and record the responses as directed under *Procedure:* the capacity factor, k', for the ciprofloxacin peak is between 1.5 and 6, the column efficiency is not less than 500 theoretical plates, the tailing factor for the analyte peak is not less than 0.9 and not more than 2.0, and the relative standard deviation for replicate injections is not more than 2%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the areas for the ciprofloxacin peaks. Calculate the quantity, in mg, of ciprofloxacin ($C_{17}H_{18}FN_3O_3$) in each mL of the Ophthalmic Solution taken by the formula:

$$(331.34/367.81)(50C/V)(r_U/r_S),$$

in which 331.34 and 367.81 are the molecular weights of ciprofloxacin and anhydrous ciprofloxacin hydrochloride, respectively; C is the concentration, in mg per mL, of USP Ciprofloxacin Hydrochloride RS in the *Standard preparation*, calculated on the anhydrous basis; V is the volume, in mL, of Ophthalmic Solution taken; and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Ciprofloxacin Tablets

» Ciprofloxacin Tablets contain Ciprofloxacin Hydrochloride equivalent to not less than 90.0 percent and not more than 110.0 percent of the labeled amount of ciprofloxacin ($C_{17}H_{18}FN_3O_3$).

Packaging and storage—Preserve in well-closed containers.

USP Reference standards ⟨11⟩—*USP Ciprofloxacin Hydrochloride RS. USP Ciprofloxacin Ethylenediamine Analog RS.*

Identification—

A: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

B: Place a number of Tablets, equivalent to about 1500 mg of ciprofloxacin, in a suitable flask containing about 750 mL of water, and sonicate for about 20 minutes. Dilute with water to 1000 mL, and mix. Centrifuge a portion of this suspension, and use the clear supernatant obtained as the test solution. Dissolve a quantity of USP Ciprofloxacin Hydrochloride RS in water to obtain a Standard solution containing 1.5 mg per mL. Proceed as directed for *Identification* test *B* under *Ciprofloxacin Hydrochloride*, starting with "Separately apply, as 1-cm bands, 5 µL each," except to use 10 µL each of the test solution and the Standard solution: the specified result is obtained.

Dissolution ⟨711⟩—

Medium: 0.01 N hydrochloric acid; 900 mL.

Apparatus 2: 50 rpm.

Time: 30 minutes.

Procedure—Determine the amount of ciprofloxacin hydrochloride ($C_{17}H_{18}FN_3O_3 \cdot HCl$) dissolved by employing UV absorption at the wavelength of maximum absorbance at about 276 nm on filtered portions of the solution under test, suitably diluted with *Dissolution Medium,* if necessary, in comparison with a Standard solution having a known concentration of USP Ciprofloxacin Hydrochloride RS in the same *Medium.*

Tolerances—An amount of ciprofloxacin hydrochloride ($C_{17}H_{18}FN_3O_3 \cdot HCl$) equivalent to not less than 80% *(Q)* of the labeled amount of ciprofloxacin ($C_{17}H_{18}FN_3O_3$) is dissolved in 30 minutes.

Uniformity of dosage units ⟨905⟩: meet the requirements.

Assay—

Diluent—Prepare a filtered and degassed mixture of 0.025 M phosphoric acid, previously adjusted (with triethylamine) to a pH of 2.0 ± 0.1, and acetonitrile (87 : 13).

Mobile phase—Prepare a filtered and degassed mixture of 0.025 M phosphoric acid, previously adjusted (with triethylamine) to a pH of 3.0 ± 0.1, and acetonitrile (87 : 13). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Quantitatively dissolve an accurately weighed quantity of USP Ciprofloxacin Hydrochloride RS in *Diluent* to obtain a solution having a known concentration of about 0.2 mg per mL.

Resolution solution—Dissolve a quantity of USP Ciprofloxacin Ethylenediamine Analog RS in the *Standard preparation* to obtain a solution containing about 0.05 mg per mL.

Assay preparation—Transfer 5 Tablets to a 500-mL volumetric flask, add about 400 mL of *Diluent,* and sonicate for about 20 minutes. Dilute with *Diluent* to volume, and mix. Quantitatively dilute an accurately measured volume of this solution, previously filtered through a 0.45-µm membrane filter, with *Diluent* to obtain a solution containing the equivalent of about 0.20 mg of ciprofloxacin per mL.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 278-nm detector and a 4.6-mm × 25-cm column that contains packing L1 and is operated at 30 ± 1°. The flow rate is about 1.5 mL per minute. Chromatograph the *Resolution solution,* and record the peak responses as directed for *Procedure:* the retention time for ciprofloxacin is between 6.4 and 10.8 minutes; the relative retention times are about 0.7 for ciprofloxacin ethylenediamine analog and 1.0 for ciprofloxacin; and the resolution, R, between the ciprofloxacin ethylenediamine analog peak and the ciprofloxacin peak is not less than 6. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the column efficiency, determined from the ciprofloxacin peak, is not less than 2500 theoretical plates; the tailing factor for the ciprofloxacin peak is not more than 2.0; and the relative standard deviation for replicate injections is not more than 1.5%.

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 peak areas for the major peaks. Calculate the quantity, in mg, of ciprofloxacin ($C_{17}H_{18}FN_3O_3$) in each Tablet taken by the formula:

$$(331.34/367.81)(CL/D)(r_U/r_S),$$

in which 331.34 and 367.81 are the molecular weights of ciprofloxacin and anhydrous ciprofloxacin hydrochloride, respectively; C is the concentration, in mg per mL, of USP Ciprofloxacin Hydrochloride RS in the *Standard preparation,* calculated on the anhydrous basis; L is the labeled quantity, in mg, of ciprofloxacin in each Tablet; D is the concentration, in mg per mL, of ciprofloxacin in the *Assay preparation,* based on the labeled quantity per Tablet and the extent of dilution; and r_U and r_S are the ciprofloxacin peak areas obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Cisplatin

$Cl_2H_6N_2Pt$ 300.04

Platinum, diamminedichloro-, (*SP*-4-2)-.

cis-Diamminedichloroplatinum [*15663-27-1*].

» Cisplatin contains not less than 98.0 percent and not more than 102.0 percent of $Cl_2H_6N_2Pt$, calculated on the anhydrous basis.

Caution—Cisplatin is potentially cytotoxic. Great care should be taken to prevent inhaling particles and exposing the skin to it.

Packaging and storage—Preserve in tight containers. Protect from light.

USP Reference standards ⟨11⟩—*USP Cisplatin RS. USP Transplatin RS. USP Potassium Trichloroammineplatinate RS.*

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Identification—

A: The retention ti
the *Assay preparatio*
preparation as obtainec

B: *Infrared Absorp*

C: *Spray reagent*—
hydrochloric acid, and
that all of the solids dis
90 mL of water. Mix
precipitate that is forme
at least 1 week.

Procedure—Prepare
per mL and a Standard s
per mL, both in din
quantities of each solu
coated with a 0.25-mm
(see *Chromatography*
chromatographic charr
equilibrated for 30 mini
of acetone and 1 N nit
distance of about 8 cm fi
to air-dry. Complete th
about 100° for 1 minute
an oven at about 100° f
solution of potassium io
spots: the principal sp
appearance and R_F valu

Crystallinity ⟨695⟩: r

Water, *Method I* ⟨921⟩:

UV purity ratio—[NOT
hydrochloric acid and n
and dry before use. Do
acetone or pressurized
light, and use within 1 h
mg of ground Cisplatin
hydrochloric acid to
alternately stir at a hig
seconds until complet
frequently to remove pa
UV absorption spectrur
0.1 N hydrochloric aci
absorbance at the maxin
246 nm is not less than

Limit of trichloroamm

Mobile phase—Trans
volumetric flask, dissol
Degas, and filter throug
this solution is 5.9 ± 0.
the *Mobile phase,* if
requirements.

Standard preparatio
solve a suitable quantity
RS, accurately weighed
saline TS to obtain a sol
6 µg per mL. Use withi

Test preparation—[N
about 50 mg of Cisp
volumetric flask, and d
dissolve by stirring by r
4 hours.

Chromatographic s
liquid chromatograph
4.6-mm × 25-cm colun
about 2 mL per minute
and record the peak r
resolution, R, between
platinate peak is not les
for replicate injections

Procedure—Separate
Standard preparation a
graph, record the chrom
due to trichloroammin

USP 28

omolyn Sodium RS.
ɔum of the *Assay prepara-*
ɔits maxima and minima at
ɔolution of USP Cromolyn

:ts the requirements.

ons of Inhalation Solution
Sodium RS in a mixture of
acetone (6 : 4 : 1) contain-
A) and 0.1 mg per mL
yer chromatographic plate
ɔith a 0.25-mm layer of
ɔw the spots to dry, and
tem consisting of a mixture
tic acid (9 : 9 : 2) until the
ɪs of the length of the plate.
:hamber, mark the solvent
ocate the spots on the plate
light: the R_F value of the
ɔn Solution corresponds to
n A. Any spot in the
on Solution moving ahead
nse than the spot in the
olution B (1.0%).

l *Standard preparation—*
omolyn Sodium.
r an accurately measured
lent to about 25 mg of
having a concentration of
ɪis solution into a 100-mL
um phosphate buffer, dilute

the absorbances of the
ɔaration in 1-cm cells at
: at about 326 nm, with a
00 aqueous solution of *pH*
:. Calculate the quantity, in
ɪhalation Solution taken by

ɔer mL, of USP Cromolyn
V is the volume, in mL, of
are the absorbances of the
ɔaration and the *Standard*

Solution

lution is an aqueous
contains not less than
110.0 percent of the
It may contain suitable

ɪt, light-resistant containers.
Cromolyn Sodium RS.
ɪts for *Identification* test *B*

ɪuirements of the test for
ɔdium Inhalation Solution,
"Inhalation Solution."

ɔare as directed in the *Assay*

Assay preparation—Transfer 4 mL of Nasal Solution to a 100-mL volumetric flask, dilute with water to volume, and mix. Transfer an aliquot of this solution, equivalent to 8 mg of cromolyn sodium, to a 250-mL volumetric flask. Add 2.5 mL of *pH 7.4 Sodium phosphate buffer*, dilute with water to volume, and mix.

Standard preparation—Prepare as directed in the *Assay* under *Cromolyn Sodium*.

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Cromolyn Sodium Inhalation Solution*.

Cromolyn Sodium Ophthalmic Solution

» Cromolyn Sodium Ophthalmic Solution is a sterile, aqueous solution of Cromolyn Sodium. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of $C_{23}H_{14}Na_2O_{11}$. It may contain suitable antimicrobial and stabilizing agents.

Packaging and storage—Preserve in tight, light-resistant, single-dose or multiple-dose containers. Ophthalmic Solution that is packaged in multiple-dose containers contains a suitable antimicrobial agent.

USP Reference standards ⟨11⟩—*USP Cromolyn Sodium RS.*

Identification—It meets the requirements for *Identification* test *B* under *Cromolyn Sodium*.

Sterility ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 4.0 and 7.0.

Related compounds—It meets the requirements of the test for *Related compounds* under *Cromolyn Sodium Inhalation Solution*, "Ophthalmic Solution" being read in place of "Inhalation Solution."

Assay—

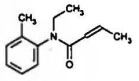
pH 7.4 Sodium phosphate buffer—Prepare as directed in the *Assay* under *Cromolyn Sodium*.

Assay preparation—Transfer 4 mL of Ophthalmic Solution to a 100-mL volumetric flask, dilute with water to volume, and mix. Transfer an aliquot of this solution, equivalent to 8 mg of cromolyn sodium, to a 250-mL volumetric flask. Add 2.5 mL of *pH 7.4 Sodium phosphate buffer*, dilute with water to volume, and mix.

Standard preparation—Prepare as directed in the *Assay* under *Cromolyn Sodium*.

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Cromolyn Sodium Inhalation Solution*.

Crotamiton



$C_{13}H_{17}NO$ 203.28
2-Butenamide, *N*-ethyl-*N*-(2-methylphenyl)-.
N-Ethyl-*o*-crotonotoluidide [*483-63-6*].

» Crotamiton is a mixture of *cis* and *trans* isomers containing not less than 97.0 percent and not more than 103.0 percent of $C_{13}H_{17}NO$.

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

USP Reference standards ⟨11⟩—*USP Crotamiton RS.*

Identification—

A: *Infrared Absorption* ⟨197F⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 20 µg per mL.

Medium: cyclohexane.

C: To about 10 mL of a saturated solution in water add a few drops of potassium permanganate TS: a brown color is produced, and a brown precipitate is formed on standing.

Specific gravity ⟨841⟩: between 1.008 and 1.011 at 20°.

Refractive index ⟨831⟩: between 1.540 and 1.543 at 20°.

Residue on ignition ⟨281⟩: not more than 0.1%.

Bound halogen—Place 4 drops in a 3-mm (ID) test tube, and add calcium oxide to a height of 1 cm. Heat the tube in a flame, starting from the top, until the reaction is complete, then ignite for a short time. Transfer the contents to a beaker containing 10 mL of water, acidify with nitric acid, and filter. To the filtrate add 0.2 mL of silver nitrate solution (1 in 60): any opalescence obtained is not more than that obtained from a blank solution treated in the same manner.

Assay—Transfer about 50 mg of Crotamiton, accurately weighed, to a 100-mL volumetric flask, add cyclohexane to volume, and mix. Transfer 10.0 mL of this solution to a 250-mL volumetric flask, dilute with cyclohexane to volume, and mix. Determine the absorbance of this solution and of a solution of USP Crotamiton RS in the same medium having a known concentration of about 20 µg per mL in 1-cm cells at the wavelength of maximum absorbance at about 242 nm, with a suitable spectrophotometer, using cyclohexane as the blank. Calculate the quantity, in mg, of $C_{13}H_{17}NO$ in the Crotamiton taken by the formula:

$$2.5C(A_U/A_S),$$

in which C is the concentration, in µg per mL, of USP Crotamiton RS in the Standard solution; and A_U and A_S are the absorbances of the assay solution and the Standard solution, respectively.

Crotamiton Cream

» Crotamiton Cream contains not less than 93.0 percent and not more than 107.0 percent of the labeled amount of $C_{13}H_{17}NO$.

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

USP Reference standards ⟨11⟩—*USP Crotamiton RS.*

Identification—The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation*, both relative to the internal standard, as obtained in the *Assay preparation*.

Minimum fill ⟨755⟩: meets the requirements.

Assay—

Internal standard solution—Dissolve butyl benzoate in methanol to obtain a solution containing about 17.5 mg per mL.

Mobile phase—Prepare a suitable degassed and filtered mixture of acetonitrile and water (3 : 2).

Standard solution—Dissolve a suitable quantity of USP Crotamiton RS, accurately weighed, in methanol to obtain a solution having a known concentration of about 1 mg per mL.

Standard preparation—Pipet 10 mL of *Standard solution* and 5 mL of *Internal standard solution* into a 50-mL volumetric flask, dilute with methanol to volume, and mix.

Assay preparation—Transfer an accurately weighed portion of Crotamiton Cream, equivalent to about 50 mg of crotamiton, to a tared 50-mL volumetric flask. Add about 25 mL of methanol, and shake and sonicate to disperse the cream. Dilute with methanol to volume, and mix. Filter about 20 mL through moderately retentive filter paper. Pipet 10 mL of the clear filtrate and 5 mL of *Internal standard solution* into a 50-mL volumetric flask, dilute with methanol to volume, and mix.

Procedure—Inject equal volumes of the *Standard preparation* and the *Assay preparation* into a liquid chromatograph (see *Chromatography* ⟨621⟩) equipped with a 254-nm detector and a 4.6-mm × 25-cm stainless steel column that contains packing L1. In a suitable

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Uniformity of dosage units ⟨905⟩: meet the requirements.

Assay—

Mobile phase—Prepare a suitable filtered and degassed mixture of water, acetonitrile, methanol, and methanesulfonic acid (48 : 28 : 24 : 0.2), and adjust with diethylamine to a pH of 3.6. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Cyclobenzaprine Hydrochloride RS in 0.1 N hydrochloric acid, and dilute quantitatively, and stepwise if necessary, with 0.1 N hydrochloric acid to obtain a solution having a known concentration of about 0.05 mg per mL.

Assay preparation—Weigh and finely powder not fewer than 20 Tablets. Transfer an accurately weighed portion of the powder, equivalent to about 10 mg of cyclobenzaprine hydrochloride, to a 200-mL volumetric flask, add 150 mL of 0.1 N hydrochloric acid, and shake by mechanical means for 30 minutes. Dilute with 0.1 N hydrochloric acid to volume, mix, and filter.

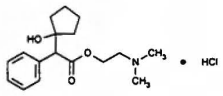
Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 290-nm detector and a 4.6-mm × 10-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed for *Procedure:* the capacity factor, k', for the analyte peak is not less than 2.0; the column efficiency determined from the analyte peak is not less than 1000 theoretical plates; the tailing factor for the analyte peak is not more than 2; 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 major peaks. Calculate the quantity, in mg, of $C_{20}H_{21}N \cdot HCl$ in the portion of Tablets taken by the formula:

$$200C(r_U/r_S),$$

in which C is the concentration, in mg per mL, of USP Cyclobenzaprine Hydrochloride 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.

Cyclopentolate Hydrochloride



$C_{17}H_{25}NO_3 \cdot HCl$ 327.85

Benzeneacetic acid, α-(1-hydroxycyclopentyl)-, 2-(dimethylamino)-ethyl ester, hydrochloride, (±)-.

