directed under the Thin-layer Chromatography. Spot 5 μL each of the sample solution and the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with *n*-butyl acetate to a distance of about 15 cm, and air-dry the plate. Spray evenly a solution of sulfuric acid in methanol (1 in 10) on the plate, and heat the plate at 105°C for 10 minutes: 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, 2 hours).

**Residue on ignition** Not more than 0.1% (1 g).

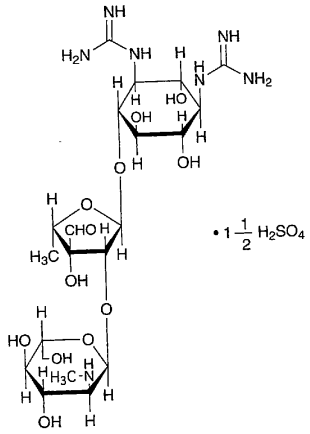
**Assay** Weigh accurately about 0.05 g each of Spironolactone and Spironolactone Reference Standard, previously dried at 105°C for 2 hours, dissolve in methanol to make exactly 250 mL. Pipet 5 mL each of these solutions, add methanol to make exactly 100 mL, and use these solutions as the sample solution and the standard solution, respectively. Perform the test with these solutions as directed under the Ultraviolet-visible Spectrophotometry, and determine the absorbances, $A_T$ and $A_S$, of the sample solution and the standard solution at 238 nm.

Amount (mg) of $C_{24}H_{32}O_4S$
= amount (mg) of Spironolactone Reference Standard
$\times \frac{A_T}{A_S}$

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

# Streptomycin Sulfate

硫酸ストレプトマイシン



$C_{21}O_{39}N_7O_{12}.1\frac{1}{2}H_2SO_4$: 728.69
*O*-2-Deoxy-2-methylamino-α-L-glucopyranosyl-(1→2)-*O*-5-deoxy-3-*C*-formyl-α-L-lyxofuranosyl-(1→4)-*N*,*N*′-diamidino-D-streptamine sesquisulfate [*3810-74-0*]

Streptomycin Sulfate conforms to the requirements of Streptomycin Sulfate in the Requirements for Antibiotic Products of Japan.

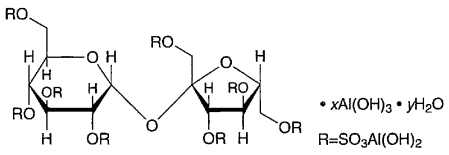
**Description** Streptomycin Sulfate occurs as a white to light yellowish white powder.

It is freely soluble in water, very slightly soluble in ethanol (95), and practically insoluble in diethyl ether.

# Sucralfate

## Aluminum Sucrose Sulfate Ester

スクラルファート



$C_{12}H_{30}Al_8O_{51}S_8.xAl(OH)_3.yH_2O$
[*54182-58-0*]

Sucralfate contains not less than 17.0% and not more than 21.0% of aluminum (Al: 26.98) and not less than 34.0% and not more than 43.0% of sucrose octasulfate ester ($C_{12}H_{22}O_{35}S_8$: 982.80), calculated on the dried basis.

**Description** Sucralfate occurs as a white powder. It is odorless and tasteless.

It is practically insoluble in water, in hot water, in ethanol (95) and in diethyl ether.

It dissolves in dilute hydrochloric acid and in sulfuric acid-sodium hydroxide TS.

**Identification** **(1)** To 0.05 g of Sucralfate in a small test tube add 0.05 g of fresh pieces of sodium, and melt by careful heating. Immerse the test tube immediately in 100 mL of water, break the test tube, shake well, and filter. To 5 mL of the filtrate add 1 drop of sodium pentacyanonitrosylferrate (III) TS: a red-purple color develops.

**(2)** Dissolve 0.040 g of Sucralfate in 2 mL of dilute sulfuric acid, and add gently 2 mL of anthrone TS to make 2 layers: a blue color develops at the zone of contact, and gradually changes to blue-green.

