Identification—

A: *Infrared Absorption* ⟨197M⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 20 µg per mL.

Medium: alcohol.

Absorptivities at 243 nm, calculated on the dried basis, do not differ by more than 3.0%.

Specific rotation ⟨781S⟩: between +87° and +95°.

Test solution: 10 mg per mL, in dioxane.

Loss on drying ⟨731⟩—Dry it at 105° for 3 hours: it loses not more than 1.0% of its weight.

Residue on ignition ⟨281⟩: not more than 0.2%.

Chromatographic purity—

Diluent—Prepare a mixture of water, tetrahydrofuran, acetonitrile, and acetic acid (47:25:25:3).

Mobile phase—Prepare a filtered and degassed mixture of water, tetrahydrofuran, and formic acid (745:255:1). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard solution—Dissolve an accurately weighed quantity of USP Methylprednisolone Hemisuccinate RS in *Diluent* to obtain a solution having a final concentration of about 0.02 mg per mL.

Test solution—Prepare a solution of Methylprednisolone Hemisuccinate in *Diluent* containing about 1 mg per mL. Shake or sonicate to aid in solubilization.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4.6-mm × 20-cm column that contains 5-µm packing L1. The flow rate is about 1.0 mL per minute. Chromatograph the *Standard solution*, and record the peak responses as directed for *Procedure:* the column efficiency determined from the methylprednisolone hemisuccinate peak is not less than 5000; and the relative standard deviation for replicate injections is not more than 5.0%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard solution* and the *Test solution* into the chromatograph, record the chromatograms, and measure all of the peak areas. Calculate the percentage of each impurity in the portion of Methylprednisolone Hemisuccinate taken by the formula:

$$100(C_S / C_U)(r_i / r_S)$$

in which C_S is the concentration, in mg per mL, of USP Methylprednisolone Hemisuccinate RS in the *Standard solution;* C_U is the concentration, in mg per mL, of Methylprednisolone Hemisuccinate in the *Test solution;* r_i is the peak area for each impurity obtained from the *Test solution;* and r_S is the peak area of methylprednisolone hemisuccinate obtained from the *Standard solution:* not more than 1.0% of any individual impurity is found, and not more than 2.0% of total impurities is found.

Assay—

Internal standard solution—Dissolve USP Fluorometholone RS in tetrahydrofuran to obtain a solution containing about 6 mg per mL.

Mobile phase—Prepare a solution containing a mixture of butyl chloride, water-saturated butyl chloride, tetrahydrofuran, methanol, and glacial acetic acid (475:475:70:35:30).

Standard preparation—Transfer about 40 mg of USP Methylprednisolone Hemisuccinate RS, accurately weighed, to a 100-mL volumetric flask. Add 5.0 mL of *Internal standard solution.* Dilute with chloroform containing 3% glacial acetic acid to volume, and mix to dissolve the powder.

Assay preparation—Using about 40 mg of Methylprednisolone Hemisuccinate, accurately weighed, prepare as directed for *Standard preparation*.

Procedure—Using a suitable microsyringe or sampling valve, inject separately suitable portions, between 4 and 8 µL, of the *Standard preparation* and the *Assay preparation* into a suitable high-pressure liquid chromatograph (see *Chromatography* ⟨621⟩) of the general type equipped with a detector for monitoring UV absorption at about 254 nm, equipped with a suitable recorder, capable of providing column pressure up to about 1000 psi and fitted with a 4-mm × 30-cm stainless steel column that contains packing L3. In a suitable chromatogram, the resolution, *R,* between methylprednisolone hemisuccinate and the internal standard is not less than 2.0; and six replicate injections of the *Standard preparation* show a coefficient of variation of not more than 2.0%. Calculate the quantity, in mg, of $C_{26}H_{34}O_8$ in the portion of Methylprednisolone Hemisuccinate taken by the formula:

$$100C(R_U / R_S)$$

in which *C* is the concentration, in mg per mL, of USP Methylprednisolone Hemisuccinate RS in the *Standard preparation;* and R_U and R_S are the peak area ratios of methylprednisolone hemisuccinate to the internal standard obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Methylprednisolone Sodium Succinate

$C_{26}H_{33}NaO_8$ 496.53

Pregna-1,4-diene-3,20-dione, 21-(3-carboxy-1-oxopropoxy)-11,17-dihydroxy-6-methyl-, monosodium salt, (6α,11β)-.

