8 cm in diameter, purplish green-brown to green-brown in color. Spike, with calyx-tubes containing small labiate flower or often fruits; sometimes leaves under spike; leaf, linear or small lanceolate; stem, prismatic, purple-brown in color. Under a magnifying glass, it reveals short hairs.

It has a characteristic aroma and slightly cool feeling on keeping in the mouth.

Identification To 1 g of pulverized Schizonepeta Spike add 10 mL of ethyl acetate, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (3:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes. After cooling for more than 10 minutes under an adequate humidity, examine under ultraviolet light (main wavelength: 365 nm): two spots, one is a blue fluorescent spot with an *R*f value of about 0.5 and the another is a yellow fluorescent spot with an *R*f value of about 0.1, are observed.

Total ash <*5.05*> Not more than 11.0%.

Acid-insoluble ash <*5.05*> Not more than 3.0%.

Extract content <*5.05*> Dilute ethanol-soluble extract: not less than 8.0%.

Containers and storage Containers—Well-closed containers.

Scopolia Rhizome

Scopoliae Rhizoma

ロートコン

Scopolia Rhizome is the rhizome with root of *Scopolia japonica* Maximowicz, *Scopolia carniolica* Jacquin or *Scopolia parviflora* Nakai (*Solanaceae*).

When dried, it contains not less than 0.29% of total

alkaloids [hyoscyamine ($C_{17}H_{23}NO_3$: 289.37) and scopolamine ($C_{17}H_{21}NO_4$: 303.35)].

Description Chiefly irregularly branched, slightly curved rhizome, about 15 cm in length, about 3 cm in diameter, occasionally longitudinally cut; externally grayish brown, with wrinkles; rhizome has constrictions and nodes; rarely, remains of stem at the apex; stem scars at upper side of each node; roots or root scars on both sides and lower surface of rhizome; fractured surface granular, grayish white to light brown in color; cortex a little lighter in color.

Odor characteristic.

Under a microscope <*5.01*>, xylem reveals groups of vessels arranged stepwise between medullary rays; xylem sieve tubes accompanied to the groups of vessels; parenchyma cells contain starch grains, and sometimes sand crystals of calcium oxalate.

Identification **(1)** To 1 g of pulverized Scopolia Rhizome add 10 mL of diethyl ether and 0.5 mL of ammonia TS, shake for 30 minutes, and filter. Wash the residue with 10 mL of diethyl ether, transfer the filtrate and the washing to a separator, add 20 mL of diluted sulfuric acid (1 in 50), shake well, and drain off the acid extract into another separator. Render the solution slightly alkaline with ammonia TS, add 10 mL of diethyl ether, shake well, transfer the diethyl ether layer to a porcelain dish, and evaporate the solvent on a water bath. To the residue add 5 drops of fuming nitric acid, and evaporate the mixture on a water bath to dryness. Cool, dissolve the residue in 1 mL of *N*,*N*-dimethylformamide, and add 5 to 6 drops of tetraethylammonium hydroxide TS: a red-purple to purple color develops.

(2) Place 2.0 g of pulverized Scopolia Rhizome in a glass-stoppered centrifuge tube, add 30 mL of ammonia TS, sonicate for 5 minutes, and centrifuge. Transfer the supernatant liquid to a separator, add 40 mL of ethyl acetate, and shake. Drain off the ethyl acetate layer, add 3 g of anhydrous sodium sulfate to the ethyl acetate, shake, and filter after the ethyl acetate becomes clear. Evaporate the solvent of the filtrate under low pressure (in vacuo), dissolve the residue in 1 mL of ethanol (95), and use this solution as the sample solution. Separately, dissolve 2 mg of Atropine Sulfate RS or atropine sulfate hydrate for thin-layer chromatography and 1 mg of Scopolamine Hydrobromide RS or scopolamine hydrobromide hydrate for thin-layer chromatography in 1 mL each of ethanol (95), and use these solutions as standard solution (1) and standard solution (2), respectively. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution, standard solutions (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, water and ammonia water (28) (90:7:3) to a distance of about 10 cm, and dry the plate at 80°C for 10 minutes. After cooling, spray evenly Dragendorff's TS for spraying on the plate: two principal spots obtained from the sample solution and each spot from the standard solutions show the same color tone and the same *R*f value.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Scopolia Rhizome according to Method 3, and perform the test. Prepare the control solution with 4.5 mL of Standard Lead Solution (not more than 15 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Scopolia Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 7.0%.

Assay Weigh accurately about 0.7 g of pulverized Scopolia Rhizome, previously dried at 60°C for 8 hours, in a glass-stoppered centrifuge tube, and moisten with 15 mL of ammonia TS. To this add 25 mL of diethyl ether, stopper the centrifuge tube tightly, shake for 15 minutes, centrifuge, and separate the diethyl ether layer. To the residue add 25 mL of diethyl ether, proceed in the same manner, and repeat this procedure twice. Combine all the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 5 mL of the mobile phase, add exactly 3 mL of the internal standard solution, and add the mobile phase to make 25 mL. Filter this solution through a filter of a porosity of not more than 0.8 μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 25 mg of Atropine Sulfate RS (separately determine the loss on drying <*2.41*> under the same conditions as Atropine Sulfate Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as standard stock solution A. Weigh accurately about 25 mg of Scopolamine Hydrobromide RS (separately determine the loss on drying <*2.41*> under the same conditions as Scopolamine Hydrobromide Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as standard stock solution B. Pipet 5 mL of standard stock solution A and 1 mL of standard stock solution B, add exactly 3 mL of the internal standard solution, then add 25 mL of the mobile phase, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Calculate the ratios, Q_{TA} and Q_{SA}, of the peak area of hyoscyamine (atropine), and the ratios, Q_{TS} and Q_{SS}, of the peak area of scopolamine to that of the internal standard in each solution, calculate the amounts of hyoscyamine and scopolamine by the following equation, and designate the total as the amount of total alkaloids.

$$\text{Amount (mg) of hyoscyamine } (C_{17}H_{23}NO_3) = M_{SA} \times Q_{TA}/Q_{SA} \times 1/5 \times 0.855$$

$$\text{Amount (mg) of scopolamine } (C_{17}H_{21}NO_4) = M_{SS} \times Q_{TS}/Q_{SS} \times 1/25 \times 0.789$$

M_{SA}: Amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

M_{SS}: amount (mg) of Scopolamine Hydrobromide RS taken, calculated on the dried basis

Internal standard solution—A solution of brucine dihydrate in the mobile phase (1 in 2500).

Operating conditions—

Detector: An ultraviolet absorption spectrometer (wavelength: 210 nm).

Column: A stainless steel column 4 mm in inside diameter and 15 cm in length, packed with octadesilcylanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: Dissolve 6.8 g of potassium dihydrogenphosphate in 900 mL of water, add 10 mL of triethylamine, adjust with phosphoric acid to pH 3.5, and add water to make 1000 mL. To 9 parts of this solution add 1 part of acetonitrile.

Flow rate: Adjust so that the retention time of scopolamine is about 8 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, scopolamine, atropine and the internal standard are

eluted in this order with the resolution between the peaks of scopolamine and atropine being not less than 11, and with the resolution between the peaks of atropine and the internal standard being not less than 4.

Containers and storage Containers—Well-closed containers.

Scopolia Extract

ロートエキス

Scopolia Extract contains not less than 0.90% and not more than 1.09% of total alkaloids [hyoscyamine ($C_{17}H_{23}NO_3$: 289.37) and scopolamine ($C_{17}H_{21}NO_4$: 303.35)].

Method of preparation Extract the coarse powder of Scopolia Rhizome with 35 vol% ethanol, Water, Purified Water or Purified Water in Containers, and prepare the viscous extract as directed under Extracts.

Description Scopolia Extract is brown to dark brown in color. It has a characteristic odor, and a bitter taste.

It dissolves in water with a slight turbidity.

Identification **(1)** Dissolve 4 g of Scopolia Extract in 10 mL of water, add 8 mL of ammonia TS and 80 mL of diethyl ether, stopper tightly, shake for 1 hour, add 2.5 g of powdered tragacanth, shake vigorously, allow to stand for 5 minutes, and separate the diethyl ether layer into a porcelain dish. Evaporate the diethyl ether on a water bath, add 5 drops of fuming nitric acid, and evaporate on a water bath to dryness. After cooling, dissolve the residue in 1 mL of *N*,*N*-dimethylformamide, and add 5 to 6 drops of tetraethylammonium hydroxide TS: a red-purple to purple color develops.

(2) Mix 0.5 g of Scopolia Extract with 30 mL of ammonia TS in a flask, and transfer the mixture to a separator. Add 40 mL of ethyl acetate to the separator, and shake the mixture. After drain off the ethyl acetate layer, add 3 g of anhydrous sodium sulfate to the ethyl acetate, shake, and filter after the ethyl acetate becomes clear. Evaporate the solvent of the filtrate under low pressure (in vacuo), dissolve the residue in 1 mL of ethanol (95), and use this solution as the sample solution. Proceed as directed in Identification (2) under Scopolia Rhizome.

Purity Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Scopolia Extract as directed in the Extracts (4), and perform the test (not more than 30 ppm).

Assay Weigh accurately about 0.4 g of Scopolia Extract, place in a glass-stoppered centrifuge tube, add 15 mL of ammonia TS, and shake. Add 25 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge, and separate the diethyl ether layer. Repeat this procedure twice with the aqueous layer, using 25 mL each of diethyl ether. Combine all the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 5 mL of the mobile phase, add exactly 3 mL of the internal standard solution, and add the mobile phase to make 25 mL. Proceed as directed under Scopolia Rhizome.

Amount (mg) of hyoscyamine ($C_{17}H_{23}NO_3$)
$= M_{SA} \times Q_{TA}/Q_{SA} \times 1/5 \times 0.855$

Amount (mg) of scopolamine ($C_{17}H_{21}NO_4$)
$= M_{SS} \times Q_{TS}/Q_{SS} \times 1/25 \times 0.789$

M_{SA}: Amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

M_{SS}: Amount (mg) of Scopolamine Hydrobromide RS taken, calculated on the dried basis

Internal standard solution—A solution of brucine dihydrate in the mobile phase (1 in 2500).

Containers and storage Containers—Tight containers.
Storage—Light-resistant, and in a cold place.

Scopolia Extract Powder

ロートエキス散

Scopolia Extract Powder contains not less than 0.085% and not more than 0.110% of total alkaloids [hyoscyamine ($C_{17}H_{23}NO_3$: 289.37) and scopolamine ($C_{17}H_{21}NO_4$: 303.35)].

Method of preparation

Scopolia Extract		100 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity	
	To make	1000 g

To Scopolia Extract add 100 mL of Purified Water or Purified Water in Containers, then warm and soften the mixture with stirring. Cool, add 800 g of starch, Lactose Hydrate or their mixture little by little, and mix well. Dry preferably at a low temperature, and dilute with a sufficient additional quantity of starch, Lactose Hydrate or their mixture to make 1000 g of homogeneous powder.

Description Scopolia Extract Powder is a brownish yellow to grayish yellow-brown powder. It has a faint, characteristic odor and a slightly bitter taste.

Identification **(1)** To 20 g of Scopolia Extract Powder add 15 mL of water and 8 mL of ammonia TS, mix homogeneously, add 100 mL of diethyl ether and 7 g of sodium chloride, stopper tightly, shake for 1 hour, add 5 g of powdered tragacanth, and shake vigorously. Allow to stand for 5 minutes, take the clearly separated diethyl ether layer, and filter. Proceed with the filtrate as directed in the Identification (1) under Scopolia Extract.

(2) Place 5.0 g of Scopolia Extract Powder in a glass-stoppered centrifuge tube, add 30 mL of ammonia TS, sonicate for 5 minutes, and centrifuge. Transfer the supernatant liquid to a separator, add 40 mL of ethyl acetate, and shake. Drain off the ethyl acetate layer, add 3 g of anhydrous sodium sulfate to the ethyl acetate, shake, and filter after the ethyl acetate becomes clear. Evaporate the solvent of the filtrate under low pressure (in vacuo), dissolve the residue in 1 mL of ethanol (95), and use this solution as the sample solution. Proceed as directed in the Identification (2) under Scopolia Rhizome.

Assay Weigh accurately about 4 g of Scopolia Extract Powder, place in a glass-stoppered centrifuge tube, add 15 mL of ammonia TS, and shake. Add 25 mL of diethyl ether, stopper tightly, shake for 15 minutes, centrifuge to take the diethyl ether layer. To the aqueous layer add 25 mL of diethyl ether, proceed in the same manner, and repeat this procedure three times. Combine all the extracts, and evaporate the solvent on a water bath. Dissolve the residue in 5 mL of the mobile phase, add exactly 3 mL of the internal stand-

ard solution, and add the mobile phase to make 25 mL. Filter this solution through a membrane filter with a pore size not exceeding 0.8 μm, discard the first 2 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 25 mg of Atropine Sulfate RS (separately determine the loss on drying <*2.41*> under the same manner as Atropine Sulfate Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as standard stock solution A. Weigh accurately about 25 mg of Scopolamine Hydrobromide RS (separately determine the loss on drying <*2.41*> under the same manner as Scopolamine Hydrobromide Hydrate), dissolve in the mobile phase to make exactly 25 mL, and use this solution as standard stock solution B. Pipet 5 mL of the standard stock solution A and 1 mL of the standard stock solution B, add exactly 3 mL of the internal standard solution, then add the mobile phase to make 25 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Calculate the ratios, Q_{TA} and Q_{SA}, of the peak area of hyoscyamine (atropine), and ratios, Q_{TS} and Q_{SS}, of the peak area of scopolamine to that of the internal standard in each solution, calculate the amounts of hyoscyamine and scopolamine by the following equation, and designate the total as the amount of total alkaloids.

Amount (mg) of hyoscyamine ($C_{17}H_{23}NO_3$)
$= M_{SA} \times Q_{TA}/Q_{SA} \times 1/5 \times 0.855$

Amount (mg) of scopolamine ($C_{17}H_{21}NO_4$)
$= M_{SS} \times Q_{TS}/Q_{SS} \times 1/25 \times 0.789$

M_{SA}: Amount (mg) of Atropine Sulfate RS taken, calculated on the dried basis

M_{SS}: Amount (mg) of Scopolamine Hydrobromide RS taken, calculated on the dried basis

Internal standard solution—A solution of brucine dihydrate in the mobile phase (1 in 2500).

Operating conditions—

Detector: An ultraviolet absorption spectrometer (wavelength: 210 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of a solution obtained by dissolving 6.8 g of potassium dihydrogenphosphate in 900 mL of water, adding 10 mL of triethylamine, adjusting the pH to 3.5 with phosphoric acid, and adding water to make 1000 mL, and acetonitrile (9:1).

Flow rate: Adjust so that the retention time of scopolamine is about 8 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, scopolamine, atropine and the internal standard are eluted in this order with the resolution between the peaks of scopolamine and atropine being not less than 11, and the resolution between the peaks of atropine and the internal standard being not less than 4.

Containers and storage Containers—Tight containers.

Scopolia Extract and Carbon Powder

ロートエキス·カーボン散

Method of preparation

Scopolia Extract	5 g
Medicinal Carbon	550 g
Natural Aluminum Silicate	345 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
To make	1000 g

Prepare before use as directed under Powders, with the above ingredients. May be prepared with an equivalent amount of Scopolia Extract Powder in place of Scopolia Extract.

Description Scopolia Extract and Carbon Powder is easily dustable and black in color. It is tasteless.

Containers and storage Containers—Well-closed containers.

Compound Scopolia Extract and Diastase Powder

複方ロートエキス·ジアスターゼ散

Method of preparation

Scopolia Extract	8 g
Diastase	200 g
Precipitate Calcium Carbonate	300 g
Sodium Bicarbonate	250 g
Magnesium Oxide	100 g
Powdered Gentian	50 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
To make	1000 g

Prepare before use as directed under Powders, with the above ingredients. May be prepared with an equivalent amount of Scopolia Extract Powder in place of Scopolia Extract.

Description Compound Scopolia Extract and Diastase Powder is light yellow in color. It has a bitter taste.

Containers and storage Containers—Well-closed containers.

Scopolia Extract and Ethyl Aminobenzoate Powder

ロートエキス·アネスタミン散

Scopolia Extract and Ethyl Aminobenzoate Powder contains not less than 22.5% and not more than 27.5% of ethyl aminobenzoate ($C_9H_{11}NO_2$: 165.19).

Method of preparation

Scopolia Extract	10 g
Ethyl Aminobenzoate	250 g
Magnesium Oxide	150 g
Sodium Bicarbonate	500 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
	To make 1000 g

Prepare as directed under Powders, with the above ingredients. May be prepared with an equivalent amount of Scopolia Extract Powder in place of Scopolia Extract.

