ing line of a suitable thin-layer chromatographic plate (see *Chromatography* ⟨621⟩) coated with a 0.25-mm layer of chromatographic silica gel mixture. Develop the chromatogram in a suitable chamber with a freshly prepared solvent system consisting of a mixture of *n*-hexane, chloroform, methanol, and ammonium hydroxide (60:30:10:1) until the solvent front has moved about three-fourths of the length of the plate. Remove the plate from the chamber, and allow the solvent to evaporate. Locate the spots on the plate by spraying with *Dragendorff's reagent:* the R_F value of one of the principal spots obtained from the test solution corresponds to that obtained from the Standard solution.

Bacterial endotoxins ⟨85⟩**—**It contains not more than 0.10 USP Endotoxin Unit per mg of miconazole.

pH ⟨791⟩: between 3.7 and 5.7.

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

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

Assay—

Mobile phase—Dissolve 5.0 g of ammonium acetate in 200 mL of water, add 300 mL of acetonitrile and 500 mL of methanol, mix, filter, and degas. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Miconazole RS in *Mobile phase* and dilute quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution having a known concentration of about 0.5 mg per mL. Transfer 10.0 mL of this solution to a 100-mL volumetric flask, dilute with *Mobile phase* to volume, and mix to obtain a *Standard preparation* having a known concentration of about 50 µg per mL.

Resolution solution—Dissolve suitable quantities of USP Miconazole RS and dibutyl phthalate in *Mobile phase* to obtain a solution containing about 50 µg of each per mL.

Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 50 mg of miconazole, to a 100-mL volumetric flask, dilute with *Mobile phase* 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.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 230-nm detector and a 4.6-mm × 30-cm column that contains packing L7. The flow rate is about 2 mL per minute. Chromatograph the *Resolution solution* and the *Standard preparation*, and record the peak responses as directed under *Procedure:* the resolution, *R,* between the dibutyl phthalate and miconazole peaks is not less than 5.0, the tailing factor for the miconazole peak is not more than 1.3, and the relative standard deviation for replicate injections of the *Standard preparation* is not more than 2.0%. The relative retention times are about 0.7 for dibutyl phthalate and 1.0 for miconazole.

Procedure—[NOTE—Allow the chromatograph to run for at least 16 to 18 minutes between injections to allow for elution of all components associated with the Injection vehicle.] 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 miconazole ($C_{18}H_{14}Cl_4N_2O$) in each mL of the Injection taken by the formula:

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

in which *C* is the concentration, in µg per mL, of USP Miconazole 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.

Miconazole Nitrate

$C_{18}H_{14}Cl_4N_2O \cdot HNO_3$ 479.14

1*H*-Imidazole, 1-[2-(2,4-dichlorophenyl)-2-[(2,4-dichlorophenyl)methoxy]ethyl]-, mononitrate.

1-[2,4-Dichloro-β-[(2,4-dichlorobenzyl)oxy]phenethyl]imidazole mononitrate [22832-87-7].

» Miconazole Nitrate contains not less than 98.0 percent and not more than 102.0 percent of $C_{18}H_{14}Cl_4N_2O \cdot HNO_3$, calculated on the dried basis.

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

USP Reference standards ⟨11⟩**—**
USP Econazole Nitrate RS
USP Miconazole Nitrate RS

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: *Ultraviolet Absorption* ⟨197U⟩—

Solution: 400 µg per mL.

Medium: 0.1 N hydrochloric acid in isopropyl alcohol (1 in 10).

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 0.2%.

Related compounds—

Mobile phase—Prepare a filtered and degassed mixture of 0.2 M ammonium acetate, methanol, and acetonitrile (38:32:30). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Resolution solution—Dissolve accurately weighed quantities of USP Miconazole Nitrate RS and USP Econazole Nitrate RS in *Mobile phase* to obtain a solution having known concentrations of about 25 µg of each per mL.

Test solution—Transfer 100 mg of Miconazole Nitrate to a 10-mL volumetric flask, add *Mobile phase* to volume, and mix.

Diluted test solution—Dilute an accurately measured volume of the *Test solution* quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution containing 25 µg of miconazole nitrate per mL.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 235-nm detector and a 4.6-mm × 10-cm column that contains 3-µm packing L1. The flow rate is about 2 mL per minute. Chromatograph the *Resolution solution*, and record the peak responses as directed for *Procedure:* the relative retention times are about 0.5 for econazole and 1.0 for miconazole; the resolution, *R,* between econazole and miconazole is not less than 10; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 10 µL) of the *Test solution* and *Diluted test solution* into the chromatograph, record the chromatograms for a time that is 1.2 times the retention time of the main peak, and measure the responses of all peaks, excluding the peak representing nitrate ion and any peak producing a response less than 0.2 times the response of the main peak. The response of any individual peak, other than the main peak in the chromatogram of the *Test solution*, is not greater than that of the main peak in the chromatogram of the *Diluted test solution* (0.25%), and the sum of the responses of all peaks, other than the main peak in the chromatogram of the *Test solution*, is not greater than twice the response of the main peak in the chromatogram of the *Diluted test solution* (0.5%).

Assay—Dissolve about 350 mg of Miconazole Nitrate, accurately weighed, in 50 mL of glacial acetic acid, and titrate with 0.1 N perchloric acid VS, determining the endpoint potentiometrically, using a glass-calomel electrode system. Perform a blank determination, and make any necessary correction. Each

mL of 0.1 N perchloric acid is equivalent to 47.92 mg of $C_{18}H_{14}Cl_4N_2O \cdot HNO_3$.