11β,17,21-Trihydroxy-6α-methylpregna-1,4-diene-3,20-dione 21-(sodium succinate) [2375-03-3].

» Methylprednisolone Sodium Succinate contains not less than 97.0 percent and not more than 103.0 percent of $C_{26}H_{33}NaO_8$, calculated on the dried basis.

Packaging and storage—Preserve in tight, light-resistant containers.

USP Reference standards ⟨11⟩—

USP Methylprednisolone Hemisuccinate RS

Identification—

A: Transfer about 100 mg to a separator, dissolve in 10 mL of water, add 1 mL of 3 N hydrochloric acid, and extract immediately with 50 mL of chloroform. Filter the chloroform extract through cotton, evaporate on a steam bath to dryness, and dry in vacuum at 60° for 3 hours: the IR absorption spectrum of a mineral oil dispersion of the residue so obtained exhibits maxima only at the same wavelengths as that of a similar preparation of USP Methylprednisolone Hemisuccinate RS.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 20 µg per mL.

Medium: methanol.

Absorptivities at 243 nm, calculated on the dried basis, do not differ by more than 3.0%.

C: It responds to the flame test for *Sodium* ⟨191⟩.

Specific rotation ⟨781S⟩: between +96° and +104°.

Test solution: 10 mg per mL, in alcohol.

Loss on drying ⟨731⟩—Dry it at 105° for 3 hours: it loses not more than 3.0% of its weight.

Sodium content—Dissolve, with gentle heating, about 1 g of it, accurately weighed, in 75 mL of glacial acetic acid. Add 20 mL of dioxane, then add 1 drop of crystal violet TS, and titrate with 0.1 N perchloric acid VS to a blue-green endpoint. Perform a blank determination, and make any necessary correction. Each mL of 0.1 N perchloric acid is equivalent to 2.299 mg of Na. Not less than 4.49% and not more than 4.77%, calculated on the dried basis, is found.

Assay—

Standard preparation—Proceed as directed under *Assay for Steroids* ⟨351⟩, using USP Methylprednisolone Hemisuccinate RS to obtain a solution having a known concentration of about 12.5 µg per mL.

Assay preparation—Weigh accurately about 100 mg of Methylprednisolone Sodium Succinate, dissolve it in alcohol to make 200.0 mL, and mix. Pipet 5 mL of this solution into a 200-mL volumetric flask, add alcohol to volume, and mix. Pipet 20 mL of the resulting solution into a glass-stoppered, 50-mL conical flask.

Procedure—To each of the flasks containing the *Assay preparation* and the *Standard preparation*, and to a similar flask containing 20.0 mL of alcohol, to provide the blank, add 2.0 mL of a solution prepared by dissolving 50 mg of blue tetrazolium in 10 mL of alcohol, and mix. Then to each flask add 4.0 mL of a mixture of 1 volume of tetramethylammonium hydroxide TS and 9 volumes of alcohol. Mix, allow to stand in the dark for 90 minutes, add 1.0 mL of glacial acetic acid, mix, and proceed as directed for *Procedure* under *Assay for Steroids* ⟨351⟩, beginning with "Concomitantly determine the absorbances." Calculate the quantity, in mg, of $C_{26}H_{33}NaO_8$ in the portion of Methylprednisolone Sodium Succinate taken by the formula:

$$8.37C(A_U / A_S).$$

Methylprednisolone Sodium Succinate for Injection

» Methylprednisolone Sodium Succinate for Injection is a sterile mixture of Methylprednisolone Sodium Succinate with suitable buffers. It may be prepared from Methylprednisolone Sodium Succinate or from Methylprednisolone Hemisuccinate with the aid of Sodium Hydroxide or Sodium Carbonate. It contains the equivalent of not less than 90.0 percent and not more than 110.0 percent of the labeled amount of methylprednisolone ($C_{22}H_{30}O_5$) in the volume of constituted solution designated on the label.

Packaging and storage—Preserve in *Containers for Sterile Solids* as described under *Injections* ⟨1⟩.

