

(3) To 0.1 g of Dantrolene Sodium add 20 mL of water and 2 drops of acetic acid (100), shake well, and filter: the filtrate responds to the Qualitative Tests (1) for sodium salt.

Purity (1) Alkali—To 0.7 g of Dantrolene Sodium add 10 mL of water, shake well, and centrifuge or filter through a membrane filter. To 5 mL of the supernatant or the filtrate add 45 mL of water, 3 drops of phenolphthalein TS and 0.10 mL of 0.1 mol/L hydrochloric acid VS: a red color is not produced.

(2) **Heavy metals**—Proceed with 1.0 g of Dantrolene Sodium 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).

(3) **Related Substances**—Dissolve 0.050 g of Dantrolene Sodium in 20 mL of tetrahydrofuran and 2 mL of acetic acid (100), add ethanol (99.5) to make 100 mL, and use this solution as the sample solution. Pipet 1 mL of the sample solution, add ethanol (99.5) to make exactly 50 mL, and use this solution as the standard solution. Perform the test with 10 μ L of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions. Determine each peak area from these solutions by the automatic integration method: the total area of all peaks other than the peak of dantrolene from the sample solution is not larger than the peak area of dantrolene from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 300 nm).

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

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of hexane, acetic acid (100) and ethanol (99.5) (90:10:9).

Flow rate: Adjust the flow rate so that the retention time of dantrolene is about 8 minutes.

Selection of column: Dissolve 5 mg of Dantrolene Sodium and 0.1 g of theophylline in 20 mL of tetrahydrofuran and 2 mL of acetic acid (100), and add ethanol (99.5) to make 100 mL. To 10 mL of this solution add ethanol (99.5) to make 100 mL. Proceed with 10 μ L of this solution under the above operating conditions, and calculate the resolution. Use a column giving elution of theophylline and dantrolene in this order with the resolution between these peaks being not less than 6.

Detection sensitivity: Adjust the detection sensitivity so that the peak height of dantrolene from 10 μ L of the standard solution is 10% to 40% of the full scale.

Time span of measurement: About twice as long as the retention time of dantrolene, after the solvent peak.

Water 14.5 - 17.0% (0.2 g, direct titration).

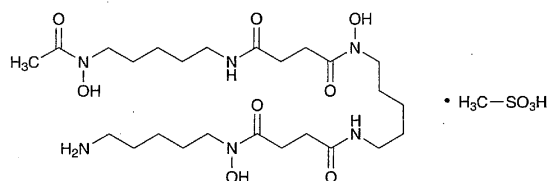
Assay Weigh accurately about 0.7 g of Dantrolene Sodium, dissolve in 180 mL of a mixture of propylene glycol and acetone (1:1), and titrate with 0.1 mol/L perchloric acid VS (potentiometric titration). Perform a blank determination, and make any necessary correction.

Each mL of 0.1 mol/L perchloric acid VS
= 33.624 mg of C₁₄H₉N₄NaO₅

Containers and storage Containers—Tight containers.

Deferoxamine Mesilate

メシル酸デフェロキサミン



C₂₅H₄₈N₆O₈·CH₄O₃S: 656.79

N-[5-(Acetylhydroxyamino)pentyl]-*N'*-(5-{3-[(5-aminopentyl)hydroxycarbonyl]propanoylamino}pentyl)-*N'*-hydroxysuccinamide monomethanesulfonate [138-14-7]

Deferoxamine Mesilate contains not less than 98.0% and not more than 102.0% of C₂₅H₄₈N₆O₈·CH₄O₃S, calculated on the anhydrous basis.

Description Deferoxamine Mesilate occurs as a white to pale yellowish white, crystalline powder.

It is freely soluble in water, and practically insoluble in ethanol (99.5), in 2-propanol and in diethyl ether.

Melting point: about 147°C (with decomposition).

Identification (1) To 5 mL of a solution of Deferoxamine Mesilate (1 in 500) add 1 drop of iron (III) chloride TS: a deep red color develops.

(2) To 0.05 g of Deferoxamine Mesilate add 0.2 g of sodium hydroxide, melt by heating over a small flame, and heat further for 2 to 3 seconds. To the residue add 0.5 mL of water, acidify with dilute hydrochloric acid, and warm: the gas evolved changes moistened potassium iodate-starch paper to blue.

(3) Determine the infrared absorption spectrum of Deferoxamine Mesilate as directed in the potassium bromide disk method under the Infrared Spectrophotometry, and compare the spectrum with the Reference Spectrum or the spectrum of Deferoxamine Mesilate Reference Standard: both spectra exhibit similar intensities of absorption at the same wave numbers.

pH Dissolve 1.0 g of Deferoxamine Mesilate in 10 mL of water: the pH of this solution is between 3.5 and 5.5.

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

(2) **Chloride**—Perform the test with 1.0 g of Deferoxamine Mesilate. Prepare the control solution with 0.90 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.032%).

(3) **Sulfate**—Perform the test with 0.6 g of Deferoxamine Mesilate. Prepare the control solution with 0.50 mL of 0.005 mol/L sulfuric acid VS (not more than 0.040%).

(4) **Heavy metals**—Proceed with 2.0 g of Deferoxamine Mesilate according to Method 4, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 10 ppm).

(5) **Arsenic**—Prepare the test solution with 1.0 g of Deferoxamine Mesilate according to Method 3, and perform the test using Apparatus B. Use a solution of magnesi-

um nitrate hexahydrate in ethanol (95) (1 in 10) (not more than 2 ppm).