Description Scopolia Extract and Ethyl Aminobenzoate Powder is slightly brownish white in color. It has a slightly bitter taste, leaving a sensation of numbness on the tongue.

Identification **(1)** To 2 g of Scopolia Extract and Ethyl Aminobenzoate Powder add 20 mL of diethyl ether, shake, and filter through a glass filter (G4). Wash the residue with three 10-mL portions of diethyl ether, combine the filtrate and the washings, evaporate to dryness, and perform the following test with the residue (ethyl aminobenzoate).

(i) Dissolve 0.01 g of the residue in 1 mL of dilute hydrochloric acid and 4 mL of water: the solution responds to Qualitative Tests <*1.09*> for primary aromatic amines.

(ii) Dissolve 0.1 g of the residue in 5 mL of water with the aid of dilute hydrochloric acid added dropwise, and add iodine TS dropwise: a brown precipitate is produced.

(iii) Warm 0.05 g of the residue with 2 drops of acetic acid (31) and 5 drops of sulfuric acid: the odor of ethyl acetate is perceptible.

(2) To the diethyl ether-insoluble residue obtained in (1) add 30 mL of water, shake gently, and filter: the filtrate responds to Qualitative Tests <*1.09*> for sodium salt and for bicarbonate.

(3) To the water-insoluble residue obtained in (2) add 10 mL of dilute hydrochloric acid, shake, and filter: the filtrate responds to Qualitative Tests <*1.09*> for magnesium salt.

(4) Place 30 g of Scopolia Extract and Ethyl Aminobenzoate Powder in a glass-stoppered conical flask, add 100 mL of water, shake for 30 minutes, and filter immediately by suction through a glass filter (G3). Transfer the residue in the flask to the same glass filter with the filtrate, and filter the residue by suction while pressing vigorously the residue on the same glass filter. Place 75 mL of the filtrate in a 300-mL beaker, and add cautiously 10 mL of diluted sulfuric acid (1 in 3). Add 0.2 mL of bromocresol green TS to this solution, and add dilute sulfuric acid dropwise while shaking thoroughly, until the color of the solution changes from green to yellow-green. After cooling, place this solution in a separator, wash with two 25-mL portions of a mixture of hexane and diethyl ether (1:1) by shaking well, and place the aqueous layer in another separator. Make slightly alkaline with ammonia TS, add immediately 30 mL of diethyl ether, and shake well. Wash the diethyl ether layer with two 10-mL portions of a saturated solution of sodium chloride, separate the diethyl ether layer, add 3 g of anhydrous sodium sulfate, shake, and filter through a pledget of cotton. Evaporate the filtrate to dryness, dissolve the residue in 0.2 mL of ethanol (95), and use this solution as the sample solution. Separately, dissolve 2 mg of Atropine Sulfate RS or atropine sulfate hydrate for thin-layer chromatography and 1 mg of Scopolamine Hydrobromide RS or scopolamine hydrobromide hydrate for thin-layer chromatography in 1 mL each of ethanol (95), and use these solutions as standard solution (1) and standard solution (2), respectively. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution, standard solution (1) and (2) on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, water and ammonia solution (28) (90:7:3) to a distance of about 10 cm, and dry the plate at 80°C for 10 minutes. After cooling, spray evenly Dragendorff's TS for spraying on the plate: two principal spots obtained from the sample solution show the same color tone and the same *R*f value with each spot from the standard solutions, respectively.

Assay Weigh accurately about 0.3 g of Scopolia Extract and Ethyl Aminobenzoate Powder, transfer to a Soxhlet extractor, extract with 100 mL of diethyl ether for 1 hour, and evaporate the solvent on a water bath. Dissolve the residue in 25 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 100 mL. Pipet 5 mL of this solution, add water to make exactly 250 mL, and use this solution as the sample solution. Weigh accurately about 75 mg of Ethyl Aminobenzoate RS, previously dried in a desiccator (silica gel) for 3 hours, dissolve in 25 mL of 1 mol/L hydrochloric acid TS, and add water to make exactly 100 mL. Pipet 5 mL of this solution, add water to make exactly 250 mL, and use this solution as the standard solution. Pipet 5 mL each of the sample solution and standard solution, to each add 10 mL of 1 mol/L hydrochloric acid TS, then add 1 mL of a solution of sodium nitrite (1 in 200), prepared before use, and allow to stand for 5 minutes with occasional shaking. Add 5 mL of ammonium amidosulfate TS, shake well, and allow to stand for 10 minutes. Add 2 mL of *N*-*N*-diethyl-*N'*-1-naphthylethylenediamine oxalate-acetone TS, mix immediately, and add water to make exactly 50 mL. Allow to stand for 2 hours, determine the absorbances, A_T and A_S, of these solutions at 550 nm, as directed under Ultraviolet-visible Spectrophotometry <*2.24*> using a blank prepared in the same manner with 5 mL of water in place of the sample solution.

$$\text{Amount (mg) of ethyl aminobenzoate } (C_9H_{11}NO_2) = M_S \times A_T/A_S$$

M_S: Amount (mg) of Ethyl Aminobenzoate RS taken

Containers and storage Containers—Well-closed containers.

Scopolia Extract and Tannic Acid Suppositories

ロートエキス·タンニン坐剤

Method of preparation

Scopolia Extract	0.5 g
Tannic Acid	1 g
Cacao Butter or a suitable base	a sufficient quantity

Prepare 10 suppositories as directed under Suppositories, with the above ingredients.

Description Scopolia Extract and Tannic Acid Suppositories are light brown in color.

Identification **(1)** To 2 Scopolia Extract and Tannic Acid Suppositories add 20 mL of diethyl ether, and dissolve the base of suppositories with shaking for 10 minutes. Shake thoroughly the mixture with 15 mL of water, separate the aqueous layer, and filter. To the filtrate add 10 mL of chloroform, shake well, and separate the chloroform layer. Take 5 mL of the chloroform solution, add 5 mL of ammonia TS, shake, and allow to stand: the ammonia layer shows a blue-green fluorescence.

(2) To 1 mL of the aqueous layer obtained in (1) after extraction with diethyl ether, add 2 drops of iron (III) chloride TS: a blue-black color develops. Allow to stand: a blue-black precipitate is formed (tannic acid).

Containers and storage Containers—Well-closed containers.

Scutellaria Root

Scutellariae Radix

オウゴン

Scutellaria Root is the root of *Scutellaria baicalensis* Georgi (*Labiatae*), from which the periderm has been removed.

It contains not less than 10.0% of baicalin ($C_{21}H_{18}O_{11}$: 446.36), calculated on the basis of dried material.

Description Cone-shaped, cylindrical, semitubular or flattened root, 5 – 20 cm in length, 0.5 – 3 cm in diameter; externally yellow-brown, with coarse and marked longitudinal wrinkles, and with scattered scars of lateral root and remains of brown periderm; scars of stem or remains of stem at the crown; sometimes central portion of xylem rotted, often forming a hollow; hard in texture and easily broken; fractured surface fibrous and yellow in color.

Almost odorless; taste, slightly bitter.

Under a microscope ⟨*5.01*⟩, a transverse section reveals a remaining cork layer 6 – 20 cells thick, coritical layer composed of parenchyma, sclerenchyma cells scattered in cortex; xylem composed of parenchyma, vessels and small amount of xylem fibers observed in xylem; vessels usually in groups and arranged in tangential direction, radial direction or in irregular form; in case where central portion of xylem rotted, cork layer observed around hollow; parenchyma cells of coritical layer and xylem contain simple and compound starch grains.

Identification **(1)** Heat gently 0.5 g of pulverized Scutellaria Root with 20 mL of diethyl ether under a reflux condenser for 5 minutes, cool, and filter. Evaporate the solvent of the filtrate, dissolve the residue in 10 mL of ethanol (95), and to 3 mL of the solution add 1 to 2 drops of dilute iron (III) chloride TS: a grayish green color develops, and it changes to purple-brown.

(2) To 1 g of pulverized Scutellaria Root add 25 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Baicalin RS or baicalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals ⟨*1.07*⟩—Proceed with 3.0 g of pulverized Scutellaria Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic ⟨*1.11*⟩—Prepare the test solution with 0.40 g of pulverized Scutellaria Root according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 6.0%.

Assay Weigh accurately about 0.5 g of pulverized Scutellaria Root, add 30 mL of diluted methanol (7 in 10), heat under a reflux condenser for 30 minutes, and cool. Transfer the mixture to a glass-stoppered centrifuge tube, centrifuge, and separate the supernatant liquid. Wash the vessel for the reflux extraction with 30 mL of diluted methanol (7 in 10), transfer the washings to the glass-stoppered centrifuge tube, centrifuge after shaking for 5 minutes, and separate the supernatant liquid. To the residue add 30 mL of diluted methanol (7 in 10), shake for 5 minutes, centrifuge, and separate the supernatant liquid. Combine all the extracts, add diluted methanol (7 in 10) to make exactly 100 mL, then pipet 2 mL of this solution, add diluted methanol (7 in 10) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 5$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica

gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 146) and acetonitrile (18:7).

Flow rate: Adjust so that the retention time of baicalin is about 6 minutes.

System suitability—

System performance: Dissolve 1 mg of Baicalin RS and 2 mg of methyl parahydroxybenzoate for resolution check in methanol to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, baicalin and methyl parahydroxybenzoate are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Scutellaria Root

Scutellariae Radix Pulverata

オウゴン末

Powdered Scutellaria Root is the powder of Scutellaria Root.

It contains not less than 10.0% of baicalin ($C_{21}H_{18}O_{11}$: 446.36), calculated on the basis of dried material.

Description Powdered Scutellaria Root occurs as a yellow-brown powder. It is almost odorless, and has a slight, bitter taste.

Under a microscope <*5.01*>, Powdered Scutellaria Root reveals fragments of parenchyma cells containing small amount of simple and compound starch grains, fragments of short reticulate vessel elements and fusiform, stick-like and ellipsoidal to spherical sclerenchyma cells; also a few fragments of spiral vessels and xylem fibers are observed.

Identification **(1)** Heat gently 0.5 g of Powdered Scutellaria Root with 20 mL of diethyl ether under a reflux condenser for 5 minutes, cool, and filter. Evaporate the filtrate, dissolve the residue in 10 mL of ethanol (95), and to 3 mL of the solution add 1 to 2 drops of dilute iron (III) chloride TS: a grayish green color develops, and it changes to purple-brown later.

(2) To 1 g of Powdered Scutellaria Root add 25 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Baicalin RS or baicalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Scutellaria Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Scutellaria Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Scutellaria Root does not show crystals of calcium oxalate.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Assay Weigh accurately about 0.5 g of Powdered Scutellaria Root, add 30 mL of diluted methanol (7 in 10), heat under a reflux condenser for 30 minutes, and cool. Transfer the mixture to a glass-stoppered centrifuge tube, centrifuge, and separate the supernatant liquid. Wash the vessel for the reflux extraction with 30 mL of diluted methanol (7 in 10), transfer the washings to the glass-stoppered centrifuge tube, centrifuge after shaking for 5 minutes, and separate the supernatant liquid. To the residue add 30 mL of diluted methanol (7 in 10), shake for 5 minutes, centrifuge, and separate the supernatant liquid. Combine all the extracts, add diluted methanol (7 in 10) to make exactly 100 mL, then pipet 2 mL of this solution, add diluted methanol (7 in 10) to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of the solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 5$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 146) and acetonitrile (18:7).

Flow rate: Adjust so that the retention time of baicalin is about 6 minutes.

System suitability—

System performance: Dissolve 1 mg of Baicalin RS and 2 mg of methyl parahydroxybenzoate for resolution check in methanol to make 100 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, baicalin and methyl parahydroxybenzoate are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operat-

ing conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Senega

Senegae Radix

セネガ

Senega is the root of *Polygala senega* Linné or *Polygala senega* Linné var. *latifolia* Torrey et Gray (*Polygalaceae*).

Description Slender, conical root often branched, 3 – 10 cm in length; main root 0.5 – 1.5 cm in diameter; externally light grayish brown to grayish brown; with many longitudinal wrinkles and sometimes with twisted protruding lines; tuberously enlarged crown, with remains of stems and red buds; branched rootlets twisted; a transverse section reveals grayish brown cortex and yellowish white xylem; usually round, and sometimes cuneate to semicircular; cortex on the opposite side is thickened.

Odor, characteristic, resembling the aroma of methyl salicylate; taste, sweet at first but leaving an acrid taste.

Under a microscope <*5.01*>, a transverse section of the main root reveals a cork layer consisting of several cell rows of light brown cork cells; secondary cortex composed of parenchyma cells and phloen, traversed by medullary rays, 1 to 3 cells wide; medullary rays on xylem not distinct. Its parenchyma cells contain oil droplets, but starch grains and calcium oxalate crystals are absent.

Identification **(1)** Shake vigorously 0.5 g of pulverized Senega with 10 mL of water: a lasting fine foam is produced.

(2) Shake 0.5 g of pulverized Senega with 30 mL of water for 15 minutes, and filter. Take 1 mL of the filtrate, mix with 50 mL of water, and determine the absorption spectrum of the solution as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum at about 317 nm.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Senega according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Senega according to Method 4, and perform the test (not more than 5 ppm).

(3) Stem—When perform the test of foreign matter <*5.01*>, the amount of the stems contained in Senega does not exceed 2.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than the stems is not more than 1.0%.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Containers and storage Containers—Well-closed containers.

Powdered Senega

Senegae Radix Pulverata

セネガ末

Powdered Senega is the powder of Senega.

Description Powdered Senega occurs as a light brown powder, and has a characteristic odor resembling the aroma of methyl salicylate; taste, sweet at first, but later acrid.

Under a microscope <*5.01*>, Powdered Senega reveals fragments of pitted vessels, reticulate vessels and tracheids; fragments of xylem fibers with oblique pits; fragments of xylem parenchyma cells with simple pits; fragments of phloem parenchyma containing oily droplets; fragments of exodermis often composed of cells suberized and divided into daughter cells; oily droplets stained red by sudan III TS. The parenchyma cells of Powdered Senega do not contain starch grains and crystals of calcium oxalate.

Identification **(1)** Shake vigorously 0.5 g of Powdered Senega with 10 mL of water: a lasting fine foam is produced.

(2) Shake 0.5 g of Powdered Senega with 30 mL of water for 15 minutes, and filter. Take 1 mL of the filtrate, mix with 50 mL of water, and determine the absorption spectrum of the solution as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum at about 317 nm.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Senega according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Senega according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, stone cells, starch grains or crystals of calcium oxalate are not observed.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 30.0%.

Containers and storage Containers—Well-closed containers.

Senega Syrup

セネガシロップ

Method of preparation

Senega, in moderately fine cutting	40 g
Sucrose	780 g
10 vol% Ethanol	a sufficient quantity
Purified Water or Purified Water in Containers	a sufficient quantity
	To make 1000 mL

Add 400 mL of 10 vol% ethanol to Senega, and macerate for one or two days. Filter the extract, wash the residue with a small amount of 10 vol% Ethanol, filter, and combine the

filtrate of the extracts and washings until total volume measures about 500 mL. Dissolve Sucrose in the mixture, by warming if necessary, and dilute to 1000 mL with Purified Water or Purified Water in Containers. May be prepared with an appropriate quantity of Ethanol and Purified Water or Purified Water in Containers in place of 10 vol% Ethanol.

Description Senega Syrup is a yellow-brown, viscous liquid. It has a characteristic odor resembling methyl salicylate and a sweet taste.

Identification Add 5 mL of water to 1 mL of Senega Syrup, and shake: lasting small bubbles are produced.

Containers and storage Containers—Tight containers.

Senna Leaf

Sennae Folium

センナ

Senna Leaf is the leaflets of *Cassia angustifolia* Vahl or *Cassia acutifolia* Delile (*Leguminosae*).

It contains not less than 1.0% of total sennosides [sennoside A ($C_{42}H_{38}O_{20}$: 862.74) and sennoside B ($C_{42}H_{38}O_{20}$: 862.74)], calculated on the basis of dried material.

Description Lanceolate to narrow lanceolate leaflets, 1.5 – 5 cm in length, 0.5 – 1.5 cm in width, light grayish yellow to light grayish yellow-green in color; margin entire, apex acute, base asymmetric, petiole short; under a magnifying glass, vein marked, primary lateral veins running toward the apex along the margin and joining the lateral vein above; lower surface having slight hairs.

Odor slight; taste, bitter.

Under a microscope <*5.01*>, a transverse section of Senna Leaf reveals epidermis with thick cuticle, with numerous stomata, and with thick-walled, warty unicellular hairs; epidermal cells are often separated into two loculi by a septum which is in parallel with the surface of the leaf, and contain mucilage in the inner loculus; palisade of a single cellular layer under each epidermis; spongy tissue, consisting of 3 to 4 cellular layers, and containing clustered or solitary crystals of calcium oxalate; cells adjacent to vascular bundle, forming crystal cell rows.

