

g, methanol, 20 mL, 100 mm).

**Purity** Other steroids—Dissolve 0.20 g of Prednisolone Acetate in exactly 10 mL of a mixture of chloroform and methanol (9:1), and use this solution as the sample solution. Separately, dissolve 0.020 g each of prednisolone, cortisone acetate and hydrocortisone acetate in exactly 10 mL of a mixture of chloroform and methanol (9:1). Pipet 1 mL of this solution, add a mixture of chloroform and methanol (9:1) to make exactly 10 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 5  $\mu$ L each of the sample solution and the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of dichloromethane, diethyl ether, methanol and water (385:75:40:6) to a distance of about 15 cm, and air-dry the plate. Examine under ultraviolet light (wavelength: 254 nm): the spots from the sample solution corresponding to those from the standard solution are not more intense than the spots from the standard solution, and any spot from the sample solution other than the principal spot and the spots from prednisolone, cortisone acetate and hydrocortisone acetate does not appear.

**Loss on drying** Not more than 1.0% (0.5 g, 105°C, 3 hours).

**Residue on ignition** Not more than 0.1% (0.5 g).

**Assay** Dissolve about 0.01 g each of Prednisolone Acetate and Prednisolone Acetate Reference Standard, previously dried and accurately weighed, in 60 mL each of methanol, add exactly 2 mL each of the internal standard solution, then add methanol to make 100 mL, and use these solutions as the sample solution and the standard solution. Perform the test with 10  $\mu$ 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 height of prednisolone acetate to that of the internal standard.

$$\begin{aligned} &\text{Amount (mg) of } C_{23}H_{30}O_6 \\ &= \text{amount (mg) of Prednisolone Acetate} \\ &\quad \text{Reference Standard} \times \frac{Q_T}{Q_S} \end{aligned}$$

**Internal standard solution**—A solution of butyl parahydroxybenzoate in methanol (3 in 1000).

**Operating conditions**—

**Detector:** An ultraviolet absorption photometer (wavelength: 254 nm).

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

**Column temperature:** A constant temperature of about 25°C.

**Mobile phase:** A mixture of water and acetonitrile (3:2).

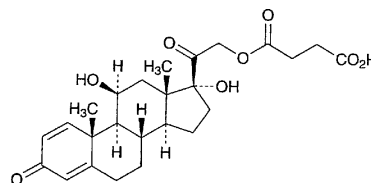
**Flow rate:** Adjust the flow rate so that the retention time of prednisolone acetate is about 10 minutes.

**Selection of column:** Proceed with 10  $\mu$ L of the standard solution under the above operating conditions, and calculate the resolution. Use a column giving elution of prednisolone acetate and the internal standard in this order with the resolution between these peaks being not less than 10.

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

## Prednisolone Succinate

コハク酸プレドニゾロン



$C_{25}H_{32}O_8$ : 460.52

11 $\beta$ ,17,21-Trihydroxypregna-1,4-diene-3,20-dione  
21-(hydrogen succinate) [2920-86-7]

Prednisolone Succinate, when dried, contains not less than 97.0% and not more than 103.0% of  $C_{25}H_{32}O_8$ .

**Description** Prednisolone Succinate occurs as a white, fine, crystalline powder. It is odorless.

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

Melting point: about 205°C (with decomposition).

**Identification (1)** To 2 mg of Prednisolone Succinate add 2 mL of sulfuric acid, and allow to stand for 2 to 3 minutes: a deep red color, without fluorescence, develops. To this solution add 10 mL of water cautiously: the color disappears and a gray, flocculent precipitate is formed.

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

**Optical rotation**  $[\alpha]_D^{20}$ : +114 – +120° (after drying, 0.067 g, methanol, 10 mL, 100 mm).

**Purity** Other steroids—Dissolve 0.10 g of Prednisolone Succinate in methanol to make exactly 10 mL, and use this solution as the sample solution. Separately, dissolve 0.030 g of prednisolone in methanol to make exactly 10 mL. Pipet 1 mL of the solution, add methanol to make exactly 10 mL, and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 5  $\mu$ L of the sample solution and the standard solution on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of ethyl acetate and ethanol (95) (2:1) to a distance of about 10 cm, and air-dry the plate. Examine the plate 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, in vacuum, phosphorus (V) oxide, 60°C, 6 hours).

