

and the internal standard in this order with the resolution between these peaks being not less than 4.

Containers and storage Containers—Tight containers.

Codeine Phosphate Tablets

リン酸コデイン錠

Codeine Phosphate Tablets contains not less than 93% and not more than 107% of the labeled amount of codeine phosphate ($C_{18}H_{21}NO_3 \cdot H_3PO_4 \cdot \frac{1}{2}H_2O$; 406.37).

Method of preparation Prepare as directed under Tablets, with Codeine Phosphate.

Identification To a quantity of powdered Codeine Phosphate Tablets, equivalent to 0.1 g of Codeine Phosphate according to the labeled amount, add 20 mL of water, shake, and filter. To 2 mL of the filtrate add water to make 100 mL, and determine the absorption spectrum as directed under the Ultraviolet-visible Spectrophotometry: it exhibits a maximum between 283 nm and 287 nm.

Assay Weigh accurately and powder not less than 20 Codeine Phosphate Tablets. Weigh accurately a portion of the powder, equivalent to about 0.1 g of codeine phosphate ($C_{18}H_{21}NO_3 \cdot H_3PO_4 \cdot \frac{1}{2}H_2O$), add 30 mL of water, shake, add 20 mL of diluted dilute sulfuric acid (1 in 20), treat the mixture with ultrasonic waves for 10 minutes, and add water to make exactly 100 mL. Filter this solution, then pipet 5 mL of the filtrate, add exactly 10 mL of the internal standard solution and water to make 20 mL, and use this solution as the standard solution. Separately, weigh accurately about 0.05 g of codeine phosphate for assay, separately determined its water content in the same manner as Codeine Phosphate, dissolve in water to make exactly 100 mL, then 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 20 μ L each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and calculate the ratios, Q_T and Q_S , of the peak area of codeine to that of the internal standard.

$$\begin{aligned} & \text{Amount (mg) of codeine phosphate} \\ & (C_{18}H_{21}NO_3 \cdot H_3PO_4 \cdot \frac{1}{2}H_2O) \\ & = \text{amount (mg) of codeine phosphate for assay,} \\ & \text{calculated on the anhydrous basis} \\ & \times 1.0227 \times \frac{Q_T}{Q_S} \times 2 \end{aligned}$$

Internal standard solution—A solution of etilefrine hydrochloride (3 in 10,000).

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 280 nm).

Column: A stainless steel column about 4 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (about 5 μ m in particle diameter).

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

Mobile phase: Dissolve 1.0 g of sodium lauryl sulfate in 500 mL of diluted phosphoric acid (1 in 1000), and adjust the pH to 3.0 with sodium hydroxide TS. To 240 mL of this solution add 70 mL of tetrahydrofuran, and mix.

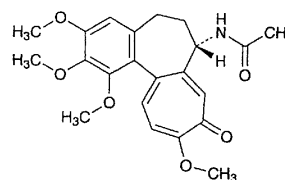
Flow rate: Adjust the flow rate so that the retention time of codeine is about 10 minutes.

Selection of column: Proceed with 20 μ L of the standard solution under the above operating conditions, and calculate the resolution. Use a column giving elution of codeine and the internal standard in this order with the resolution between these peaks being not less than 4.

Containers and storage Containers—Tight containers.

Colchicine

コルヒチン



$C_{22}H_{25}NO_6$; 399.44

N-[(7*S*)-(5,6,7,9)-Tetrahydro-1,2,3,10-tetramethoxy-9-oxobenzo[*a*]heptalen-7-yl)]acetamide [64-86-8]

Colchicine, when dried, contains not less than 96.0% of $C_{22}H_{25}NO_6$.

Description Colchicine occurs as a yellowish white powder. It is odorless.

It is freely soluble in acetic anhydride and in ethanol (95), sparingly soluble in water, and very slightly soluble in diethyl ether.

It is colored by light.

Identification (1) To 1 mL of a solution of Colchicine in ethanol (95) (1 in 20) add 1 drop of iron (III) chloride TS: a dark reddish orange color is produced.

(2) Mix 1 mg of Colchicine with 2 drops of sulfuric acid in a porcelain dish: a yellow color is produced. On the addition of 1 drop of nitric acid: the color of the solution changes from blue-green through purple to yellow. Add 5 mL of sodium hydroxide TS: the color of the solution changes to reddish.

(3) Dissolve 0.01 g of Colchicine in 0.5 mL of dilute hydrochloric acid and 10 mL of water, and boil under a reflux condenser for 1 hour. Add 40 mL of warm water, shake, and filter while warm. Cool, and extract the filtrate with 20 mL of chloroform. Filter the chloroform extract, and evaporate on a water bath to dryness. Dissolve the residue in 0.5 mL of 1,4-dioxane, and add 10 mL of diethyl ether. Collect the crystals on a filter, wash with small portions of diethyl ether, and dry at 105°C for 1 hour: the crystals so obtained melt between 176°C and 179°C.

Optical rotation $[\alpha]_D^{20}$: -230 to -245° (after drying, 0.1 g, ethanol (95), 10 mL, 100 mm).

Purity (1) Chloroform—To 0.010 g of Colchicine add 2 mL of sodium hydroxide TS and 1 drop of aniline, and heat the mixture while shaking: no odor of phenyl isocyanide (toxic) is perceptible.