Miconazole Nitrate Cream

» Miconazole Nitrate Cream contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of miconazole nitrate ($C_{18}H_{14}Cl_4N_2O \cdot HNO_3$).

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

Labeling—Cream that is packaged and labeled for use as a vaginal preparation shall be labeled Miconazole Nitrate Vaginal Cream.

USP Reference standards ⟨11⟩**—**
USP Miconazole Nitrate 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*.

Minimum fill ⟨755⟩**:** meets the requirements.

Assay—

Buffer solution—Transfer 10 mL of triethylamine to a suitable flask, dilute with 1000 mL of water, adjust with phosphoric acid to a pH of about 2.5, and mix.

Mobile phase—Prepare a filtered and degassed mixture of *Buffer solution,* methanol, acetonitrile, and tetrahydrofuran (8:5:4:3). Make adjustments if necessary (see *System Suitablility* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Miconazole Nitrate RS and benzoic acid in *Mobile phase,* and dilute quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution having a known concentration of about 0.28 and 0.02 mg per mL for miconazole nitrate and benzoic acid, respectively.

Assay preparation—Transfer an accurately weighed quantity of Cream, equivalent to about 14 mg of miconazole nitrate, to a 50-mL volumetric flask, dissolve in and dilute with *Mobile phase* to volume, and mix. Sonicate in a water bath at 40° to 45° until the sample is completely dispersed, and mix. Cool the solution to below room temperature, mix, and pass a portion of the solution through a 0.45-µm teflon filter into an HPLC vial.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 225-nm detector and 4.6-mm × 25-cm column that contains packing L11. The flow rate is about 1.0 mL per minute. The column temperature is maintained at 45°. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the column efficiency for miconazole nitrate peak is not less than 7500 theoretical plates; the tailing factor for miconazole nitrate peak is not more than 2.0; and the relative standard deviation for replicate injections of miconazole nitrate is not more than 2.0%. The resolution between miconazole nitrate and benzoic acid is not less than 13.

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 miconazole nitrate ($C_{18}H_{14}Cl_4N_2O \cdot HNO_3$) in the portion of Cream taken by the formula:

$$50C(r_U / r_S)$$

in which C is the concentration, in mg per mL, of USP Miconazole Nitrate 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.

Miconazole Nitrate Topical Powder

» Miconazole Nitrate Topical Powder contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of miconazole nitrate ($C_{18}H_{14}Cl_4N_2O \cdot HNO_3$).

Packaging and storage—Preserve in well-closed containers.

USP Reference standards ⟨11⟩**—**
USP Miconazole Nitrate RS

Identification—Transfer a portion of Topical Powder, equivalent to about 100 mg of miconazole nitrate to a 50-mL beaker, disperse in 40 mL of methanol, and mix for a minimum of 5 minutes. Allow to settle for 5 to 10 minutes, and filter into a 100-mL beaker. Evaporate on a steam bath to dryness. Dry the residue at 105° for 10 minutes: the IR absorption spectrum of a potassium bromide dispersion of the residue so obtained exhibits maxima only at the same wavelengths as that of a similar preparation of USP Miconazole Nitrate RS.

Microbial enumeration tests ⟨61⟩ **and Tests for specified microorganisms** ⟨62⟩**—**The total count does not exceed 100 microorganisms per g, and tests for *Staphylococcus aureus*, and *Pseudomonas aeruginosa*, are negative.

Minimum fill ⟨755⟩: meets the requirements.

Assay—

Internal standard solution—Dissolve cholestane in chloroform to obtain a solution having a concentration of about 0.5 mg per mL.

Standard preparation—Dissolve an accurately weighed quantity of USP Miconazole Nitrate RS in a mixture of chloroform and methanol (1:1) to obtain a solution having a known concentration of about 0.8 mg per mL. Transfer 5.0 mL of this solution to a test tube, add 2.0 mL of *Internal standard solution*, and evaporate at a temperature not higher than 40° with the aid of a current of nitrogen to dryness. Dissolve the residue in 2.0 mL of a mixture of chloroform and methanol (1:1), and mix to obtain a *Standard preparation* having a known miconazole nitrate concentration of about 2 mg per mL.

Assay preparation—Transfer an accurately weighed portion of Topical Powder, equivalent to about 20 mg of miconazole nitrate, to a stoppered 50-mL centrifuge tube. Add 25.0 mL of methanol, and shake by mechanical means for 30 minutes to dissolve the miconazole nitrate. Centrifuge to obtain a clear supernatant. Transfer 5.0 mL of this solution to a test tube, add 2.0 mL of *Internal standard solution,* and evaporate at a temperature not higher than 40° with the aid of a current of nitrogen to dryness. Dissolve the residue in 2.0 mL of a mixture of chloroform and methanol (1:1).

Chromatographic system (see *Chromatography* ⟨621⟩)—The gas chromatograph is equipped with a flame-ionization detector and a 1.2-m × 2-mm glass column containing 3 percent phase G32 on support S1A. The injection port, detector, and column are maintained at temperatures of about 250°, 300°, and 250°, respectively. Helium is used as the carrier gas, at a flow rate of about 50 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed under *Procedure:* the resolution, *R,* between the cholestane and miconazole nitrate peaks is not less than 2.0, and the relative standard deviation for replicate injections is not more than 3.0%.

Procedure—Separately inject equal volumes (about 5 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 for cholestane and miconazole nitrate are about 0.5 and 1.0, respectively. Calculate the quantity, in mg, of miconazole nitrate