Aerosol. Calculate the quantity, in mg, of $C_9H_{13}NO_3$ in the Inhalation Aerosol 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 Injection

» Epinephrine Injection is a sterile solution of Epinephrine in Water for Injection prepared with the aid of Hydrochloric Acid or other suitable buffers. It contains not less than 90.0 percent and not more than 115.0 percent of the labeled amount of epinephrine ($C_9H_{13}NO_3$).

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

Labeling—The label indicates that the Injection 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 Endotoxin RS
USP Epinephrine Bitartrate RS

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 Injection *(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: It responds to the *Identification* test under *Epinephrine Nasal 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*.

Bacterial endotoxins ⟨85⟩**—**It contains not more than 357.0 USP Endotoxin Units per mg of epinephrine.

pH ⟨791⟩**:** between 2.2 and 5.0.

Total acidity—Transfer 5.0 mL of Injection to a flask, add 10 mL of water, and titrate with 0.01 N sodium hydroxide VS to a pH of 7.40. Perform a blank determination, and make any necessary correction. Not more than 25.0 mL of 0.01 N sodium hydroxide is required.

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

Assay—

Mobile phase—To 1 L of 0.05 M monobasic sodium phosphate add about 519 mg of sodium 1-octanesulfonate and about 45 mg of edetate disodium, and mix. Adjust by the dropwise addition of phosphoric acid, if necessary, to a pH of 3.8. Mix 85 volumes of this solution with 15 volumes of methanol. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

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

Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 1 mg of epinephrine, to a 10-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

System suitability preparation—Dissolve 10 mg of dopamine hydrochloride in 100 mL of the *Standard preparation*, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 280-nm detector and a 4.6-mm × 15-cm column that contains packing L7. The flow rate is about 2 mL per minute. Chromatograph the *Standard preparation* and the *System suitability preparation*, and record the peak responses as directed for *Procedure:* the relative retention times are about 1.0 for epinephrine and 2.0 for dopamine hydrochloride; the resolution, *R,* between epinephrine and dopamine hydrochloride is not less than 3.5; 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, 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 Injection taken by the formula:

$$(183.20/333.29)(10)(C/V)(r_U / r_S)$$

in which 183.20 and 333.29 are the molecular weights of epinephrine and epinephrine bitartrate, respectively; *C* is the concentration, in mg per mL, of USP Epinephrine Bitartrate RS in the *Standard preparation; V* is the volume, in mL, of Injection taken; and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Epinephrine Inhalation Solution

» Epinephrine Inhalation Solution is a sterile solution of Epinephrine in Purified Water prepared with the aid of Hydrochloric Acid. It contains, in each 100 mL, not less than 0.9 g and not more than 1.15 g of $C_9H_{13}NO_3$.

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

Labeling—The label indicates that the Inhalation 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 Inhalation Solution as the *Test solution*, proceed as directed for *Color and clarity* under *Epinephrine Injection*.

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 speci-

fied 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 1 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