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 blackish 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 the Thin-layer Chromatography. Spot 5 μL each of the sample solution and 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 glacial acetic acid (10:10:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): one of the spots from the sample solution and a blue-violet spot from the standard solution show the same color tone and *Rf* value.

Purity Foreign matter—The amount of receptacle, peduncle and other foreign matter contained in Schisandra Fruit does not exceed 1.0%.

Total ash Not more than 5.0%.

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, 5 - 10 cm in length, 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 2 g of pulverized Schizonepeta Spike add 20 mL of water, shake well, and distill. To 3 mL of the distillate add 2 or 3 drops of 2,4-dinitrophenylhydrazine-ethanol TS: an orange-red precipitate is formed.

Total ash Not more than 11.0%.

Acid-insoluble ash Not more than 3.0%.

Extract content Dilute ethanol-soluble extract: not less than 8.0%.

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, or Purified Water, 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 filtrate to dryness under reduced pressure, 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 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 water layer, using 25 mL each of diethyl ether. Combine the extracts, and evaporate the diethyl ether 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 ($C_{17}H_{23}NO_3$)
= amount (mg) of Atropine Sulfate Reference Standard, calculated on the dried basis

$$\times \frac{Q_{TA}}{Q_{SA}} \times 0.855$$

Amount (mg) of scopolamine ($C_{17}H_{21}NO_4$)
= amount (mg) of Scopolamine Hydrobromide Reference Standard, calculated on the dried basis
$\times \frac{Q_{TS}}{Q_{SS}} \times \frac{1}{25} \times 0.789$

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 or their mixture	a sufficient quantity
To make	1000 g

To Scopolia Extract add 100 mL of Purified Water, then warm and soften the mixture with stirring. Cool, add 800 g of starch, Lactose 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 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, 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 filtrate to dryness under reduced pressure, 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 4.0 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. Repeat this procedure three times with the water layer, using 25-mL portions of diethyl ether. Combine the extracts, and evaporate the diethyl ether 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. Filter this solution through a membrane 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 0.025 g of Atropine Sulfate Reference Standard (separately determine its loss on drying), dissolve in the mobile phase to make exactly 25 mL, and use this solution as standard stock solution A. Weigh accurately about 0.025 g of Scopolamine Hydrobromide Reference Standard (separately determine its loss on drying), 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 exactly 25 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions. Determine 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 ($C_{17}H_{23}NO_3$)
= amount (mg) of Atropine Sulfate Reference Standard, calculated on the dried basis
$\times \frac{Q_{TA}}{Q_{SA}} \times \frac{1}{5} \times 0.855$

Amount (mg) of scopolamine ($C_{17}H_{21}NO_4$)
= amount (mg) of Scopolamine Hydrobromide Reference Standard, calculated on the dried basis
$\times \frac{Q_{TS}}{Q_{SS}} \times \frac{1}{25} \times 0.789$

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 octadesilcylanized 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 the flow rate so that the retention time of scopolamine is about 8 minutes.

Selection of column: Proceed with 10 μL of the standard solution under the above operating conditions, and determine the resolution. Use a column giving elution of scopolamine, atropine and the internal standard in this order with the resolution between the peaks of scopolamine and atropine being not less than 11, and the resolution between the