USP Reference standards ⟨11⟩**—**
USP Endotoxin RS
USP Fluorometholone RS
USP Methylprednisolone RS
USP Methylprednisolone Hemisuccinate RS

Constituted solution—At the time of use, it meets the requirements for *Constituted Solutions* under *Injections* ⟨1⟩.

Identification—It meets the requirements of *Identification* test *A* under *Methylprednisolone Sodium Succinate*.

Bacterial endotoxins ⟨85⟩**—**It contains not more than 0.17 USP Endotoxin Unit per mg of methylprednisolone.

pH ⟨791⟩: between 7.0 and 8.0, in a solution containing about 50 mg of methylprednisolone sodium succinate per mL.

Loss on drying ⟨731⟩**—**Dry it at 105° for 3 hours: it loses not more than 2.0% of its weight.

Particulate matter ⟨788⟩: meets the requirements for small-volume injections.

Free methylprednisolone—Using the chromatograms obtained in the *Assay*, measure the areas of the peaks from the internal standard and free methylprednisolone. Calculate the ratio of the area of the free methylprednisolone peak to that of the internal standard in the chromatogram obtained from the *Standard preparation*, S_S, and the same ratio in the chromatogram obtained from the *Assay preparation*, S_U. Calculate the quantity, in mg, of free methylprednisolone in the *Assay preparation* taken by the formula:

$$100C(S_U / S_S)$$

in which C is the concentration, in mg per mL, of USP Methylprednisolone RS in the *Standard preparation;* and S_U and S_S are the ratios as defined above. The amount of free methylprednisolone is not more than 6.6% of the labeled amount of methylprednisolone.

Other requirements—It meets the requirements for *Sterility Tests* ⟨71⟩, *Uniformity of Dosage Units* ⟨905⟩, and *Labeling* under *Injections* ⟨1⟩.

Assay—

Internal standard solution—Prepare a solution of USP Fluorometholone RS in tetrahydrofuran containing about 3 mg per mL.

Mobile phase—Prepare a filtered mixture of butyl chloride, water-saturated butyl chloride, tetrahydrofuran, methanol, and glacial acetic acid (95:95:14:7:6). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Weigh accurately about 32.5 mg of USP Methylprednisolone Hemisuccinate RS, and transfer it to a 50-mL volumetric flask. Add by pipet 5.0 mL of *Internal standard solution* and 5.0 mL of a solution of glacial acetic acid in chloroform (3 in 100) containing in each mL an accurately known quantity of about 0.30 mg of USP Methylprednisolone RS. Dilute with glacial acetic acid in chloroform (3 in 100) to volume, and mix.

Assay preparation—Mix the constituted solutions prepared from the contents of 10 vials of Methylprednisolone Sodium Succinate for Injection. Transfer an accurately measured volume of the resulting constituted solution, equivalent to about 50 mg of methylprednisolone, to a suitable flask containing 10.0 mL of *Internal standard solution*, and dilute with glacial acetic acid in chloroform (3 in 100) to 100.0 mL. Shake thoroughly for 5 minutes, then allow the phases to separate, discarding the upper phase.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 3.9-mm × 30-cm column that contains packing L3. The flow rate is about 1.0 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed for *Procedure:* the order of elution of peaks is the internal standard peak, methylprednisolone hemisuccinate peak, and successive smaller peaks of free methylprednisolone and methylprednisolone 17-hemisuccinate.

Procedure—Separately inject equal volumes (about 6 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the peak areas for the internal standard, methylprednisolone hemisuccinate, and methylprednisolone 17-hemisuccinate. Calculate the quantity, in mg, of methylprednisolone ($C_{22}H_{30}O_5$) in the portion of constituted solution taken by the formula:

$$0.789(100C)(R_U / R_S)$$

in which 0.789 is the ratio of the molecular weight of methylprednisolone to that of methylprednisolone hemisuccinate; C is the concentration, in mg per mL, of USP Methylprednisolone Hemisuccinate RS in the *Standard preparation;* and R_U and R_S are the ratios of the sum of the peak areas for methylprednisolone hemisuccinate and methylprednisolone 17-hemisuccinate to the peak area of the internal standard obtained from the *Standard preparation* and the *Assay preparation*, respectively. To this quantity add the amount, in mg, of free methylprednisolone found in the test for *Free methylprednisolone.*