2-(Dimethylamino)ethyl (±)-1-hydroxy-α-phenylcyclopentaneacetate hydrochloride [*5870-29-1*].

» Cyclopentolate Hydrochloride contains not less than 98.0 percent and not more than 102.0 percent of $C_{17}H_{25}NO_3 \cdot HCl$, calculated on the dried basis.

Packaging and storage—Preserve in tight containers, and store in a cold place.

USP Reference standards ⟨11⟩—*USP Cyclopentolate Hydrochloride RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: A solution (1 in 500) responds to the tests for *Chloride* ⟨191⟩.

pH ⟨791⟩: between 4.5 and 5.5, in a solution (1 in 100).

Loss on drying ⟨731⟩—Dry it at 105° for 4 hours: it loses not more than 0.5% of its weight.

Residue on ignition ⟨281⟩: not more than 0.05%.

Chromatographic purity—

Buffer solution, Mobile phase, and *Chromatographic system*—Prepare as directed under *Assay.*

Test preparation—Use the *Assay preparation.*

Procedure—Inject a volume (about 20 µL) of the *Test preparation* into the chromatograph, record the chromatogram obtained for a period of not less than twice the retention time of cyclopentolate, and measure the peak responses. Calculate the percentage of each peak, other than the solvent peak and the cyclopentolate peak, in the specimen of Cyclopentolate Hydrochloride taken by the same formula:

$$100r_i/r_t,$$

in which r_i is the response of each peak and r_t is the sum of the responses of all of the peaks, excluding that of the solvent peak: not more than 1.0% individual impurity and not more than 2.0% total impurities are found.

Assay—

Buffer solution—Dissolve 660 mg of dibasic ammonium phosphate in 1000 mL of water. Adjust with phosphoric acid to a pH of 3.0 ± 0.1, and mix.

Mobile phase—Prepare a suitable filtered and degassed mixture of acetonitrile and *Buffer solution* (7 : 3). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Cyclopentolate Hydrochloride RS in water, dilute quantitatively, and stepwise if necessary, with water, and mix to obtain a solution having a known concentration of about 0.1 mg per mL.

Assay preparation—Transfer about 100 mg of Cyclopentolate Hydrochloride, accurately weighed, to a 100-mL volumetric flask, dilute with water to volume, and mix. Transfer 5.0 mL of this solution to a 50-mL volumetric flask, dilute with water to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 220-nm detector and a 4.6-mm × 15-cm column that contains packing L15. The flow rate is about 2 mL per minute. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the column efficiency determined from the analyte peak is not less than 3000 theoretical plates, the tailing factor for the analyte peak is not more than 2.0, and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 20 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 $C_{17}H_{25}NO_3 \cdot HCl$ in the portion of Cyclopentolate Hydrochloride taken by the formula:

$$1000C(r_U/r_S),$$

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

Cyclopentolate Hydrochloride Ophthalmic Solution

» Cyclopentolate Hydrochloride Ophthalmic Solution is a sterile, aqueous solution of Cyclopentolate Hydrochloride. It may contain suitable buffers and other additives. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of $C_{17}H_{25}NO_3 \cdot HCl$.

Packaging and storage—Preserve in tight containers, and store at controlled room temperature.

USP Reference standards ⟨11⟩—*USP Cyclopentolate Hydrochloride RS.*

Identification—Place in a 125-mL separator a volume of Ophthalmic Solution, equivalent to about 50 mg of cyclopentolate hydrochloride, and place in a second separator about 50 mg of USP Cyclopentolate Hydrochloride RS dissolved in 5 mL of water. Treat each solution as

follows. Add 1 g of potassium carbonate, and extract with two 10-mL portions of ether. Pass the ether extracts through ether-washed filter paper, collect the filtrate in a small beaker, and evaporate to dryness: the residue so obtained responds to *Identification* test *A* under *Cyclopentolate Hydrochloride*.

Sterility ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 3.0 and 5.5.

Assay—

Buffer solution, Mobile phase, Standard preparation, and *Chromatographic system*—Proceed as directed in the *Assay* under *Cyclopentolate Hydrochloride*.

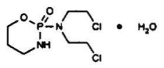
Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 10 mg of cyclopentolate hydrochloride, to a 100-mL volumetric flask, dilute with water to volume, and mix.

Procedure—Proceed as directed in the *Assay* under *Cyclopentolate Hydrochloride*. Calculate the quantity, in mg, of cyclopentolate hydrochloride ($C_{17}H_{25}NO_3 \cdot HCl$) in each mL of the Ophthalmic Solution taken by the formula:

$$100(C/V)(r_U/r_S),$$

in which V is the volume, in mL, of Ophthalmic Solution taken, and the other terms are as defined therein.

Cyclophosphamide



$C_7H_{15}Cl_2N_2O_2P \cdot H_2O$ 279.10

2*H*-1,3,2-Oxazaphosphorin-2-amine, *N*,*N*-bis(2-chloroethyl)tetrahydro-, 2-oxide, monohydrate, (±).

(±)-2-[Bis(2-chloroethyl)amino]tetrahydro-2*H*-1,3,2-oxazaphosphorine 2-oxide monohydrate [*6055-19-2*].

Anhydrous 261.09 [*50-18-0*].

» Cyclophosphamide contains not less than 97.0 percent and not more than 103.0 percent of $C_7H_{15}Cl_2N_2O_2P$, calculated on the anhydrous basis.

Caution—Great care should be taken in handling Cyclophosphamide, as it is a potent cytotoxic agent.

Packaging and storage—Preserve in tight containers, at a temperature between 2° and 30°.

USP Reference standards ⟨11⟩—*USP Cyclophosphamide RS*.

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation*, both relative to the internal standard, as obtained in the *Assay*.

pH ⟨791⟩: between 3.9 and 7.1, in a solution (1 in 100), determined 30 minutes after its preparation.

Water, *Method I* ⟨921⟩: between 5.7% and 6.8%.

Heavy metals ⟨231⟩—Dissolve 1.0 g in 25 mL of water, and filter if necessary: the limit is 0.002%.

Assay—

Mobile phase—Prepare a suitable, degassed solution of water and acetonitrile (70 : 30).

Internal standard solution—Dissolve about 185 mg of ethylparaben in 250 mL of alcohol in a 1000-mL volumetric flask, dilute with water to volume, and mix.

Standard preparation—Transfer an accurately weighed quantity of USP Cyclophosphamide RS, equivalent to about 25 mg of anhydrous cyclophosphamide, to a 50-mL volumetric flask, add about 25 mL of water, and shake to dissolve the USP Reference Standard. Add 5.0 mL of *Internal standard solution*, dilute with water to volume, and mix to obtain a *Standard preparation* having a known concentration of about 0.5 mg of anhydrous cyclophosphamide per mL.

Assay preparation—Transfer an accurately weighed quantity of Cyclophosphamide, equivalent to about 200 mg of anhydrous cyclophosphamide, to a 200-mL volumetric flask, add about 50 mL of water, shake for about 5 minutes, dilute with water to volume, and mix. Pipet 25 mL of this solution and 5 mL of *Internal standard solution* into a 50-mL volumetric flask, dilute with water to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 195-nm detector and a 3.9-mm × 30-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. Chromatograph six replicate injections of the *Standard preparation*, and record the peak responses as directed for *Procedure*: the relative standard deviation is not more than 2%, and the resolution factor between cyclophosphamide and ethylparaben is not less than 2.

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. The relative retention times are about 0.7 for cyclophosphamide and 1.0 for ethylparaben. Calculate the quantity, in mg, of $C_7H_{15}Cl_2N_2O_2P$ in the Cyclophosphamide taken by the formula:

$$400C(R_U/R_S),$$

in which C is the concentration, in mg per mL, of anhydrous cyclophosphamide in the *Standard preparation*, as determined from the concentration of USP Cyclophosphamide RS corrected for moisture content by a titrimetric water determination; and R_U and R_S are the ratios of the peak responses of cyclophosphamide to those of ethylparaben in the *Assay preparation* and the *Standard preparation*, respectively.

Cyclophosphamide for Injection

» Cyclophosphamide for Injection is a sterile mixture of Cyclophosphamide with or without a suitable diluent. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of anhydrous cyclophosphamide ($C_7H_{15}Cl_2N_2O_2P$).

Packaging and storage—Preserve in *Containers for Sterile Solids* as described under *Injections* ⟨1⟩. Storage at a temperature not exceeding 25° is recommended. It will withstand brief exposure to temperatures up to 30°, but is to be protected from temperatures above 30°.

USP Reference standards ⟨11⟩—*USP Cyclophosphamide RS. USP Endotoxin RS.*

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

Identification—

A: It responds to the *Thin-layer Chromatographic Identification Test* ⟨201⟩, a solution of it in chloroform, equivalent to 20 mg of cyclophosphamide per mL, filtered if necessary, being used as the test solution. Apply 5 µL of the test solution and the Standard solution, use a solvent system consisting of a mixture of chloroform, methanol, and ammonium hydroxide (75 : 20 : 5), and visualize the spots by placing the plate in an iodine chamber.

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation*, both relative to the internal standard, as obtained in the *Assay*.

Bacterial endotoxins ⟨85⟩—It contains not more than 0.20 USP Endotoxin Unit per mg of cyclophosphamide.

pH ⟨791⟩: between 3.0 and 9.0, but the range does not exceed 3 pH units, in a solution containing the equivalent of 20 mg of anhydrous cyclophosphamide per mL, determined 30 minutes after its preparation.

USP 28

Other requirements—I
⟨71⟩, *Uniformity of D*
Injections ⟨1⟩.

Assay—

Mobile phase, Interna
tion—Prepare as directed

Assay preparation—A
mide for Injection, equ
cyclophosphamide, and p
the *Assay* under *Cycloph*

Chromatographic sys
graphic system in the *As*

Procedure—Proceed a
Cyclophosphamide. Calc
in the portion of Cyclo
formula:

in which the terms are as

Cyclophospha

» Cyclophosphamide
percent and not more
amount of anh
($C_7H_{15}Cl_2N_2O_2P$).

Packaging and storage—
temperature not exceedi
withstand brief exposure
protected from temperatur

USP Reference standard

Identification—

A: Extract a portion
about 50 mg of cyclophos
about 2 mL of the chlorofo
potassium bromide, evapor
last trace of solvent in a s
prepare a potassium bromi
of the potassium bromide
between 6.5 and 14 µm, o
similar preparation of USP

B: The retention time
the *Assay preparation* corre
Standard preparation, as o

Disintegration ⟨701⟩: 3
Uncoated Tablets.

Uniformity of dosage unit

Procedure for content un

Perchloric acid solution
water, and dilute with wate

4-(p-Nitrobenzyl)pyridine
benzyl)pyridine in 200 mL

Sodium hydroxide solutio
1000 mL of diluted alcohol

Procedure—Place 1 Tabl
that the final concentration
about two-thirds full of wa
disintegrated, dilute with w
first 10 mL of the filtrate. P
tubes 2.0 mL of the filtrate,
2.0 mL of the Standard solut
weighed quantity of USP
diluting quantitatively and
having a known concentrati
tube as follows. Add 0.7 m
heat at 95° for 10 minutes.
mix, add 1.6 mL of *4-(p-Nitr*
at 95° for 10 minutes. Co
solution, and mix. Within 4 n
solutions in 1-cm cells at the

Remove the plate from the developing tank, mark the solvent front, and allow the spots to dry. Spray the plate with dilute sulfuric acid (1 in 2), and heat at 105° until brown or black spots appear: the R_F value of the principal spot obtained from the test specimen corresponds to that obtained from the Reference Standard.

Bacterial endotoxins ⟨85⟩—It contains not more than 31.3 USP Endotoxin Units per mg of dexamethasone phosphate.

pH ⟨791⟩: between 7.0 and 8.5.

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

Assay—

Mobile phase—Prepare a suitable degassed solution of 0.01 M monobasic potassium phosphate in a mixture of methanol and water (1 : 1) which, at ambient temperature and at a flow rate of about 1.6 mL per minute, gives a retention time of about 5 minutes for dexamethasone phosphate.

Standard preparation—[NOTE—Prepare this solution at the time of use.] Dissolve an accurately weighed quantity of USP Dexamethasone Phosphate RS in *Mobile phase* to obtain a solution having a known concentration of about 80 µg per mL.

Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 8 mg of dexamethasone phosphate, to a 100-mL volumetric flask. Dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4-mm × 30-cm column that contains packing L1. Chromatograph five replicate injections of the *Standard preparation*, and record the peak responses as directed under *Procedure:* the relative standard deviation is not more than 1.5%.

Procedure—By means of a suitable sampling valve, separately inject equal volumes (about 20 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 $C_{22}H_{30}FO_8P$ in each mL of the Injection taken by the formula:

$$0.1(C/V)(r_U/r_S),$$

in which C is the concentration, in µg per mL, of USP Dexamethasone Phosphate RS in the *Standard preparation*, V is the volume, in mL, of Injection taken, and r_U and r_S are the peak responses at equivalent retention times obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Dexamethasone Sodium Phosphate Ophthalmic Ointment

» Dexamethasone Sodium Phosphate Ophthalmic Ointment is a sterile ointment containing an amount of dexamethasone sodium phosphate ($C_{22}H_{28}FNa_2O_8P$) equivalent to not less than 90.0 percent and not more than 115.0 percent of the labeled amount of dexamethasone phosphate ($C_{22}H_{30}FO_8P$).

Packaging and storage—Preserve in collapsible ophthalmic ointment tubes.

USP Reference standards ⟨11⟩—*USP Dexamethasone RS. USP Dexamethasone Phosphate RS.*

Identification—The *Assay preparation*, prepared as directed in the *Assay*, responds to the *Identification* test under *Dexamethasone Sodium Phosphate Cream.*

Minimum fill ⟨755⟩: meets the requirements.

Sterility ⟨71⟩: meets the requirements.

Metal particles—It meets the requirements of the test for *Metal Particles in Ophthalmic Ointments* ⟨751⟩.

Assay—

Alcohol-aqueous phosphate buffer, 0.05 M Phosphate buffer, Mobile phase, Standard preparation, and *Chromatographic system*—Prepare as directed in the *Assay* under *Dexamethasone Sodium Phosphate Cream.*

Assay preparation—Using an accurately weighed portion of Ophthalmic Ointment, prepare as directed in the *Assay* under *Dexamethasone Sodium Phosphate Cream.*

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Dexamethasone Sodium Phosphate Cream.* Calculate the quantity, in mg, of $C_{22}H_{30}FO_8P$ in the portion of Ophthalmic Ointment taken by the formula:

$$0.1C(r_U/r_S).$$

Dexamethasone Sodium Phosphate Ophthalmic Solution

» Dexamethasone Sodium Phosphate Ophthalmic Solution is a sterile, aqueous solution of Dexamethasone Sodium Phosphate. It contains an amount of dexamethasone sodium phosphate ($C_{22}H_{28}FNa_2O_8P$) equivalent to not less than 90.0 percent and not more than 115.0 percent of the labeled amount of dexamethasone phosphate ($C_{22}H_{30}FO_8P$).

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

USP Reference standards ⟨11⟩—*USP Dexamethasone RS. USP Dexamethasone Phosphate RS.*

Identification—The *Assay preparation*, prepared as directed in the *Assay*, responds to the *Identification* test under *Dexamethasone Sodium Phosphate Cream.*

pH ⟨791⟩: between 6.6 and 7.8.

Sterility ⟨71⟩: meets the requirements.

Assay—

Mobile phase, Standard preparation, and *Chromatographic system*—Prepare as directed in the *Assay* under *Dexamethasone Sodium Phosphate Injection.*

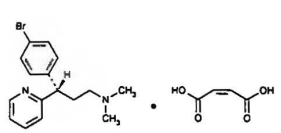
Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 8 mg of dexamethasone phosphate, to a 100-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Dexamethasone Sodium Phosphate Injection.* Calculate the quantity, in mg, of $C_{22}H_{30}FO_8P$ in each mL of the Ophthalmic Solution taken by the formula:

$$0.1(C/V)(r_U/r_S),$$

in which V is the volume, in mL, of Ophthalmic Solution taken.

Dexbrompheniramine Maleate



$C_{16}H_{19}BrN_2 \cdot C_4H_4O_4$ 435.32

2-Pyridinepropanamine, γ-(4-bromophenyl)-*N*,*N*-dimethyl-, (*S*)-, (*Z*)-2-butenedioate (1 : 1).

(+)-2-[*p*-Bromo-α-[2-(dimethylamino)ethyl]benzyl]pyridine maleate (1 : 1) [*2391-03-9*].

» Dexbromphen
98.0 percent ar
$C_{16}H_{19}BrN_2 \cdot C_4H$

Packaging and stor
USP Reference stan
ate RS.

Identification—
A: *Infrared Absc*
B: *Ultraviolet Ab*
Solution: 35 µg p
Medium: methan
Absorptivities at 2
differ by more than 3.

Specific rotation ⟨78
Test solution: 50 r

Loss on drying ⟨731⟩
than 0.5% of its weigh

Residue on ignition ⟨

Related compounds—
Test solution—Disso
Maleate in 5 mL of me
Chromatographic sy
chromatograph is equip
mm × 1.2-m glass co
S1AB. The column tem
injection port and detec
250°. The carrier gas is
a retention time of 6 to
the *Test solution,* record
area as directed unde
dexbrompheniramine m
Procedure—Inject a v
the chromatograph. Rec
less than twice the reten
and measure the areas
extraneous peaks (excep
observed) does not exce

Organic volatile impur
ments.

