tion and methanol (55:45). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Resolution solution—Prepare a solution in methanol containing about 0.4 mg of USP Dihydrocodeine Bitartrate RS and 0.6 mg of hydrocodone bitartrate per mL. Prepare a mixture of this solution and water (1:1).

Standard preparation—Transfer about 20 mg of USP Dihydrocodeine Bitartrate RS, accurately weighed, to a 50-mL volumetric flask, add 25 mL of methanol, swirl to dissolve, dilute with water to volume, and mix.

Assay preparation—Transfer about 20 mg of Dihydrocodeine Bitartrate, accurately weighed, to a 50-mL volumetric flask, add 25 mL of methanol, swirl to dissolve, dilute with water to volume, and mix.

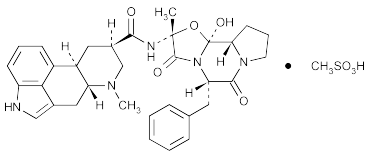
Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 280-nm detector and a 4.6-mm × 25-cm column that contains 5- µm packing L3. The flow rate is about 1.5 mL per minute. Chromatograph the *Resolution solution*, and record the responses as directed under *Procedure:* the relative retention times are about 0.8 for hydrocodone and 1.0 for dihydrocodeine, and the resolution, *R*, between the hydrocodone and dihydrocodeine peaks is not less than 1.8. Chromatograph the *Standard preparation*, and record the responses as directed under *Procedure:* the column efficiency determined from the dihydrocodeine peak is not less than 900 theoretical plates, the tailing factor for the dihydrocodeine peak is not more than 1.7, and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 25 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the responses for the major peaks. Calculate the quantity, in mg, of $C_{18}H_{23}NO_3 \cdot C_4H_6O_6$ in the portion of Dihydrocodeine Bitartrate taken by the formula:

$$50C(r_U / r_S)$$

in which *C* is the concentration, in mg per mL, of USP Dihydrocodeine Bitartrate RS in the *Standard preparation*, and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Dihydroergotamine Mesylate



$C_{33}H_{37}N_5O_5 \cdot CH_4O_3S$ 679.78

Ergotaman-3′,6′,18-trione, 9,10-dihydro-12′-hydroxy-2′-methyl-5′-(phenylmethyl)-, (5′α)-, monomethanesulfonate (salt).

Dihydroergotamine monomethanesulfonate [6190-39-2].

» Dihydroergotamine Mesylate contains not less than 97.0 percent and not more than 103.0 percent of $C_{33}H_{37}N_5O_5 \cdot CH_4O_3S$, calculated on the dried basis.

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

USP Reference standards ⟨11⟩**—**
USP Dihydroergotamine Mesylate RS

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 50 µg per mL.

Medium: 70% alcohol.

Absorptivities at 280 nm, calculated on the dried basis, do not differ by more than 3.0%.

C: The principal spot from the *Test preparation* found in the test for *Related alkaloids* corresponds in R_F value to that obtained from the *Standard preparation*.

Specific rotation ⟨781S⟩**:** between −16.7° and −22.7°.

Test solution: 25 mg per mL, in a mixture of chloroform, alcohol, and ammonium hydroxide (10:10:1).

pH ⟨791⟩**:** between 4.4 and 5.4, in a solution (1 in 1000).

Loss on drying ⟨731⟩**—**Dry it in vacuum at 100 ° to constant weight: it loses not more than 4.0% of its weight.

Related alkaloids—

Solvent mixture—Mix 10 volumes of chloroform, 10 volumes of methanol, and 1 volume of ammonium hydroxide.

Test solution—Prepare a solution of Dihydroergotamine Mesylate in *Solvent mixture* to contain 20 mg per mL.