**(3)** Dissolve 0.5 g of Sucralfate in 10 mL of dilute hydrochloric acid: the solution responds to the Qualitative Tests for aluminum.

**Purity** **(1)** Clarity and color of solution—Dissolve 1.0 g of Sucralfate in 10 mL of dilute sulfuric acid: the solution is clear and colorless.

**(2)** Chloride—Dissolve 0.5 g of Sucralfate in 30 mL of dilute nitric acid, and heat gently to boiling. After cooling, add water to make 100 mL, and to 10 mL of this solution add 3 mL of dilute nitric acid and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution with 0.70 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.50%).

**(3)** Heavy metals—Dissolve 1.0 g of Sucralfate in 20 mL of a solution of sodium chloride (1 in 5) and 1 mL of dilute hydrochloric acid, and to this solution add 2 mL of dilute acetic acid and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution as follows: evaporate 1 mL of dilute hydrochloric acid on a water bath to dryness, and add 20 mL of a solution of

sodium chloride (1 in 5), 2 mL of dilute acetic acid, 2.0 mL of Standard Lead Solution and water to make 50 mL (not more than 20 ppm).

**(4)** Arsenic—Dissolve 1.0 g of Sucralfate in 5 mL of dilute hydrochloric acid, use this solution as the test solution, and perform the test using Apparatus B (not more than 2 ppm).

**(5)** Free aluminum—To 3.0 g of Sucralfate add 50 mL of water, heat in a water bath for 5 minutes, cool, and filter. Wash the residue with four 5-mL portions of water, combine the filtrate with the washings, add 2 mL of dilute hydrochloric acid, and heat in a water bath for 30 minutes. After cooling, neutralize the solution with sodium hydroxide TS, add water to make exactly 100 mL, and use this solution as the sample solution. Pipet 50 mL of the sample solution, add exactly 25 mL of 0.05 mol/L disodium dihydrogen ethylenediamine tetraacetate VS and 20 mL of acetic acid-ammonium acetate buffer solution, pH 4.5, and boil for 5 minutes. After cooling, add 50 mL of ethanol (95), and titrate the excess disodium dihydrogen ethylenediamine tetraacetate with 0.05 mol/L zinc acetate VS until the color of the solution changes from green-purple through purple to red (indicator: 3 mL of dithizone TS). Perform a blank determination (not more than 0.2%).

Each mL of 0.05 mol/L disodium dihydrogen ethylenediamine tetraacetate VS
= 1.3491 mg of Al

**(6)** Related substances—Proceed with 50 $\mu$L of the sample solution obtained in the Assay (2) Sucrose octasulfate ester as directed in the Assay (2) Sucrose octasulfate ester, and perform the test as directed under the Liquid Chromatography. Determine the peak area of sucrose octasulfate ester from the sample solution and that of a related substance with the relative retention time about 0.7 to the peak of sucrose octasulfate ester by the automatic integration method, and calculate the ratio of the peak area of the related substance to that of sucrose octasulfate ester: it is not more than 0.1.

Detection sensitivity: Adjust the detection sensitivity so that the peak height of sucrose octasulfate ester from 50 $\mu$L of the standard solution obtained in the Assay (2) Sucrose octasulfate ester composes 60% to 100% of the full scale.

**Loss on drying** Not more than 14.0% (1 g, 105°C, 3 hours).

**Acid-consuming capacity** Weigh accurately about 0.25 g of Sucralfate, previously dried, place in a 200-mL glass-stoppered conical flask, add exactly 100 mL of 0.1 mol/L hydrochloric acid VS, stopper the flask tightly, and shake at 37 ± 2°C for exactly 1 hour (150 shakings per minute, amplitude: 20 mm). After cooling in water for 5 minutes, pipet 10 mL of the supernatant liquid, and titrate the excess acid with 0.1 mol/L sodium hydroxide VS until the pH becomes 3.5. Perform a blank determination. The amount of 0.1 mol/L hydrochloric acid VS consumed per g of Sucralfate is not less than 130 mL.

