

$A_T$  and  $A_S$ , of berberine in each solution.

$$\begin{aligned} & \text{Amount (mg) of } C_{20}H_{19}NO_5 \\ &= \text{amount (mg) of Berberine Chloride Reference} \\ & \quad \text{Standard, calculated on the dehydrated basis} \\ & \quad \times \frac{A_T}{A_S} \times \frac{353.37}{371.82} \end{aligned}$$

**Operating conditions—**

**Detector:** An ultraviolet absorption photometer (wavelength: 345 nm).

**Column:** A stainless steel column about 4 mm in inside diameter and about 25 cm in length, packed with octadecylsilylated silica gel for liquid chromatography (5  $\mu$ m in particle diameter).

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

**Mobile phase:** Dissolve 3.4 g of monobasic potassium phosphate and 1.7 g of sodium lauryl sulfate in 1000 mL of a mixture of water and acetonitrile (1:1).

**Flow rate:** Adjust the flow rate so that the retention time of berberine is about 10 minutes.

**Selection of column:** Dissolve each 1 mg of berberine chloride and palmatin chloride in the mobile phase to make 10 mL. Proceed with 10  $\mu$ L of this solution under the above operating conditions, and calculate the resolution. Use a column giving elution of palmatin and berberine in this order with the resolution between these peaks being not less than 1.5.

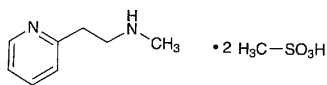
**System repeatability:** When the test is repeated five times with the standard solution under the above operating conditions, the relative standard deviation of the peak areas of berberine is not more than 1.5%.

**Containers and storage** Containers—Tight containers.

Storage—Light-resistant.

## Betahistine Mesilate

メシル酸ベタヒスチン



$C_8H_{12}N_2 \cdot 2CH_4O_3S$ : 328.41

*N*-Methyl-*N*-[2-(pyridin-2-yl)ethyl]amine  
dimethanesulfonate [5638-76-6, Betahistine]

Betahistine Mesilate, when dried, contains not less than 98.0% of  $C_8H_{12}N_2 \cdot 2CH_4O_3S$ .

**Description** Betahistine Mesilate occurs as white crystals or crystalline powder. It is odorless, or has a faint, characteristic odor and a bitter taste.

It is very soluble in water, freely soluble in methanol and in acetic acid (100), sparingly soluble in ethanol (99.5), very slightly soluble in acetic anhydride, and practically insoluble in diethyl ether.

It is hygroscopic.

**Identification (1)** To 5 mL of a solution of Betahistine Mesilate (1 in 10) add 15 mL of sodium hydroxide TS and

20 mL of chloroform, and shake. Separate the chloroform layer, wash with 10 mL of water, and use the chloroform layer as the sample solution. Take 5 mL of the sample solution, evaporate the solvent under reduced pressure by warming, dissolve the residue in 1 mL of water, add 1 mL of acetaldehyde and 0.5 mL of sodium pentacyanonitrosylferrate (III) TS, and shake: a blue to blue-purple color develops.

(2) Determine the absorption spectrum of a solution of Betahistine Mesilate in 0.1 mol/L methanesulfonic acid TS (1 in 20,000) as directed under the Ultraviolet-visible Spectrophotometry, and compare the spectrum with the Reference Spectrum: both spectra exhibit similar intensities of absorption at the same wavelengths.

(3) To 0.03 g of Betahistine Mesilate add 0.1 g of sodium nitrate and 0.1 g of anhydrous sodium carbonate, mix well, and heat gradually. After cooling, dissolve the residue in 2 mL of dilute hydrochloric acid and 10 mL of water, filter if necessary, and to the filtrate add 1 mL of barium chloride TS: a white precipitate is formed.

**Melting point** 110–114°C (after drying).

**Purity (1)** Clarity and color of solution—Dissolve 1.0 g of Betahistine Mesilate in 10 mL of water: the solution is clear and colorless.

(2) Chloride—Perform the test with 1.0 g of Betahistine Mesilate. Prepare the control solution with 0.40 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.014%).

(3) Heavy metals—Proceed with 1.0 g of Betahistine Mesilate according to Method 4, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

(4) Arsenic—Prepare the test solution with 1.0 g of Betahistine Mesilate according to Method 3, and perform the test using Apparatus B (not more than 2 ppm).

(5) Related substances—Dissolve 0.20 g of Betahistine Mesilate in 10 mL of methanol, and use this solution as the sample solution. Pipet 1 mL of the sample solution, add methanol to make exactly 200 mL, and use this solution as the standard solution. 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 solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of chloroform, methanol and ammonia solution (28) (40:10:1) to a distance of about 10 cm, and air-dry the plate. Allow to stand for 5 minutes in iodine vapor: the spots other than the principal spot from the sample solution are not more intense than the spot from the standard solution.

**Loss on drying** Not more than 1.0% (1 g, in vacuum, phosphorus (V) oxide, 70°C, 24 hours).

**Residue on ignition** Not more than 0.10% (1 g).

**Assay** Weigh accurately about 0.2 g of Betahistine Mesilate, previously dried, dissolve in 1 mL of acetic acid (100), add 50 mL of acetic anhydride, and titrate with 0.1 mol/L perchloric acid VS (potentiometric titration). Perform a blank determination, and make any necessary correction.

$$\begin{aligned} & \text{Each mL of 0.1 mol/L perchloric acid VS} \\ &= 16.421 \text{ mg of } C_8H_{12}N_2 \cdot 2CH_4O_3S \end{aligned}$$

**Containers and storage** Containers—Tight containers.