Assay—Dissolve about
accurately weighed, in 5
crystal violet TS, and titra
endpoint. Perform a blan
correction. Each mL of 0
mg of $C_{16}H_{19}BrN_2 \cdot C_4H_4C$

Dexbrompheniramine Pseudoephedrin

» Dexbrompheniramine
Sulfate Oral Solution
and not more than 11
of dexbrompheniramin
and pseudoephedrine s

USP Reference standards
ate RS. USP Pseudoephedri

Identification—
A: The retention time of
maleate in the chromatogram
that in the chromatogram of
the *Assay.*
B: The retention time
sulfate in the chromatogram
that in the chromatogram of t
the *Assay.*
C: A solution of it respo
D: Transfer a volume of
of dexbrompheniramine male

ick Test

Test conforms to the
ıg biologics (650.1 to
is a sterile solution of
ducts of growth of the
'rium diphtheriae) of
ot less than 400 MLD
ıL or 400,000 MRD
er mL in guinea pigs.
s of the U.S. Standard
tested in guinea pigs.

emperature between 2° and

not later than 1 year after
orage (5°, 1 year).

Toxoids

Adsorbed conforms to
cerning biologics (see
ıspension prepared by
or adsorbed diphtheria
:tanus toxoid, and an
ı toxoids are used. The
e proportions of the
ı immunizing dose of
scribed in the labeling,
equirements for those
than 0.02 percent of

emperature between 2° and

not later than 2 years after
orage (5°, 1 year).

well shaken before use and

e

CH_3 • HCl

l-hydroxy-2-(methylami-
ochloride, (±)-.
ethyl]benzyl alcohol 3,4-
93-8].

ıins not less than 98.5
101.5 percent of
ıe dried basis.

Packaging and storage—Preserve in tight containers.

USP Reference standards ⟨11⟩—*USP Dipivefrin Hydrochloride RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation*, as obtained in the *Assay.*

C: A solution (1 in 100) meets the requirements of the tests for *Chloride* ⟨191⟩.

Melting range ⟨741⟩: between 155° and 165°, but the range between beginning and end of melting does not exceed 2°.

Loss on drying ⟨731⟩—Dry it in a suitable vacuum drying tube over phosphorus pentoxide at 60° for 6 hours: it loses not more than 1.0% of its weight.

Residue on ignition ⟨281⟩: not more than 0.3%.

Heavy metals, *Method I* ⟨231⟩: not more than 0.0015%.

Iron ⟨241⟩: not more than 5 ppm.

Standard iron solution—Use the *Standard Iron Solution* prepared as directed under *Iron* ⟨241⟩.

Hydroxylamine solution—Dissolve 5 g of hydroxylamine hydrochloride in 50 mL of water.

Triazine solution—Dissolve 125 mg of 2,4,6-tri-(2-pyridyl)-*S*-triazine in 100 mL of methanol.

Standard solution—Into a 50-mL color-comparison tube pipet 1 mL of *Standard iron solution*, add 42.0 mL of water, and mix.

Test solution—Into a 50-mL color-comparison tube add 2.0 g of Dipivefrin Hydrochloride, 43.0 mL of water, and mix.

Procedure—To each of the tubes containing the *Standard solution* and the *Test solution,* add 5.0 mL of *Hydroxylamine solution,* 2.0 mL of *Triazine solution,* and mix: the color of the solution from the *Test solution* is not darker than that of the solution from the *Standard solution.*

Assay—

Mobile phase—Prepare a mixture of acetonitrile, 0.014 M sodium dodecyl sulfate, and glacial acetic acid (24 : 15 : 1).

Standard preparation—Dissolve a suitable quantity of USP Dipivefrin Hydrochloride RS, accurately weighed, in 0.0015 N hydrochloric acid to obtain a solution having a known concentration of about 5 mg per mL.

Assay preparation—Prepare as directed for *Standard preparation,* using 500 mg of Dipivefrin Hydrochloride, accurately weighed, in place of the Reference Standard.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4-mm × 30-cm column that contains packing L1. The flow rate is about 2 mL per minute. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the column efficiency is not less than 500 theoretical plates; the tailing factor for the major peak is not more than 1.5; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph by means of a suitable microsyringe or sampling valve, record the chromatograms, and measure the responses for the major peaks. Calculate the quantity, in mg, of $C_{19}H_{29}NO_5 \cdot HCl$ in the portion of Dipivefrin Hydrochloride taken by the formula:

$$100C(r_U / r_S),$$

in which C is the concentration, in mg per mL, of USP Dipivefrin Hydrochloride 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.

Dipivefrin Hydrochloride Ophthalmic Solution

» Dipivefrin Hydrochloride Ophthalmic Solution is a sterile, aqueous solution of Dipivefrin Hydrochloride. It contains not less than 90.0 percent and not more than 115.0 percent of the labeled amount of $C_{19}H_{29}NO_5 \cdot HCl$. It contains a suitable antimicrobial agent and may contain stabilizers, suitable buffers, and chelating agents.

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

USP Reference standards ⟨11⟩—*USP Dipivefrin Hydrochloride RS.*

Identification—It meets the requirements for *Identification* test *B* under *Dipivefrin Hydrochloride.*

Sterility Tests ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 2.5 and 3.5.

Assay—

Mobile phase and *Chromatographic system*—Prepare as directed in the *Assay* under *Dipivefrin Hydrochloride.*

Standard preparation—Dissolve a suitable quantity of USP Dipivefrin Hydrochloride RS, accurately weighed, in 0.0015 N hydrochloric acid to obtain a solution having a known concentration of about 1 mg per mL.

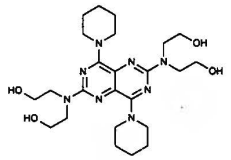
Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 25 mg of dipivefrin hydrochloride, to a 25-mL volumetric flask, dilute with 0.0015 N hydrochloric acid to volume, if necessary, and mix.

Procedure—Proceed as directed in the *Assay* under *Dipivefrin Hydrochloride.* Calculate the quantity, in mg, of $C_{19}H_{29}NO_5 \cdot HCl$ in each mL of the Ophthalmic Solution taken by the formula:

$$(25C / V)(r_U / r_S),$$

in which C is the concentration, in mg per mL, of USP Dipivefrin Hydrochloride RS in the *Standard preparation;* V is the volume, in mL, of Ophthalmic Solution taken; and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Dipyridamole



$C_{24}H_{40}N_8O_4$ 504.63

Ethanol, 2,2′,2″,2‴-[4,8-di-1-piperidinylpyrimido[5,4-*d*]pyrimidine-2,6-diyl)dinitrilo]tetrakis-.

2,2′,2″,2‴-[4,8-Dipiperidinopyrimido[5,4-*d*]pyrimidine-2,6-diyl)dinitrilo]tetraethanol [*58-32-2*].

» Dipyridamole contains not less than 98.0 percent and not more than 102.0 percent of $C_{24}H_{40}N_8O_4$, calculated on the dried basis.

Packaging and storage—Preserve in tight, light-resistant containers. Store at room temperature.

USP Reference standards ⟨11⟩—*USP Dipyridamole RS.*

Identification, *Infrared Absorption* ⟨197K⟩.

Melting range ⟨741⟩: between 162° and 168°, but the range between beginning and end of melting does not exceed 2°.

Loss on drying ⟨731⟩—Dry it at 105° for 3 hours: it loses not more than 0.2% of its weight.

Chloride—Dissolve 500 mg in 5 mL of alcohol and 2 mL of 2 N nitric acid, and add 1 mL of silver nitrate TS: no turbidity or precipitate is produced.

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Emedastine Ophthalmic Solution

» Emedastine Ophthalmic Solution is a sterile, aqueous solution containing an amount of Emedastine Difumarate equivalent to not less than 90.0 percent and not more than 110.0 percent of the labeled amount of emedastine ($C_{17}H_{26}N_4O$).

Packaging and storage—Preserve in tight, light-resistant containers, in a refrigerator or at controlled room temperature.

USP Reference standards ⟨11⟩—*USP Emedastine Difumarate RS.*

Identification—The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

Sterility ⟨71⟩—It meets the requirements when tested as directed for *Membrane Filtration* under *Test for Sterility of the Product to be Examined.*

pH ⟨791⟩: between 5.0 and 8.0.

Assay—

Buffer solution—Dissolve 13.8 g of monobasic sodium phosphate and 10 mL of triethylamine in 800 mL of water. Adjust with phosphoric acid to a pH of 5.7, dilute with water to 1000 mL, and mix.

Mobile phase—Prepare a filtered and degassed mixture of *Buffer solution* and acetonitrile (83 : 17). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Emedastine Difumarate RS in *Mobile phase* to obtain a solution having a known concentration of about 0.057 mg of emedastine per mL.

System suitability solution—Add 50 µL of 30 percent hydrogen peroxide to 2 mL of *Standard preparation,* and heat at 100° for 30 minutes. Add another 2 mL of *Standard preparation,* mix, and use immediately.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution into a suitable volumetric flask to obtain a solution having a known concentration of about 0.057 mg of emedastine per mL.

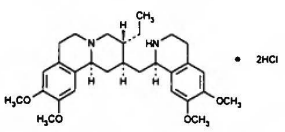
Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 280-nm detector and a 3.9-mm × 15-cm column that contains packing L7. The flow rate is about 1.0 mL per minute. Chromatograph the *System suitability solution,* and record the peak responses as directed for *Procedure:* the relative retention times are about 1.0 for emedastine and 1.2 for emedastine *N*-oxide; the resolution, *R,* between emedastine and emedastine *N*-oxide is not less than 1.5; the column efficiency determined from the emedastine peak is not less than 1000 theoretical plates; and the tailing factor is not more than 2.0. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the peak responses for emedastine. Calculate the quantity, in mg, of emedastine ($C_{17}H_{26}N_4O$) in each mL of the Ophthalmic Solution taken by the formula:

$$(302.42/534.57)C(V_1/V_2)(r_U/r_S),$$

in which 302.42 and 534.57 are the molecular weights of emedastine and emedastine difumarate, respectively; C is the concentration, in mg per mL, of USP Emedastine Difumarate RS in the *Standard preparation;* V_1 is the volume, in mL, of the volumetric flask used to prepare the *Assay preparation;* V_2 is the volume, in mL, of Ophthalmic Solution taken; and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Emetine Hydrochloride



$C_{29}H_{40}N_2O_4 \cdot 2HCl$ 553.56

Emetan, 6′,7′,10,11-tetramethoxy-, dihydrochloride.

Emetine dihydrochloride [*316-42-7*].

» Emetine Hydrochloride is the hydrochloride of an alkaloid obtained from Ipecac, or prepared by methylation of cephaeline, or prepared synthetically. It contains not less than 98.0 percent and not more than 101.5 percent of $C_{29}H_{40}N_2O_4 \cdot 2HCl$, calculated on the anhydrous basis.

Packaging and storage—Preserve in tight, light-resistant containers. Store at 25°, excursions permitted between 15° and 30°.

USP Reference standards ⟨11⟩—*USP Cephaeline Hydrobromide RS. USP Emetine Hydrochloride RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 50 µg per mL.

Medium: 0.5 N sulfuric acid.

C: A solution (1 in 20) responds to the tests for *Chloride* ⟨191⟩.

Water, *Method I* ⟨921⟩: between 15.0% and 19.0%.

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

Acidity—Dissolve 100 mg in 10 mL of water, add 1 drop of methyl red TS, and titrate with 0.020 N sodium hydroxide: not more than 0.5 mL is required to produce a yellow color.

Limit of cephaeline—[NOTE—Conduct this test in subdued light until after the chromatogram has been completely developed.]

Standard preparation—Dissolve 23 mg of USP Cephaeline Hydrobromide RS in 100.0 mL of methanol.

Test preparation—Dissolve 100 mg of Emetine Hydrochloride in 10.0 mL of methanol.

Spray reagent—Dissolve 300 mg of *p*-nitroaniline in 25 mL of 2 N hydrochloric acid, and cool to about 4°. Slowly add 5 mL of sodium nitrite solution (1 in 25), maintaining the temperature at about 4°. Freshly prepare the solution for each test.

Procedure—Apply 10-µL portions of the *Standard preparation* and the *Test preparation,* respectively, to a suitable thin-layer chromatographic plate coated with a 0.25-mm layer of chromatographic silica gel. Place the plate in a chromatographic tank containing a mixture of 9 volumes of chloroform and 1 volume of diethylamine, and develop the chromatogram until the solvent front has moved about 12 cm. Remove the plate from the tank, and allow the plate to air-dry for 20 minutes. Spray the dried plate with 2.5 N sodium hydroxide solution, and dry at 50° for 5 minutes. Then spray the plate with *Spray reagent:* any cephaeline spot from the *Test preparation* is not larger or more intense than that produced by the *Standard preparation* (2%).

Assay—Dissolve about 150 mg of Emetine Hydrochloride, accurately weighed, in 5 mL of glacial acetic acid, warming, if necessary. Allow the solution to cool, add 10 mL of dioxane, 5 mL of mercuric acetate TS, and 3 drops of crystal violet TS, and titrate with 0.1 N perchloric acid VS. Perform a blank determination, and make any necessary correction. Each mL of 0.1 N perchloric acid is equivalent to 27.68 mg of $C_{29}H_{40}N_2O_4 \cdot 2HCl$.

Identification—It meets the requirements for the *Identification* test under *Epinephrine Nasal Solution*.

Sterility ⟨71⟩: meets the requirements.

Assay—Pipet 10 mL of Inhalation Solution into a 125-mL separator, and extract the solution with two 10-mL portions of chloroform. Proceed as directed in the *Assay* under *Epinephrine Nasal Solution*, beginning with "Rinse the stopper and mouth of the separator," but use for the acetylation 1.05 g of sodium bicarbonate and 0.50 mL of acetic anhydride, and extract the acetylated product with six 15-mL portions of chloroform instead of the 25-mL portions specified therein, and use 15.0 mL of chloroform instead of 5.0 mL in the determination of the specific rotation.

Epinephrine Nasal Solution

» Epinephrine Nasal Solution is a solution of Epinephrine in Purified Water prepared with the aid of Hydrochloric Acid. It contains, in each 100 mL, not less than 90 mg and not more than 115 mg of $C_9H_{13}NO_3$.

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

Labeling—The label indicates that the Nasal Solution is not to be used if its color is pinkish or darker than slightly yellow or if it contains a precipitate.

Color and clarity—Using the Nasal Solution as the *Test solution*, proceed as directed for *Color and clarity* under *Epinephrine Injection*.

Identification—To 5 mL of pH 4.0 acid phthalate buffer (see *Buffer Solutions* in the section *Reagents, Indicators, and Solutions*) add 0.5 mL of Nasal Solution and 1.0 mL of 0.1 N iodine. Mix, and allow to stand for 5 minutes. Add 2 mL of sodium thiosulfate solution (1 in 40): a deep red color is produced.

Assay—Pipet 30 mL of Nasal Solution into a 125-mL separator, add 25 mL of chloroform, shake vigorously for 1 minute, allow the liquids to separate, and discard the chloroform. Wash twice more with chloroform, separating and discarding the lower layer as completely as possible each time. Rinse the stopper and mouth of the separator with a few drops of water. Add 0.2 mL of starch TS, then while swirling the separator add iodine and potassium iodide TS dropwise until the blue color formed persists, and immediately add just sufficient 0.1 N sodium thiosulfate to discharge the blue color. [NOTE—Proceed with the assay from this point without delay.]

Add to the liquid in the separator 2.10 g of sodium bicarbonate, preventing it from coming in contact with the mouth of the separator, and swirl until most of the bicarbonate has dissolved. By means of a 1-mL syringe that is not fitted with a needle, rapidly inject 1.0 mL of acetic anhydride directly into the contents of the separator. Immediately insert the stopper in the separator, and shake vigorously until the evolution of carbon dioxide has ceased (7 to 10 minutes), releasing the pressure as necessary through the stopcock. Allow to stand for 5 minutes, and extract the solution with six 25-mL portions of chloroform, filtering each extract through a small pledget of cotton, previously washed with chloroform, into a beaker.

Evaporate the combined chloroform extracts on a steam bath in a current of air to about 3 mL, transfer the residue by means of small portions of chloroform to a tared 50-mL beaker, and heat again to evaporate the solvent completely. Heat further at 105° for 30 minutes, cool in a desiccator, and weigh the residue of triacetylepinephrine. Add 5.0 mL of chloroform, cover the beaker, gently swirl the contents until the residue has completely dissolved, and determine the specific rotation, R, using a 200-mm semimicro polarimeter tube.

Calculate the quantity, in mg, of $C_9H_{13}NO_3$ in the volume of Nasal Solution taken by the formula:

$$(183.20/309.32)(W)(0.5 + 0.5R/93),$$

in which 183.20 and 309.32 are the molecular weights of epinephrine and triacetylepinephrine, respectively; and W is the weight, in mg, and R is the specific rotation (in degrees, without regard to the sign), of the isolated triacetylepinephrine.