**Assay** **(1)** Aluminum—Weigh accurately about 1 g of Sucralfate, dissolve in 10 mL of dilute hydrochloric acid by warming on a water bath, cool, and add water to make exactly 250 mL. Pipet 25 mL of this solution, add exactly 25 mL of 0.05 mol/L disodium dihydrogen ethylenediamine tetraacetate VS and 20 mL of acetic acid-ammonium acetate buffer solution, pH 4.5, and boil for 5 minutes. After cooling, add 50 mL of ethanol (95), and titrate the excess disodium dihydrogen ethylenediamine tetraacetate with 0.05 mol/L zinc acetate VS until the color of the solution changes from green-purple through purple to red (indicator: 3 mL of dithizone TS). Perform a blank determination.

Each mL of 0.05 mol/L disodium dihydrogen ethylenediamine tetraacetate VS
= 1.3491 mg of Al

**(2)** Sucrose octasulfate ester—Weigh accurately about 0.55 g of Sucralfate, add exactly 10 mL of sulfuric acid-sodium hydroxide TS, shake vigorously, and dissolve with ultrasonic wave at below 30°C for 5 minutes. To this solution add 0.1 mol/L sodium hydroxide VS to make exactly 25 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.25 g of Potassium Sucrose Octasulfate Reference Standard, add the mobile phase to make exactly 25 mL, and use this solution as the standard solution. Prepare rapidly the sample solution and the standard solution, and perform the test immediately. Pipet 50 $\mu$L each of the sample solution and the standard solution, and perform the test as directed under the Liquid Chromatography according to the following conditions. Determine the peak areas, $A_T$ and $A_S$, of sucrose octasulfate ester from each solution.

Amount (mg) of sucrose octasulfate ester ($C_{12}H_{22}O_{35}S_8$)
= amount (mg) of Potassium Sucrose Octasulfate Reference Standard, calculated on the anhydrous basis

$$\times \frac{A_T}{A_S} \times 0.7633$$

*Operating conditions—*

Detector: A differential refractometer.

Column: A stainless steel column about 4 mm in inside diameter and about 30 cm in length, packed with aminopropylsilanized silica gel for liquid chromatography (about 8 $\mu$m in particle diameter).

Column temperature: Room temperature.

Mobile phase: Dissolve a suitable amount (26 to 132 g) of ammonium sulfate in 1000 mL of water, and adjust with phosphoric acid to a pH of 3.5. Allow a solution of Potassium Sucrose Octasulfate Reference Standard in dilute hydrochloric acid (1 in 100) to stand at 60°C for 10 minutes, cool, and perform the test immediately. Adjust the amount of ammonium sulfate in the mobile phase so that the peak of a related substance with the relative retention time being about 0.7 to that of sucrose octasulfate ester almost returns to the base line, and the peak of sucrose octasulfate ester elutes most rapidly.

Flow rate: Adjust the flow rate so that the retention time of sucrose octasulfate ester is between 6 and 11 minutes.

Selection of column: Allow a solution of Potassium Sucrose Octasulfate Reference Standard in dilute hydrochloric acid (1 in 100) to stand at 60°C for 10 minutes, cool, and proceed immediately with 50 $\mu$L of this solution under the above operating conditions. Use a column with a resolution being not less than 1.5 between sucrose octasulfate ester and a related substance with the relative retention time being about 0.7 to sucrose octasulfate ester.

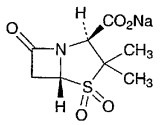
System repeatability: Repeat the test 6 times with the standard solution under the above operating conditions: the rela-

tive standard deviation of the peak area of sucrose octasulfate ester is not more than 2.0%.

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

# Sulbactam Sodium

スルバクタムナトリウム



$C_8H_{10}NNaO_5S$: 255.22
Monosodium (2*S*,5*R*)-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylate 4,4-dioxide
[*69388-84-7*]

Sulbactam Sodium contains not less than 875 μg (potency) per mg, calculated on the anhydrous basis. The potency of Sulbactam Sodium is expressed as mass (potency) of sulbactam ($C_8H_{11}NO_5S$: 233.24).

