

(100:60:23:17) to a distance of about 10 cm, and air-dry the plate. Spray evenly a solution of sulfuric acid in ethanol (99.5) (1 in 10) on the plate, and heat at 120°C for 30 minutes. Examine under ultraviolet light (main wavelength: 254 nm): 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 0.5% (1 g, 105°C, 4 hours).

Residue on ignition Not more than 0.05% (1 g).

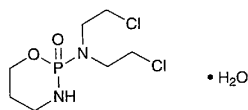
Assay Weigh accurately about 0.5 g of Cyclophosphamide Hydrochloride, previously dried, dissolve in 50 mL of a mixture of acetic anhydride and acetic acid (100) (4:1), and titrate with 0.1 mol/L perchloric acid VS until the color of the solution changes from purple through blue-green to yellow-green (indicator: 2 drops of crystal violet TS). Perform a blank determination, and make any necessary correction.

Each mL of 0.1 mol/L perchloric acid VS
= 32.785 mg of $C_{17}H_{25}NO_3 \cdot HCl$

Containers and storage Containers—Tight containers.

Cyclophosphamide

シクロホスファミド



$C_7H_{15}Cl_2N_2O_2P \cdot H_2O$: 279.10
N,N-Bis(2-chloroethyl)tetrahydro-2*H*-1,3,2-oxazaphosphorin-2-amine 2-oxide monohydrate
[6055-19-2]

Cyclophosphamide contains not less than 97.0% of $C_7H_{15}Cl_2N_2O_2P \cdot H_2O$.

Description Cyclophosphamide occurs as white crystals or crystalline powder. It is odorless.

It is very soluble in acetic acid (100), freely soluble in ethanol (95), in acetic anhydride and in chloroform, and soluble in water and in diethyl ether.

Melting point: 45 – 53°C

Identification (1) Dissolve 0.1 g of Cyclophosphamide in 10 mL of water, and add 5 mL of silver nitrate TS: no precipitate is produced. Then boil this solution: a white precipitate is produced. Collect the precipitate, and add dilute nitric acid to a portion of this precipitate: it does not dissolve. Add excess ammonia TS to another portion of the precipitate: it dissolves.

(2) Add 1 mL of diluted sulfuric acid (1 in 25) to 0.02 g of Cyclophosphamide, and heat until white fumes are evolved. After cooling, add 5 mL of water, and shake. Neutralize with ammonia TS, then acidify with dilute nitric acid: this solution responds to the Qualitative Tests (2) for phosphate.

Purity (1) Clarity and color of solution—Dissolve 0.20 g of Cyclophosphamide in 10 mL of water: the solution is

clear and colorless.

(2) Chloride—Perform the test with 0.40 g of Cyclophosphamide at a temperature not exceeding 20°C. Prepare the control solution with 0.40 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.036%).

(3) Heavy metals—Proceed with 1.0 g of Cyclophosphamide according to Method 1, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

Water 5.5 – 7.0% (0.5 g, direct titration).

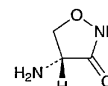
Assay Weigh accurately about 0.3 g of Cyclophosphamide, add 15 mL of hydrogen chloride-ethanol TS, and heat in a water bath under a reflux condenser for 3.5 hours while protecting from moisture. Distil the ethanol under reduced pressure. Dissolve the residue in 40 mL of a mixture of acetic anhydride and acetic acid (100) (7:3), and titrate with 0.1 mol/L perchloric acid-dioxane VS (indicator: 2 drops of crystal violet TS) until the color of the solution changes from blue through green to yellow. Perform a blank determination, and make any necessary correction.

Each mL of 0.1 mol/L perchloric acid-dioxane VS
= 13.955 mg of $C_7H_{15}Cl_2N_2O_2P \cdot H_2O$

Containers and storage Containers—Tight containers.
Storage—Not exceeding 30°C.

Cycloserine

サイクロセリン



$C_3H_6N_2O_2$: 102.09
(4*R*)-4-Aminoisoxazolidin-3-one [68-41-7]

Cycloserine contains not less than 900 μg (potency) per mg, calculated on the dried basis. The potency of Cycloserine is expressed as mass (potency) of cycloserine ($C_3H_6N_2O_2$).

Description Cycloserine occurs as white to light yellowish white, crystals or crystalline powder.

It is soluble in water, and sparingly soluble in ethanol (95).

Identification Determine the infrared absorption spectrum of Cycloserine, previously dried, as directed in the potassium bromide disk method under the Infrared Spectrophotometry, and compare the spectrum with the Reference Spectrum or the spectrum of previously dried Cycloserine Reference Standard: both spectra exhibit similar intensities of absorption at the same wave numbers.

Optical rotation $[\alpha]_D^{20}$: +108 – +114° (2.5 g calculated on the dried basis, 2 mol/L sodium hydroxide TS, 50 mL, 100 mm).

pH Dissolve 1.0 g of Cycloserine in 20 mL of water: the pH of the solution is between 5.0 and 7.4.