Standard solution and *Standard dilutions*—Prepare a solution of USP Dihydroergotamine Mesylate RS in *Solvent mixture* to contain 20 mg per mL *(Standard solution).* Prepare a series of dilutions of the *Standard solution* in *Solvent mixture* to contain 0.40 mg, 0.20 mg, and 0.10 mg per mL *(Standard dilutions).*

Procedure—In a suitable chromatographic chamber arranged for thin-layer chromatography place a volume of a solvent system consisting of a mixture of chloroform and alcohol (9:1) sufficient to develop the chromatogram, cover, and allow to equilibrate for 30 minutes. Apply 5- µL portions of the *Test solution,* the *Standard solution,* and each of the three *Standard dilutions* to a suitable thin-layer chromatographic plate coated with a 0.25-mm layer of chromatographic silica gel. Allow the spots to dry, and develop the chromatogram until the solvent front has moved about three-fourths of the length of the plate. Remove the plate from the developing chamber, mark the solvent front, and allow the solvent to evaporate. Locate the spots on the plate by lightly spraying with a solution prepared by dissolving 800 mg of *p*-dimethylaminobenzaldehyde in a cooled mixture of 80 g of alcohol and 20 g of sulfuric acid. The R_F value of the principal spot obtained from the *Test solution* corresponds to that obtained from the *Standard solution*. Estimate the concentration of any other spots observed in the lane for the *Test solution* by comparison with the *Standard dilutions*. The spots from the 0.40-, 0.20-, and 0.10-mg-per-mL dilutions are equivalent to 2.0%, 1.0%, and 0.50% of impurities, respectively. The sum of the impurities is not greater than 2.0%.

Assay—

Diluent 1—Prepare a solution of 0.1 mL of phosphoric acid in 1000 mL of water.

Diluent 2—Prepare a mixture of *Diluent 1* and acetonitrile (60:40).

Solution A—Prepare a filtered and degassed mixture of water, 25 percent ammonia water, and 98% formic acid (1000:10:5). Adjust the pH to 8.50.

Solution B—Prepare a filtered and degassed mixture of acetonitrile and *Solution A* (80:20).

Mobile phase—Use variable mixtures of *Solution A* and *Solution B* as directed for *Chromatographic system*. Make adjustments to either solution as necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Dihydroergotamine Mesylate RS in acetonitrile, and dilute quantitatively, and stepwise if necessary, with *Diluent 1* to obtain a solution having a known concentration of about 0.6 mg per mL. [NOTE—The final ratio of acetonitrile and *Diluent 1* should be similar to the final ratio obtained in the *Assay preparation.*]

Assay preparation—Transfer about 30 mg of Dihydroergotamine Mesylate, accurately weighed, to a 50-mL volumetric flask, dissolve in 20 mL of acetonitrile, dilute with *Diluent 1* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 280-nm detector and 4.0-mm × 25-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. The chromatograph is programmed as follows.

Time (minutes)	Solution A (%)	Solution B (%)	Elution
0	60	40	equilibration
0–12	60→50	40→50	linear gradient
12–20	50→15	50→85	linear gradient
20–25	15	85	isocratic
24–25	15→60	85→40	linear gradient
25–31	60	40	re-equilibration

Chromatograph the *Standard preparation,* and record the peak areas as directed for *Procedure:* the tailing factor is between 0.8 and 1.5; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 10 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the areas for the analyte peaks. Calculate the quantity, in mg, of $C_{33}H_{37}N_5O_5 \cdot CH_4O_3S$ in the portion of Dihydroergotamine Mesylate taken by the formula:

$$50C(r_U / r_S)$$

in which C is the concentration, in mg per mL, of USP Dihydroergotamine Mesylate RS in the *Standard preparation;* and r_U and r_S are the peak areas obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Dihydroergotamine Mesylate Injection

» Dihydroergotamine Mesylate Injection is a sterile solution of Dihydroergotamine Mesylate in Water for Injection. It contains not less than 90.0 percent and not more than 110.0 per cent of the labeled amount of $C_{33}H_{37}N_5O_5 \cdot CH_4O_3S$.

Packaging and storage—Preserve in single-dose containers, preferably of Type I glass, protected from light.