Identification **(1)** Macerate 0.5 g of pulverized Senna Leaf in 10 mL of diethyl ether for 2 minutes, and filter. Add 5 mL of ammonia TS to the filtrate: a yellow-red color is produced in the aqueous layer. To the residue of maceration add 10 mL of water, and macerate for 2 minutes. Filter, and add 5 mL of ammonia TS: a yellow-red color is produced in the aqueous layer.

(2) To 2 g of pulverized Senna Leaf add 40 mL of a mixture of tetrahydrofuran and water (7:3), shake for 30 minutes, and centrifuge. Transfer the supernatant liquid to a separator, add 13 g of sodium chloride, and shake for 30 minutes. Separate the aqueous layer with undissolved sodium chloride, and adjust to pH 1.5 with 1 mol/L hydrochloric acid TS. Transfer this solution to another separator, shake with 30 mL of tetrahydrofuran for 10 minutes, separate the tetrahydrofuran layer, and use the tetrahydrofuran layer as the sample solution. Separately, dissolve 1 mg of Sennoside A RS or sennoside A for thin-layer chromatography in 1 mL of a mixture of tetrahydrofuran and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (40:40:30:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red fluorescent spot from the standard solution.

Purity **(1)** Rachis and fruit—When perform the test of foreign matter <*5.01*>, the amount of rachis and fruits contained in Senna Leaf does not exceed 5.0%.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than rachis and fruits contained in Senna Leaf does not exceed 1.0%.

(3) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 12.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Assay Weigh accurately about 0.5 g of pulverized Senna Leaf in a glass-stoppered centrifuge tube, add 25 mL of diluted methanol (7 in 10), shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 10 mL of diluted methanol (7 in 10), shake for 10 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure once more, combine all the extracts, add diluted methanol (7 in 10) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Sennoside A RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in a solution of sodium hydrogen carbonate (1 in 100) to make exactly 20 mL, and use this solution as standard stock solution (1). Weigh accurately about 10 mg of Sennoside B RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in a solution of sodium hydrogen carbonate (1 in 100) to make exactly 20 mL, and use this solution as standard stock solution (2). Pipet 5 mL of the standard stock solution (1) and 10 mL of the standard stock solution (2), add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{Ta} and A_{Sa}, of sennoside A, and the peak areas, A_{Tb} and A_{Sb}, of sennoside B in each solution, calculate the amounts of sennoside A and sennoside B by the following equations, and designate the total as the amount of total sennosides.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_{Sa} \times A_{Ta}/A_{Sa} \times 1/4$$

$$\text{Amount (mg) of sennoside B } (C_{42}H_{38}O_{20}) = M_{Sb} \times A_{Tb}/A_{Sb} \times 1/2$$

M_{Sa}: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

M_{Sb}: Amount (mg) of Sennoside B RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 2.45 g of tetra-*n*-heptylammonium bromide in 1000 mL of a mixture of diluted 1 mol/L acetic acid-sodium acetate buffer solution (pH 5.0) (1 in 10) and acetonitrile (17:8).

Flow rate: Adjust so that the retention time of sennoside A is about 26 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, sennoside B and sennoside A are eluted in this order with the resolution between these peaks being not less than 15, and the number of theoretical plates of the peak of sennoside A being not less than 8000.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Senna Leaf

Sennae Folium Pulveratum

センナ末

Powdered Senna Leaf is the powder of Senna Leaf. It contains not less than 1.0% of total sennosides [sennoside A ($C_{42}H_{38}O_{20}$: 862.74) and sennoside B ($C_{42}H_{38}O_{20}$: 862.74)], calculated on the basis of dried material.

Description Powdered Senna Leaf occurs as a light yellow to light grayish yellow-green powder. It has a slight odor and a bitter taste.

Under a microscope <*5.01*>, Powdered Senna Leaf reveals fragments of vessels and vein tissue accompanied with crystal cell rows; fragments of thick-walled, bent, unicellular hairs; fragments of palisade and spongy tissue; clustered and solitary crystals of calcium oxalate, 10 to 20 μm in diameter.

Identification **(1)** Macerate 0.5 g of Powdered Senna Leaf in 10 mL of diethyl ether for 2 minutes, and filter. Add 5 mL of ammonia TS to the filtrate: a yellow-red color is produced in the aqueous layer. To the residue of maceration add 10 mL of water, and macerate for 2 minutes. Filter, and add 5 mL of ammonia TS: a yellow-red color is produced in the aqueous layer.

(2) To 2 g of Powdered Senna Leaf add 40 mL of a mixture of tetrahydrofuran and water (7:3), shake for 30 minutes, and centrifuge. Transfer the supernatant liquid to a separator, add 13 g of sodium chloride, and shake for 30 minutes. Separate the aqueous layer with undissolved sodium chloride, and adjust to pH 1.5 with 1 mol/L hydrochloric acid TS. Transfer this solution to another separator, shake with 30 mL of tetrahydrofuran for 10 minutes, separate the tetrahydrofuran layer, and use the tetrahydrofuran layer as the sample solution. Separately, dissolve 1 mg of Sennoside A RS or sennoside A for thin-layer chromatography in 1 mL of a mixture of tetrahydrofuran and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solutions on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (40:40:30:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red fluorescent spot from the standard solution.

Purity **(1)** Foreign matter <*5.01*>—Under a microscope, stone cells and thick fibers are not observable.

(2) Total BHC's and total DDT's <*5.01*>—Not more than 0.2 ppm, respectively.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 12.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Assay Weigh accurately about 0.5 g of Powdered Senna Leaf in a glass-stoppered centrifuge tube, add 25 mL of diluted methanol (7 in 10), shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 10 mL of diluted methanol (7 in 10), shake for 10 minutes, centrifuge, and separate the supernatant liquid. Repeat this procedure once more, combine all the extracts, add diluted methanol (7 in 10) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Sennoside A RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in a solution of sodium hydrogen carbonate (1 in 100) to make exactly 20 mL, and use this solution as standard stock solution (1). Weigh accurately about 10 mg of Sennoside B RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in a solution of sodium hydrogen carbonate (1 in 100) to make exactly 20 mL, and use this solution as standard stock solution (2). Pipet 5 mL of the standard stock solution (1) and 10 mL of the standard stock solution (2), add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{Ta} and A_{Sa}, of sennoside A, and the peak areas, A_{Tb} and A_{Sb}, of sennoside B in each solution, calculate the amounts of sennoside A and sennoside B by the following equations, and designate the total as the amount of total sennoside.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_{Sa} \times A_{Ta}/A_{Sa} \times 1/4$$

$$\text{Amount (mg) of sennoside B } (C_{42}H_{38}O_{20}) = M_{Sb} \times A_{Tb}/A_{Sb} \times 1/2$$

M_{Sa}: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

M_{Sb}: Amount (mg) of Sennoside B RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: Dissolve 2.45 g of tetra-*n*-heptylammonium bromide in 1000 mL of a mixture of diluted 1 mol/L acetic acid-sodium acetate buffer solution (pH 5.0) (1 in 10) and acetonitrile (17:8).

Flow rate: Adjust so that the retention time of sennoside A is about 26 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, sennoside B and sennoside A are eluted in this order with the resolution between these peaks being not less than 15, and the number of theoretical plates of the peak of sennoside A being not less than 8000.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of sennoside A is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Sesame

Sesami Semen

ゴマ

Sesame is the seed of *Sesamum indicum* Linné (*Pedaliaceae*).

Description Ovate to spatulate seed, 3 – 4 mm in length, about 2 mm in width, about 1 mm in thickness; externally dark brown to black, rarely light brown to brown. Under a magnifying glass, thin ridges are observed on edges. 100 seeds weigh about 0.2 – 0.3 g.

Odorless; taste, slightly sweet and oily.

Under a microscope <*5.01*>, transverse section reveals a seed coat consisting of palisade epidermis and flattened parenchyma; in the interior, endosperm and cotyledon; epidermal cells contain orbicular crystals of calcium oxalate and black pigment; parenchymatous cells of endosperm and cotyledon contain oil drops and aleurone grains.

Identification Grind a suitable amount of Sesame. To 1.0 g of the ground add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of sesamin for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate and acetic acid (100) (10:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Sesame Oil

Oleum Sesami

ゴマ油

Sesame Oil is the fixed oil obtained from the seeds of *Sesamum indicum* Linné (*Pedaliaceae*).

Description Sesame Oil is a clear, pale yellow oil. It is odorless or has a faint, characteristic odor, and has a bland taste.

It is miscible with diethyl ether and with petroleum ether.

It is slightly soluble in ethanol (95).

It congeals between 0°C and −5°C.

Congealing point of the fatty acids: 20 – 25°C

Identification To 1 mL of Sesame Oil add 0.1 g of sucrose and 10 mL of hydrochloric acid, and shake for 30 seconds: the acid layer becomes light red and changes to red on standing.

Specific gravity <*1.13*> d_{25}^{25}: 0.914 – 0.921

Acid value <*1.13*> Not more than 0.2.

Saponification value <*1.13*> 187 – 194

Unsaponifiable matters <*1.13*> Not more than 2.0%.

Iodine value <*1.13*> 103 – 118

Containers and storage Containers—Tight containers.

Shakuyakukanzoto Extract

芍薬甘草湯エキス

Shakuyakukanzoto Extract contains not less than 50 mg and not more than 150 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 40 mg and not more than 120 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Peony Root	6 g	5 g
Glycyrrhiza	6 g	5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Shakuyakukanzoto Extract occurs as a light brown powder or brown viscous extract. It has slightly an odor, and a sweet taste.

Identification (1) Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 10 mL of water, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the

plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(2) Shake 0.5 g of the dry extract (or 1.5 g of the viscous extract) with 10 mL of water, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) according to Method 3, and perform the test (not more than 2 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 8.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Paeoniflorin—Weigh accurately about 0.2 g of the dry extract (or an amount of the viscous extract, equivalent to 0.2 g of dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(2) Glycyrrhizic acid—Weigh accurately about 0.2 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.2 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Shimbuto Extract

真武湯エキス

Shimbuto Extract contains not less than 26 mg and not more than 78 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), not less than 0.5 mg and not more than 2.0 mg (for preparation prescribed 0.8 g of Ginger) or not less than 0.6 mg and not more than 2.4 mg (for preparation prescribed 1 g of Ginger) or not less than 0.9 mg and not more than 3.6 mg (for preparation prescribed 1.5 g of Ginger) of [6]-gingerol, and not less than 0.7 mg (for preparation prescribed 1 g of Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride) or not less than 0.2 mg (for preparation prescribed 1 g of Powdered Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride, or as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride) or not less than 0.1 mg (for preparation prescribed 1 g of Powdered Processed Aconite Root 2) of total alkaloids (as benzoylmesaconine hydrochloride and 14-benzoylhypacomine hydrochloride) or not less than 0.1 mg (for preparation prescribed 0.5 g of Powdered Processed Aconite Root 1) of total alkaloids (as benzoylmesaconine hydrochloride and 14-anisoylaconine hydrochloride, or as benzoylmesaconine hydrochloride and benzoylhypaconine hydrochloride), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Poria Sclerotium	5 g	5 g	5 g	4 g
Peony Root	3 g	3 g	3 g	3 g
Atractylodes Rhizome	3 g	—	3 g	—
Atractylodes Lancea Rhizome	—	3 g	—	3 g
Ginger	1 g	1 g	0.8 g	1.5 g
Processed Aconite Root (Processed Aconite Root 1)	1 g	—	—	—
Powdered Processed Aconite Root (Powdered Processed Aconite Root 1)	—	1 g	—	0.5 g
Powdered Processed Aconite Root (Powdered Processed Aconite Root 2)	—	—	1 g	—

Prepare a dry extract as directed under Extracts, according to the prescription 1) to 4), using the crude drugs shown above.

Description Shimbuto Extract occurs as light yellow-brown to brown powder. It has a characteristic odor and a hot and bitter taste.

Identification **(1)** To 2.0 g of Shimbuto Extract, add 10 mL of water, shake, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(2) For preparation prescribed Atractylodes Rhizome—To 1.0 g of Shimbuto Extract, add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly diluted sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Atractylodes Rhizome).

(3) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of Shimbuto Extract, add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(4) To 1.0 g of Shimbuto Extract, add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(5) To 3.0 g of Shimbuto Extract, add 20 mL of diethyl ether and 2 mL of ammonia TS, shake for 10 minutes, centrifuge, and take the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of aceto-

nitrile to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of benzoylmesaconine hydrochloride for thin-layer chromatography in 10 mL of ethanol (99.5), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 10 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Processed Aconite Root or Powdered Processed Aconite Root).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Shimbuto Extract as directed in the Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of Shimbuto Extract according to Method 3, and perform the test (not more than 3 ppm).

(3) Aconitum diester alkaloids (aconitine, jesaconitine, hypaconitine and mesaconitine)—Weigh accurately 1.0 g of Shimbuto Extract, add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, proceed in the same manner, and repeat the procedure twice. Combine all the extracts, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue with exactly 10 mL of a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1). Centrifuge this solution, and use the supernatant liquid as the sample solution. Separately, pipet 1 mL of aconitum diester alkaloids standard solution for purity, add a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 40 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions: the heights of the peaks corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the sample solution are not higher than the respective heights corresponding to aconitine, jesaconitine, hypaconitine and mesaconitine from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for aconitine, hypaconitine and mesaconitine; 254 nm for jesaconitine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of mesaconitine is about 31 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of aconitum diester alkaloids standard solution for purity under the above operating conditions, using 254 nm, mesaconitine, hypaconitine, aconitine and jesaconitine are eluted in this order, and each resolution between these peaks is not less than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, using 231 nm, the relative standard deviation of the peak height of mesaconitine is not more than 1.5%.

Loss on drying <*2.41*> Not more than 7.0% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%.

Assay **(1)** Paeoniflorin—Weigh accurately about 0.5 g of Shimbuto Extract, add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(2) [6]-gingerol—Weigh accurately about 0.5 g of Shimbuto Extract, add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of [6]-gingerol for assay, dissolve in diluted methanol to make exactly 100 mL. Pipet 5 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of [6]-gingerol in each solution.

Amount (mg) of [6]-gingerol = $M_S \times A_T/A_S \times 1/20$

M_S: Amount (mg) of [6]-gingerol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 282 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (620:380:1).

Flow rate: 1.0 mL per minute (the retention time of [6]-gingerol is about 15 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of [6]-gingerol are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of [6]-gingerol is not more than 1.5%.

(3) Total alkaloids—Weigh accurately about 1 g of Shimbuto Extract, add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. Centrifuge this solution, remove the diethyl ether layer, then add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and take the diethyl ether layer. To the aqueous layer, add 1.0 mL of ammonia TS and 20 mL of diethyl ether, and repeat the above process twice more. Combine all the extracts, and evaporate the solvent under low pressure (in vacuo). Dissolve the residue with a mixture of phosphate buffer solution for processed aconite root and acetonitrile (1:1) to make exactly 10 mL. Centrifuge this solution, and use the supernatant liquid as the sample solution. Perform the test with exactly 20 μL each of the sample solution and the aconitum monoester alkaloids standard solution TS for assay as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine, A_{TM} and A_{SM}, A_{TH} and A_{SH}, as well as A_{TA} and A_{SA}, in each solution, respectively.

Amount (mg) of benzoylmesaconine hydrochloride = $C_{SM} \times A_{TM}/A_{SM} \times 10$

Amount (mg) of benzoylhypaconine hydrochloride = $C_{SH} \times A_{TH}/A_{SH} \times 10$

Amount (mg) of 14-anisoylaconine hydrochloride = $C_{SA} \times A_{TA}/A_{SA} \times 10$

C_{SM}: Concentration (mg/mL) of benzoylmesaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SH}: Concentration (mg/mL) of benzoylhypaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

C_{SA}: Concentration (mg/mL) of 14-anisoylaconine hydrochloride for assay in aconitum monoester alkaloids standard solution TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 231 nm for benzoylmesaconine and benzoylhypaconine; 254 nm for 14-anisoylaconine).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of phosphate buffer solution for processed aconite root and tetrahydrofuran (183:17).

Flow rate: 1.0 mL per minute (the retention time of benzoylmesaconine is about 15 minutes).

System suitability—

System performance: When the procedure is run with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of benzoylmesaconine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 20 μL of the aconitum monoester alkaloids standard solution TS for assay under the above operating conditions, the relative standard deviation of the peak areas of benzoylmesaconine, benzoylhypaconine and 14-anisoylaconine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Shosaikoto Extract

小柴胡湯エキス

Shosaikoto Extract contains not less than 2 mg and not more than 8 mg of saikosaponin b_2, not less than 80 mg and not more than 240 mg of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Bupleurum Root	7 g	6 g
Pinellia Tuber	5 g	5 g
Ginger	1 g	1 g
Scutellaria Root	3 g	3 g
Jujube	3 g	3 g
Ginseng	3 g	3 g
Glycyrrhiza	2 g	2 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Shosaikoto Extract occurs as a light brown to grayish brown powder or black-grayish brown viscous extract. It has a slight odor, and a sweet first then slightly pungent and bitter taste.