Epinephrine Ophthalmic Solution

» Epinephrine Ophthalmic Solution is a sterile, aqueous solution of Epinephrine prepared with the aid of Hydrochloric Acid. It contains not less than 90.0 percent and not more than 115.0 percent of the labeled amount of $C_9H_{13}NO_3$. It contains a suitable antibacterial agent and may contain an anti-oxidant, suitable buffers, and chelating and tonicity-adjusting agents.

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

Labeling—The label indicates that the Ophthalmic Solution is not to be used if its color is pinkish or darker than slightly yellow or if it contains a precipitate.

USP Reference standards ⟨11⟩—*USP Epinephrine Bitartrate RS.*

Color and clarity—Using the Ophthalmic Solution as the *Test solution*, proceed as directed for *Color and clarity* under *Epinephrine Injection*.

Identification—

A: The UV absorption spectrum of the *Assay preparation* prepared as directed in the *Assay* exhibits maxima and minima at the same wavelengths as that of a similar solution of USP Epinephrine Bitartrate RS.

B: A solution (1 in 2) is levorotatory.

Sterility ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 2.2 and 4.5.

Assay—

pH 5.8 Buffer—Mix 1 volume of 1 M dibasic potassium phosphate with 9 volumes of 1 M monobasic potassium phosphate. Adjust by the addition of small volumes of either solution to a pH of 5.80 ± 0.05.

Standard preparation—Dissolve a suitable quantity of USP Epinephrine Bitartrate RS, accurately weighed, in 0.1 N hydrochloric acid to obtain a solution having a known concentration of about 40 µg of epinephrine per mL.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 20 mg of epinephrine, to a 250-mL beaker containing 2.0 mL of *pH 5.8 Buffer*. Add 9 g of chromatographic siliceous earth, and mix. Carefully transfer the mixture to a 45- × 2.2-cm chromatographic tube containing a pledget of glass wool at the bottom, and tap the column gently to effect packing. Dry-wash the beaker with about 1 g of chromatographic siliceous earth, add to the column, and plug the top with a pledget of glass wool. Wash the column with 100 mL of water-washed ether, and discard the eluant. Add 10.0 mL of 0.1 N hydrochloric acid to a 125-mL separator, and place the separator under the column. To about 100 mL of water-washed ether add 1 mL of bis(2-ethylhexyl) phosphoric acid, and elute the column with this solution, collecting the eluate in the separator. Extract the epinephrine into the aqueous acid layer, and carefully transfer the aqueous layer to a 500-mL volumetric flask. Shake the ether layer with two 50-mL portions of 0.1 N hydrochloric acid, add the acidic aqueous extracts to the volumetric flask, dilute with 0.1 N hydrochloric acid to volume, and mix.

Procedure—Concomitantly determine the absorbances of the *Assay preparation* and the *Standard preparation* at the wavelength of maximum absorbance at about 280 nm, with a suitable spectrophotometer, using 0.1 N hydrochloric acid as the blank. Calculate the quantity, in mg, of $C_9H_{13}NO_3$ in each mL of the Ophthalmic Solution taken by the formula:

$$0.5(C/V)(A_U/A_S),$$

in which C is the concentration, in µg per mL, of epinephrine in the *Standard preparation*; V is the volume, in mL, of Ophthalmic Solution taken; and A_U and A_S are the absorbances of the *Assay preparation* and the *Standard preparation*, respectively.

...halation

...ation Aerosol is ...phrine Bitartrate ...ainer. It contains ... than 110.0 percent ... bitartrate ($C_9H_{13}NO_3$) ...

...small, nonreactive, ligh... ...h metered-dose valves ...

...pinephrine Bitartrate RS ...

...eaker, and deliver 3 spray... ...e water, actuating the valv... ...the beaker. Filter, and to... ...drochloric acid (1 in 12)... ...nd for 5 minutes, and add... ...own color is produced. ...by pressing the tip against... ...Cover the spot with 2 or... ...line and 1 volume of acet... ...duced.

...ntire contents: meets ... ...rs under *Aerosols, Nas...* ... *Powder Inhalers* ⟨601⟩, ...

...lution—Prepare as direct...

...curately weighed quantity ...ly prepared sodium bisulfi... ...ively and stepwise with ... ...essary to obtain a solut... ...15 µg per mL. ...nimum recommended do... ...he inhaler as directed. Rin... ...four 5.0-mL portions of ...n (1 in 500), and transfer ...-mL centrifuge tube. Add ...ake vigorously for 1 min... ...ear supernatant as directed...

...s, transfer the *Test prepa...* ...*ion*, and 20.0 mL of wate... ...) µL of *Ferro-citrate soluti...* Concomitantly determine ...tometer, in 5-cm cells, of ...the *Standard preparation*, ...e at about 530 nm, against ... $C_9H_{13}NO_3 \cdot C_4H_6O_6$ contai... ...ula:

$$...A_S),$$

...per mL, of USP Epinephr... ...*on; N* is the number of sp... ...nmended dose; and A_U and ... ...n the *Test preparation* and ...

...phrine Bitartrate Inhalati... ...*rticle size* under *Isoproter...* ...e limits of the test.

...solution—Prepare as direct...

...lirected under *Delivered d...*

...able specimen beaker is ... ...s inside bottom surface ha... ...aerosol valve stem du... ...entrapment and side-of-... ...specimen.] Place 20 mL ...

...loroform in a suitable 100-mL beaker. Prime the valve of ...pinephrine Bitartrate Inhalation Aerosol by alternately shaking ...d firing it 10 times through its oral inhalation actuator. Accurately ...eigh the Aerosol, shake it, and immediately deliver a single spray ...nder the surface of the chloroform, actuating the valve by pressing ...e tip into the indentation in the bottom of the beaker. Raise the ...erosol above the surface of the chloroform, and shake it gently ...reparatory to delivering another spray similarly under the surface of ...e chloroform. Deliver a total of 3 sprays in this manner. Rinse the ...alve stem and ferrule with about 2 mL of chloroform, collecting the ...nsing with the specimen in the beaker. Allow the Aerosol to dry, ...eigh it, and determine the total weight of the 3 sprays. Transfer the ...olution to a centrifuge tube with the aid of two 3-mL portions of ...hloroform, and add 10.0 mL of freshly prepared sodium bisulfite ...olution (1 in 500). Insert the stopper, shake vigorously for 1 minute, ...entrifuge for 5 minutes, and use the clear supernatant as the *Assay ...eparation*.

...Procedure—Transfer 5.0 mL each of the *Standard preparation* and ...e *Assay preparation* to separate test tubes. To each tube add 100 µL ...f *Ferro-citrate solution* and 1.0 mL of *Buffer solution*, and mix. Concomitantly determine the absorbances of the solutions in 1-cm ...ells at the wavelength of maximum absorbance at about 530 nm, ...with a suitable spectrophotometer, using water as the blank. Calculate ...e quantity, in mg, of $C_9H_{13}NO_3 \cdot C_4H_6O_6$ in each mL of the Aerosol ...aken by the formula:

$$(0.01Cd/W)(A_U/A_S),$$

...in which C is the concentration, in µg per mL, of USP Epinephrine ...Bitartrate RS in the *Standard preparation*, d is the density, in g per ...mL, of the Aerosol, determined as directed for d in the *Procedure* in ...e *Assay* under *Isoproterenol Sulfate Inhalation Aerosol*, W is the ...weight, in g, of the specimen taken, and A_U and A_S are the absorbances ...of the solutions from the *Assay preparation* and the *Standard ...reparation*, respectively.

...pinephrine Bitartrate Ophthalmic ...olution

...Epinephrine Bitartrate Ophthalmic Solution is a sterile, ...uffered, aqueous solution of Epinephrine Bitartrate. It ...ontains an amount of epinephrine bitartrate equivalent ...o not less than 90.0 percent and not more than 115.0 ...ercent of the labeled amount of epinephrine ($C_9H_{13}NO_3$). It contains a suitable antibacterial agent ...nd may contain suitable preservatives.

...ckaging and storage—Preserve in small, well-filled, tight, light-...sistant containers.

...abeling—The label indicates that the Ophthalmic Solution is not to ...e used if its color is pinkish or darker than slightly yellow or if it ...contains a precipitate.

...SP Reference standards ⟨11⟩—*USP Epinephrine Bitartrate RS.*

...olor and clarity—Using the Ophthalmic Solution as the *Test ...olution*, proceed as directed for *Color and clarity* under *Epinephrine ...njection*.

...H ⟨791⟩: between 3.0 and 3.8.

...ther requirements—It responds to the *Identification* test under *...pinephrine Nasal Solution*, and meets the requirements under *...terility Tests* ⟨71⟩.

Assay—

pH 2.5 Buffer—Transfer 6.8 g of monobasic potassium phosphate and 1.1 g of sodium 1-octanesulfonate to a 1-liter volumetric flask. Dissolve in water, dilute with water to volume, and mix. Adjust the solution with phosphoric acid to a pH of 2.5 ± 0.1.

Mobile phase—Prepare a filtered and degassed mixture of *pH 2.5 Buffer* and acetonitrile (850 : 150). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Epinephrine Bitartrate RS in water, and dilute quantitatively, and stepwise if necessary, with water to obtain a solution having a known concentration of about 0.1 mg per mL.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, freshly mixed and free from air bubbles, equivalent to about 50 mg of epinephrine bitartrate, to a 500-mL volumetric flask, dilute with water to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 280-nm detector and a 3.2-mm × 25-cm column that contains packing L1. The flow rate is about 1 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed under *Procedure:* the column efficiency determined from the analyte peak is not less than 2000 theoretical plates, the tailing factor for the analyte peak is not more than 2.5, and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 50 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 epinephrine ($C_9H_{13}NO_3$) in each mL of the Ophthalmic Solution taken by the formula:

$$(183.20/333.29)(500C/V)(r_U/r_S),$$

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

Epinephrine Bitartrate for Ophthalmic Solution

» Epinephrine Bitartrate for Ophthalmic Solution is a sterile, dry mixture of Epinephrine Bitartrate and suitable antioxidants, prepared by freeze-drying. It contains an amount of epinephrine bitartrate equivalent to not less than 90.0 percent and not more than 110.0 percent of the labeled amount of epinephrine ($C_9H_{13}NO_3$).

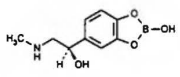
Packaging and storage—Preserve in *Containers for Sterile Solids* as described under *Injections* ⟨1⟩.

Completeness of solution ⟨641⟩—A 100-mg portion dissolves in 5 mL of water to yield a clear solution.

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

Other requirements—A solution of it responds to the *Identification* test under *Epinephrine Nasal Solution*, and meets the requirements of the *Assay* under *Epinephrine Bitartrate Ophthalmic Solution*. It meets also the requirements under *Sterility Tests* ⟨71⟩ and *Uniformity of Dosage Units* ⟨905⟩.

Epinephryl Borate Ophthalmic Solution



$C_9H_{12}BNO_4$ 209.01

1,3,2-Benzodioxaborole-5-methanol, 2-hydroxy-α-[(methylamino)-methyl]-, (*R*)-.

(–)-3,4-Dihydroxy-α-[(methylamino)methyl]benzyl alcohol, cyclic 3,4-ester with boric acid [*5579-16-8*].

» Epinephryl Borate Ophthalmic Solution is a sterile solution in water of Epinephrine as a borate complex. It contains an amount of epinephryl borate ($C_9H_{12}BNO_4$) equivalent to not less than 90.0 percent and not more than 115.0 percent of the labeled amount of epinephrine ($C_9H_{13}NO_3$). It contains a suitable antibacterial agent and one or more suitable preservatives and buffering agents.

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

Labeling—The label indicates that the Ophthalmic Solution is not to be used if its color is pinkish or darker than slightly yellow or if it contains a precipitate.

Color and clarity—

Standard solution—Transfer 2.0 mL of 0.100 N iodine VS to a 500-mL volumetric flask, dilute with water to volume, and mix.

Procedure—Visually examine a portion of the Ophthalmic Solution *(Test solution)* in a suitable clear glass test tube against a white background: it is not pinkish, and it contains no precipitate. If any yellow color is observed in the *Test solution*, concomitantly determine the absorbances of the *Test solution* and the *Standard solution* in 1-cm cells with a suitable spectrophotometer set at 460 nm: the absorbance of the *Test solution* does not exceed that of the *Standard solution*.

Identification—

A: To 5 mL of pH 4.0 acid phthalate buffer (see *Buffer Solutions* in the section *Reagents, Indicators, and Solutions*) add 0.5 mL of Ophthalmic Solution and 1 mL of 0.1 N iodine. Mix, allow to stand for 5 minutes, and add 2 mL of 0.1 N sodium thiosulfate: a deep red color is produced.

B: To 5 mL in a porcelain evaporating dish add 5 drops of sulfuric acid and 5 mL of methanol: the ignited mixture burns with a green-bordered flame.

Sterility ⟨71⟩: meets the requirements.

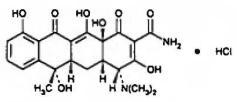
pH ⟨791⟩: between 5.5 and 7.6.

Assay—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 100 mg of epinephrine, to a 250-mL separator. Dilute with water to 30 mL, and adjust with dilute hydrochloric acid (1 in 12) to a pH of 4.0 ± 0.2. Add 25 mL of carbon tetrachloride, shake vigorously for 1 minute, allow the phases to separate, and discard the carbon tetrachloride washing. In the same manner, wash with two additional 25-mL portions of carbon tetrachloride, and discard the washings. Rinse the stopper and the mouth of the separator with 2 to 3 mL of water such that the rinsings enter the separator and combine with the solution under assay. Add 0.2 mL of starch TS, and, while swirling the separator, add iodine and potassium iodide TS dropwise until the blue color persists. Immediately add a volume of 0.1 N sodium thiosulfate just sufficient to discharge the blue color. [NOTE—Proceed with the assay from this point without delay.] Add 2.10 g of sodium bicarbonate through a dry powder funnel to prevent the powder from coming in contact with the mouth of the separator, and swirl to dissolve most of the solid. By means of a syringe fitted with a suitable pipet, rapidly inject 1.0 mL of acetic anhydride directly into the contents of the separator. Swirl the unstoppered separator gently for 3 minutes to allow carbon dioxide to escape. Insert the stopper, and shake gently until the evolution of carbon dioxide has ceased (7 to 10 minutes), releasing the pressure through the stopcock as necessary. Allow to stand for 5 minutes. Extract with six 25-mL portions of chloroform, shaking for 1 minute each time, filtering each extract through a small pledget of chloroform-saturated cotton and collecting the extracts in a 400-mL beaker. Add several glass beads, and evaporate on a steam bath to about 3 mL. With the aid of 15 to 20 mL of chloroform, transfer the residue to a tared 50-mL beaker, and evaporate on the steam bath to dryness. Dry the residue at 105° for 30 minutes, cool in a desiccator, and weigh the triacetylepinephrine so obtained. Transfer 10.0 mL of chloroform to the beaker, and gently swirl to dissolve the residue, dislodging the semisolid residue from the glass surface, if necessary, with a small metal spatula. Determine the angular rotation of the solution in a 100-mm polarimeter tube. Calculate the quantity, in mg, of epinephrine ($C_9H_{13}NO_3$) in the volume of Ophthalmic Solution taken by the formula:

$$(183.20/309.32)(W)(0.5 + 0.5R/93),$$

in which 183.20 and 309.32 are the molecular weights of epinephrine and triacetylepinephrine, respectively; W is the weight, in mg, of the isolated triacetylepinephrine; and R is the specific rotation, in degrees, of the triacetylepinephrine solution.

Epitetracycline Hydrochloride



$C_{22}H_{24}N_2O_8 \cdot HCl$ 480.90

2-Naphthacenecarboxamide, 4-(dimethylamino)-1,4,4a,5,5a,6,11,12a-octahydro-3,6,10,12,12a-pentahydroxy-6-methyl-1,11-dioxo-, monohydrochloride, [4*R*-(4α,4a*β*,5a*β*,6α,-12a*β*)]-.

(4*R*,4a*S*,5a*S*,6*S*,12a*S*) 4-(dimethylamino)-1,4,4a,5,5a,6,11,12a-octahydro-3,6,10,12,12a-pentahydroxy-6-methyl-1,11-dioxo-2-naphthacenecarboxamide monohydrochloride [*23313-80-6*].

» Epitetracycline Hydrochloride contains not less than 70.0 percent of $C_{22}H_{24}N_2O_8 \cdot HCl$.

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

USP Reference standards ⟨11⟩—*USP Tetracycline Hydrochloride RS*.

pH ⟨791⟩: between 2.3 and 4.0, in a solution containing 10 mg per mL.

Loss on drying ⟨731⟩—Dry about 100 mg in a capillary-stoppered bottle in vacuum at a pressure not exceeding 5 mm of mercury at 60° for 3 hours: it loses not more than 6.0% of its weight.

4-Epianhydrotetracycline ⟨226⟩—Dissolve about 250 mg, accurately weighed, in 10 mL of 0.1 N hydrochloric acid, and adjust with 6 N ammonium hydroxide to a pH of 7.8. Transfer this solution with the aid of *EDTA buffer* to a 50-mL volumetric flask, dilute with *EDTA buffer* to volume, and mix. Use this solution, without delay, as the *Test solution:* not more than 2.0% is found.

Assay—

Edetate disodium solution, *Stationary phase*, *Alkaline methanol solution*, *Column support*, *Chromatographic column*, and *Standard preparation*—Prepare as directed in the section *Epitetracycline hydrochloride content and Assay for tetracycline hydrochloride* under *Tetracycline Hydrochloride for Topical Solution*.