USP Reference standards ⟨11⟩**—**
USP Dihydroergotamine Mesylate RS
USP Endotoxin RS

Identification—Dilute 2 mL of Injection with water to 25 mL: the UV absorption spectrum of the solution so obtained exhibits maxima and minima at the same wavelengths as that of a similar solution of USP Dihydroergotamine Mesylate RS , concomitantly measured.

Bacterial endotoxins ⟨85⟩**—**It contains not more than 175.0 USP Endotoxin Units per mg of dihydroergotamine mesylate.

pH ⟨791⟩: between 3.4 and 4.9.

Other requirements—It meets the requirements under *Injections* ⟨1⟩.

Assay—

Reagent preparation—Dissolve 250 mg of *p*-dimethylaminobenzaldehyde in a cooled mixture of 130 mL of sulfuric acid and 70 mL of water, and add 0.40 mL of ferric chloride solution (1 in 20).

Standard preparation—Dissolve in tartaric acid solution (1 in 100) a suitable quantity of USP Dihydroergotamine Mesylate RS, accurately weighed, and dilute quantitatively and stepwise with the same solvent to obtain a solution having a known concentration of about 50 µg per mL.

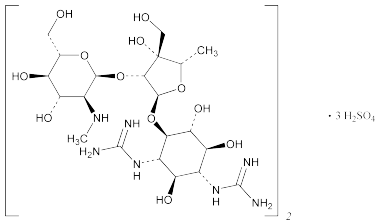
Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 5 mg of dihydroergotamine mesylate, to a 100-mL volumetric flask, dilute with tartaric acid solution (1 in 100) to volume, and mix.

Procedure—Transfer 5.0 mL each of the *Standard preparation,* the *Assay preparation*, and tartaric acid solution (1 in 100) to provide the blank, to separate 50-mL conical flasks. Add 10.0 mL of the *Reagent preparation* to each, shake, and allow to stand for 30 minutes. Concomitantly determine the absorbances of the solutions in 1-cm cells at the wavelength of maximum absorbance at about 585 nm, with a suitable spectrophotometer, using the blank to set the instrument. Calculate the quantity, in mg, of $C_{33}H_{37}N_5O_5 \cdot CH_4O_3S$ in each mL of the Injection taken by the formula:

$$(0.1C / V)(A_U / A_S)$$

in which C is the concentration, in µg per mL, of USP Dihydroergotamine Mesylate RS in the *Standard preparation, V* is the volume, in mL, of Injection taken, and A_U and A_S are the absorbances of the solutions from the *Assay preparation* and the *Standard preparation,* respectively.

Dihydrostreptomycin Sulfate



$(C_{21}H_{41}N_7O_{12})_2 \cdot 3H_2SO_4$ 1461.44
Dihydrostreptomycin sulfate (2:3) (salt) [5490-27-7].

» Dihydrostreptomycin Sulfate has a potency equivalent to not less than 650 µg of dihydrostreptomycin ($C_{21}H_{41}N_7O_{12}$) per mg, except that if it is labeled as being cr ystalline, it has a potency equivalent to not less than 725 µg of dihydrostreptomycin per mg, or if it is labeled as being solely for oral use, it has a potency equivalent to not less than 450 µg of dihydrostreptomycin per mg.

Packaging and storage—Preserve in tight containers.

Labeling—Label it to indicate that it is intended for veterinar y use only. If it is cr ystalline, it may be so labeled. If it is intended solely for oral use, it is so labeled. Where it is intended for use in preparing injectable dosage forms, the label states that it is sterile or must be subjected to further processing during the preparation of injectable dosage forms.

USP Reference standards ⟨11⟩**—**
USP Dihydrostreptomycin Sulfate RS
USP Endotoxin RS
USP Streptomycin Sulfate RS

Identification—

A: To a solution of 4 mg in 2 mL of water, add 0.5 mL of 1 N hydrochloric acid, and heat in a water bath for 20 minutes. Remove the tube from the bath, and add 1.0 mL of a 1 in 200 solution of 1-naphthol in 1 N sodium hydroxide. Heat again for 10 minutes, cool briefly in an ice bath, and add water to make