Identification **(1)** Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solu-

tion. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 15 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(3) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Scutellaria Root).

(4) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Ginsenoside Rb_1 RS or ginsenoside Rb_1 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Ginseng).

(5) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, then add 5 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <1.07>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to about 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <1.11>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <2.41> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <5.01> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separaterly, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

$$\text{Amount (mg) of saikosaponin } b_2 = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000

and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute (the retention time of baicalin is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of baicalein for resolution check in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and baicalein is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution be-

tween the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Shoseiryuto Extract

小青竜湯エキス

Shoseiryuto Extract contains not less than 8 mg and not more than 24 mg of the total alkaloids (ephedrine and pseudoephedrine), not less than 26 mg and not more than 78 mg of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 14 mg and not more than 42 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Ephedra Herb	3 g	3 g
Peony Root	3 g	3 g
Processed Ginger	3 g	—
Ginger	—	3 g
Glycyrrhiza	3 g	3 g
Cinnamon Bark	3 g	3 g
Asiasarum Root	3 g	3 g
Schisandra Fruit	3 g	3 g
Pinellia Tuber	6 g	6 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Shoseiryuto Extract occurs as a light brown to brown powder or black-brown viscous extract. It has a characteristic odor and a acid first then pungent taste.

Identification **(1)** Shake 1.0 g of dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol and shake, centrifuge, and use the 1-butanol layer as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid(100) (4:4:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly ninhydrin-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: a red-purple spot is observed at an *R*f value of about 0.5 (Ephedra Herb).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(3) For preparation prescribed Processed Ginger—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-shogaol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 1 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of cyclohexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Processed Ginger).

(4) For preparation prescribed Ginger—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of [6]-gingerol for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and spray water: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-green to grayish green spot from the standard solution (Ginger).

(5) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

(6) Perform the test according to the following i) or ii) (Cinnamon Bark).

i) Put 10 g of the dry extract (or 30 g of the viscous extract) in a 300-mL hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect the apparatus for essential oil determination, and heat under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to

the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use the layer as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

ii) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution.

(7) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of asarinin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown spot from the standard solution (Asiasarum Root).

(8) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of sodium hydroxide TS, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of schisandrin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-purple spot from the standard solution (Schisandra Fruit).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Cadmium—Take 5.0 g of the dry extract (or an amount of the viscous extract, equivalent to 5.0 g of the dried substance) in a platinum, quartz or porcelain crucible, heat weakly, then incinerate by ignition at 450°C. After cooling, add a small amount of 2 mol/L nitric acid TS to the residue, filter if necessary, wash the crucible several times with small portions of 2 mol/L nitric acid TS, combine the filtrate and washings, add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the sample solution. Separately, to 5.0 mL of Standard Cadmium Solution add 2 mol/L nitric acid TS to make exactly 20 mL, and use this solution as the standard solution. Perform the test with the sample solution and standard solution as directed under Atomic Absorption Spectrophotometry <*2.23*>: the absorbance of the sample solution is not more than that of the standard solution (not more than 1 ppm).

Gas: Combustible gas—Acetylene or hydrogen.
Supporting gas—Air.
Lamp: Cadmium hollow-cathode lamp.
Wavelength: 228.8 nm.

(3) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 12.0%, calculated on the dried basis.

Assay **(1)** Total alkaloids (ephedrine and pseudoephedrine)—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of dried substance), add 20 mL of diethyl ether, shake, then add 3.0 mL of 0.1 mol/L hydrochloric acid TS, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ethler layer. To the aqueous layer add 1.0 mL of ammonia TS and 20 mL of diethyl ether, shake for 30 minutes, centrifuge, and separate the diethyl ether layer. In addition, repeat twice in the same manner for the aqueous layer using 1.0 mL of ammonia TS and 20 mL of diethyl ether. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL. Centrifuge this solution, and use the supernatant liquid as the sample solution. Separately, weigh accurately about 10 mg of ephedrine hydrochloride for assay of crude drugs, previously dried at 105°C for 3 hours, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine the peak areas, A_{TE} and A_{TP}, of ephedrine and pseudoephedrine obtained from the sample solution, and the peak area, A_S, of ephedrine from the standard solution.

Amount (mg) of total alkaloids (ephedrine and pseudoephedrine)
$= M_S \times (A_{TE} + A_{TP})/A_S \times 1/10 \times 0.819$

M_S: Amount (mg) of ephedrine hydrochloride for assay of crude drugs taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 5 g of sodium lauryl sulfate add 350 mL of acetonitrile, shake, and add 650 mL of water and 1 mL of phosphoric acid to dissolve lauryl sulfate.

Flow rate: 1.0 mL per minute (the retention time of ephedrine is about 27 minutes).

System suitability—

System performance: Dissolve 1 mg each of ephedrine hydrochloride for assay of crude drugs and pseudoephedrine hydrochloride in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, pseudoephedrine and ephedrine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of ephedrine is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS, (separately determined the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

Amount (mg) of paeoniflorin ($C_{23}H_{28}O_{11}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Glycyrrhizic acid—Perform the test according to the following i) or ii).

i) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS, (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

Amount (mg) of glycyrrhizic acid ($C_{42}H_{62}O_{16}$)
$= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5. Dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 50 mL of methanol. To 2 mL of this solution add 2 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peaks of glycyrrhizic acid and (*E*)-cinnamaldehyde is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

ii) Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the su-

pernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Proceed as directed in the operating conditions in i).

System suitability—

System repeatability: Proceed as directed in the system suitability in i).

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

Containers and storage Containers—Tight containers.

Simple Ointment

単軟膏

Method of preparation

Yellow Beeswax	330 g
Fixed oil	a sufficient quantity
	To make 1000 g

Prepare as directed under Ointments, with the above ingredients.

Description Simple Ointment is yellow in color. It has a slight, characteristic odor.

Containers and storage Containers—Tight containers.

Sinomenium Stem and Rhizome

Sinomeni Caulis et Rhizoma

ボウイ

Sinomenium Stem and Rhizome is the climbing stem and rhizome of *Sinomenium acutum* Rehder et Wilson (*Menispermaceae*), usually cut transversely.

Description Round or elliptic sections, 0.2 – 0.4 cm in thickness, 1 – 4.5 cm in diameter; cortex on both fractured surfaces, light brown to dark brown; in xylem, grayish brown vessel portions and dark brown medullary rays lined alternately and radially; flank, dark gray, with longitudinal wrinkles and warty protrusions.

Almost odorless; taste, bitter.

Under a microscope <5.01>, a transverse section reveals extremely thick-walled stone cells in primary cortex and pericycle; irregular-sized vessels lined nearly stepwise in the vessel portion; cells of medullary ray mostly not lignified, and extremely thick-walled and large stone cells scattered here and there; primary cortex containing needle crystals of calcium oxalate; medullary rays containing starch gains, mainly simple grain, 3 – 20 μm in diameter, and small needle crystals of calcium oxalate.

Identification To 1.0 g of pulverized Sinomenium Stem and Rhizome add 5 mL of methanol, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of sinomenine for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <2.03>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, 1-propanol, water and acetic acid (100) (3:3:2:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, air-dry the plate, and spray evenly sodium nitrite TS on the plate: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and the same *R*f value. Further, a spot with the same color tone appears immediately below the spot.

Total ash <5.01> Not more than 7.0%.

Acid-insoluble ash <5.01> Not more than 0.5%.

Containers and storage Containers—Well-closed containers.

Smilax Rhizome

Smilacis Rhizoma

サンキライ

Smilax Rhizome is the rhizome of *Smilax glabra* Roxburgh (*Liliaceae*).

Description Flattened and irregular cylindrical tuber, often with node-like branches; usually 5 – 15 cm in length, 2 – 5 cm in diameter; the outer surface grayish yellow-brown to yellow-brown, and the upper surface scattered with knotty remains of stem; transverse section irregular elliptical to obtuse triangular, whitish to reddish white, consisting of extremely thin cortex and mostly of stele.

Odor, slight; almost tasteless.

Under a microscope <5.01>, a transverse section reveals a 2- to 3-cell-wide cork layer, with extremely narrow cortical layer, usually consisting of a 2- to 4-cell-wide, thick-walled parenchyma cells, showing large mucilage cells here and there; mucilage cell containing raphides of calcium oxalate; stele consisting chiefly of parenchyma cells, and scattered with vascular bundles; parenchyma cells containing starch grains composed mostly of simple grains, 12 – 36 μm in diameter, and sometimes mixed with 2- to 4-compound grains.

Purity **(1)** Heavy metals <1.07>—Proceed with 3.0 g of pulverized Smilax Rhizome according to Method 3, and per-

form the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Smilax Rhizome according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 5.0%.

Containers and storage Containers—Well-closed containers.

Powdered Smilax Rhizome

Smilacis Rhizoma Pulveratum

サンキライ末

Powdered Smilax Rhizome is the powder of Smilax Rhizome.

Description Powdered Smilax Rhizome occurs as a light yellow-brown powder, and has a slight odor, and is practically tasteless.

Under a microscope <*5.01*>, Powdered Smilax Rhizome reveals starch grains and fragments of parenchyma cells containing them; fragments of raphides of calcium oxalate contained in mucilage masses; fragments of lignified parenchyma cells of cortical layer; fragments of cork cells and scalariform vessels; starch grains composed mostly of simple grains, and mixed with a few 2- to 4-compound grains 12 – 36 μm in diameter.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Smilax Rhizome according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Smilax Rhizome according to Method 4, and perform the test (not more than 5 ppm).

(3) Foreign matter—Under a microscope <*5.01*>, Powdered Smilax Rhizome does not show a large quantity of stone cells or thick-walled fibers.

Total ash <*5.01*> Not more than 5.0%.

Containers and storage Containers—Well-closed containers.

Sodium Bicarbonate and Bitter Tincture Mixture

苦味重曹水

Method of preparation

Sodium Bicarbonate	30 g
Bitter Tincture	20 mL
Water, Purified Water or Purified Water in Containers	a sufficient quantity
To make	1000 mL

Prepare before use, with the above ingredients.

Description Sodium Bicarbonate and Bitter Tincture Mixture is a clear, yellowish liquid, having a bitter taste.

Containers and storage Containers—Tight containers.

Anhydrous Sodium Sulfate

Sal Mirabilis Anhydricus

無水ボウショウ

Na_2SO_4: 142.04
[*7757-82-6*]

Anhydrous Sodium Sulfate is mainly sodium sulfate (Na_2SO_4) containing no water of crystallization.

It, when dried, contains not less than 99.0% of sodium sulfate (Na_2SO_4).

Description Anhydrous Sodium Sulfate occurs as white, crystals or powder. It is odorless and has a salty and slightly bitter taste.

It is freely soluble in water, and practically insoluble in ethanol (99.5).

Identification **(1)** A solution of Anhydrous Sodium Sulfate (1 in 20) responds to Qualitative Tests <*1.09*> (1) for sodium salt.

(2) A solution of Anhydrous Sodium Sulfate (1 in 20) responds to Qualitative Tests <*1.09*> (1) for sulfate.

Purity **(1)** Acidity or alkalinity—Dissolve 0.5 g of Anhydrous Sodium Sulfate in 5 mL of freshly boiled and cooled water: the solution is clear and colorless, and neutral.

(2) Chloride <*1.03*>—Perform the test with 0.5 g of previously dried Anhydrous Sodium Sulfate. Prepare the control solution with 0.5 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.036%).

(3) Heavy metals <*1.07*>—Proceed with 2.0 g of previously dried Anhydrous Sodium Sulfate according to Method 1, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 10 ppm).

(4) Arsenic <*1.11*>—Prepare the test solution with 1.0 g of previously dried Anhydrous Sodium Sulfate according to Method 1, and perform the test (not more than 2 ppm).

Loss on drying <*2.41*> Not more than 0.5% (4 g, 105°C, 4 hours).

Assay Weigh accurately about 0.4 g of previously dried Anhydrous Sodium Sulfate, dissolve in 200 mL of water, add 1 mL of hydrochloric acid, boil, and gradually add 8 mL of barium chloride TS. Heat the solution in a water bath for 1 hour. After cooling, filter through a filter paper for quantitative analysis (No.5C), wash the residue on the filter paper with water until the washings do not give the turbidity with silver nitrate TS. After drying the residue together with the paper, ignite at 500 – 800°C to constant mass, and weigh the mass of the residue as the amount of barium sulfate ($BaSO_4$: 233.39).

Amount (mg) of sodium sulfate (Na_2SO_4)
= amount (mg) of barium sulfate ($BaSO_4$) × 0.609

Containers and storage Containers—Well-closed containers.

Sodium Sulfate Hydrate

Sal Mirabilis

ボウショウ

$Na_2SO_4.10H_2O$: 322.19
[*7727-73-3*]

Sodium Sulfate Hydrate is mainly decahydrate of sodium sulfate (Na_2SO_4).

It, when dried, contains not less than 99.0% of sodium sulfate (Na_2SO_4: 142.04).

Description Sodium Sulfate Hydrate occurs as colorless or white, crystals or crystalline powder. It is odorless and has a cooling and salty taste.

It is freely soluble in water, and practically insoluble in ethanol (99.5).

It is quickly efflorescent in air, soluble in its own water of crystallization at about 33°C and lost the water at 100°C.

Identification **(1)** A solution of Sodium Sulfate Hydrate (1 in 20) responds to Qualitative Tests <*1.09*> (1) for sodium salt.

(2) A solution of Sodium Sulfate Hydrate (1 in 20) responds to Qualitative Tests <*1.09*> (1) for sulfate.

Purity **(1)** Acidity or alkalinity—Dissolve 0.5 g of Sodium Sulfate Hydrate in 5 mL of freshly boiled and cooled water: the solution is clear and colorless, and neutral.

(2) Chloride <*1.03*>—Perform the test with 0.5 g of previously dried Sodium Sulfate Hydrate. Prepare the control solution with 0.5 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.036%).

(3) Heavy metals <*1.07*>—Proceed with 2.0 g of previously dried Sodium Sulfate Hydrate according to Method 1, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 10 ppm).

(4) Arsenic <*1.11*>—Prepare the test solution with 1.0 g of previously dried Sodium Sulfate Hydrate according to Method 1, and perform the test (not more than 2 ppm).

Loss on drying <*2.41*> 51.0 – 57.0% (4 g, 105°C, 4 hours).

Assay Weigh accurately about 0.4 g of previously dried Sodium Sulfate Hydrate, dissolve in 200 mL of water, add 1 mL of hydrochloric acid, boil, and gradually add 8 mL of barium chloride TS. Heat the solution in a water bath for 1 hour. After cooling, filter through a filter paper for quantitative analysis (No.5C), wash the residue on the filter paper with water until the washings do not give the turbidity with silver nitrate TS. After drying the residue together with the paper, ignite at 500 – 800°C to constant mass, and weigh the mass of the residue as the amount of barium sulfate ($BaSO_4$: 233.39).

Amount (mg) of sodium sulfate (Na_2SO_4)
= amount (mg) of barium sulfate ($BaSO_4$) × 0.609

Containers and storage Containers—Well-closed containers.

Sophora Root

Sophorae Radix

クジン

Sophora Root is the root of *Sophora flavescens* Aiton (*Leguminosae*) or often such root from which the periderm has been removed.

Description Cylindrical root, 5 – 20 cm in length, 2 – 3 cm in diameter; externally dark brown to yellow-brown, with distinct longitudinal wrinkles, and with laterally extended lenticels; root without periderm, externally yellow-white, with somewhat fibrous surface; under a magnifying glass, the transverse section, light yellow-brown; cortex, 0.1 – 0.2 cm in thickness, slightly tinged with dark color near cambium, forming a crack between xylem.

Odor, slight; taste, extremely bitter and lasting.

Identification To 1.0 g of pulverized Sophora Root add 10 mL of methanol, shake for 10 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethanol (99.5), ethyl acetate and ammonia solution (28) (3:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: two brown spots appear at an *R*f value of about 0.5.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Sophora Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Sophora Root according to Method 4, and perform the test (not more than 5 ppm).

(3) Stem—When perform the test of foreign matter <*5.01*>, the amount of its stems contained in Sophora Root does not exceed 10.0%.

(4) Foreign matter <*5.01*>—The amount of foreign matter other than stems is not more than 1.0%.

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Sophora Root

Sophorae Radix Pulverata

クジン末

Powdered Sophora Root is the powder of Sophora Root.

Description Powdered Sophora Root occurs as a light brown powder. It has a slight odor, and an extremely bitter and lasting taste.

