

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 0.020 g of atropine sulfate for thin-layer chromatography and 0.010 g of scopolamine hydrobromide in 10 mL each of ethanol (95), and use these solutions as standard solution (1) and standard solution (2). Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 10  $\mu$ L each of the sample solution and the standard solutions on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of chloroform, methanol, acetone and ammonia solution (28) (73:15:10:2) 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 from the sample solution show the same in color tone and *R<sub>f</sub>* value with each yellow-red 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 diethyl ether 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 0.075 g of Ethyl Aminobenzoate Reference Standard, 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 the 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*-(1-naphthyl)-*N'*-diethylethylenediamine oxalate-acetone TS, mix immediately, and add water to make exactly 50 mL. Allow to stand for 2 hours, determine the absorbances, *A<sub>T</sub>* and *A<sub>S</sub>*, of these solutions at 550 nm, as directed under the Ultraviolet-visible Spectrophotometry using a blank prepared in the same manner with 5 mL of water in place of the sample solution.

$$\begin{aligned} & \text{Amount (mg) of ethyl aminobenzoate (C}_9\text{H}_{11}\text{NO}_2\text{)} \\ &= \text{amount (mg) of Ethyl Aminobenzoate Reference} \\ & \quad \text{Standard} \\ & \quad \times \frac{A_T}{A_S} \end{aligned}$$

**Containers and storage** Containers—Well-closed containers.

## Scopolia Extract, Papaverine and Ethyl Aminobenzoate Powder

ロートエキス・パパベリン・アネスタミン散

Scopolia Extract, Papaverine and Ethyl Aminobenzoate Powder contains not less than 10.8% and not more than 13.2% of ethyl aminobenzoate (C<sub>9</sub>H<sub>11</sub>NO<sub>2</sub>: 165.19).

### Method of preparation

Scopolia Extract	15 g
Papaverine Hydrochloride	15 g
Ethyl Aminobenzoate	120 g
Starch, Lactose or their mixture	a sufficient quantity
To make 1000 g	

Prepare as directed under Powders, with the above ingredients. May be prepared with Scopolia Extract Powder in place of Scopolia Extract.

**Description** Scopolia Extract, Papaverine and Ethyl Aminobenzoate Powder is brownish yellow to grayish yellow-brown in color. It has a slightly bitter taste, leaving a sensation of numbness on the tongue.

**Identification (1)** To 4 g of Scopolia Extract, Papaverine 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 the Qualitative Tests 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 20 mL of chloroform, shake well, filter, and further wash the residue with 10 mL of chloroform. Combine the filtrate and the washing, transfer this solution to a separator, and add 10 mL of 0.1 mol/L hydrochloric acid TS. After shaking, separate the chloroform layer, add 2 g of anhydrous sodium sulfate, shake, and filter through a pledget of cotton. Evaporate the filtrate to dryness, dry the residue at 105°C for 3 hours, and perform the following tests (papaverine hydrochloride):

(i) To 1 mg of the residue add 1 drop of formaldehyde solution-sulfuric acid TS: a colorless or light yellow-green color, changing to red-purple, is produced.

(ii) Dissolve 1 mg of the residue in 3 mL of acetic anhydride and 5 drops of sulfuric acid, heat in a water bath for 1 minute, and view under ultraviolet light: the solution shows a yellow-green fluorescence.

(3) Place 20 g of Scopolia Extract, Papaverine and Ethyl Aminobenzoate Powder in a glass-stoppered conical flask, add 80 mL of water, shake for 15 minutes, and filter by suc-

tion through a glass filter (G3). Transfer 60 mL of the filtrate to a separator, add 0.5 mL of 1 mol/L hydrochloric acid TS, and extract with three 20-mL portions of chloroform by shaking. Make the aqueous layer 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, and 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 the solution as the sample solution. Dissolve 0.020 g of atropine sulfate for thin-layer chromatography, 0.010 g of scopolamine hydrobromide and 0.020 g of papaverine hydrochloride in 10 mL each of ethanol (95), and use these solutions as standard solutions (1), (2) and (3). Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 10  $\mu$ L each of the sample solution and the standard solutions on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of chloroform, methanol, acetone and ammonia solution (28) (73:15:10:2) to a distance of about 10 cm, and dry the plate at 80°C for 20 minutes. After cooling, spray Dragendorff's TS for spraying upon the plate evenly: three yellow-red principal spots obtained from the sample solution and the corresponding spots from standard solutions (1), (2) and (3) show the same *R<sub>f</sub>* values.

**Assay** Weigh accurately about 0.6 g of Scopolia Extract, Papaverine and Ethyl Aminobenzoate Powder, transfer to a Soxhlet extractor, and extract with 100 mL of diethyl ether for 1 hour, and evaporate the diethyl ether 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. Separately, weigh accurately about 0.075 g of Ethyl Aminobenzoate Reference Standard, 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 the standard solution, add 10 mL of 1 mol/L hydrochloric acid TS to each, 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*-(1-naphthyl)-*N'*-diethylethylenediamine oxalate-acetone TS, mix immediately, and add water to make exactly 50 mL. Allow to stand for 2 hours, and determine the absorbances,  $A_T$  and  $A_S$ , of these solutions at 550 nm as directed under the Ultraviolet-visible Spectrophotometry using a blank prepared in the same manner with 5 mL of water in place of the sample solution.

$$\begin{aligned} & \text{Amount (mg) of ethyl aminobenzoate (C}_9\text{H}_{11}\text{NO}_2\text{)} \\ &= \text{amount (mg) of Ethyl Aminobenzoate Reference} \\ & \quad \text{Standard} \\ & \quad \times \frac{A_T}{A_S} \end{aligned}$$

**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 water 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 bluish-black color develops. Allow to stand: a bluish-black precipitate is formed (tannic acid).

**Containers and storage** Containers—Well-closed containers.

## Compound Scopolia Extract and Tannic Acid Ointment

複方ロートエキス・タンニン軟膏

### Method of preparation

Scopolia Extract	100 g
Tannic Acid	30 g
<i>d</i> - or <i>dl</i> -Camphor	10 g
Ichthammol	100 g
Simple Ointment or a suitable ointment base	a sufficient quantity

To make 1000 g

Prepare as directed under Ointments, with the above ingredients.

**Description** Compound Scopolia Extract and Tannic Acid Ointment is brownish in color.

**Identification (1)** Shake 3 g of Compound Scopolia Extract and Tannic Acid Ointment with 20 mL of diethyl ether for 10 minutes to dissolve the base of suppositories. Shake thoroughly the mixture with 15 mL of water, separate the water 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,