Assay preparation—Transfer about 22 mg of Epitetracycline Hydrochloride, accurately weighed, to a 25-mL volumetric flask, add 1 mL of methanol, and swirl to dissolve. Dilute with *Stationary phase* to volume, and mix. Transfer 2.0 mL of this solution to a 10-mL volumetric flask, dilute with *Stationary phase* to volume, and mix. Pipet 2.0 mL of this solution into the *Chromatographic column*, and allow it to penetrate the *Column support*. Add 20 mL of benzene to the solvent reservoir, and collect the eluate at the rate of about 1 mL per minute. When the benzene level reaches the top of the *Column support*, add 60 mL of chloroform to the solvent reservoir, and

continue collecting eluate
of the *Column support*, d
mixture of butyl alcohol
eluate in a 10-mL gradua
cylinder with a low-acti
collecting eluate until the
volumetric flask is the *As*

Procedure—Add 2.0
Standard preparation an
chloroform to volume, an
preparation, determine t
wavelength of maximum
spectrophotometer, usin
quantity, in mg, of $C_{22}H$
chloride taken by the for

in which W is the weight
RS taken, P is the poten
Hydrochloride RS, and A
from the *Assay prepara*
tively.

Equilin

$C_{18}H_{20}O_2$ 268.35
Estra-1,3,5(10),7-tetraen
3-Hydroxyestra-1,3,5(10

» Equilin contains
more than 103.0 pe
dried basis.

Packaging and storage
USP Reference standa
Clarity of solution—
hydroxide in a 125-mL
solution is complete, t
comparison tube: the so

Identification—
A: *Infrared Absorp*
B: *Ultraviolet Abso*
Solution: 50 µg per
Medium: alcohol.

Specific rotation ⟨781S
Test solution: 20 m

Loss on drying ⟨731⟩—
not more than 0.5% of i

Residue on ignition ⟨2

Assay—
Mobile phase—Prepa
35 volumes of acetonitr
Internal standard so
obtain a solution having
Standard preparatio
Equilin RS, accurately
obtain a solution having
mL.
Assay preparation—
weighed, to a 50-mL v
ternal standard soluti
Chromatographic sy
mid chromatograph i

with water to 50.0 mL to obtain a solution having a known concentration of about 100 µg per mL. Prepare this solution fresh.

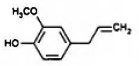
Assay preparation—Transfer a portion of Ophthalmic Solution, equivalent to 50 mg of eucatropine hydrochloride, to a 100-mL volumetric flask, and dilute with water to volume. Dilute 10.0 mL of this solution with water to 50.0 mL.

Procedure—Transfer duplicate 2-mL portions of the *Standard preparation* and of the *Assay preparation* to separate glass-stoppered, 40-mL centrifuge tubes. To one set of two tubes add 3 mL of water and 1 mL of sodium hydroxide solution (1 in 100). Heat these tubes in a boiling water bath for 10 minutes, and allow to cool to room temperature. To the remaining set of tubes, which provide the blanks for the *Standard preparation* and *Assay preparation*, respectively, add 4 mL of water. To each tube add 2 mL of approximately 0.2 M ceric sulfate in diluted sulfuric acid (prepared by dissolving 12.6 g of ceric ammonium sulfate in 50 mL of water and 3 mL of sulfuric acid, and diluting with water to 100 mL) and 20.0 mL of isooctane. Shake by mechanical means for 15 minutes, allow the layers to separate, and remove the isooctane from each tube. Concomitantly determine the absorbances of the isooctane solutions from the hydrolyzed aliquots in 1-cm cells at the wavelength of maximum absorbance at about 242 nm, with a suitable spectrophotometer, against the respective blanks. Calculate the quantity, in mg, of $C_{17}H_{25}NO_3 \cdot HCl$ in the portion of Ophthalmic Solution taken by the formula:

$$0.5C(A_U/A_S),$$

in which C is the concentration, in µg per mL, of USP Eucatropine Hydrochloride RS in the *Standard preparation*, and A_U and A_S are the absorbances of the solutions from the *Assay preparation* and the *Standard preparation*, respectively.

Eugenol



$C_{10}H_{12}O_2$ 164.20
Phenol, 2-methoxy-4-(2-propenyl)-.
4-Allyl-2-methoxyphenol [*97-53-0*].

» Eugenol is obtained from Clove Oil and from other sources.

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

Solubility in 70 percent alcohol—One volume dissolves in 2 volumes of 70 percent alcohol.

Specific gravity ⟨841⟩: between 1.064 and 1.070.

Distilling range, *Method II* ⟨721⟩—Not less than 95% distils between 250° and 255°.

Refractive index ⟨831⟩: between 1.540 and 1.542 at 20°.

Heavy metals, *Method II* ⟨231⟩: 0.004%.

Hydrocarbons—Dissolve 1 mL in 20 mL of 0.5 N sodium hydroxide in a stoppered, 50-mL tube, add 18 mL of water, and mix: a clear mixture results immediately, but it may become turbid when exposed to air.

Limit of phenol—Shake 1 mL with 20 mL of water, filter, and add 1 drop of ferric chloride TS to 5 mL of the clear filtrate: the mixture exhibits a transient grayish green color but not a blue or violet color.

Factor IX Complex

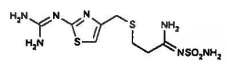
» Factor IX Complex conforms to the regulations of the federal Food and Drug Administration concerning biologics (see *Biologics* ⟨1041⟩). It is a sterile, freeze-dried powder consisting of partially purified Factor IX fraction, as well as concentrated Factors II, VII, and X fractions, of venous plasma obtained from healthy human donors. It contains no preservative. It meets the requirements of the test for potency in having not less than 80 percent and not more than 120 percent of the potency stated on the label in Factor IX Units by comparison with the U.S. Factor IX Standard or with a working reference that has been calibrated with it.

Packaging and storage—Preserve in hermetic containers in a refrigerator.

Expiration date—The expiration date is not later than 2 years from the date of manufacture.

Labeling—Label it with a warning that it is to be used within 4 hours after constitution, and to state that it is for intravenous administration and that a filter is to be used in the administration equipment.

Famotidine



$C_8H_{15}N_7O_2S_3$ 337.45
Propanimidamide, *N'*-(aminosulfonyl)-3-[[[2-[(diaminomethylene)amino]-4-thiazolyl]methyl]thio]-.
[1-Amino-3-[[[2-[(diaminomethylene)amino]-4-thiazolyl]methyl]thio]propylidene] sulfamide [*76824-35-6*].

» Famotidine contains not less than 98.5 percent and not more than 101.0 percent of $C_8H_{15}N_7O_2S_3$, calculated on the dried basis.

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

USP Reference standards ⟨11⟩—*USP Famotidine RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 25 µg per mL.

Medium: phosphate buffer.

Absorptivities at 265 nm, calculated on the dried basis, do not differ by more than 3.0%. [NOTE—Prepare the phosphate buffer as follows. Adjust 250 mL of 0.02 M phosphoric acid with sodium hydroxide solution (1 in 10) to a pH of 2.5, dilute with water to 500 mL, and mix.]

Loss on drying ⟨731⟩—Dry it at a pressure between 1 and 5 mm of mercury at 80° for 5 hours: it loses not more than 0.5% of its weight.

Residue on ignition ⟨281⟩: not more than 0.1%.

Heavy metals, *Method II* ⟨231⟩: not more than 0.001%.

Chromatographic purity—

Adsorbent: 0.25-mm layer of chromatographic silica gel mixture.

Test solution—Transfer about 200 mg of Famotidine, accurately weighed, to a 10-mL volumetric flask, add 2 mL of methanol, and shake for 10 minutes. Add 0.1 mL of glacial acetic acid, stir until dissolved, dilute with methanol to volume, and mix.

Standard solutions—Dissolve an accurately weighed portion of USP Famotidine RS in a mixture of methanol and glacial acetic acid (100 : 1) to obtain *Standard solution 1* having a known concentration

of 0.2 mg per mL. Dilute a portion of this solution, accurately measured, with a mixture of methanol and glacial acetic acid (100 : 1) to obtain *Standard solution 2* containing 65 µg of USP Famotidine RS per mL.

Developing solvent system: a mixture of ethyl acetate, methanol, toluene, and ammonium hydroxide (40 : 25 : 20 : 2).

Procedure—Separately apply 5 µL of the *Test solution* and 5 µL of each *Standard solution* to a plate, and dry under a stream of nitrogen. Proceed as directed for *Thin-Layer Chromatography* under *Chromatography* ⟨621⟩. Examine the plate under short-wavelength UV light, and compare the intensities of any secondary spots observed in the chromatogram of the *Test solution* with those of the principal spots in the chromatograms of the *Standard solutions:* no secondary spot from the chromatogram of the *Test solution* is larger in size or more intense than the principal spot obtained from *Standard solution 2* (0.3%); and the sum of the intensities of the secondary spots obtained from the *Test solution* corresponds to not more than 1.0% (*Standard solution 1*).

Organic volatile impurities, *Method IV* ⟨467⟩: meets the requirements.

Solvent—Use dimethyl sulfoxide.

Assay—Dissolve about 250 mg of Famotidine, accurately weighed, in 80 mL of glacial acetic acid, and titrate with 0.1 N perchloric acid VS (see *Titrimetry* ⟨541⟩), using a suitable anhydrous electrode system. Any aqueous electrolyte solution contained in the electrodes employed should be removed, the electrode rendered anhydrous and filled with 0.1 N lithium perchlorate in acetic anhydride. Perform a blank determination and make any necessary correction. Each mL of 0.1 N perchloric acid is equivalent to 16.87 mg of $C_8H_{15}N_7O_2S_3$.

Famotidine Tablets

» Famotidine Tablets contain not less than 90.0 percent and not more than 110.0 percent of the labeled amount of famotidine ($C_8H_{15}N_7O_2S_3$).

Packaging and storage—Preserve in well-closed, light-resistant containers. Store at controlled room temperature.

USP Reference standards ⟨11⟩—*USP Famotidine RS.*

Identification—

A: (See *Thin-Layer Chromatographic Identification Test* ⟨201⟩.)

Developing solvent—Prepare a mixture of ethyl acetate, methanol, toluene, and ammonium hydroxide (40 : 25 : 20 : 2).

Standard solution—Dissolve USP Famotidine RS in glacial acetic acid to obtain a solution having a concentration of 4 mg per mL.

Test solution—Transfer a portion of finely powdered Tablets, equivalent to about 40 mg of famotidine, to a 10-mL volumetric flask. Dissolve in glacial acetic acid with the aid of sonication, dilute with glacial acetic acid to volume, and centrifuge to get a clear liquid.

Procedure—Apply separately 10 µL each of the *Standard solution* and the *Test solution* to a suitable thin-layer chromatographic plate coated with a 0.25-mm layer of chromatographic silica gel mixture, allow the spots to dry, and develop the plate in a paper-lined chromatographic chamber equilibrated with *Developing solvent* for about 1 hour prior to use. Allow the chromatogram to develop until the solvent front has moved about 15 cm. Remove the plate, air-dry, and examine the plate under short-wavelength UV light: the principal spot from the *Test solution* corresponds in appearance and R_F value to that of the *Standard solution.*

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

Dissolution ⟨711⟩—

Medium: pH 4.5, 0.1 M phosphate buffer; prepared by dissolving 13.6 g of monobasic potassium phosphate in 1 L of water; 900 mL.

Apparatus 2: 50 rpm.

Time: 30 minutes.

Procedure—Determine the amount of $C_8H_{15}N_7O_2S_3$ dissolved from UV absorption at the wavelength of maximum absorbance at about 265 nm, using filtered portions of the solution under test, suitably diluted with *Medium* if necessary, in comparison with a Standard solution having a known concentration of USP Famotidine RS in the same *Medium.*

Tolerances—Not less than 75% *(Q)* of the labeled amount of $C_8H_{15}N_7O_2S_3$ is dissolved in 30 minutes.

Related compounds—

Buffer solution, Mobile phase, Diluent, System suitability solution, Standard preparation, Assay preparation, and *Chromatographic system*—Proceed as directed in the *Assay.*

Standard solution—Use the *Standard preparation.*

Test solution—Use the *Assay preparation.*

Procedure—Separately inject a volume (about 50 µL) of the *Test solution* into the chromatograph, record the chromatograms, and measure the responses for the major peaks. Calculate the percentage of each impurity in the portion of Tablets taken by the formula:

$$100(1/F)C(D/LN)(r_i/r_s),$$

in which F is the relative response factor for each impurity peak (see *Table 1* for values); C is the concentration, in mg per mL, of USP Famotidine RS in the *Standard solution; L* is the labeled amount, in mg, of famotidine in each Tablet; N is the number of tablets taken to prepare the *Test solution; D* is the dilution factor used to prepare the *Test solution; r_i* is the peak area obtained for each individual impurity in the *Test solution;* and r_s is the peak area for famotidine in the *Standard solution.*

Table 1

Relative Retention Time	Relative Response Factor (F)	Name	Limit (%)
0.38	1.0	Famotidine related compound A[1]	1.0
0.65	1.0	Famotidine related compound B[2]	0.5
0.85	1.0	Famotidine related compound C[3]	0.5
1.21	1.3	Famotidine related compound D[4]	0.5

[1] 3-[2-(diaminomethyleneamino)-1,3-thiazol-4-ylmethylsulfinyl]-N-sulfamoyl-propanamidine

[2] 3-[2-(diaminomethyleneamino)-1,3-thiazol-4-ylmethylthio]-propanoic acid

[3] 3-[2-(diaminomethyleneamino)-1,3-thiazol-4-ylmethylthio]-N-sulfamoyl-propanamide

[4] 3-[2-(diaminomethyleneamino)-1,3-thiazol-4-ylmethylthio]-propanamide

In addition to not exceeding the limits for each impurity in *Table 1,* not more than 1.5% of total impurities is found.

Uniformity of dosage units ⟨905⟩: meet the requirements.

Assay—

Buffer solution—Dissolve 13.6 g of sodium acetate trihydrate in 750 mL of water. Add 1 mL of triethylamine, adjust with glacial acetic acid to a pH of 6.0, and dilute with water to 1 L.

Mobile phase—Prepare a mixture of *Buffer solution* and acetonitrile (93 : 7), mix, and degas. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Diluent—Dissolve 6.8 g of monobasic potassium phosphate in 750 mL of water, adjust with 1 M potassium hydroxide to a pH of 6.0, and dilute with water to 1 L.

System suitability stock solution—Transfer 10 mg of famotidine to a 50-mL volumetric flask, add 1 mL of 0.1 N hydrochloric acid, heat at 80° for 30 minutes, and cool to room temperature. Add 2 mL of 0.1 N sodium hydroxide, heat at 80° for 30 minutes, cool to room temperature, and neutralize by adding 1 mL of 0.1 N hydrochloric acid. Dilute with *Diluent* to volume. Transfer 10 mL of this solution to a separate 50-mL volumetric flask containing 5 mg of famotidine dissolved in 8 mL of methanol. Dilute with *Diluent* to volume.

System suitability solution—Transfer about 1 mL of *System suitability stock solution* to a suitable container, add 1 mL of *Diluent* and 1 drop of hydrogen peroxide solution, and mix well. [NOTE—Prepare fresh daily.]

Standard preparation—Transfer about 10 mg of USP Famotidine RS, accurately weighed, into a 100-mL volumetric flask, add 20 mL

USP 28

of methanol, and soni
volume, and mix.

Assay preparation—
volumetric flask. Add
Tablets. Add 200 mL o
300 rpm for 1 hour. Di
Quantitatively dilute a
obtain a solution contai

Chromatographic sy
liquid chromatograph i
4.6-mm × 15-cm colu
temperature is maintain
minute. Chromatograph
the famotidine peak and
products listed in *Table*
Procedure: the resolutio
C and famotidine is nc
famotidine and famotidi
1.3; and the capacity fa
2.0. Chromatograph the
responses as directed fo
for replicate injections i

Procedure—Separatel
Standard preparation a
ograph, record the chrom
major peaks. Calcula
($C_8H_{15}N_7O_2S_3$) in each T

in which C is the concent
in the *Standard preparat*
the *Assay preparation; N*
Assay preparation; and *r*
Assay preparation and t

Felodipine

H_3C

$C_{18}H_{19}Cl_2NO_4$ 384.26
3,5-Pyridinedicarboxylic
2,6-dimethyl-, ethyl
(±)-Ethyl methyl 4-(2,3-
3,5-pyridinedicarbox

» Felodipine contains
more than 101.0 perc
the dried basis.

Packaging and storage—
and store at controlled roo

USP Reference standard

Color of solution—Pre
concentration of 20 mg p
cm cell at the wavelength
methanol being used as th

Identification—

A: *Infrared Absorptio*

B: The retention time
Assay preparation corr
Standard preparation, as

Loss on drying ⟨731⟩—D
not more than 0.5% of its weight.

Standard preparation—Prepare as directed in the *Assay* under *Fluorescein Sodium*.

Assay preparation—Remove 1 Strip from its package, taking care not to allow any portion of the tip to adhere to the packaging material, transfer to a 100-mL volumetric flask, add 50 mL of water, shake the flask vigorously, and dilute with water to volume. Shake occasionally, and, after 1 hour, mix the contents of the flask. Transfer an aliquot (V) of this solution, equivalent to about 100 µg of fluorescein sodium, to a 200-mL volumetric flask, dilute with water to volume, and mix. Transfer 3 mL of the resulting solution to a 100-mL volumetric flask containing 20 mL of pH 9.0 alkaline borate buffer (see *Buffer Solutions* in the section *Reagents, Indicators, and Solutions*), dilute with water to volume, and mix.