Under a microscope <*5.01*>, Powdered Sophora Root reveals mainly starch grains and fragments of parenchyma

cells containing them, fibers, bordered pitted vessels, reticulate vessels; a few fragments of corky tissue and solitary crystals of calcium oxalate. Starch grains usually composed of 2- to 4-compound grains 15 – 20 μm in diameter, and simple grains 2 – 5 μm in diameter.

Identification To 1.0 g of Powdered Sophora Root add 10 mL of methanol, shake for 10 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethanol (99.5), ethyl acetate, and ammonia solution (28) (3:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: two brown spots appear at an *R*f value of about 0.5.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Sophora Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Sophora Root according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 6.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Soybean Oil

Oleum Sojae

ダイズ油

Soybean Oil is the fixed oil obtained from the seeds of *Glycine max* Merrill (*Leguminosae*).

Description Soybean Oil is a clear, pale yellow oil. It is odorless or has a slight odor, and has a bland taste.

It is miscible with diethyl ether and with petroleum ether.

It is slightly soluble in ethanol (95), and practically insoluble in water.

It congeals between −10°C and −17°C.

Congealing point of the fatty acids: 22 – 27°C

Specific gravity <*1.13*> d_{25}^{25}: 0.916 – 0.922

Acid value <*1.13*> Not more than 0.2.

Saponification value <*1.13*> 188 – 195

Unsaponifiable matter <*1.13*> Not more than 1.0%.

Iodine value <*1.13*> 126 – 140

Containers and storage Containers—Tight containers.

Sweet Hydrangea Leaf

Hydrangeae Dulcis Folium

アマチャ

Sweet Hydrangea Leaf is the leaf and twig of *Hydrangea macrophylla* Seringe var. *thunbergii* Makino (*Saxifragaceae*), usually crumpled.

Description Usually wrinkled and contracted leaf, dark green to dark yellow-green in color. When soaked in water and smoothed out, it is lanceolate to acuminately ovate, 5 – 15 cm in length, 2 – 10 cm in width; margin serrated, base slightly wedged; coarse hair on both surfaces, especially on the veins; lateral veins not reaching the margin but curving upwards and connecting with each other; petiole short and less than one-fifth of the length of lamina.

Odor, slight; taste, characteristically sweet.

Identification To 1.0 g of pulverized Sweet Hydrangea Leaf add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 2 mg of sweet hydrangea leaf dihydroisocoumarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of diethyl ether, hexane and formic acid (5:5:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two of the several spots obtained from the sample solution have the same color tone and *R*f value with the spots from the standard solution.

Purity **(1)** Stem—When perform the test of foreign matter <*5.01*>, the amount of stems contained in Sweet Hydrangea Leaf does not exceed 3.0%.

(2) Foreign matter <*5.01*>—The amount of foreign matter other than stems contained in Sweet Hydrangea Leaf does not exceed 1.0%.

Loss on drying <*5.01*> Not more than 13.0% (6 hours).

Total ash <*5.01*> Not more than 12.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Containers and storage Containers—Well-closed containers.

Powdered Sweet Hydrangea Leaf

Hydrangeae Dulcis Folium Pulveratum

アマチャ末

Powdered Sweet Hydrangea Leaf is the powder of Sweet Hydrangea Leaf.

Description Powdered Sweet Hydrangea Leaf occurs as a dark yellow-green powder, and has a faint odor and a characteristic, sweet taste.

Under a microscope <*5.01*>, Powdered Sweet Hydrangea Leaf reveals fragments of epidermis with wavy lateral cell wall; stomata with two subsidiary cells; unicellular and thin-

walled hair with numerous protrusions of the surface, 150 – 300 μm in length; fragments of palisade tissue and spongy tissue; fragments of vascular bundle and mucilage cells containing raphides of calcium oxalate 50 – 70 μm in length.

Identification To 1.0 g of Powdered Sweet Hydrangea Leaf add 10 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 2 mg of sweet hydrangea leaf dihydroisocoumarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of diethyl ether, hexane and formic acid (5:5:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two of the several spots obtained from the sample solution have the same color tone and *R*f value with the spots from the standard solution.

Purity Foreign matter <*5.01*>—Under a microscope, Powdered Sweet Hydrangea Leaf does not show stone cells, a large quantity of fibers or starch grains.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 12.0%.

Acid-insoluble ash <*5.01*> Not more than 2.5%.

Containers and storage Containers—Well-closed containers.

Swertia Herb

Swertiae Herba

センブリ

Swertia Herb is the whole herb of *Swertia japonica* Makino (*Gentianaceae*) collected during the blooming season.

It contains not less than 2.0% of swertiamarin ($C_{16}H_{22}O_{10}$: 374.34), calculated on the basis of dried material.

Description Herb, 10 – 50 cm in length, having flowers, opposite leaves, stems, and, usually, with short, lignified roots; stems square, about 2 mm in diameter, often with branches; the leaves and stems dark green to dark purple or yellow-brown in color; the flowers white to whitish, and the roots yellow-brown. When smoothed by immersing in water, leaves, linear or narrow lanceolate, 1 – 4 cm in length, 0.1 – 0.5 cm in width, entire, and sessile; corolla split deeply as five lobes; the lobes narrow, elongated ellipse shape, and under a magnifying glass, with two elliptical nectaries juxtaposed at the base of the inner surface; the margin of lobe resembles eyelashes; the five stamens grow on the tube of the corolla and stand alternately in a row with corolla-lobes; peduncle distinct.

Odor, slight; taste, extremely bitter and persisting.

Identification To 0.5 g of pulverized Swertia Herb add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Swertiamarin RS or swertiamarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and *R*f value.

Purity Foreign matter <*5.01*>—The amount of straw and other foreign matters contained in Swertia Herb is not more than 1.0%.

Loss on drying <*5.01*> Not more than 12.0% (6 hours).

Total ash <*5.01*> Not more than 6.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 20.0%.

Assay Weigh accurately about 1 g of moderately fine powder of Swertia Herb in a glass-stoppered centrifuge tube, add 40 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 40 mL of methanol, and proceed in the same manner. Combine the extracts, and add methanol to make exactly 100 mL. Pipet 5 mL of the solution, add the mobile phase to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Swertiamarin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in methanol to make exactly 20 mL. Pipet 5 mL of the solution, add the mobile phase to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of swertiamarin in each solution.

$$\text{Amount (mg) of swertiamarin } (C_{16}H_{22}O_{10}) = M_S \times A_T/A_S \times 5$$

M_S: Amount (mg) of Swertiamarin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 238 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water and acetonitrile (91:9).

Flow rate : Adjust so that the retention time of swertiamarin is about 12 minutes.

System suitability—

System performance: Dissolve 1 mg each of Swertiamarin RS and theophylline in the mobile phase to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, theophylline and swertiamarin are eluted in this order with the resolution of these peaks being not less than 10.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of swertiamarin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Swertia Herb

Swertiae Herba Pulverata

センブリ末

Powdered Swertia Herb is the powder of Swertia Herb.

It contains not less than 2.0% of swertiamarin ($C_{16}H_{22}O_{10}$: 374.34), calculated on the basis of dried material.

Description Powdered Swertia Herb occurs as a grayish yellow-green to yellow-brown powder. It has a slight odor, and extremely bitter, persistent taste.

Under a microscope ⟨*5.01*⟩, Powdered Swertia Herb reveals xylem tissues with fibers (components of stems and roots); assimilation tissues (components of leaves and calyces); striated epidermis (components of stems and peduncles); tissues of corollas and filaments with spiral vessels; cells of anthers and their inner walls; spherical pollen grains with granular patterns (components of flowers), about 30 μm in diameter; starch grains are simple grain, about 6 μm in diameter, and very few.

Identification To 0.5 g of Powdered Swertia Herb add 10 mL of methanol, shake for 5 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of Swertiamarin RS or swertiamarin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution and the spot from the standard solution show the same color tone and *R*f value.

Purity Foreign matter—Under a microscope ⟨*5.01*⟩, crystals of calcium oxalate, a large quantity of starch grains and groups of stone cells are not observable.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 6.5%.

Acid-insoluble ash ⟨*5.01*⟩ Not more than 2.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 20.0%.

Assay Weigh accurately about 1 g of Powdered Swertia Herb in a glass-stoppered centrifuge tube, add 40 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 40 mL of methanol, and proceed in the same manner. Combine the extracts, and add methanol to make exactly 100 mL. Pipet 5 mL of the solution, add the mobile phase to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Swertiamarin RS (separately determine the water ⟨*2.48*⟩ by coulometric titration, using 10 mg), dissolve in methanol to make exactly 20 mL. Pipet 5 mL of the solution, add the mobile phase to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of swertiamarin in each solution.

$$\text{Amount (mg) of swertiamarin } (C_{16}H_{22}O_{10}) = M_S \times A_T/A_S \times 5$$

M_S: Amount (mg) of Swertiamarin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 238 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water and acetonitrile (91:9).

Flow rate: Adjust so that the retention time of swertiamarin is about 12 minutes.

System suitability—

System performance: Dissolve 1 mg each of Sweriamarin RS and theophylline in the mobile phase to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, theophylline and swertiamarin are eluted in this order with the resolution of these peaks being not less than 10.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of swertiamarin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Swertia and Sodium Bicarbonate Powder

センブリ·重曹散

Method of preparation

Powdered Swertia Herb	30 g
Sodium Bicarbonate	700 g
Starch, Lactose Hydrate or their mixture	a sufficient quantity
	To make 1000 g

Prepare as directed under Powders, with the above ingredients.

Description Swertia and Sodium Bicarbonate Powder occurs as a light grayish yellow powder, having a bitter taste.

Identification (1) To 10 g of Swertia and Sodium Bicarbonate Powder add 10 mL of ethanol (95), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 2 mg of Swertiamarin RS or swertiamarin for thin-layer chromatography in 1 mL of ethanol (95), and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Proceed as directed in the Identifica-

tion under Powdered Swertia Herb.

(2) To 0.5 g of Swertia and Sodium Bicarbonate Powder add 10 mL of water. After stirring, centrifuge the mixture with 500 revolutions per minute. Smear, using a small glass rod, the slide glass with a small amount of the precipitate, add 1 drop of a mixture of water and glycerin (1:1), and put a cover glass on it so that the tissue section spreads evenly without overlapping each other, taking precaution against inclusion of bubbles, and use this as the preparation for microscopic examination. If the precipitate separates into two layers, proceed with the upper layer in the same manner, and use as the preparation for microscopic examination. Heat the preparation for microscopic examination in a short time: the preparation reveals the yellow-green to yellow-brown, approximately spherical pollen grains with granular patterns under a microscope <*5.01*>. The pollen grains are 25 – 34 μm in diameter.

(3) The supernatant liquid obtained in (2) by centrifuging responds to Qualitative Tests <*1.09*> (1) for bicarbonate.

Containers and storage Containers—Well-closed containers.

Toad Cake

Bufonis Crustum

センソ

Toad Cake is the parotoid secretion of *Bufo gargarizans* Cantor or *Bufo melanostictus* Schneider (*Bufonidae*).

When dried, it contains not less than 5.8% of bufo steroid (bufalin, cinobufagin and resibufogenin).

Description A round disk with slightly dented bottom and protuberant surface, about 8 cm in diameter, about 1.5 cm in thickness, the mass of one disk being about 80 to 90 g; or a round disk with almost flattened surfaces on both sides, about 3 cm in diameter, and about 0.5 cm in thickness, the mass of one disk being about 8 g; externally red-brown to black-brown, somewhat lustrous, approximately uniform and horny, hard in texture, and difficult to break; fractured surface nearly flat, and edges of broken pieces red-brown and translucent.

Odorless.

Identification To 0.3 g of pulverized Toad Cake add 3 mL of acetone, shake for 10 minutes, filter, and use the filtrate as the sample solution. Separately, dissolve 1 mg of resibufogenin for thin-layer chromatography in 2 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of cyclohexane and acetone (3:2) to a distance of about 10 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the spot from the standard solution.

Total ash <*5.01*> Not more than 5.0%.

Acid-insoluble ash <*5.01*> Not more than 2.0%.

Assay Weigh accurately about 0.5 g of pulverized Toad Cake, previously dried in a desiccator (silica gel) for 24 hours, add 50 mL of methanol, heat under a reflux condenser for 1 hour, cool, and filter. Wash the residue with 30 mL of methanol, and combine the washing and filtrate. To this solution add methanol to make exactly 100 mL. Pipet 10 mL of this solution, add exactly 5 mL of the internal standard solution, add methanol to make 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg, about 20 mg and about 20 mg of bufalin for assay, cinobufagin for assay and resibufogenin for assay, respectively, previously dried in a desiccator (silica gel) for 24 hours, and dissolve in methanol to make exactly 100 mL. Pipet 10 mL of this solution, proceed in the same manner as the sample solution, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Calculate the ratios, Q_{TB} and Q_{SB}, of the peak area of bufalin, Q_{TC} and Q_{SC}, of the peak area of cinobufagin, and Q_{TR} and Q_{SR}, of the peak area of resibufogenin, respectively, to that of the internal standard, and designate the total amount as an amount of bufosteroid.

Amount (mg) of bufalin = $M_{SB} \times Q_{TB}/Q_{SB}$

Amount (mg) of cinobufagin = $M_{SC} \times Q_{TC}/Q_{SC}$

Amount (mg) of resibufogenin = $M_{SR} \times Q_{TR}/Q_{SR}$

M_{SB}: Amount (mg) of bufalin for assay taken
M_{SC}: Amount (mg) of cinobufagin for assay taken
M_{SR}: Amount (mg) of resibufogenin for assay taken

Internal standard solution—A solution of indometacin in methanol (1 in 4000).

Operating conditions—

Detector: An ultraviolet spectrophotometer (wavelength: 300 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 30 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (11:9).

Flow rate: Adjust so that the retention time of the internal standard is 16 to 19 minutes.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, bufalin, cinobufagin, resibufogenin and the internal standard are eluted in this order with the resolution among these peaks being not less than 1.5.

Containers and storage Containers—Well-closed containers.

Tokakujokito Extract

桃核承気湯エキス

Tokakujokito Extract contains not less than 38 mg and not more than 152 mg of amygdalin, not less than 1 mg and not more than 4 mg of (*E*)-cinnamic acid, not less than 3 mg of sennosides A ($C_{42}H_{38}O_{20}$: 862.74) or not less than 9 mg of rhein, and not less than 10 mg and not more than 30 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Peach Kernel	5 g	5 g	5 g
Cinnamon Bark	4 g	4 g	4 g
Rhubarb	3 g	3 g	3 g
Glycyrrhiza	1.5 g	1.5 g	1.5 g
Anhydrous Sodium Sulfate	1 g	0.9 g	
Sodium Sulfate			2 g

Prepare a dry extract as directed under Extracts, according to the prescription 1) to 3), using the crude drugs shown above. Or, prepare a dry extract by adding Light Anhydrous Silicic Acid to an extractive, prepared as directed under Extracts, according to the prescription 2), using the crude drugs shown above.

Description Tokakujokito Extract occurs as a green-yellow-brown to dark brown powder. It has characteristic odor and, salty, slightly astringent, and then slightly sweet taste.

Identification **(1)** To 1.0 g of Tokakujokito Extract add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 2 mg of amygdalin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-propanol, ethyl acetate and water (4:4:3) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 10 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the green-brown spot from the standard solution (Peach Kernel).

(2) Perform the test according to the following (i) or (ii) (Cinnamon Bark).

(i) Put 10 g of Tokakujokito Extract in a 300-mL of hard-glass flask, add 100 mL of water and 1 mL of silicone resin, connect the apparatus for essential oil determination, and heat to boil under a reflux condenser. The graduated tube of the apparatus is to be previously filled with water to the standard line, and 2 mL of hexane is added to the graduated tube. After heating under reflux for 1 hour, separate the hexane layer, and use this solution as the sample solution. Separately, dissolve 1 mg of (*E*)-cinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 2,4-dinitrophenylhydrazine TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-orange spot from the standard solution.

(ii) To 2.0 g of Tokakujokito Extract add 10 mL of water, shake, then add 5 mL of hexane, shake, centrifuge, and use the hexane layer as the sample solution. Separately, dissolve 1 mg of (*E*)-2-methoxycinnamaldehyde for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 40 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane and ethyl acetate (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the bluish white fluorescent spot from the standard solution.

(3) To 1.0 g of Tokakujokito Extract add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg of rhein for thin-layer chromatography in 10 mL of acetone, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the orange fluorescent spot from the standard solution (Rhubarb).

(4) To 1.0 g of Tokakujokito Extract add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Tokakujokito Extract as directed in Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of Tokakujokito Extract according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 8.0% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not less than 20.0% and more than 40.0%.