Purity (1) Heavy metals—Proceed with 1.0 g of Cycloserine

ine 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).

(2) Condensation products—Dissolve 0.02 g of Cycloserine in sodium hydroxide TS to make exactly 50 mL, and determine the absorbance of this solution at 285 nm as directed under the Ultraviolet-visible Spectrophotometry: not more than 0.8.

Loss on drying Not more than 1.5% (0.5 g, reduced pressure, 60°C, 3 hours).

Residue on ignition Not more than 0.5% (1 g).

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—*Bacillus subtilis* ATCC 6633

(2) Culture medium—Use the medium i in 1) Medium for test organism [5] under (1) Agar media for seed and base layer. Adjust the pH of the medium so that it will be 6.0 to 6.1 after sterilization.

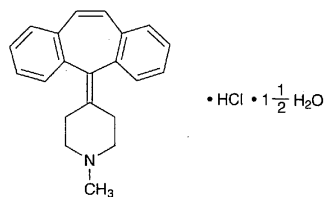
(3) Standard solution—Weigh accurately an amount of Cycloserine Reference Standard, previously dried at 60°C for 3 hours under reduced pressure of not exceeding 0.67 kPa, equivalent to about 0.040 g (potency), dissolve in water to make exactly 100 mL, and use this solution as the standard stock solution. Keep the standard stock solution at 5°C or below and use within 24 hours. 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 100 µg (potency) and 50 µg (potency), 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 Cycloserine equivalent to about 0.040 g (potency), dissolve in water to make exactly 100 mL. Take exactly a suitable amount of this solution, add phosphate buffer solution, pH 6.0 to make solutions so that each mL contains 100 µg (potency) and 50 µg (potency), and use these solutions as the high concentration sample solution and the low concentration sample solution, respectively.

Containers and storage Containers—Tight containers.

Cyproheptadine Hydrochloride

塩酸シプロヘプタジン



C₂₁H₂₁N.HCl.1½H₂O: 350.88

4-(5*H*-Dibenzo[*a, d*]cyclohepten-5-ylidene)-1-methylpiperidine monohydrochloride sesquihydrate
[41354-29-4]

Cyproheptadine Hydrochloride, when dried, contains not less than 98.5% of C₂₁H₂₁N.HCl: 323.86

Description Cyproheptadine Hydrochloride occurs as a white to pale yellow, crystalline powder. It is odorless, and has a slightly bitter taste.

It is freely soluble in methanol and in acetic acid (100), soluble in chloroform, sparingly soluble in ethanol (95), slightly soluble in water, and practically insoluble in diethyl ether.

Identification (1) Dissolve 0.1 g of Cyproheptadine Hydrochloride in 10 mL of methanol, apply 1 drop of this solution on filter paper, air-dry, and examine under ultraviolet light (main wavelength: 254 nm): the solution shows a pale blue fluorescence.

(2) Weigh 0.1 g of Cyproheptadine Hydrochloride, transfer to a separator, dissolve in 5 mL of chloroform, add 4 mL of water and 1 mL of sodium carbonate TS, and shake. Transfer the chloroform layer to another separator, and wash with 4 mL of water by shaking well. Filter the chloroform layer through absorbent cotton moistened previously with chloroform, and evaporate the filtrate to dryness. Dissolve the residue in 8 mL of dilute ethanol by warming at 65°C. Rub the inner wall of the container with a glass rod while cooling until crystallization begins, and allow to stand for 30 minutes. Collect the crystals, and dry at 80°C for 2 hours: the crystals melt between 111°C and 115°C.

(3) Determine the absorption spectrum of a solution of Cyproheptadine Hydrochloride in ethanol (95) (1 in 100,000) 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.

(4) A saturated solution of Cyproheptadine Hydrochloride responds to the Qualitative Tests (2) for chloride.

Purity (1) Acid—Dissolve 2.0 g of Cyproheptadine Hydrochloride in 25 mL of methanol, and add 1 drop of methyl red TS and 0.30 mL of 0.1 mol/L sodium hydroxide VS: a yellow color develops.

(2) Heavy metals—Proceed with 1.0 g of Cyproheptadine Hydrochloride according to Method 2, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 20 ppm).

Loss on drying 7.0–9.0% (1 g, in vacuum at a pressure not exceeding 0.67 kPa, 100°C, 5 hours).

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

Assay Weigh accurately about 0.5 g of Cyproheptadine Hydrochloride, previously dried, and dissolve in 20 mL of acetic acid (100) by warming at 50°C. After cooling, add 40 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.

Each mL of 0.1 mol/L perchloric acid VS
= 32.387 mg of C₂₁H₂₁N.HCl

Containers and storage Containers—Well-closed containers.