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Fluorescein Sodium*. Calculate the quantity, in mg, of fluorescein sodium ($C_{20}H_{10}Na_2O_5$) in the Strip taken by the formula:

$$(333)(C/V)(I_U/I_S),$$

in which C is the concentration, in µg per mL, of fluorescein sodium in the *Standard preparation*; V is the volume of the aliquot of solution taken for the *Assay preparation*; and I_U and I_S are the fluorescence intensities observed for the *Assay preparation* and the *Standard preparation*, respectively. Calculate the average content from the individual assays of not less than 10 Strips.

Fluorescein Sodium and Benoxinate Hydrochloride Ophthalmic Solution

» Fluorescein Sodium and Benoxinate Hydrochloride Ophthalmic Solution is a sterile aqueous solution of Fluorescein Sodium and Benoxinate Hydrochloride. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amounts of fluorescein sodium ($C_{20}H_{10}Na_2O_5$) and benoxinate hydrochloride ($C_{17}H_{28}N_2O_3 \cdot HCl$). It contains a suitable preservative.

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

USP Reference standards ⟨11⟩—*USP Benoxinate Hydrochloride RS. USP Diacetylfluorescein RS.*

Identification—

A: It responds to *Identification* test *A* under *Fluorescein Sodium*.

B: The relative retention times of the major peaks in the chromatogram of the *Assay* correspond to those in the chromatograms of the *Standard fluorescein sodium preparation* and the *Standard benoxinate hydrochloride preparation* as obtained in the *Assay*.

Sterility ⟨71⟩—It meets the requirements when tested as directed for *Membrane Filtration* under *Test for Sterility of the Product to be Examined*.

pH ⟨791⟩: between 4.3 and 5.3.

Assay—

Mobile phase—Dissolve 100 mg of sodium 1-pentanesulfonate in 1 mL of glacial acetic acid in a 2000-mL volumetric flask. Add 600 mL of acetonitrile and 10 mL of triethanolamine, dilute with water to volume, and mix. Adjust with phosphoric acid to a pH of 3, and pass through a filter having a 0.5-µm or finer porosity. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard fluorescein sodium preparation—Transfer about 55 mg of USP Diacetylfluorescein RS, accurately weighed, to a 50-mL volumetric flask containing 5 mL of alcohol. Add 1 mL of 2.5 N sodium hydroxide, and heat on a steam bath at about the boiling temperature for 20 minutes, with frequent swirling. Cool, dilute with water to volume, and mix. Transfer 10.0 mL of this solution to a 100-mL volumetric flask, dilute with *Mobile phase* to volume, and mix. This solution contains the equivalent of about 0.1 mg of fluorescein sodium per mL.

Standard benoxinate hydrochloride preparation—Quantitatively dissolve an accurately weighed quantity of USP Benoxinate Hydrochloride RS in *Mobile phase*, and if necessary dilute quantitatively and stepwise with *Mobile phase* to obtain a solution having a known concentration of about 0.1J mg per mL, J being the ratio of the labeled amount, in mg, of benoxinate hydrochloride to the labeled amount, in mg, of fluorescein sodium in each mL of Ophthalmic Solution.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 5 mg of fluorescein sodium, to a 50-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4-mm × 30-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. Chromatograph the *Standard fluorescein sodium preparation* and the *Standard benoxinate hydrochloride preparation*, and record the peak responses as directed for *Procedure*: the tailing factor for each analyte peak is not more than 2.0, and the relative standard deviation for replicate injections of each *Standard preparation* is not more than 2.0%.

Procedure—[NOTE—Use peak areas where peak responses are indicated.] Separately inject equal volumes (about 25 µL) of the *Standard fluorescein sodium preparation*, the *Standard benoxinate hydrochloride preparation*, and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the peak responses. Calculate the quantity, in mg, of fluorescein sodium ($C_{20}H_{10}Na_2O_5$) in each mL of Ophthalmic Solution taken by the formula:

$$(376.28/416.39)(W/10V)(r_U/r_S),$$

in which 376.28 and 416.39 are the molecular weights of fluorescein sodium and diacetylfluorescein, respectively; W is the quantity, in mg, of USP Diacetylfluorescein RS taken to prepare the *Standard fluorescein sodium preparation*; V is the volume, in mL, of Ophthalmic Solution taken; and r_U and r_S are the fluorescein peak responses obtained from the *Assay preparation* and the *Standard fluorescein sodium preparation*, respectively. Calculate the quantity, in mg, of benoxinate hydrochloride ($C_{17}H_{28}N_2O_3 \cdot HCl$) in each mL of Ophthalmic Solution taken by the formula:

$$50(C/V)(r_U/r_S),$$

in which C is the concentration, in mg per mL, of USP Benoxinate Hydrochloride RS in the *Standard benoxinate hydrochloride preparation*; V is the volume, in mL, of Ophthalmic Solution taken, and r_U and r_S are the benoxinate peak responses obtained from the *Assay preparation* and the *Standard benoxinate hydrochloride preparation*, respectively.

Fluorescein Sodium and Proparacaine Hydrochloride Ophthalmic Solution

» Fluorescein Sodium and Proparacaine Hydrochloride Ophthalmic Solution is a sterile aqueous solution of Fluorescein Sodium and Proparacaine Hydrochloride. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amounts of fluorescein sodium ($C_{20}H_{10}Na_2O_5$) and proparacaine hydrochloride ($C_{16}H_{26}N_2O_3 \cdot HCl$). It contains a suitable preservative.

Packaging and storage—Preserve in tight, light-resistant containers, preferably of Type I amber glass, and store in a refrigerator.

Labeling—Label it to state that it is to be stored in a refrigerator before and after the container is opened.

USP Reference standards ⟨11⟩—*USP Diacetylfluorescein RS. USP Proparacaine Hydrochloride RS.*

Identification—

A: It responds to *Identification* test *A* under *Fluorescein Sodium*.

B: It responds to the *Identification* test under *Proparacaine Hydrochloride Ophthalmic Solution*.

Sterility ⟨71⟩—It meets the requirements when tested as directed for *Membrane Filtration* under *Test for Sterility of the Product to be Examined*.

pH ⟨791⟩: between 4.0 and 5.2.

Assay for fluorescein sodium—

Standard preparation—Prepare as directed in the *Assay* under *Fluorescein Sodium.*

Assay preparation—Using Ophthalmic Solution, prepare as directed in the *Assay* under *Fluorescein Sodium.*

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Fluorescein Sodium.* Calculate the quantity, in mg, of fluorescein sodium ($C_{20}H_{10}Na_2O_5$) in the volume of Ophthalmic Solution taken by the formula:

$$3333C(I_U/I_S),$$

in which the terms are as defined therein.

Assay for proparacaine hydrochloride—

Standard preparation—Prepare as directed for Standard preparation in the *Assay* under *Proparacaine Hydrochloride Ophthalmic Solution.*

Assay preparation—Using Ophthalmic Solution, prepare as directed for *Assay preparation* under *Proparacaine Hydrochloride Ophthalmic Solution.*

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Proparacaine Hydrochloride Ophthalmic Solution.* Calculate the quantity, in mg, of proparacaine hydrochloride ($C_{16}H_{26}N_2O_3 \cdot HCl$) in each mL of Ophthalmic Solution taken by the formula:

$$100(C/V)(r_U/r_S),$$

in which the terms are as defined therein.

Fludeoxyglucose F 18 Injection

» Fludeoxyglucose F 18 Injection is a sterile, aqueous solution, suitable for intravenous administration, of 2-deoxy-2-[^{18}F]fluoro-D-glucose in which a portion of the molecules are labeled with radioactive ^{18}F (see *Radiopharmaceuticals for Positron Emission Tomography—Compounding* ⟨823⟩). It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of ^{18}F expressed in MBq (mCi) per mL at the time indicated in the labeling. It may contain suitable preservatives and/or stabilizing agents.

Specific activity: no carrier added.

Packaging and storage—Preserve in single-dose or multiple-dose containers that are adequately shielded.

Labeling—Label it to include the following, in addition to the information specified for *Labeling* under *Injection* ⟨1⟩: the time and date of calibration; the amount of ^{18}F as fludeoxyglucose expressed as total MBq (mCi) per mL, at time of calibration; the expiration time and date; the name and quantity of any added preservative or stabilizer; and the statement "Caution—Radioactive Material." The labeling indicates, that in making dosage calculations, correction is to be made for radioactive decay. The radioactive half-life of ^{18}F is 109.7 minutes. The label indicates "Do not use if cloudy or if it contains particulate matter."

USP Reference standards ⟨11⟩—*USP Endotoxin RS. USP Fludeoxyglucose RS. USP Fludeoxyglucose Related Compound A RS. USP Fludeoxyglucose Related Compound B RS.*

Identification—

A: *Radionuclidic identity*—Its half-life, determined using a suitable detector system (see *Radioactivity* ⟨821⟩), is between 105 and 115 minutes.

B: *Radiochemical identity*—The R_F value of Fludeoxyglucose F 18 in the chromatogram of the *Test solution* corresponds to that in the chromatogram of the *Standard solution*, as obtained in the *Radiochemical purity* test.

Bacterial endotoxins ⟨85⟩ (see *Sterilization and Sterility Assurance* under *Radiopharmaceuticals for Positron Emission Tomography—Compounding* ⟨823⟩)—It contains not more than $175/V$ USP Endotoxin Unit per mL of the Injection, in which V is the maximum administered total dose, in mL, at the expiration time.

pH ⟨791⟩: between 4.5 and 7.5.

Radiochemical purity—

Standard solution—Dissolve 10 mg of USP Fludeoxyglucose RS in 100 mg of acetonitrile and water (95 : 5). (The USP Fludeoxyglucose RS that is specified in this test is nonradioactive 2-deoxy-2-fluoro-D-glucose [molecular weight 182.15].)

Test solution—Use the Injection.

Procedure—Apply a volume of Injection, appropriately diluted such that it provides a count rate suitable for the radioactivity detection system being utilized, to an activated silica gel thin-layer chromatographic plate (see *Chromatography* ⟨621⟩). Apply about 10 µg of the *Standard solution* to the same chromatographic plate. Develop the chromatogram in a solvent system consisting of a mixture of acetonitrile and water (95 : 5) until the solvent has moved about three-fourths of the length of the plate. Remove the plate, and allow the chromatogram to dry. Determine the radioactivity distribution by scanning the chromatogram with a suitable collimated radiation detector. Determine the location of the Fludeoxyglucose by spraying the developed chromatographic plate with 2 N sulfuric acid and heating the plate at 110° for 10 minutes: the R_F value of Fludeoxyglucose F 18 (determined by radiochromatogram scanning) corresponds to that of the *Standard solution* (about 0.4); the radioactivity of Fludeoxyglucose F 18 is not less than 90% of the total radioactivity.

Radionuclidic purity—Using a suitable gamma-ray spectrometer (see *Selection of a Counting Assembly* under *Radioactivity* ⟨821⟩), count an appropriate aliquot of the Injection for a period of time sufficient to collect a gamma spectrum. The resultant gamma spectrum should be analyzed for the presence of identifiable photopeaks which are not characteristic of ^{18}F emissions. Not less than 99.5% of the observed gamma emissions should correspond to the 0.511 MeV, 1.022 MeV, or Compton scatter peaks of ^{18}F.

Chemical purity—[NOTE—The methods and limits described in this section relate to potential impurities associated with the acid-hydrolysis method of synthesis for the Injection. Specific examples include aminopolyether (Kryptofix®) and 2-chloro-2-deoxy-D-glucose. If methods of synthesis that may result in different impurities are used, the presence of unlabeled ingredients, reagents, and by-products specific to the process must be controlled, and their potential for physiological or pharmacological effects must be considered (see *Radiopharmaceuticals for Positron Emission Tomography—Compounding* ⟨823⟩). Any ingredients with toxic potential must be within appropriate limits, and conformance with these limits is to be demonstrated by the use of one or more validated limit tests.]

LIMIT OF AMINOPOLYETHER—[NOTE—This test must be performed for Fludeoxyglucose F 18 produced by any route of synthesis that uses this reagent.]

Absorbent: 0.25-mm layer of chromatographic silica gel.[1]

Test solution: Use the Injection.

Standard solution—Dissolve an accurately weighed quantity of USP Fludeoxyglucose Related Compound A RS in saline TS to obtain a solution having a known concentration of 50 µg per mL.

Application volume: about 1 µL.

Developing solvent system: a mixture of methanol and 30% ammonium hydroxide (9 : 1).

Procedure—Proceed as directed for *Thin-Layer Chromatography* under *Chromatography* ⟨621⟩. Place the plate in a chamber containing iodine crystals. Develop the plate until a spot is visible on the chromatogram of the *Standard solution:* the size and intensity of the spot obtained from the *Test solution* does not exceed that obtained from the *Standard solution.*

LIMIT OF 2-CHLORO-2-DEOXY-D-GLUCOSE—[NOTE—This test is performed when the nucleophilic synthesis includes hydrolysis with hydrochloric acid or the use of anionic exchange resins in the chloride form to trap fluoride ^{18}F released from the target prior to its use in the synthesis of Fludeoxyglucose F 18.]

Mobile phase—Dissolve about 16 g of 50% sodium hydroxide solution in 1000 mL of water, filter, and degas by sparging with helium.

[1] Available from Alltech Associates, Inc., 2051 Waukegan Rd., Deerfield, IL 60015 as Machery Nagel SILG/UV 254 4 × 8 cm, Alltech catalog No. 805021.

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System suitability solu
tities of USP Fludeoxyglu
Compound B RS in *Mobil*
concentrations of 1.0 mg
Standard solution—Dis
USP Fludeoxyglucose Rel
solution having a known c
Test solution—Use the I
Chromatographic syste
liquid chromatograph is
detector and a 4.0-mm
packing L46. The flow rat
Chromatograph the *Stand*
solution, and record the pea
resolution, R, between flud
compound B is not less tha
for replicate injections is n
Procedure—Separately ir
Standard solution and the
record the chromatograms, a
Calculate the quantity, in m
mL of the Injection taken b

in which C is the conc
Fludeoxyglucose Related Cc
and r_U and r_S are the 2-chlorc
from the *Test solution* and t
more than 1 mg is found in t
produced.

Residual solvents—

Standard solutions—Prepa
acetonitrile, and dehydrated
0.1%, 0.01%, and 0.1%, resp
Test solutions—Use the Inj
Chromatographic system
chromatograph is equipped
splitless injector system, ar
column coated with a 0.2
stationary phase. The carrier
mL per minute. (Nitrogen n
chromatograph is programmec
maintained at 40° for 2 minute
rate of 20° per minute to 1
minutes. The injection port an
at 250° and 300°, respectivel
record the identity peak resp
resolution, R, between any tw
the relative standard deviation
5%.
Procedure—Separately injec
Standard solutions and the *T*
record the chromatograms,
Calculate the percentage of a
injection by the formula:

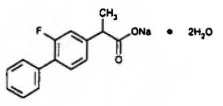
$$C$$

in which C is the percentage of
solution; and r_i and r_S are the p
obtained from the *Test solution*
respectively: not more than 0.0
than 0.5% of ether is found; ar
found.

Other requirements—It meets
⟨1⟩, except that the Injection ma
completion of the test for *Steril*
within 24 hours of final manufac
to the recommendation of *Volun*

Assay for radioactivity—Usin
directed under *Radioactivity* ⟨8
MBq (or mCi) per mL, of the In

Flurbiprofen Sodium



$C_{15}H_{12}FNaO_2 \cdot 2H_2O$ 302.27

[1,1'-Biphenyl]-4-acetic acid, 2-fluoro-α-methyl, sodium salt dihydrate, (±)-.

Sodium (±)-2-(2-fluoro-4-biphenylyl)propionate dihydrate.

Anhydrous 266.25

» Flurbiprofen Sodium contains not less than 97.0 percent and not more than 103.0 percent of $C_{15}H_{12}FNaO_2 \cdot 2H_2O$.

Packaging and storage—Preserve in well-closed containers.

USP Reference standards ⟨11⟩—*USP Flurbiprofen RS. USP Flurbiprofen Sodium RS. USP Flurbiprofen Related Compound A RS.*

Identification—

A: *Infrared Absorption* ⟨197M⟩—

Test specimen: previously dried.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 10 µg per mL.

Medium: pH 6.0 buffer consisting of 2.42 g of monobasic sodium phosphate and 0.66 g of dibasic sodium phosphate dissolved in water to make 1000 mL.

Absorptivities at 246 nm, calculated on the dried basis, do not differ by more than 3.0%.

C: The residue obtained by igniting it meets the requirements of the tests for *Sodium* ⟨191⟩.

Specific rotation ⟨781S⟩: between –0.45° and +0.45°.

Test solution: 50 mg per mL, in methanol.

Loss on drying ⟨731⟩—Dry about 0.3 g of it in vacuum at a pressure not exceeding 1 mm of mercury over phosphorus pentoxide in a suitable drying tube at 60° for 18 hours: it loses not less than 11.3% and not more than 12.5% of its weight.

Heavy metals, *Method II* ⟨231⟩: 0.001%.