Assay **(1)** Amygdalin—Weigh accurately about 0.5 g of

Tokakujokito Extract, add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, elute through a column prepared previously with 2 g of polyamide for column chromatography using water to make exactly 20 mL of effluent , and use this solution as the sample solution. Separately, weigh accurately about 10 mg of amygdalin for assay, previously dried in a desiccator (silica gel) for 24 hours or more, and dissolve in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of amygdalin in each solution.

$$\text{Amount (mg) of amygdalin} = M_S \times A_T/A_S \times 4$$

M_S: Amount (mg) of amygdalin for assay taken

Operation conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 45°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and methanol (5:1).

Flow rate: 0.8 mL per minute (the retention time of amygdalin is about 12 minutes).

Systemic suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of amygdalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of amygdalin is not more than 1.5%.

(2) (*E*)-Cinnamic acid—Conduct this procedure using light-resistant vessels. Weigh accurately about 0.5 g of Tokakujokito Extract, add 20 mL of diethyl ether and 10 mL of water, shake for 10 minutes, centrifuge, and separate the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, proceed in the same manner as above, and repeat this procedure two more times. Combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue in diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-cinnamic acid for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 10 mL of this solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of (*E*)-cinnamic acid in each solution.

$$\text{Amount (mg) of (}E\text{)-cinnamic acid} = M_S \times A_T/A_S \times 1/20$$

M_S: Amount (mg) of (*E*)-cinnamic acid for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 273 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (800:200:1).

Flow rate: 1.0 mL per minute (the retention time of (*E*)-cinnamic acid is about 22 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of (*E*)-cinnamic acid are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of (*E*)-cinnamic acid is not more than 1.5%.

(3) Sennoside A—Weigh accurately about 0.5 g of Tokakujokito Extract, add 20 mL of ethyl acetate and 10 mL of water, shake for 10 minutes, centrifuge, remove the ethyl acetate layer, then add 20 mL of ethyl acetate, proceed in the same manner as above, and remove the ethyl acetate layer. To the aqueous layer obtained add 10 mL of methanol, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and separate the supernatant liquid. Combine all the supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of Sennoside A RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 200 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of sennoside A in each solution.

$$\text{Amount (mg) of sennoside A } (C_{42}H_{38}O_{20}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Sennoside A RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (840:160:1).

Flow rate: 1.0 mL per minute (the retention time of sennoside A is about 20 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of sennoside A are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak

area of sennoside A is not more than 1.5%.

(4) Rhein—Weigh accurately about 0.5 g of Tokakujokito Extract, add 80 mL of water, shake, and add water to make exactly 100 mL. Pipet 5 mL of this solution, add 20 mL of iron (III) chloride TS, heat under a reflux condenser for 30 minutes, add 3 mL of hydrochloric acid, and heat in addition under a reflux condenser for 30 minutes. After cooling, extract three times with 25 mL each of diethyl ether, combine all the extracts, evaporate the solvent under low pressure (in vacuo), dissolve the residue to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg of rhein for assay, and dissolve in acetone to make exactly 100 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of rhein in each solution.

$$\text{Amount (mg) of rhein} = M_S \times A_T/A_S \times 4/5$$

M_S: Amount (mg) of rhein for assay taken, calculated on the basis of the content obtained by qNMR

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 278 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 50°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (650:350:1).

Flow rate: 1.0 mL per minute (the retention time of rhein is about 17 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of rhein are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of rhein is not more than 1.5%.

(5) Glycyrrhizic acid—Weigh accurately about 0.5 g of Tokakujokito Extract, add 20 mL of ethyl acetate and 10 mL of water, and shake for 10 minutes. After centrifugation remove the ethyl acetate layer, add 20 mL of ethyl acetate, proceed in the same manner as described above, and remove the ethyl acetate layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.

Tokishakuyakusan Extract

当帰芍薬散エキス

Tokishakuyakusan Extract contains not less than 0.6 mg and not more than 2.4 mg of (*E*)-ferulic acid, not less than 34 mg and not more than 102 mg (for preparation prescribed 4 g of Peony Root) or not less than 51 mg and not more than153 mg (for preparation prescribed 6 g of Peony Root) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), and not less than 0.4 mg of atractylenolide III (for preparation prescribed Atractylodes Rhizome) or not less than 0.1 mg of atractylodin (for preparation prescribed Atractylodes Lancea Rhizome), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)	4)
Japanese Angelica Root	3 g	3 g	3 g	3 g
Cnidium Rhizome	3 g	3 g	3 g	3 g
Peony Root	6 g	6 g	4 g	4 g
Poria Sclerotium	4 g	4 g	4 g	4 g
Atractylodes Rhizome	4 g	4 g	4 g	—
Atractylodes Lancea Rhizome	—	—	—	4 g
Alisma Tuber	4 g	5 g	4 g	4 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the preparation 1) to 4), using the crude drugs shown above.

Description Tokishakuyakusan Extract is a light brown to

brown powder or black-brown viscous extract. It has a characteristic odor, and a slight sweet taste at first and a bitter taste later.

Identification **(1)** Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 15 mL of water and 5 mL of 0.1 mol/L hydrochloric acid TS, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root; Cnidium Rhizome).

(2) Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 5 minutes: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple spot from the standard solution (Peony Root).

(3) For preparation prescribed Atractylodes Rhizome—Shake 1.0 g of the dry extract (or 3.0 g of the viscous extract) with 10 mL of water, add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenolide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Atractylodes Rhizome).

(4) For preparation prescribed Atractylodes Lancea Rhizome—Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 10 mL of water, add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 0.5 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(5) Shake 2.0 g of the dry extract (or 6.0 g of the viscous extract) with 20 mL of water and 2 mL of ammonia solution (28), add 20 mL of a mixture of hexane and ethyl acetate (20:1), shake, and centrifuge. Separate the upper layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of methanol to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of alisol A for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl formate, water and formic acid (30:1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid-acetic acid TS on the plate, heat the plate at 105°C for 5 minutes, allow to cool, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Alisma Tuber).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 9.5% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 10.0%, calculated on the dried basis.

Assay **(1)** (*E*)-Ferulic acid—Conduct this procedure without exposure to light, using light-resistant vessels. Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of (*E*)-ferulic acid for assay, and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 2 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of (*E*)-ferulic acid in each solution.

$$\text{Amount (mg) of (}E\text{)-ferulic acid} = M_S \times A_T/A_S \times 1/50$$

M_S: Amount (mg) of (*E*)-ferulic acid for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 320 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 7.8 g of sodium dihydrogen phosphate in 1000 mL of water, and add 2 mL of phosphoric acid. To 850 mL of this solution add 150 mL of acetonitrile.

Flow rate: 1.0 mL per minute [the retention time of (*E*)-ferulic acid is about 10 minutes].

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of (*E*)-ferulic acid are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of (*E*)-ferulic acid is not more than 1.5%.

(2) Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <2.48> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute (the retention time of paeoniflorin is about 9 minutes).

System suitability—

System performance: Dissolve 1 mg of albiflorin in 10 mL of the standard solution. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(3) Atractylenolide III—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of atractylenolide III for assay, previously dried in a desiccator (silica gel) for more than 24 hours, and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of atractylenolide III in each solution.

$$\text{Amount (mg) of atractylenolide III} = M_S \times A_T/A_S \times 1/40$$

M_S: Amount (mg) of atractylenolide III for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 210 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (550:450:1).

Flow rate: 1.0 mL per minute (the retention time of atractylenolide III is about 10 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of atractylenolide III are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of atractylenolide III is not more than 1.5%.

(4) Atractylodin—Conduct this procedure without exposure to light, using light-resistant vessels. Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with exactly 10 μL each of the sample solution and atractylodin TS for assay as directed under Liquid Chromatography <2.01> according to the following conditions, and determine the peak areas, A_T and A_S, of atractylodin in each solution.

$$\text{Amount (mg) of atractylodin} = C_S \times A_T/A_S \times 50$$

C_S: Concentration (mg/mL) of atractylodin in atractylodin TS for assay

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 330 mL of a mixture of water and phosphoric acid (55:1) add 670 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of atractylodin is about 13 minutes).

System suitability—

System performance: When the procedure is run with 10 μL of atractylodin TS for assay under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of atractylodin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of atractylodin TS for assay under the above operating conditions, the relative standard deviation of the peak area of atractylodin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Tragacanth

Tragacantha

トラガント

Tragacanth is the exudation obtained from the trunks of *Astragalus gummifer* Labillardiére or other species of the same genus (*Leguminosae*).

Description Tragacanth occurs as curved, flattened or lamellate fragments, 0.5 – 3 mm in thickness. It is white to light yellow in color, translucent, and horny in texture. It is easily broken, and swells in water.

Odorless; tasteless and mucilaginous.

Identification **(1)** To 1 g of pulverized Tragacanth add 50 mL of water: a nearly uniform, slightly turbid mucilage is formed.

(2) To pulverized Tragacanth add dilute iodine TS, and examine the mixture microscopically <*5.01*>: a few blue-colored starch grains are observable.

Purity Karaya gum—Boil 1 g of Tragacanth with 20 mL of water until a mucilage is formed, add 5 mL of hydrochloric acid, and again boil the mixture for 5 minutes: no light red to red color develops.

Total ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Well-closed containers.

Powdered Tragacanth

Tragacantha Pulverata

トラガント末

Powdered Tragacanth is the powder of Tragacanth.

Description Powdered Tragacanth occurs as a white to yellowish white powder. It is odorless, tasteless and mucilaginous.

Under a microscope <*5.01*>, it, immersed in olive oil or liquid paraffin, reveals numerous angular fragments with a small amount of the circular or irregular lamellae or of starch grains. Starch grains are spherical to elliptical, mostly simple and occasionally 2- to 4-compound grains, simple grain, 3 – 25 μm in diameter. The fragments are swollen and altered with water.

Identification **(1)** To 1 g of Powdered Tragacanth add 50 mL of water: a nearly uniform, slightly turbid mucilage is formed.

(2) To Powdered Tragacanth add dilute iodine TS, and examine the mixture microscopically <*5.01*>: a few blue-colored starch grains are observable.

Purity Karaya gum—Boil 1 g of Powdered Tragacanth with 20 mL of water until a mucilage is formed, add 5 mL of hydrochloric acid, and again boil the mixture for 5 minutes: no light red to red color develops.

Total ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Tight containers.

Tribulus Fruit

Tribuli Fructus

シツリシ

Tribulus Fruit is the fruit of *Tribulus terrestris* Linné (*Zygophyllaceae*).

Description Pentagonal star shaped fruit, composed of five mericarps, 7 – 12 mm in diameter, often each mericarp separated; externally grayish green to grayish brown; a pair of longer and shorter spines on surface of each mericarp, the longer spine 3 – 7 mm in length, the shorter one 2 – 5 mm in length, numerous small processes on ridge; pericarp hard in texture, cut surface light yellow; each mericarp contains 1 – 3 seeds.

Almost odorless; taste, mild at first, followed by bitterness.

Under a microscope <*5.01*>, a transverse section reveals epicarp composed of an epidermis; mesocarp composed of parenchyma and sclerenchyma layer; endocarp composed of several-cellular-layered fiber cells; a single-layer of cell between mesocarp and endocarp contain solitary crystals of calcium oxalate; cotyledons of seed contain oil drops and aleurone grains, and occasionally starch grains.

Identification To 2 g of pulverized Tribulus Fruit add 5 mL of methanol, shake for 10 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography, develop the plate with a mixture of ethyl acetate and water (40:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): a bluish white fluorescent spot appears at an *R*f value of about 0.4.

Purity **(1)** Peduncle—When perform the test of foreign matter <*5.01*>, the amount of peduncle contained in Tribulus Fruit does not exceed 4.0%.

(2) Foreign matters <*5.01*>—Not more than 1.0% of foreign matters other than peduncle.

Loss on drying <*5.01*> Not more than 11.0% (6 hours).

Total ash <*5.01*> Not more than 13.0%.

Acid-insoluble ash <*5.01*> Not more than 1.5%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 8.5%.

Containers and storage Containers—Well-closed containers.

Trichosanthes Root

Trichosanthis Radix

カロコン

Trichosanthes Root is the root of *Trichosanthes kirilowii* Maximowicz, *Trichosanthes kirilowii* Maximowicz var. *japonica* Kitamura or *Trichosanthes bracteata* Voigt (*Cucurbitaceae*), from which the cork layer is mostly removed.

Description Irregular cylindrical root, 5 – 10 cm in length, 3 – 5 cm in diameter, often cut lengthwise; externally light yellow-white, and with irregular pattern of vascular bundles appearing as brownish yellow lines; fractured surface somewhat fibrous and light yellow in color; under a magnifying glass, the transverse section reveals wide medullary rays and brownish yellow spots or small holes formed by vessels.

Almost odorless; taste, slightly bitter.

Identification To 2.0 g of pulverized Trichosanthes Root add 5 mL of methanol, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of hexane, ethyl acetate and acetic acid (100) (20:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 10 minutes, and examine under ultraviolet light (main wavelength: 365 nm): a light yellow to light yellow-green fluorescent spot appears at an *R*f value of about 0.4.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Trichosanthes Root according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Trichosanthes Root according to Method 4, and perform the test (not more than 5 ppm).

Total ash <*5.01*> Not more than 4.0%.

Containers and storage Containers—Well-closed containers.

Turmeric

Curcumae Longae Rhizoma

ウコン

Turmeric is the rhizome of *Curcuma longa* Linné (*Zingiberaceae*) with or without cork layers, usually with the application of blanching.

It contains not less than 1.0% and not more than 5.0% of total curcuminoids (curcumin, demethoxycurcumin and bisdemethoxycurcumin), calculated on the basis of dried material.

Description Turmeric is a main rhizome or a lateral rhizome; main rhizome, nearly ovoid, about 3 cm in diameter, about 4 cm in length; lateral rhizome, cylindrical, with round tips, curved, about 1 cm in diameter, 2 - 6 cm in length; both main and lateral rhizomes with cyclic nodes; rhizome with cork layer, yellow-brown, lustrous; rhizome without cork layer, dark yellow-red, with yellow-red powders on surface; hard in texture, not easily broken; transversely cut surface yellow-brown to red-brown, lustrous like wax.

Odor, characteristic; taste, slightly bitter and stimulant, it colors a saliva yellow on chewing.

Under a microscope <*5.01*>, a transverse section reveals the outermost layer to be composed of a cork layer 4 – 10 cells thick; sometimes a cork layer partly remains; cortex and stele, divided by an endodermis, composed of parenchyma, vascular bundles scattered; oil cells scattered in parenchyma; parenchymatous cells contain yellow substances, sandy and solitary crystals of calcium oxalate, and gelatinized starch.

Identification **(1)** To 0.5 g of pulverized Turmeric, add 20 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (11:9:1) to a distance about 7 cm, and air-dry the plate: a yellow spot appears at an *R*f value of about 0.4.

(2) To 0.2 g of pulverized Turmeric, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), centrifuge after shaking for 20 minutes. Perform the test with the supernatant liquid as directed in the Assay, and determine the peak areas of curcumin, demethoxycurcumin and bisdemethoxycurcumin: the peak area of curcumin is larger than the peak area of demethoxycurcumin and is larger than 0.69 times the peak area of bisdemethoxycurcumin.

Purity **(1)** Heavy metals <*1.07*>—Proceed with 3.0 g of pulverized Turmeric according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of pulverized Turmeric according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 17.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 9.0%.

Assay Weigh accurately about 0.2 g of pulverized Turmeric, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), shake for 20 minutes, centrifuge, and separate the supernatant liquid. To the residue, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), and proceed in the same manner as described above. Combine all the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of curcumin for assay, and dissolve in methanol to make exactly 50 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as described under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_{TC}, A_{TD} and A_{TD} of curcumin, demethoxycurcumin and bisdemethoxycurcumin in the sample solution as well as the peak area A_S of curcumin in the standard solution.

Amount (mg) of total curcuminoids (curcumin, demethoxycurcumin and bisdemethoxycurcumin)
$= M_S \times (A_{TC} + A_{TD} + A_{TB} \times 0.69)/A_S \times 1/5$

M_S: Amount (mg) of curcumin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 245 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilianized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (56:43:1).

Flow rate: 1.0 mL per minute (the retention time of curcumin is about 11 minutes).

System suitability—

System performance: Dissolve 1 mg each of curcumin for assay, demethoxycurcumin and bisdemethoxycurcumin in methanol to make 5 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, bisdemethoxycurcumin, demethoxycurcumin and curcumin are eluted in this order with the resolution among these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of curcumin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Powdered Turmeric

Curcumae Longae Rhizoma Pulveratum

ウコン末

Powdered Turmeric is the powder of Turmeric.

It contains not less than 1.0% and not more than 5.0% of total curcuminoids (curcumin, demethoxycurcumin and bisdemethoxycurcumin), calculated on the basis of dried material.