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

**Assay** Weigh accurately about 0.01 g each of Prednisolone Succinate and Prednisolone Succinate Reference Standard,

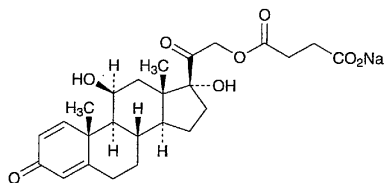
previously dried, and dissolve each in methanol to make exactly 100 mL. Pipet 5 mL each of these solutions, add methanol to make exactly 50 mL, and use these solutions as the sample solution and the standard solution. Determine the absorbances,  $A_T$  and  $A_S$ , of the sample solution and the standard solution at 242 nm as directed under the Ultraviolet-visible Spectrophotometry.

$$\begin{aligned} & \text{Amount (mg) of } C_{25}H_{32}O_8 \\ &= \text{amount (mg) of Prednisolone Succinate Reference Standard} \\ & \times \frac{A_T}{A_S} \end{aligned}$$

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

## Prednisolone Sodium Succinate for Injection

注射用コハク酸プレドニゾロンナトリウム



$C_{25}H_{31}NaO_8$ : 482.50  
Monosodium 11 $\beta$ ,17,21-trihydroxypregna-1,4-diene-3,20-dione 21-succinate [1715-33-9]

Prednisolone Sodium Succinate for Injection is a preparation for injection which is dissolved before used. It contains not less than 72.4% and not more than 83.2% of prednisolone sodium succinate ( $C_{25}H_{31}NaO_8$ : 482.50), and the equivalent of not less than 90% and not more than 110% of the labeled amount of prednisolone ( $C_{21}H_{28}O_5$ : 360.44).

The amount should be stated as the amount of prednisolone ( $C_{21}H_{28}O_5$ ).

**Method of preparation** Prepare as directed under Injections, with Prednisolone Succinate and Dried Sodium Carbonate or Sodium Hydroxide.

It contains a suitable buffer agent.

**Description** Prednisolone Sodium Succinate for Injection occurs as a white powder or porous, friable mass.

It is freely soluble in water.

It is hygroscopic.

**Identification** (1) To 2 mg of Prednisolone Sodium Succinate for Injection add 2 mL of sulfuric acid, and allow to stand for 2 to 3 minutes: a deep red color, without fluorescence, develops. To this solution add 10 mL of water cautiously: the color disappears and a gray, flocculent precipitate is formed.

(2) Dissolve 0.01 g of Prednisolone Sodium Succinate for Injection in 1 mL of methanol, add 1 mL of Fehling's TS, and heat: an orange to red precipitate is formed.

(3) Dissolve 0.1 g of Prednisolone Sodium Succinate for

Injection in 2 mL of sodium hydroxide TS, allow to stand for 10 minutes, and filter. Add 1 mL of dilute hydrochloric acid to the filtrate, shake, and filter if necessary. Adjust the solution with diluted ammonia TS (1 in 10) to a pH of about 6, and add 2 to 3 drops of iron (III) chloride TS: a brown precipitate is formed.

(4) Prednisolone Sodium Succinate for Injection responds to the Qualitative Tests (1) for sodium salt.

**pH** Dissolve 1.0 g of Prednisolone Sodium Succinate for Injection in 40 mL of water: the pH of the solution is between 6.5 and 7.2.

**Purity** Clarity and color of solution—Dissolve 0.25 g of Prednisolone Sodium Succinate for Injection in 10 mL of water: the solution is clear and colorless.

**Loss on drying** Not more than 2.0% (0.15 g, in vacuum, phosphorus (V) oxide, 60°C, 3 hours).

**Assay** Take a quantity of sealed containers of Prednisolone Sodium Succinate for Injection, equivalent to about 0.10 g of prednisolone ( $C_{21}H_{28}O_5$ ), and dissolve the contents in a suitable amount of diluted methanol (1 in 2), and transfer to a 100-mL volumetric flask. Wash each container with diluted methanol (1 in 2), collect the washings in the volumetric flask, and add diluted methanol (1 in 2) to make volume. Pipet 4 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL. Pipet 5 mL of this solution, add exactly 5 mL of the internal standard solution, mix, and use this solution as the sample solution. Separately, weigh accurately about 0.025 g of Prednisolone Succinate Reference Standard, previously dried in a desiccator for 6 hours (in vacuum, phosphorus (V) oxide, 60°C), dissolve in methanol to make exactly 25 mL. Pipet 5 mL of this solution, add diluted methanol (1 in 2) to make exactly 50 mL. Pipet 5 mL of this solution, add exactly 5 mL of the internal standard solution, mix, and use this solution as the standard solution. Perform the test with 10  $\mu$ L 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 prednisolone succinate to that of the internal standard.

Amount (mg) of prednisolone sodium succinate ( $C_{25}H_{31}NaO_8$ )

= amount (mg) of Prednisolone Succinate Reference Standard

$$\times \frac{Q_T}{Q_S} \times 5 \times 1.0477$$

Amount (mg) of prednisolone ( $C_{21}H_{28}O_5$ )

= amount (mg) of prednisolone sodium succinate ( $C_{25}H_{31}NaO_8$ )  $\times 0.7470$

**Internal standard solution**—A solution of propyl parahydroxybenzoate in diluted methanol (1 in 2) (1 in 25,000).

**Operating conditions**—

**Detector:** An ultraviolet absorption photometer (wavelength: 254 nm).

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

**Column temperature:** A constant temperature of about 25°C.