Limit of flurbiprofen related compound A—

Diluent, Mobile phase, and *System suitability preparation*—Proceed as directed in the *Assay.*

Standard solution—Use *Standard flurbiprofen related compound A preparation,* prepared as directed in the *Assay.*

Test solution—Use the *Assay preparation.*

Chromatographic system—Proceed as directed in the *Assay,* except that chromatograph the *Standard solution* instead of the *Standard preparation.*

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard solution* and the *Test solution* into the chromatograph, record the chromatograms, and measure the areas for the major peaks. Calculate the percentage of flurbiprofen related compound A in the portion of Flurbiprofen Sodium taken by the formula:

$$200(C/W)(r_U/r_S),$$

in which C is the concentration, in µg per mL, of USP Flurbiprofen Related Compound A RS in the *Standard solution; W* is the weight, in mg, of the portion of Flurbiprofen Sodium taken to prepare the *Test solution;* and r_U and r_S are the peak areas for flurbiprofen related compound A obtained from the *Test solution* and the *Standard solution,* respectively: not more than 1.5% is found.

Organic volatile impurities, *Method I* ⟨467⟩: meets the requirements.

Assay—

Diluent—Mix 500 mL of methanol and 250 mL of water.

Mobile phase—Prepare a filtered and degassed mixture of acetonitrile, water, and glacial acetic acid (50 : 49 : 1). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard flurbiprofen related compound A preparation—Dissolve an accurately weighed quantity of USP Flurbiprofen Related Compound A RS in methanol to obtain a stock solution having a known concentration of about 150 µg per mL. Transfer 1.0 mL of this solution to a 200-mL volumetric flask, dilute with *Diluent* to volume, and mix.

Standard preparation—Dissolve an accurately weighed quantity of USP Flurbiprofen RS in methanol to obtain a stock solution having a known concentration of about 1 mg per mL. Transfer 5.0 mL of this solution to a 100-mL volumetric flask, dilute with *Diluent* to volume, and mix.

System suitability preparation—Transfer 5 mL of the stock solution used to prepare the *Standard preparation* and 2 mL of the stock solution used to prepare the *Standard flurbiprofen related compound A preparation* to a 100-mL volumetric flask, dilute with *Diluent* to volume, and mix.

Assay preparation—Transfer about 100 mg of Flurbiprofen Sodium, accurately weighed, to a 100-mL volumetric flask, dissolve in and dilute with methanol to volume, and mix. Transfer 5.0 mL of this solution to a second 100-mL volumetric flask, dilute with *Diluent* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4.0-mm × 25-cm column that contains 10-µm packing L7. The flow rate is about 2 mL per minute. Chromatograph the *System suitability preparation,* and record the peak responses as directed for *Procedure:* the resolution, R, between flurbiprofen related compound A and flurbiprofen is not less than 1.0. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the tailing factor is not more than 2.5; and the relative standard deviation for replicate injections is not more than 1.0%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the areas for the major peaks. Calculate the percentage of $C_{15}H_{12}FNaO_2 \cdot 2H_2O$ in the portion of Flurbiprofen Sodium taken by the formula:

$$200(302.27/244.27)(C/W)(r_U/r_S),$$

in which 302.27 and 244.27 are the molecular weights of flurbiprofen sodium dihydrate and anhydrous flurbiprofen, respectively; C is the concentration, in µg per mL, of USP Flurbiprofen RS in the *Standard preparation; W* is the weight, in mg, of the portion of Flurbiprofen Sodium taken to prepare the *Assay preparation;* and r_U and r_S are the flurbiprofen peak responses obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Flurbiprofen Sodium Ophthalmic Solution

» Flurbiprofen Sodium Ophthalmic Solution contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of flurbiprofen sodium ($C_{15}H_{12}FNaO_2 \cdot 2H_2O$).

Packaging and storage—Preserve in tight containers.

USP Reference standards ⟨11⟩—*USP Flurbiprofen RS. USP Flurbiprofen Related Compound A RS.*

Identification—The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

pH ⟨791⟩: between 6.0 and 7.0.

Antimicrobial effectiveness ⟨51⟩: meets the requirements.

Sterility ⟨71⟩—It meets the requirements when tested as directed for *Membrane Filtration* under *Test for Sterility of the Product to be Examined.*

Assay—

Diluent, Mobile phase, Standard flurbiprofen related compound A preparation, Standard preparation, and *System suitability preparation*—Proceed as directed in the *Assay* under *Flurbiprofen Sodium.*

Assay preparation—Use the undiluted Ophthalmic Solution.

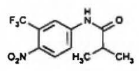
Chromatographic system—Proceed as directed in the *Assay* under *Flurbiprofen Sodium*, using a 4.0-mm × 5-cm guard column that contains 5-µm packing L1.

Procedure—Separately inject equal volumes (about 15 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the areas for the major peaks. Calculate the quantity of flurbiprofen sodium ($C_{15}H_{12}FNaO_2 \cdot 2H_2O$) in each mL of the Ophthalmic Solution taken by the formula:

$$(302.27/244.27)C(r_U/r_S),$$

in which 302.27 and 244.27 are the molecular weights of flurbiprofen sodium dihydrate and anhydrous flurbiprofen, respectively; C is the concentration, in mg per mL, of USP Flurbiprofen 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.

Flutamide



$C_{11}H_{11}F_3N_2O_3$ 276.21

Propanamide, 2-methyl-N-[4-nitro-3-trifluoromethyl)-phenyl]-. α,α,α-Trifluoro-2-methyl-4′-nitro-*m*-propionotoluidide [*13311-84-7*].

» Flutamide contains not less than 98.0 percent and not more than 101.0 percent of $C_{11}H_{11}F_3N_2O_3$, calculated on the dried basis.

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

USP Reference standards ⟨11⟩—*USP Flutamide RS. USP o-Flutamide RS.*

Identification—

A: *Infrared Absorption* ⟨197M⟩.

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

Melting range ⟨741⟩: between 110° and 114°, but the range between beginning and end of melting does not exceed 2°.

Loss on drying ⟨731⟩—Dry it in vacuum at 60° for 3 hours: it loses not more than 0.5% of its weight.

Residue on ignition ⟨281⟩: not more than 0.1%.

Heavy metals, *Method II* ⟨231⟩: not more than 10 ppm.

Related compounds—

Mobile phase and *System suitability solution*—Prepare as directed in the *Assay.*

Standard solution—Use the *Standard preparation,* prepared as directed in the *Assay.*

Test solution—Use the *Assay preparation,* prepared as directed in the *Assay.*

Detector sensitivity solution—Transfer 1.0 mL of the *Standard solution* into a 100-mL volumetric flask, dilute with a mixture of water and acetonitrile (4 : 1) to volume, and mix. Dilute quantitatively, and stepwise if necessary, with a mixture of water and acetonitrile (4 : 1) to obtain a solution having a known concentration of about 0.1 µg per mL.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 240-nm detector and a 4.6-mm × 25-cm column that contains packing L1. The column temperature is maintained at 25 ± 5°. The flow rate is about 1.0 mL per minute. Chromatograph the *System suitability solution,* and record the peak area responses as directed for *Procedure:* the relative retention times are about 1.4 for *o*-flutamide and 1.0 for flutamide; and the resolution, *R,* between flutamide and *o*-flutamide is not less than 6.0. Chromatograph the *Detector sensitivity solution,* and record the peak area responses as directed for *Procedure:* the relative standard deviation for replicate injections is not more than 10.0% for flutamide.

Procedure—Inject a volume (about 20 µL) of the *Test solution* into the chromatograph, record the chromatogram, and measure the peak area responses. Calculate the percentage of each impurity in the portion of Flutamide taken by the formula:

$$100(1/F)(r_i/r_s),$$

in which F is the relative response factor of the impurities according to the table below; r_i is the peak area response for each impurity; and r_s is the sum of the responses of all the peaks: the impurities meet the requirements tabulated below.

Compound name	Relative retention time	Relative Response Factor *(F)*	Limit (%)
4-Nitro-3-trifluoromethylacetanilide	0.42	1.06	0.2
4-Nitro-3-trifluoromethylaniline	0.45	1.10	0.15
3-trifluoromethylaniline	0.63	1.10	0.2
4-Nitro-3-trifluoromethylpropionanilide	0.66	1.02	0.3
3-trifluoromethylisobutyranilide	0.80	1.95	0.2
o-Flutamide	1.40	1.78	0.2
Flutamide	1.0	1.0	—
Unknown	—	1.0	0.05
Total unknown	—	—	0.1
Total impurities	—	—	0.4

Assay—

Mobile phase—Prepare a filtered and degassed mixture of water and acetonitrile (55 : 45). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Flutamide RS in 50 mL of acetonitrile, and dilute quantitatively, and stepwise if necessary, with water to obtain a solution having a known concentration of about 0.2 mg per mL.

System suitability solution—Transfer about 50 mg of USP Flutamide RS, accurately weighed, to a 50-mL volumetric flask, dissolve in 10 mL of acetonitrile, dilute with water to volume, and mix. Transfer 1.0 mL of this solution and 5.0 mL of the *Standard preparation* into a 100-mL volumetric flask, dilute with a mixture of water and acetonitrile (4 : 1) to volume, and mix.

Assay preparation—Transfer about 50 mg of Flutamide, previously dried and accurately weighed, to a 250-mL volumetric flask. Add 50 mL of acetonitrile, and sonicate until the Flutamide dissolves. Add 150 mL of water, mix, and allow to warm to room temperature. Dilute with water to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 240-nm detector and a 4.6-mm × 25-cm column that contains packing L1. The column temperature is maintained at 25 ± 5°. The flow rate is about 1.0 mL per minute. Chromatograph the *System suitability solution,* and record the peak area responses as directed for *Procedure:* the relative retention times are about 1.4 for *o*-flutamide and 1.0 for flutamide; and the resolution, *R,* between flutamide and *o*-flutamide is not less than 6.0. Chromatograph the *Standard preparation,* and record the peak area responses as directed for *Procedure:* the tailing factor is not more than 2.0; and the relative standard deviation for replicate injections is not more than 1.5%.

Procedure—Separately inject equal volumes (about 20 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 $C_{11}H_{11}F_3N_2O_3$ in the portion of Flutamide taken by the formula:

$$250C(r_U/r_S),$$

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

USP Reference standards ⟨11⟩—*USP Gentamicin Sulfate RS.*

Identification—Shake a quantity of Ointment, equivalent to about 5 mg of gentamicin, with a mixture of 200 mL of chloroform and 5 mL of water. Allow to separate, and filter the aqueous layer: the filtrate so obtained meets the requirements of the *Identification* test under *Gentamicin Injection.*

Minimum fill ⟨755⟩: meets the requirements.

Water, *Method I* ⟨921⟩: not more than 1.0%, 20 mL of a mixture of toluene and methanol (7 : 3) being used in place of methanol in the titration vessel.

Assay—Proceed with Ointment as directed under *Antibiotics—Microbial Assays* ⟨81⟩, using an accurately weighed quantity of Ointment, equivalent to about 1 mg of gentamicin, shaken with about 50 mL of ether in a separator, and extracted with four 20-mL portions of *Buffer No. 3.* Combine the aqueous extracts, and dilute quantitatively and stepwise with *Buffer No. 3* to obtain a *Test Dilution* having a concentration assumed to be equal to the median dose level of the Standard.

Gentamicin Sulfate Ophthalmic Ointment

» Gentamicin Sulfate Ophthalmic Ointment contains the equivalent of not less than 90.0 percent and not more than 135.0 percent of the labeled amount of gentamicin.

Packaging and storage—Preserve in collapsible ophthalmic ointment tubes, and avoid exposure to excessive heat.

USP Reference standards ⟨11⟩—*USP Gentamicin Sulfate RS.*

Identification—Shake a quantity of Ophthalmic Ointment, equivalent to about 5 mg of gentamicin, with a mixture of 200 mL of chloroform and 5 mL of water. Allow to separate, and filter the aqueous layer: the filtrate so obtained meets the requirements of the *Identification* test under *Gentamicin Injection.*

Sterility ⟨71⟩: meets the requirements.

Minimum fill ⟨755⟩: meets the requirements.

Metal particles—It meets the requirements of the test for *Metal Particles in Ophthalmic Ointments* ⟨751⟩.

Other requirements—It meets the requirements of the test for *Water* and of the *Assay* in *Gentamicin Sulfate Ointment.*

Gentamicin Sulfate Ophthalmic Solution

» Gentamicin Sulfate Ophthalmic Solution is a sterile, buffered solution of Gentamicin Sulfate with preservatives. It contains the equivalent of not less than 90.0 percent and not more than 135.0 percent of the labeled amount of gentamicin.

Packaging and storage—Preserve in tight containers, and avoid exposure to excessive heat.

USP Reference standards ⟨11⟩—*USP Gentamicin Sulfate RS.*

pH ⟨791⟩: between 6.5 and 7.5.

Other requirements—It meets the requirements of the *Identification* test under *Gentamicin Injection* and meets the requirements under *Sterility Tests* ⟨71⟩, when tested as directed in the section *Membrane Filtration* in *Test for Sterility of the Product To Be Examined.*

Assay—Proceed with Ophthalmic Solution as directed in the *Assay* under *Gentamicin Injection.*

Gentamicin Sulfate and Betamethasone Acetate Ophthalmic Solution

» Gentamicin Sulfate and Betamethasone Acetate Ophthalmic Solution contains not less than 90.0 percent and not more than 125.0 percent of the labeled amount of gentamicin and contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of betamethasone acetate ($C_{24}H_{31}FO_6$).

Packaging and storage—Preserve in tight containers.

Labeling—Label it to indicate that it is for veterinary use only.

USP Reference standards ⟨11⟩—*USP Betamethasone Acetate RS. USP Gentamicin Sulfate RS.*

Identification—

A: Apply 10 µL of Ophthalmic Solution and 10 µL of a Standard solution containing 5 mg per mL of USP Gentamicin Sulfate RS in water to a thin-layer chromatographic plate (see *Chromatography* ⟨621⟩) coated with a 0.25-mm layer of chromatographic silica gel mixture. Allow the spots to dry, and in a paper-lined tank develop the chromatogram in a solvent system consisting of the lower phase mixture of dichloromethane, methanol, and ammonium hydroxide (1 : 1 : 1) 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 plate to air-dry. Locate the spots on the plate by placing it in a tank containing about 15 g of iodine crystals for 15 minutes: the R_F values of the three principal spots obtained from the test solution correspond to those obtained from the Standard solution.

B: The retention time of the major peak obtained in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation*, both relative to the internal standard, as obtained in the *Assay for betamethasone acetate.*

pH ⟨791⟩: between 5.5 and 7.0.

Sterility ⟨71⟩—It meets the requirements when tested as directed for *Membrane Filtration* in *Test for Sterility of the Product To Be Examined.*

Other requirements—It meets the requirements under *Antimicrobial Effectiveness Tests* ⟨51⟩.

Assay for gentamicin—Proceed as directed for gentamicin under *Antibiotics—Microbial Assays* ⟨81⟩, using an accurately measured volume of Ophthalmic Solution diluted quantitatively and stepwise with *Buffer No. 3* to obtain a *Test Dilution* having a concentration assumed to be equal to the median dose level of the Standard.

Assay for betamethasone acetate—

Mobile phase—Prepare a filtered and degassed mixture of water and acetonitrile (8 : 7). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Internal standard solution—Dissolve a quantity of *o*-phenylphenol in methanol to obtain a solution containing about 0.55 mg per mL.

Standard preparation—Dissolve an accurately weighed quantity of USP Betamethasone Acetate RS in methanol, and dilute quantitatively, and stepwise if necessary, with methanol to obtain a solution having a known concentration of about 0.45 mg per mL. Transfer 2.0 mL of this solution to a 10-mL volumetric flask, add 1.0 mL of *Internal standard solution*, dilute with methanol to volume, and mix to obtain a solution having a known concentration of about 0.09 mg of USP Betamethasone Acetate RS per mL.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Solution, equivalent to about 2 mg of betamethasone acetate, to a 10-mL volumetric flask. Dilute with methanol to volume, and mix. Transfer a portion of this solution to a centrifuge tube, and centrifuge. Transfer 4.0 mL of the clear supernatant to a 10-mL volumetric flask. Add 1.0 mL of *Internal standard solution*, dilute with a mixture of methanol and water (1 : 1) to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-mm detector and a 3.9-mm × 30-cm column that contains packing L1. The flow rate is about 1 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed under *Procedure:* the relative retention times are about 1.3 for *o*-phenylphenol and 1.0 for betamethasone acetate; the resolution, *R*, between the betamethasone acetate and *o*-phenylphenol peaks is not less than 3.9; 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 responses for the major peaks. Calculate the quantity, in mg, of betamethasone acetate ($C_{24}H_{31}FO_6$) in each mL of the Ophthalmic Solution taken by the formula:

$$25(C/V)(R_U/R_S),$$

in which C is the concentration, in mg per mL, of USP Betamethasone Acetate RS, calculated on the anhydrous basis, in the *Standard preparation*; V is the volume, in mL, of Ophthalmic Solution taken to prepare the *Assay preparation*; and R_U and R_S are the ratios of the betamethasone acetate peak response to the internal standard peak response obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Gentamicin Sulfate and Betamethasone Valerate Ointment

» Gentamicin Sulfate and Betamethasone Valerate Ointment contains not less than 90.0 percent and not more than 125.0 percent of the labeled amount of gentamicin and an amount of betamethasone valerate equivalent to not less than 90.0 percent and not more than 110.0 percent of the labeled amount of betamethasone ($C_{22}H_{29}FO_5$).