Description Powdered Turmeric occurs as a yellow-brown to dark yellow-brown powder. It has a characteristic odor and a bitter, stimulant taste, and colors the saliva yellow on chewing.

Under a microscope <*5.01*>, all elements are yellow in color; it reveals parenchymatous cells containing mainly masses of gelatinized starch or yellow substances, also fragments of scalariform vessels; fragments of cork layers, epidermis, thick-walled xylem parenchymatous cells, and non-glandular hairs are occasionally observed.

Identification (1) To 0.5 g of Powdered Turmeric, add 20 mL of methanol, shake for 15 minutes, filter, and use the filtrate as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (11:9:1) to a distance about 7 cm, and air-dry the plate: a yellow spot appears at an Rf value of about 0.4.

(2) To 0.2 g of Powdered Turmeric, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), centrifuge after shaking for 20 minutes. Perform the test with the supernatant liquid as directed in the Assay, and determine the peak areas of curcumin, demethoxycurcumin and bisdemethoxycurcumin: the peak area of curcumin is larger than the peak area of demethoxycurcumin and is larger than 0.69 times the peak area of bisdemethoxycurcumin.

Purity (1) Heavy metals <*1.07*>—Proceed with 3.0 g of Powdered Turmeric according to Method 3, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 10 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.40 g of Powdered Turmeric according to Method 4, and perform the test (not more than 5 ppm).

Loss on drying <*5.01*> Not more than 17.0% (6 hours).

Total ash <*5.01*> Not more than 7.5%.

Acid-insoluble ash <*5.01*> Not more than 1.0%.

Extract content <*5.01*> Dilute ethanol-soluble extract: not less than 9.0%.

Assay Weigh accurately about 0.2 g of Powdered Turmeric, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), shake for 20 minutes, centrifuge, and separate the supernatant liquid. To the residue, add 25 mL of a mixture of methanol and acetic acid (100) (99:1), and proceed in the same manner as described above. Combine all the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of curcumin for assay, and dissolve in methanol to make exactly 50 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as described under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_{TC}, A_{TD} and A_{TB} of curcumin, demethoxycurcumin and bisdemethoxycurcumin in the sample solution as well as the peak area A_S of curcumin in the standard solution.

Amount (mg) of total curcuminoids (curcumin, demethoxycurcumin and bisdemethoxycurcumin)
$= M_S \times (A_{TC} + A_{TD} + A_{TB} \times 0.69)/A_S \times 1/5$

M_S: Amount (mg) of curcumin for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 245 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilianized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of water, acetonitrile and acetic acid (100) (56:43:1).

Flow rate: 1.0 mL/per minute (the retention time of curcumin is about 11 minutes).

System suitability—

System performance: Dissolve 1 mg each of curcumin for assay, demethoxycurcumin and bisdemethoxycurcumin in methanol to make 5 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, bisdemethoxycurcumin, demethoxycurcumin and curcumin are eluted in this order with the resolution among these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of curcumin is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Turpentine Oil

Oleum Terebinthinae

テレビン油

Turpentine Oil is the essential oil distilled with steam from the wood or balsam of *Pinus* species (*Pinaceae*).

Description Turpentine Oil is a clear, colorless to pale yellow liquid. It has a characteristic odor and a pungent, bitter taste.

Turpentine Oil (1 mL) is miscible with 5 mL of ethanol (95) and this solution is neutral.

Refractive index ⟨*2.45*⟩ n_D^{20}: 1.465 – 1.478

Specific gravity ⟨*1.13*⟩ d_{20}^{20}: 0.860 – 0.875

Purity **(1)** Foreign matter—Turpentine Oil has no offensive odor. Shake 5 mL of Turpentine Oil with 5 mL of a solution of potassium hydroxide (1 in 6): the aqueous layer does not show a yellow-brown to dark brown color.

(2) Hydrochloric acid-coloring substances—Shake 5 mL of Turpentine Oil with 5 mL of hydrochloric acid, and allow to stand for 5 minutes: the hydrochloric acid layer is light yellow and not brown in color.

(3) Mineral oil—Place 5 mL of Turpentine Oil in a Cassia flask, cool to a temperature not exceeding 15°C, add dropwise 25 mL of fuming sulfuric acid while shaking, warm between 60°C and 65°C for 10 minutes, and add sulfuric acid to raise the lower level of the oily layer to the graduated portion of the neck: not more than 0.1 mL of oil separates.

Distilling range ⟨*2.57*⟩ 150 – 170°C, not less than 90 vol%.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Uncaria Hook

Uncariae Uncis Cum Ramulus

チョウトウコウ

Uncaria Hook is, hook or the hook-bearing stem of *Uncaria rhynchophylla* Miquel, *Uncaria sinensis* Haviland or *Uncaria macrophylla* Wallich (*Rubiaceae*), sometimes after being passed through hot water or steamed.

Uncaria Hook contains not less than 0.03% of total alkaloids (rhynchophylline and hirsutine), calculated on the basis of dried material.

Description Uncaria Hook is uncinate hook or short stem with opposite or single hook; the hook, 1 – 4 cm in length, curved and acuminate; externally red-brown to dark brown or grayish brown, some one with hairs, the transverse section oblong to elliptical, light brown; stem thin and prismatic square to cylindrical, 2 – 5 mm in diameter, externally, red-brown to dark brown or grayish brown; the transverse section, square to elliptical; the pith light brown, square to elliptical; hard in texture.

Odorless and practically tasteless.

Under a microscope ⟨*5.01*⟩, a transverse section of the hook reveals vascular bundles in the cortex, unevenly distributed and arranged in a ring. Parenchyma cells in the secondary cortex containing sand crystals of calcium oxalate.

Identification To 1.0 g of pulverized Uncaria Hook add 10 mL of water and 1 mL of ammonia TS, shake, then add 10 mL of diethyl ether, shake for 10 minutes, centrifuge, and use the diethyl ether layer as the sample solution. Separately, dissolve 1 mg each of rhyncophylline for thin-layer chromatography and hirsutine for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography ⟨*2.03*⟩. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (7:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly Dragendorff's TS for spraying on the plate, and air-dry the plate. Then spray evenly sodium nitrite TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with one of the spots from the standard solution.

Loss on drying ⟨*5.01*⟩ Not more than 12.0% (6 hours).

Total ash ⟨*5.01*⟩ Not more than 4.0%.

Extract content ⟨*5.01*⟩ Dilute ethanol-soluble extract: not less than 8.5%.

Assay Weigh accurately about 0.2 g of moderately fine powder of Uncaria Hook, transfer into a glass-stoppered centrifuge tube, add 30 mL of a mixture of methanol and dilute acetic acid (7:3), shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add two 10-mL portions of a mixture of methanol and dilute acetic acid (7:3), proceed in the same manner, and combine all of the supernatant liquid. To the combined liquid add a mixture of methanol and dilute acetic acid (7:3) to make exactly 50 mL, and use this as the sample solution. Separately, weigh accurately about 5 mg of rhynchophylline for assay, previously dried in a desiccator (silica gel) for 24 hours, and dissolve in a mixture of methanol and dilute acetic acid (7:3) to make exactly 100 mL. Pipet 1 mL of this solution, add a mixture of methanol and dilute acetic acid (7:3) to make exactly 10 mL, and use this solution as the standard solution (1). Separately, dissolve 1 mg of hirsutine in 100 mL of a mixture of methanol and dilute acetic acid (7:3), and use this solution as the standard solution (2). Perform the test with exactly 20 μL each of the sample solution and standard solutions (1) and (2) as directed under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions. Determine the peak areas, A_{Ta} and A_{Tb}, of rhynchophylline and hirsutine obtained from the sample solution, and the peak area, A_S, of rhynchophylline from the standard solution (1).

Amount (mg) of total alkaloids (rhynchophylline and hirsutine)
$= M_S \times (A_{Ta} + 1.405A_{Tb})/A_S \times 1/20$

M_S: Amount (mg) of rhynchophylline for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 245 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 200 mL of water, add 10 mL of acetic acid (100) and water to make 1000 mL, and add 350 mL of acetonitrile.

Flow rate: Adjust so that the retention time of rhynchophylline is about 17 minutes.

System suitability—

System performance: Dissolve 5 mg of rhynchophylline for assay in 100 mL of a mixture of methanol and dilute acetic acid (7:3). To 5 mL of this solution add 1 mL of ammonia solution (28), and reflux for 10 minutes or warm at about 50°C for 2 hours. After cooling, to 1 mL of the solution so obtained add a mixture of methanol and dilute acetic acid (7:3) to make 5 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, the peak of isorhynchophylline is appears in addition to the peak of rhynchophylline, and the resolution between these peaks is not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution (1) under the above operating conditions, the relative standard deviation of the peak areas of rhynchophylline is not more than 1.5%.

Containers and storage Containers—Well-closed containers.

Unseiin Extract

温清飲エキス

Unseiin Extract contains not less than 24 mg and not more than 72 mg (for preparation prescribed 3 g of Peony Root) or not less than 32 mg and not more than 96 mg (for preparation prescribed 4 g of Peony Root) of paeoniflorin ($C_{23}H_{28}O_{11}$: 480.46), not less than 39 mg and not more than 117 mg (for preparation prescribed 1.5 g of Scutellaria Root), or not less than 78 mg and not more than 234 mg (for preparation prescribed 3 g of Scutellaria Root) of baicalin ($C_{21}H_{18}O_{11}$: 446.36), and not less than 20 mg and not more than 60 mg of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)	3)
Japanese Angelica Root	4 g	4 g	3 g
Rehmannia Root	4 g	4 g	3 g
Peony Root	3 g	4 g	3 g
Cnidium Rhizome	3 g	4 g	3 g
Scutellaria Root	3 g	3 g	1.5 g
Gardenia Fruit	2 g	2 g	1.5 g
Coptis Rhizome	1.5 g	1.5 g	1.5 g
Phellodendron Bark	1.5 g	1.5 g	1.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) to 3), using the crude drugs shown above.

Description Unseiin Extract occurs as a yellow-brown to very dark brown powder or black-brown viscous extract. It has a slight odor, and has a slightly sweet taste at first, followed by a pungent taste.

Identification **(1)** To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 15 mL of water and 5 mL of 0.1 mol/L hydrochloric acid TS, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of butyl acetate and hexane (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root and Cnidium Rhizome).

(2) To 0.5 g of the dry extract (or 1.5 g of the viscous extract) add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of Paeoniflorin RS or paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS on the plate, and heat the plate at 105°C for 1 minute: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red-purple to purple spot from the standard solution (Peony Root).

(3) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of wogonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:10:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly iron (III) chloride-methanol TS on the plate: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-brown to grayish brown spot from the standard solution (Scutellaria Root).

(4) To 0.5 g of the dry extract (or 1.5 g of the viscous extract) add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of geniposide for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfric acid TS on the plate, and heat the plate at 105°C for 1 minute: one of the several spots ob-

tained from the sample solution has the same color tone and *R*f value with the purple to dark purple spot from the standard solution (Gardenia Fruit).

(5) To 0.5 g of the dry extract (or 1.5 g of the viscous extract) add 10 mL of methanol, shake, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of coptisine chloride for thin-layer chromatography in 5 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 2 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ammonia solution (28) and methanol (15:1:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Coptis Rhizome).

(6) To 1.0 g of dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), then add 2 mL of diethyl ether to the residue, and use this solution as the sample solution. Separately, dissolve 1 mg of limonin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 5 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, hexane and acetic acid (100) (10:5:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly vanillin-sulfuric acid-ethanol TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the purple to dark purple spot from the standard solution (Phellodendron Bark).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not more than 9.0%, calculated on the dried basis.

Assay **(1)** Paeoniflorin—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add exactly 50 mL of diluted methanol (1 in 2), shake for 15 minutes, and filter. Pipet 5 mL of the filtrate, flow through in a column packed with 2 g of polyamide for column chromatography, elute with 20 mL of water, then add 1 mL of acetic acid (100) to the eluate, add water to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Paeoniflorin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in diluted methanol (1 in 2) to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 20 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of paeoniflorin in each solution.

$$\text{Amount (mg) of paeoniflorin } (C_{23}H_{28}O_{11}) = M_S \times A_T/A_S \times 5/8$$

M_S: Amount (mg) of Paeoniflorin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 232 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20°C.

Mobile phase: A mixture of water, acetonitrile and phosphoric acid (850:150:1).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: Dissolve 1 mg each of Paeoniflorin RS and albiflorin in diluted methanol (1 in 2) to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, albiflorin and paeoniflorin are eluted in this order with the resolution between these peaks being not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of paeoniflorin is not more than 1.5%.

(2) Baicalin—Weigh accurately about 0.1 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.1 g of the dried substance), add exactly 50 mL of diluted methanol (7 in 10), shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Baicalin RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), and dissolve in methanol to make exactly 100 mL. Pipet 5 mL of this solution, add diluted methanol (7 in 10) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of baicalin in each solution.

$$\text{Amount (mg) of baicalin } (C_{21}H_{18}O_{11}) = M_S \times A_T/A_S \times 1/4$$

M_S: Amount (mg) of Baicalin RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 277 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 200) and acetonitrile (19:6).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of baicalin are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of baicalin is not more than 1.5%.

(3) Berberine—Weigh accurately about 0.2 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.2 g of the dried substance), add exactly 50 mL of the mobile phase, shake for 15 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 10 mg of Berberine Chloride RS (separately determine the water <*2.48*> in the same manner as Berberine Chloride Hydrate), dissolve in the mobile phase to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solution.

Amount (mg) of berberine chloride ($C_{20}H_{18}ClNO_4$) $= M_S \times A_T/A_S \times 1/2$

M_S: Amount (mg) of Berberine Chloride RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 345 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: Dissolve 3.4 g of potassium dihydrogen phosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

Flow rate: 1.0 mL per minute.

System suitability—

System performance: Dissolve 1 mg each of Berberine Chloride RS and palmatine chloride in the mobile phase to make 10 mL. When the procedure is run with 10 μL of this solution under the above operating conditions, palmatine and berberine are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of berberine is not more than 1.5%.

Containers and storage Containers—Tight containers.

Uva Ursi Fluidextract

ウワウルシ流エキス

Uva Ursi Fluidextract contains not less than 3.0 w/v% of arbutin.

Method of preparation Prepare an infusion from Bearberry Leaf, in coarse powder, as directed under Fluidextracts, using hot Purified Water or hot Purified Water in Containers. Remove a part of the accompanying tannin, evaporate the mixture under low pressure (in vacuo), if necessary, and add Purified Water or Purified Water in Containers to adjust the percentage. It may contain an appropriate quantity of Ethanol.

Description Uva Ursi Fluidextract is a yellow-brown to dark red-brown liquid, and has a bitter and astringent taste.

It is miscible with water and with ethanol (95).

Identification To 1 mL of Uva Ursi Fluidextract add 30 mL of a mixture of ethanol (95) and water (7:3), shake, filter, and use the filtrate as the sample solution. Proceed as directed in the Identification (2) under Bearberry Leaf.

Purity Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of Uva Ursi Fluidextract as direct under the Fluidextracts (4), and perform the test (not more than 30 ppm).

Assay Pipet 1 mL of Uva Ursi Fluidextract, add water to make exactly 100 mL, and use this solution as the sample solution. Proceed as directed in the Assay under Bearberry Leaf.

Amount (mg) of arbutin $= M_S \times A_T/A_S$

M_S: Amount (mg) of arbutin for assay taken

Containers and storage Containers—Tight containers.

Wood Creosote

Creosotum Ligni

木クレオソート

Wood Creosote is a mixture of phenols obtained from by using wood tar derived from dry distillation of stems and branches of various plants of genus *Pinus* (*Pinaceae*), genus *Cryptomeria* (*Taxodiaceae*), genus *Fagus* (*Fagaceae*), genus *Afzelia* (genus *Intsia*); (*Leguminosae*), genus *Shorea* (*Dipterocarpaceae*) or genus *Tectona* (*Verbenaceae*), followed by distillation and collection at 180 to 230°C, then further purification and then re-distillation.

Wood Creosote contains not less than 23% and not more than 35% of guaiacol ($C_7H_8O_2$: 124.14).

Description Wood Creosote is a colorless or pale yellow, clear liquid. It has a characteristic odor.

It is slightly soluble in water.

It is miscible with methanol and with ethanol (99.5).

Its saturated solution is acidic.

It is highly refractive.

It gradually changes in color by light or by air.

Identification Use the sample solution obtained in the Assay as the sample solution. Separately, dissolve 0.1 g of phenol, *p*-cresol, guaiacol, and 2-methoxy-4-methylphenol in methanol respectively, to make 100 mL. To 10 mL of each solution add methanol to make 50 mL, and use these solutions as standard solution (1), standard solution (2), standard solution (3) and standard solution (4). Perform the test with 10 μL each of the sample solution, standard solution (1), (2), (3) and (4) as directed under Liquid Chromatography <*2.01*> according to the following conditions: the main peaks obtained with the sample solution show the same retention times with those obtained with the standard solutions (1), (2), (3) and (4).