(6) Related substances—Dissolve 0.050 g of Deferoxamine Mesilate in 50 mL of the mobile phase, and use this solution as the sample solution. Pipet 3 mL of the sample solution, add the mobile phase to make exactly 50 mL, 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. Determine each peak area of both solutions by the automatic integration method: the total area of all peaks other than the peak of deferoxamine from the sample solution is not larger than the peak area of deferoxamine from the standard solution.

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 230 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 20 cm in length, packed with octadecylsilylanized silica gel for liquid chromatography (10 μ m in particle diameter).

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

Mobile phase: Dissolve 1.32 g of diammonium hydrogenphosphate, 0.37 g of disodium dihydrogen ethylenediamine tetraacetate dihydrate and 1.08 g of sodium 1-heptanesulfonate in 950 mL of water. Adjust this solution with phosphoric acid to a pH of 2.8, and to 800 mL of this solution add 100 mL of 2-propanol.

Flow rate: Adjust the flow rate so that the retention time of deferoxamine is about 15 minutes.

Selection of column: Dissolve 0.016 g of Deferoxamine Mesilate and 4 mg of methyl parahydroxybenzoate in 50 mL of the mobile phase. Proceed with 20 μ L of this solution under the above operating conditions, and calculate the resolution. Use a column giving elution of deferoxamine and methyl parahydroxybenzoate in this order with the resolution between these peaks being not less than 4.

Detection sensitivity: Adjust the detection sensitivity so that the peak height of deferoxamine from 20 μ L of the standard solution is between 5 mm and 20 mm.

Time span of measurement: About twice as long as the retention time of deferoxamine after the solvent peak.

Water Not more than 2.0% (0.2 g, direct titration).

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

Assay Weigh accurately about 0.06 g of Deferoxamine Mesilate and Deferoxamine Mesilate Reference Standard (determine the water content before using in the same manner as Deferoxamine Mesilate), dissolve each in 20 mL of water, add 10 mL of 0.05 mol/L sulfuric acid TS, and add water to make exactly 50 mL. Pipet 5 mL each of these solutions, add exactly 5 mL of 0.05 mol/L sulfuric acid TS and exactly 0.2 mL of iron (III) chloride TS, then add water to make exactly 50 mL, and use these solutions as the sample solution and the standard solution, respectively. Perform the test with these solutions as directed under the Ultraviolet-visible Spectrophotometry, using a solution prepared by adding 0.05 mol/L sulfuric acid TS to 0.2 mL of iron (III) chloride TS to make exactly 50 mL as the blank, and determine the absorbances, A_T and A_S , of each solution from the sample solution and the standard solution at 430 nm.

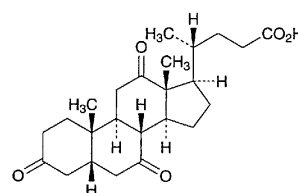
Amount (mg) of $C_{25}H_{48}N_6O_8 \cdot CH_4O_3S$
= amount (mg) of Deferoxamine Mesilate Reference Standard, calculated on the anhydrous basis

$$\times \frac{A_T}{A_S}$$

Containers and storage Containers—Tight containers.

Dehydrocholic Acid

デヒドロコール酸



$C_{24}H_{34}O_5$: 402.52
3,7,12-Trioxo-5 β -cholan-24-oic acid [81-23-2]

Dehydrocholic Acid, when dried, contains not less than 98.5% of $C_{24}H_{34}O_5$.

Description Dehydrocholic Acid occurs as a white, crystalline powder. It is odorless, and has a bitter taste.

It is sparingly soluble in 1,4-dioxane, slightly soluble in ethanol (95), and practically insoluble in water and in diethyl ether.

It dissolves in sodium hydroxide TS.

Identification (1) Dissolve 5 mg of Dehydrocholic Acid in 1 mL of sulfuric acid and 1 drop of formaldehyde solution, and allow to stand for 5 minutes. Add 5 mL of water to the solution: the solution shows a yellow color and a blue-green fluorescence.

(2) To 0.02 g of Dehydrocholic Acid add 1 mL of ethanol (95), shake, add 5 drops of 1,3-dinitrobenzene TS and 0.5 mL of a solution of sodium hydroxide (1 in 8), and allow to stand: a purple to red-purple color develops, and gradually changes to brown.

Optical rotation $[\alpha]_D^{20}$: +29 – +32° (after drying, 0.2 g, 1,4-dioxane, 10 mL, 100 mm).

Melting point 233 – 242°C

Purity (1) Odor—To 2.0 g of Dehydrocholic Acid add 100 mL of water, and boil for 2 minutes: the solution is odorless.

(2) Clarity and color of solution—To 0.10 g of Dehydrocholic Acid, previously powdered in a mortar, add 30 mL of ethanol (95), and dissolve by shaking for 10 minutes: the solution is clear and colorless.

(3) Chloride—To 2.0 g of Dehydrocholic Acid add 100 mL of water, shake for 5 minutes and filter, and use this filtrate as the sample solution. To 25 mL of the sample solution add 6 mL of dilute nitric acid, heat in a water bath for 6 minutes, filter after cooling, and collect the clear filtrate. Wash the residue with 10 mL of water, combine the washings and the filtrate, dilute with water to 50 mL, and perform the test using this solution as the test solution. Prepare