Packaging and storage—Preserve in collapsible tubes or other tight containers.

Labeling—Label it to indicate that it is for veterinary use only.

USP Reference standards ⟨11⟩—*USP Betamethasone Valerate RS. USP Beclomethasone Dipropionate RS. USP Gentamicin Sulfate RS.*

Identification—

A: Transfer an amount of Ointment, equivalent to about 15 mg of gentamicin, to a centrifuge tube, and add 10 mL of a mixture of methanol and 0.1 N hydrochloric acid (4 : 1) and 25 mL of solvent hexane. Rotate for 30 minutes, and centrifuge. Discard the upper phase. Apply 25 µL of the lower phase and 25 µL of a Standard solution containing 3 mg per mL of USP Gentamicin Sulfate RS in a mixture of methanol and 0.1 N hydrochloric acid (4 : 1) to a suitable thin-layer chromatographic plate (see *Chromatography* ⟨621⟩) coated with a 0.25-mm layer of chromatographic silica gel mixture. Allow the spots to dry, and develop the chromatogram in a solvent system consisting of the lower phase of a mixture of chloroform, methanol, and ammonium hydroxide (1 : 1 : 1) 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 spots to air-dry. Locate the spots on the plate by placing it in a tank containing about 15 g of iodine crystals for 15 minutes: the R_F values of the three principal spots obtained from the test solution correspond to those obtained from the Standard solution.

B: The retention time of the major peak obtained in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation*, both relative to the internal standard, as obtained in the *Assay for betamethasone.*

Microbial limits ⟨61⟩—It meets the requirements of the tests for absence of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella* species, and *Escherichia coli.*

Minimum fill ⟨755⟩: meets the requirements.

Assay for gentamicin—Proceed as directed for gentamicin under *Antibiotics—Microbial Assays* ⟨81⟩, using an accurately weighed quantity of Ointment, equivalent to about 3 mg of gentamicin, shaken with about 50 mL of ether in a separator and extracted with three 25-mL portions of *Buffer No. 3*. Combine the aqueous extracts, and dilute quantitatively and stepwise with *Buffer No. 3* to obtain a *Test Dilution* having a concentration assumed to be equal to the median dose level of the Standard.

USP 28

Assay for betamethason

Mobile phase—Prepa
methanol and water (475
System Suitability under
Diluent—Transfer 25 m
Add 2.5 mL of glacial ac
and mix.

Internal standard solu
methasone Dipropionate R
about 0.4 mg per mL.

Standard preparation—
USP Betamethasone Valer
and stepwise if necessary,
known concentration of ab
this solution to a stoppere
solution, and mix to obtain
of about 0.15 mg of USP I

Assay preparation—Tra
Ointment, equivalent to ab
centrifuge tube. Add 10.0
mL of *Diluent*, and shake vi
an ice–methanol bath for 1
phases. Transfer the clear su
to warm to room temperatu

Chromatographic syste
liquid chromatograph is e
4.6-mm × 15-cm column t
about 2.5 mL per minute. C
and record the peak respons
retention times are about 1.5
for betamethasone valerate;
asone valerate and beclom
than 3.5; and the relative sta
not more than 2.0%.

Procedure—Separately in
Standard preparation and t
ograph, record the chromato
major peaks. Calculate th
($C_{22}H_{29}FO_5$) in the portion o

(392.47 / 4

in which 392.47 and 47
betamethasone and betameth
concentration, in mg per mL,
the *Standard preparation*;,
betamethasone valerate peak
response obtained from the
preparation, respectively.

Gentamicin Sulfat Valerate Otic Solu

» Gentamicin Sulfate an
Solution contains not less
than 125.0 percent of the
and an amount of betam
not less than 90.0 perc
percent of the labelec
($C_{22}H_{29}FO_5$).

Packaging and storage—Pres
Labeling—Label it to indicate
USP Reference standards ⟨1
ate RS. USP Betamethasone

Identification—
A: Transfer an amount of O
gentamicin, to a centrifuge t
antity of USP Gentamicin S
ain a solution having a co
nsfer 1.0 mL of this solu

Developing solvent system: a mixture of ethyl acetate, anhydrous formic acid, and water (67 : 16.5 : 16.5).

Procedure—Proceed as directed for *Thin-Layer Chromatography* under *Chromatography* ⟨621⟩, applying the *Test solution,* the *Standard solution,* and the *Tropine reference solution.* Spray the plate with Dragendorff's reagent, followed by hydrogen peroxide TS, and immediately cover with a glass plate of the same size. Examine the plate no later than 5 to 10 minutes after spraying. In the chromatogram obtained from the *Test solution,* identify the spot corresponding to the principal spot in the chromatogram of the *Tropine reference solution:* this spot is not more intense than the spot obtained from the *Standard solution:* not more than 0.5% of tropine is found. ▲USP28

Add the following:

▲Chromatographic purity—

Buffer solution, Mobile phase, System suitability solution, and *Chromatographic system*—Proceed as directed in the *Assay.*

Standard solution—Use the *Standard preparation,* prepared as directed in the *Assay.*

Test solution—Use the *Assay preparation,* prepared as directed in the *Assay.*

Procedure—Separately inject a volume (about 7 µL) of the *Test solution* into the chromatograph, record the chromatogram, and measure the responses for the major peaks. Continue the elution for 2.2 times the retention time of the homatropine peak. Disregard the peak for the bromide ion, which appears close to the solvent peak. Calculate the percentage of each impurity in the portion of Homatropine Hydrobromide taken by the formula:

$$100(r_i / r_s),$$

in which r_i and r_s are the peak response for each impurity and the sum of all peak responses, respectively, obtained from the *Test solution.* In addition to not exceeding the limits for each impurity in *Table 1,* not more than 0.1% of any other individual impurity is found; and not more than 1.0% of total impurities is found.

Table 1

Impurity	Relative Retention Time	Limit (%)
Mandelic acid	0.3	0.1
Dehydrohomatropine	0.9	0.5
Scopolamine	1.1	0.1
Atropine	1.9	0.1

▲USP28

Change to read:

Assay—

▲*Buffer solution*—Dissolve 6.8 g of monobasic potassium phosphate and 7.0 g of sodium 1-heptanesulfonate monohydrate in 1000 mL of water, adjust with 3 M phosphoric acid to a pH of 2.7, and mix.

Mobile phase—Prepare a filtered and degassed mixture of *Buffer solution* and methanol (67 : 33).

Standard preparation—Dissolve an accurately weighed quantity of USP Homatropine Hydrobromide RS in *Mobile phase* to obtain a solution having a concentration of about 2 mg per mL.

System suitability solution—Prepare a solution of USP Scopolamine Hydrobromide RS having a concentration of about 0.1 mg per mL. Transfer 10 mL of this solution to a 100-mL volumetric flask, add 0.5 mL of the *Standard preparation,* and dilute with *Mobile phase* to volume.

Test preparation—Transfer about 100 mg of Homatropine Hydrobromide, accurately weighed, to a 50-mL volumetric flask, dissolve in and dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 210-nm detector and a 4.6-mm × 10-cm column that contains 3-µm packing L1. The flow rate is about 1.5 mL per minute. The column temperature is maintained at 40°. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the tailing factor is not more than 2.0; and the relative standard deviation for replicate injections is not more than 1.0%. Chromatograph the *System suitability solution,* and record the peak responses as directed for *Procedure:* the resolution between homatropine and scopolamine peaks is not less than 1.5.

Procedure—Separately inject equal volumes (about 7 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 $C_{16}H_{21}NO_3 \cdot HBr$ in the portion of Homatropine Hydrobromide taken by the formula:

$$50C(r_U / r_S),$$

in which C is the concentration, in mg per mL, of USP Homatropine Hydrobromide 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. ▲USP28

Homatropine Hydrobromide Ophthalmic Solution

» Homatropine Hydrobromide Ophthalmic Solution is a sterile, buffered, aqueous solution of Homatropine Hydrobromide. It contains not less than 95.0 percent and not more than 105.0 percent of the labeled amount of $C_{16}H_{21}NO_3 \cdot HBr$. It may contain suitable antimicrobial agents.

Packaging and storage—Preserve in tight containers.

USP Reference standards ⟨11⟩—*USP Homatropine Hydrobromide RS.*

Identification—

A: Proceed with Ophthalmic Solution as directed under *Identification—Organic Nitrogenous Bases* ⟨181⟩. The specified results are obtained.

B: It responds to the tests for *Bromide* ⟨191⟩.

Sterility ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 2.5 and 5.0.

Assay—

Standard preparation—Accurately weigh about 50 mg of USP Homatropine Hydrobromide RS, dissolve in water, and dilute with water in a volumetric flask to 100 mL. Dilute 10.0 mL of this solution with water to 50.0 mL to obtain a solution having a known concentration of about 100 µg per mL. Prepare this solution fresh.

Assay preparation—Transfer a portion of Ophthalmic Solution equivalent to 50 mg of homatropine hydrobromide, to a 100-mL volumetric flask, and dilute with water to volume. Dilute 10.0 mL of this solution with water to 50.0 mL.

Procedure—Transfer duplicate 2-mL portions of the *Standard preparation* and of the *Assay preparation* to separate glass-stoppered 40-mL centrifuge tubes. To one set of two tubes add 3 mL of water and 1 mL of sodium hydroxide solution (1 in 100). Heat these tubes in a boiling water bath for 20 minutes, and allow to cool to room temperature. To the remaining set of tubes, which serve as blanks for the *Standard preparation* and the *Assay preparation,* respectively, add 4 mL of water. To each tube, add 2 mL of approximately 0.2% ceric sulfate in diluted sulfuric acid (prepared by dissolving 12.6 g of ceric ammonium sulfate in 50 mL of water and 3 mL of sulfuric acid and diluting with water to 100 mL) and 20.0 mL of isooctane. Shake by mechanical means for 15 minutes, allow the layers to separate, and remove the isooctane from each tube. Concomitantly determine the absorbances of the isooctane solutions from the hydrolyzed aliquots in 1-cm cells at the wavelength of maximum absorbance at about 242 nm, with a suitable spectrophotometer, against the respective blanks. Calculate the quantity, in mg, of $C_{16}H_{21}NO_3 \cdot HBr$ in the portion of Ophthalmic Solution taken by the formula:

$$0.5C(A_U / A_S),$$

in which C is the concentration, in µg per mL, of USP Homatropine Hydrobromide RS in the *Standard preparation;* and A_U and A_S are the absorbances of the solutions from the *Assay preparation* and the *Standard preparation,* respectively.

Hydroxocobalamin Injection

» Hydroxocobalamin Injection is a sterile solution of Hydroxocobalamin in Water for Injection. It contains not less than 95.0 percent and not more than 115.0 percent of the labeled amount of hydroxocobalamin ($C_{62}H_{89}CoN_{13}O_{15}P$).

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

USP Reference standards ⟨11⟩—*USP Cyanocobalamin RS. USP Endotoxin RS.*

Identification—Dilute 3.0 mL of Injection with pH 4.0 buffer (prepared by dissolving 2.61 g of sodium acetate and 20.5 g of sodium chloride in 5.25 mL of glacial acetic acid and sufficient water to make 1500 mL of solution) to 100 mL: the UV-visible absorption spectrum of this solution exhibits maxima at 352 ± 2 nm and 525 ± 2 nm. The ratio A_{352}/A_{525} is between 2.7 and 3.3.

Bacterial endotoxins ⟨85⟩—It contains not more than 0.4 USP Endotoxin Unit per µg of hydroxocobalamin.

pH ⟨791⟩: between 3.5 and 5.0.

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

Assay—

pH 9.3 Buffer—Dissolve 23.8 g of sodium borate and 402 mg of boric acid in sufficient water to make 1500 mL of solution, and mix.

Standard preparation—Dissolve a suitable quantity of USP Cyanocobalamin RS, accurately weighed, in *pH 9.3 Buffer* and dilute quantitatively, and stepwise if necessary, to obtain a solution having a known concentration of about 30 µg per mL.

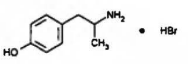
Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 5 mg of hydroxocobalamin, to a 50-mL volumetric flask containing about 25 mL of *pH 9.3 Buffer*. Add 5.0 mL of potassium cyanide solution (1 in 10,000), allow to stand at room temperature for 30 minutes, dilute with *pH 9.3 Buffer* to volume, and mix. Transfer 15.0 mL of this solution to a second 50-mL volumetric flask, dilute with *pH 9.3 Buffer* to volume, and mix.

Procedure—Concomitantly determine the absorbances of the solutions in 1-cm cells at the wavelength of maximum absorbance at about 361 nm, with a suitable spectrophotometer, using *pH 9.3 Buffer* as the blank. Calculate the quantity, in mg, of hydroxocobalamin ($C_{62}H_{89}CoN_{13}O_{15}P$) in each mL of the Injection taken by the formula:

$$(1346.36/1355.37)(0.1667C/V)(A_U/A_S),$$

in which 1346.36 and 1355.37 are the molecular weights of hydroxocobalamin and cyanocobalamin, respectively; C is the concentration, in µg per mL, of USP Cyanocobalamin 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 *Assay preparation* and the *Standard preparation*, respectively.

Hydroxyamphetamine Hydrobromide



$C_9H_{13}NO \cdot HBr$ 232.12

Phenol, 4-(2-aminopropyl)-, hydrobromide.

(±)-*p*-(2-Aminopropyl)phenol hydrobromide [*306-21-8*].

» Hydroxyamphetamine Hydrobromide contains not less than 98.0 percent and not more than 101.5 percent of $C_9H_{13}NO \cdot HBr$, calculated on the dried basis.

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

USP Reference standards ⟨11⟩—*USP Hydroxyamphetamine Hydrobromide RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: Dissolve about 500 mg of ammonium molybdate in 10 mL of sulfuric acid, and add to this solution about 2 mg of Hydroxyamphetamine Hydrobromide: an intense blue color is produced *(distinction from similar amino compounds such as amphetamine and methamphetamine, which, lacking a phenolic hydroxyl, do not undergo this reaction).*

C: Dissolve about 200 mg in 2 mL of water, and add a solution of 500 mg of potassium carbonate in 2 mL of water. Extract with two 10-mL portions of ether, allow the clear ether solution to evaporate to dryness, and dry at about 80°: the hydroxyamphetamine so obtained melts between 124° and 127° (see *Class I* under *Melting Range or Temperature* ⟨741⟩).

D: To a solution of about 10 mg of it in 10 mL of water add 1 mL of 2 N nitric acid, then add silver nitrate TS: a pale yellow precipitate is formed, and it is slightly soluble in 6 N ammonium hydroxide.

Melting range ⟨741⟩: between 189° and 192°.

Loss on drying ⟨731⟩—Dry it at 105° for 2 hours: it loses not more than 0.5% of its weight.

Residue on ignition ⟨281⟩: not more than 0.1%.

Bromide content—Accurately weigh about 400 mg, and dissolve in 50 mL of water. Add 50 mL of methanol and 10 mL of glacial acetic acid, then add eosin Y TS, and titrate with 0.1 N silver nitrate VS. Each mL of 0.1 N silver nitrate is equivalent to 7.990 mg of Br. The content of Br, calculated on the dried basis, is between 33.6% and 35.2%.

Ordinary impurities ⟨466⟩—

Test solution: methanol.

Standard solution: methanol.

Eluant: a mixture of toluene, methanol, and ammonium hydroxide (10 : 4 : 0.25).

Visualization: 1.

Assay—Dissolve about 400 mg of Hydroxyamphetamine Hydrobromide, accurately weighed, in a mixture of 10 mL of glacial acetic acid and 10 mL of mercuric acetate TS, warming slightly, if necessary, to effect solution. Add crystal violet TS, and titrate with 0.1 N perchloric acid VS. Perform a blank determination, and make any necessary correction. Each mL of 0.1 N perchloric acid is equivalent to 23[illegible] mg of $C_9H_{13}NO \cdot HBr$.

Hydroxyamphetamine Hydrobromide Ophthalmic Solution

» Hydroxyamphetamine Hydrobromide Ophthalmic Solution is a sterile, buffered, aqueous solution of Hydroxyamphetamine Hydrobromide. It contains not less than 95.0 percent and not more than 105.0 percent of the labeled amount of $C_9H_{13}NO \cdot HBr$. It contains a suitable antimicrobial agent.

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

USP Reference standards ⟨11⟩—*USP Hydroxyamphetamine Hydrobromide RS.*

Identification—

A: Dissolve about 500 mg of ammonium molybdate in 10 mL of sulfuric acid, and add 0.2 mL of Ophthalmic Solution: an intense blue color is produced *(distinction from similar amino compounds such as amphetamine and methamphetamine, which, lacking a phenolic hydroxyl, do not undergo this reaction).*

B: The dried diacetylhydroxyamphetamine obtained in the Assay melts between 96° and 100° (see *Class I* under *Melting Range or Temperature* ⟨741⟩), but the range between beginning and end of melting does not exceed 2.0°.

C: It responds to *Identification* test *D* under *Hydroxyamphetamine Hydrobromide.*

D: Dilute a volume of Ophthalmic Solution, equivalent to about 50 mg of hydroxyamphetamine hydrobromide, with 0.01 N hyd[illegible]