Operating conditions—

Proceed as directed in the operating conditions in the Assay.

Specific gravity ⟨*2.56*⟩ d_{20}^{20}: not less than 1.076.

Purity **(1)** Coal Creosote—Accurately measure 10 mL of Wood Creosote, add methanol to make exactly 20 mL, and use this solution as the sample solution. Separately, to 1 mg each of benzo[*a*]pyrene, benz[*a*]anthracene and dibenz[*a*,*h*]anthracene add a small quantity of ethyl acetate, if necessary, and add methanol to make 100 mL. To 1 mL of this solution add methanol to make 100 mL, and use this solution as the standard solution. Perform the test with exactly 1 μL each of the sample solution and standard solution as directed under Gas Chromatography ⟨*2.02*⟩ according to the following conditions: No peaks are detected with the sample solution for the retention times corresponding to benzo[*a*]pyrene, benz[*a*]anthracene and dibenz[*a*,*h*]anthracene of the standard solution. Change these conditions if any peak is detected for retention times that correspond to benzo[*a*]pyrene, benz[*a*]anthracene or dibenz[*a*,*h*]anthracene, to verify that such a peak does not belong to benzo[*a*]pyrene, benz[*a*]anthracene or dibenz[*a*,*h*]anthracene.

Operating conditions—

Detector: A mass spectrometer (EI).

Monitored ions:

Benz[*a*]anthracene: Molecular ion *m/z* 228, Fragment ion *m/z* 114	About 14 to 20 minutes
Benzo[*a*]pyrene: Molecular ion *m/z* 252, Fragment ion *m/z* 125	About 20 to 25 minutes
Dibenz[*a*,*h*]anthracene: Molecular ion *m/z* 278, Fragment ion *m/z* 139	About 25 to 30 minutes

Column: A quartz tube 0.25 mm in inside diameter and 30 m in length, with internal coating 0.25 – 0.5 μm in thickness made of 5% diphenyl-95% dimethyl polysiloxane for gas chromatography.

Column temperature: Inject sample at a constant temperature in vicinity of 45°C, then raise temperature to 240°C at the rate of 40°C per minute, maintain the temperature at 240°C for 5 minutes, then raise temperature to 300°C at the rate of 4°C per minute, then raise the temperature to 320°C at the rate of 10°C per minute, then maintain temperature at 320°C for 3 minutes.

Injection port temperature: A constant temperature in vicinity of 250°C.

Interface temperature: A constant temperature in vicinity of 300°C.

Carrier gas: Helium.

Flow rate: Adjust so that the retention time of benzo[*a*]pyrene is about 22 minutes.

Splitless.

System suitability—

Test for required detectability: Accurately measure 1 mL of standard solution and add methanol to make exactly 10 mL, and use this solution as the solution for system suitability test. When the test is performed with conditions described above with 1 μL of the solution for system suitability test, the SN ratio of each substance is not less than 3.

System performance: When the procedure is run with conditions described above with 1 μL of the solution for system suitability test, the elution takes place in order of benz[*a*]anthracene, benzo[*a*]pyrene and then dibenz[*a*,*h*]anthracene.

System repeatability: When the test is repeated 6 times with 1 μL of the solution for system suitability test under the above conditions, the relative standard deviation of the peak area of benzo[*a*]pyrene, benz[*a*]anthracene and dibenz[*a*,*h*]anthracene is respectively not more than 10%.

(2) Acenaphthene—To 0.12 g of Wood Creosote add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, dissolve 25 mg of acenaphthene in methanol to make 50 mL. To 5 mL of this solution add methanol to make 20 mL. To 2 mL of this solution add methanol to make 100 mL, and use this solution as the standard solution. Perform the test with exactly 1 μL each of the sample solution and standard solution as directed under Gas Chromatography ⟨*2.02*⟩ according to the following conditions: No peaks are detected with sample solution for the retention time corresponding to acenaphthene of the standard solution. Change these conditions if any peak is detected for the retention time corresponding to acenaphthene, to verify that such a peak does not belong to athenaphthene.

Operating conditions—

Detector: A hydrogen flame-ionization detector.

Column: A fused silica tube 0.25 mm inside diameter and 60 m in length, with internal coating 0.25 – 0.5 μm in thickness made of polymethylsiloxane for gas chromatography.

Column temperature: Perform injection at a constant temperature in vicinity of 45°C, then raise the temperature by 11.5°C per minute until reaching 160°C, then raise the temperature by 4°C per minute until reaching 180°C, then raise the temperature by 8°C until reaching 270°C, then maintain temperature at 270°C for 3 minutes.

Injection port temperature: 250°C.

Detector temperature: 250°C.

Carrier gas: Helium.

Flow rate: Adjust so that the retention time of acenaphthene is about 18 minutes.

Splitless.

System suitability—

Test for required detectability: Accurately measure 1 mL of the standard solution, add methanol to make exactly 10 mL, and use this solution as the solution for system suitability test. When the procedure is run with conditions described above with 1 μL of solution for system suitability test, the SN ratio of acenaphthene is not less than 3.

System repeatability: When the test is repeated 6 times with 1 μL of the solution for system suitability test under the above operating conditions, the relative standard deviation of the peak area of acenaphthene is not more than 6.0%.

(3) Other impurities

Add 2 mL of petroleum benzin to 1.0 mL of Wood Creosote, then add 2 mL of barium hydroxide test solution, agitate to mix and allow to stand. No blue or muddy brown color develops in the upper layer of the mixture. Furthermore, no red color develops in the lower layer.

Distilling range ⟨*2.57*⟩ 200 – 220°C, not less than 85 vol%.

Assay To about 0.1 g of Wood Creosote, accurately weighed, add methanol to make exactly 50 mL. Pipet 10 mL of this solution add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, add methanol to about 30 mg of accurately measured guaiacol for assay to make exactly 50 mL. Pipet 10 mL of this solution, add methanol to make exactly 50 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and standard solution under Liquid Chromatography ⟨*2.01*⟩ according to the following conditions, and determine the peak areas, A_T and A_S, of guaiacol in each solution.

$$\text{Amount (mg) of guaiacol} = M_S \times A_T/A_S$$

M_S: Amount (mg) of guaiacol for assay taken

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 275 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Mixture of water and acetonitrile (4:1).

Flow rate: Adjust so that the retention time of guaiacol is about 9 minutes.

System suitability—

System performance: Dissolve 2 mg each of guaiacol and phenol in methanol to make 10 mL. The procedure is run with conditions described above with 10 μL of this solution, the elution takes place in order of phenol then guaiacol, with the degree in separation of not less than 2.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of guaiacol is not more than 1.5%.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Yokukansan Extract

抑肝散エキス

Yokukansan Extract contains not less than 0.15 mg of total alkaloids (rhyncophylline and hirsutine), not less than 0.6 mg and not more than 2.4 mg of saikosaponin b_2, and not less than 10 mg and not more than 30 mg of glycyrrhizic acid ($C_{42}H_{62}O_{16}$: 822.93), per extract prepared with the amount specified in the Method of preparation.

Method of preparation

	1)	2)
Japanese Angelica Root	3 g	3 g
Uncaria Hook	3 g	3 g
Cnidium Rhizome	3 g	3 g
Atractylodes Rhizome	4 g	—
Atractylodes Lancea Rhizome	—	4 g
Poria Sclerotium	4 g	4 g
Bupleurum Root	2 g	2 g
Glycyrrhiza	1.5 g	1.5 g

Prepare a dry extract or viscous extract as directed under Extracts, according to the prescription 1) or 2), using the crude drugs shown above.

Description Yokukansan Extract is a light brown to grayish brown powder or a black-brown viscous extract. It has a slightly odor, and a slightly bitter and acid taste.

Identification **(1)** To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of diethyl ether, shake, and centrifuge. Separate the diethyl ether layer, add 10 mL of sodium hydroxide TS, shake, centrifuge, separate the diethyl ether layer, and use this layer as the sample solution. Separately, use (*Z*)-ligustilide TS for thin-layer chromatography as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of butyl acetate and hexane (2:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the blue-white fluorescent spot from the standard solution (Japanese Angelica Root; Cnidium Rhizome).

(2) To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 20 mL of water and 2 mL of ammonia TS, shake, then add 20 mL of diethyl ether, shake, and separate the diethyl ether layer. Evaporate the solvent under low pressure (in vacuo), add 1 mL of methanol to the residue, and use the solution as the sample solution. Separately, dissolve 1 mg each of rhyncophyllin for thin-layer chromatography and hirsutine for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, 1-propanol, water and acetic acid (100) (7:5:4:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): at least one of the several spots obtained from the sample solution has the same color tone and *R*f value with one of the two dark violet spots from the standard solution (Uncaria Hook).

(3) For preparation prescribed Atractylodes Rhizome—To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of diethyl ether, and shake. Separate the diethyl ether layer, evaporate the solvent under low pressure (in vacuo), dissolve the residue in 2 mL of diethyl ether, and use this solution as the sample solution. Separately, dissolve 1 mg of atractylenoide III for thin-layer chromatography in 2 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 5 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and hexane (1:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 1-naphthol-sulfuric acid TS on the plate, heat the plate at 105°C for 5 minutes, and allow to cool: one of the several spots obtained from the sample solution has the same color tone and *R*f value with the red to purple-red spot from the standard solution (Atractylodes Rhizome).

(4) For preparation prescribed Atractylodes Lancea Rhizome—To 2.0 g of the dry extract (or 6.0 g of the viscous extract) add 10 mL of water, shake, then add 25 mL of hexane, and shake. Separate the hexane layer, evaporate the solvent under low pressure (in vacuo), add 2 mL of hexane to the residue, and use this solution as the sample solution. Perform the test with the sample solution as directed under Thin-layer Chromatography <*2.03*>. Spot 20 μL of the sample solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of hexane and acetone (7:1) to a distance of about 7 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): a dark purple spot is observed at an *R*f value of about 0.5. The spot shows a greenish brown color after being sprayed evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heated at 105°C for 5 minutes, and allowed to cool (Atractylodes Lancea Rhizome).

(5) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of saikosaponin b_2 for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 10 μL of the sample solution and 2 μL of the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, ethanol (99.5) and water (8:2:1) to a distance of about 7 cm, and air-dry the plate. Spray evenly 4-dimethylaminobenzaldehyde TS for spraying on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow fluorescent spot from the standard solution (Bupleurum Root).

(6) To 1.0 g of the dry extract (or 3.0 g of the viscous extract) add 10 mL of water, shake, then add 10 mL of 1-butanol, shake, centrifuge, and use the 1-butanol layer as the sample solution. Separately, dissolve 1 mg of liquiritin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under Thin-layer Chromatography <*2.03*>. Spot 1 μL each of the sample solution and standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate, methanol and water (20:3:2) to a distance of about 7 cm, and air-dry the plate. Spray evenly dilute sulfuric acid on the plate, heat the plate at 105°C for 5 minutes, and examine under ultraviolet light (main wavelength: 365 nm): one of the several spots obtained from the sample solution has the same color tone and *R*f value with the yellow-green fluorescent spot from the standard solution (Glycyrrhiza).

Purity **(1)** Heavy metals <*1.07*>—Prepare the test solution with 1.0 g of the dry extract (or an amount of the viscous extract, equivalent to 1.0 g of the dried substance) as directed under Extracts (4), and perform the test (not more than 30 ppm).

(2) Arsenic <*1.11*>—Prepare the test solution with 0.67 g of the dry extract (or an amount of the viscous extract, equivalent to 0.67 g of the dried substance) according to Method 3, and perform the test (not more than 3 ppm).

Loss on drying <*2.41*> The dry extract: Not more than 10.0% (1 g, 105°C, 5 hours).

The viscous extract: Not more than 66.7% (1 g, 105°C, 5 hours).

Total ash <*5.01*> Not less than 10.0%, calculated on the dried basis.

Assay **(1)** Total alkaloids (rhyncophylline and hirsutine)—Weigh accurately about 1 g of the dry extract (or an amount of the viscous extract, equivalent to about 1 g of the dried substance), add 20 mL of diethyl ether, shake, then add 3 mL of 1 mol/L hydrochloric acid TS and 7 mL of water, and shake for 10 minutes, centrifuge, and remove the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, and proceed in the same manner as above. To the aqueous layer add 10 mL of sodium hydroxide TS and 20 mL of diethyl ether, shake for 10 minutes, centrifuge, and separate the diethyl ether layer. To the aqueous layer add 20 mL of diethyl ether, proceed in the same manner, and repeat this procedure twice. Combine all the extracts, evaporate the solvent under low pressure (in vacuo) at not more than 40°C, dissolve the residue in the mobile phase to make exactly 10 mL, and use this solution as the sample solution. Separately, weigh accurately about 5 mg each of rhyncophylline for assay and hirsutine for assay, dissolve in a mixture of methanol and diluted acetic acid (7:3) to make exactly 100 mL. Pipet 10 mL of this solution, add the mixture of methanol and diluted acetic acid (7:3) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_{TR} and A_{TH}, and A_{SR} and A_{SH}, of rhyncophylline and hirsutine in each solution.

Amount (mg) of total alkaloids (rhyncophylline and hirsutine)
$$= (M_{SR} \times A_{TR}/A_{SR} + M_{SH} \times A_{TH}/A_{SH}) \times 1/50$$

M_{SR}: Amount (mg) of rhyncophylline for assay taken
M_{SH}: Amount (mg) of hirsutine for assay taken

Operation conditions—

Detector: An ultraviolet absorption photometer (wavelength: 245 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: To 1 g of sodium lauryl sulfate add 600 mL of methanol, shake, then add 400 mL of water and 1 mL of phosphoric acid.

Flow rate: 1.0 mL per minute (the retention times of rhyncophylline and hirsutine are about 17 minutes and about 47 minutes, respectively).

Systemic suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peaks of rhyncophylline and hirsutine are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of rhyncophylline and hirsutine is not more than 1.5%, respectively.

(2) Saikosaponin b_2—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and separate the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, separate the supernatant liquid, combine all the supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, use saikosaponin b_2 standard TS for assay as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of saikosaponin b_2 in each solution.

Amount (mg) of saikosaponin $b_2 = C_S \times A_T/A_S \times 50$

C_S: Concentration (mg/mL) of saikosaponin b_2 in saikosaponin b_2 standard TS for assay

Operation conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of 0.05 mol/L sodium dihydrogen phosphate TS and acetonitrile (5:3).

Flow rate: 1.0 mL per minute (the retention time of saikosaponin b_2 is about 12 minutes).

Systemic suitability—

System performance: When the procedure is run with 10 μL of the standard solution under the above operating conditions, the number of theoretical plates and the symmetry factor of the peak of saikosaponin b_2 are not less than 5000 and not more than 1.5, respectively.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of saikosaponin b_2 is not more than 1.5%.

(3) Glycyrrhizic acid—Weigh accurately about 0.5 g of the dry extract (or an amount of the viscous extract, equivalent to about 0.5 g of the dried substance), add 20 mL of diethyl ether and 10 mL of water, and shake for 10 minutes. After centrifugation, remove the diethyl ether layer, add 20 mL of diethyl ether, proceed in the same manner as described above, and remove the diethyl ether layer. To the aqueous layer add 10 mL of methanol, shake for 30 minutes, centrifuge, and take the supernatant liquid. To the residue add 20 mL of diluted methanol (1 in 2), shake for 5 minutes, centrifuge, and take the supernatant liquid. Combine these supernatant liquids, add diluted methanol (1 in 2) to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 10 mg of Glycyrrhizic Acid RS (separately determine the water <*2.48*> by coulometric titration, using 10 mg), dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 10 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine the peak areas, A_T and A_S, of glycyrrhizic acid in each solution.

$$\text{Amount (mg) of glycyrrhizic acid } (C_{42}H_{62}O_{16}) = M_S \times A_T/A_S \times 1/2$$

M_S: Amount (mg) of Glycyrrhizic Acid RS taken, calculated on the anhydrous basis

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 254 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: Dissolve 3.85 g of ammonium acetate in 720 mL of water, and add 5 mL of acetic acid (100) and 280 mL of acetonitrile.

Flow rate: 1.0 mL per minute (the retention time of glycyrrhizic acid is about 15 minutes).

System suitability—

System performance: Dissolve 5 mg of monoammonium glycyrrhizinate for resolution check in 20 mL of dilute ethanol. When the procedure is run with 10 μL of this solution under the above operating conditions, the resolution between the peak having the relative retention time of about 0.9 to glycyrrhizic acid and the peak of glycyrrhizic acid is not less than 1.5.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of glycyrrhizic acid is not more than 1.5%.

Containers and storage Containers—Tight containers.