**Description** Sulbactam Sodium occurs as a white to yellowish white crystalline powder.

It is freely soluble in water, sparingly soluble in methanol, very slightly soluble in ethanol (99.5), and practically insoluble in acetonitrile.

**Identification** **(1)** Determine the infrared absorption spectrum of Sulbactam Sodium as directed in the potassium bromide disk method under the Infrared Spectrophotometry, and compare the spectrum with the Reference Spectrum: both spectra exhibit similar intensities of absorption at the same wave numbers.

**(2)** Sulbactam Sodium responds to the Qualitative Test (1) for sodium salt.

**Optical rotation** $[\alpha]_D^{20}$: +219 – +233° (1.0 g, water, 100 mL, 100 mm).

**pH** Dissolve 1.0 g of Sulbactam Sodium in 20 mL of water: the pH of the solution is between 5.2 and 7.2.

**Purity** **(1)** Clarity and color of solution—Dissolve 1.0 g of Sulbactam Sodium in 20 mL of water: the solution is clear, and colorless to pale yellow.

**(2)** Heavy metals—Proceed with 1.0 g of Sulbactam 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)** Arsenic—Prepare the test solution with 1.0 g of Sulbactam Sodium as directed in Method 3, and perform the test using Apparatus B (not more than 2 ppm).

**(4)** Sulbactam penicillamine—Weigh accurately about 0.2 g of Sulbactam Sodium, dissolve in the mobile phase to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.04 g of sulbactam sodium for sulbactam penicillamine, dissolve in 2 mL of water, add 0.5 mL of sodium hydroxide TS, allow to stand for 10 minutes at a room temperature, and add 0.5 mL of 1 mol/L hydrochloric acid TS, then add the mobile phase to make exactly 100 mL. Pipet 5 mL of this solution, add the mobile phase to make exactly 50 mL, and use this solution as the standard solution. Perform the test with 10 μL each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and determine the peak areas, $A_T$ and $A_S$, of sulbactam penicillamine by the automatic integration method: the amount of sulbactam penicillamine is not more than 1.0%.

$$\text{Amount (\%) of sulbactam penicillamine} = \frac{\text{amount (mg) of sulbactam sodium for sulbactam penicillamine}}{\text{amount (mg) of the sample}} \times \frac{A_T}{A_S} \times 5$$

*Operating conditions—*

Column, column temperature, mobile phase, and flow rate: Proceed as directed in the operating conditions in the Assay.

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

*System suitability—*

System performance: Proceed as directed in the system suitability in the Assay.

System repeatability: When the test is repeated 6 times with 10 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of sulbactam penicillamine is not more than 2.0%.

**Water** Not more than 1.0% (0.5 g, volumetric titration, direct titration).

**Assay** Weigh accurately an amount of Sulbactam Sodium and Sulbactam Reference Standard, equivalent to about 0.1 g (potency), dissolve each in a suitable amount of the mobile phase, add exactly 10 mL of the internal standard solution, then add the mobile phase to make 100 mL, and use these solutions as the sample solution and the standard solution. Perform the test with 10 μL 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 sulbactam to that of the internal standard.

$$\text{Amount [μg (potency)] of } C_8H_{11}NO_5S = \text{amount [mg (potency)] of Sulbactam Reference Standard} \times \frac{Q_T}{Q_S} \times 1000$$

*Internal standard solution*—A solution of ethyl parahydroxybenzoate in the mobile phase (7 in 1000).

*Operating conditions—*

Detector: An ultraviolet absorption photometer (wavelength: 220 nm).

Column: A stainless steel column 3.9 mm in inside diameter and 30 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (10 μm in particle diameter).

Column temperature: A constant temperature of about 35°C.

Mobile phase: To 750 mL of 0.005 mol/L tetrabutylammonium hydroxide TS add 250 mL of acetonitrile for liquid chromatography.

Flow rate: Adjust the flow rate so that the retention time of sulbactam is about 6 minutes.