(2) Colchicine—Dissolve 0.10 g of Colchicine in 10 mL of water, and to 5 mL of this solution add 2 drops of iron (III) chloride TS: no definite green color is produced.

(3) Other alkaloids—Dissolve 0.10 g of Colchicine in 20 mL of water, and to 2.0 mL of this solution add 0.5 mL of 2,4,6-trinitrophenol TS: the solution is clear.

Loss on drying Not more than 5.0% (1 g, 105°C, 3 hours).

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

Assay Weigh accurately about 0.4 g of Colchicine, previously dried, dissolve in 25 mL of acetic anhydride, and titrate with 0.05 mol/L perchloric acid VS (potentiometric titration). Perform a blank determination, and make any necessary correction.

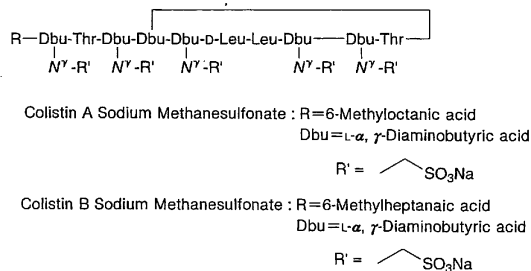
Each mL of 0.05 mol/L perchloric acid VS
= 19.972 mg of $C_{22}H_{25}NO_6$

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Colistin Sodium Methanesulfonate

コリスチンメタンスルホン酸ナトリウム



[8068-28-8, Colistin Sodium Methanesulfonate]

Colistin Sodium Methanesulfonate, when dried, contains not less than 10,000 Units per mg. The potency of Colistin Sodium Methanesulfonate is expressed as mass of colistin A ($C_{53}H_{100}N_{16}O_{13}$: 1168.46).

Description Colistin Sodium Methanesulfonate occurs as a white to light yellowish white powder.

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

Identification (1) Dissolve 0.02 g of Colistin Sodium Methanesulfonate in 2 mL of water, add 0.5 mL of sodium hydroxide TS, and add 5 drops of copper (II) sulfate TS while shaking: a blue-purple color develops.

(2) Dissolve 0.04 mg of Colistin Sodium Methanesulfonate in 1 mL of 1 mol/L hydrochloric acid TS, and add 0.5 mL of dilute iodine TS: the color of iodine disappears.

(3) Determine the infrared absorption spectrum of Colistin Sodium Methanesulfonate, previously dried, as directed in the potassium bromide disk method under the In-

frared Spectrophotometry, and compare the spectrum with the Reference Spectrum or the spectrum of dried Colistin Sodium Methanesulfonate Reference Standard: both spectra exhibit similar intensities of absorption at the same wave numbers.

(4) Colistin Sodium Methanesulfonate responds to the Qualitative Test (1) for sodium salt.

pH Dissolve 0.1 g of Colistin Sodium Methanesulfonate in 10 mL of water, and allow to stand for 30 minutes: the pH of the solution is between 6.5 and 8.5.

Purity (1) Clarity and color of solution—Dissolve 0.16 g of Colistin Sodium Methanesulfonate in 10 mL of water: the solution is clear and colorless.

(2) Heavy metals—Proceed with 1.0 g of Colistin Sodium Methanesulfonate according to Method 4, and perform the test. Prepare the control solution with 3.0 mL of Standard Lead Solution (not more than 30 ppm).

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

(4) Free colistin—Dissolve 0.08 g of Colistin Sodium Methanesulfonate in 3 mL of water, add 0.05 mL of a solution of silicotungstic acid 26-water (1 in 10), and compare the solution with the reference suspension described under the Test Methods for Plastic Containers: the turbidity is not greater than that of the reference suspension (not more than 0.25%).

Loss on drying Not more than 3.0% (0.1 g, reduced pressure, 60°C, 3 hours).

Assay Perform the test according to the Cylinder-plate method as directed under the Microbial Assay for Antibiotics according to the following conditions.

(1) Test organism—*Escherichia coli* NIHJ

(2) Culture medium—To 10.0 g of peptone, 30.0 g of sodium chloride, 3.0 g of meat extract and 20.0 g of agar add 1000 mL of water, then add a suitable amount of sodium hydroxide TS so that the pH of the medium is being 6.5 to 6.6 after sterilization, sterile, and use this as the seeded agar medium and the agar medium for base layer.

(3) Standard solution—Weigh accurately an amount of Colistin Sodium Methanesulfonate Reference Standard, previously dried, dissolve in phosphate buffer solution, pH 6.0 to make a solution containing 100,000 Units per mL, and use this solution as the standard stock solution. Keep the standard stock solution at 10°C or below and use within 7 days. Take exactly a suitable amount of the standard stock solution before use, and add phosphate buffer solution, pH 6.0 to make solutions so that each mL contains 10,000 Units and 2500 Units, and use these solutions as the high concentration standard solution and the low concentration standard solution, respectively.

(4) Sample solution—Weigh accurately an amount of Colistin Sodium Methanesulfonate, previously dried, dissolve in phosphate buffer solution, pH 6.0 to make a solution containing about 100,000 Units per mL, and use this solution as the sample stock solution. Take exactly a suitable amount of the sample stock solution, add phosphate buffer solution, pH 6.0 to make solutions so that each mL contains 10,000 Units and 2500 Units, and use these solutions as the high concentration sample solution and the low concentra-