spectrum of a potassium bromide dispersion of the crystals exhibits maxima only at the same wavelengths as that of a similar preparation of USP Prednisolone Acetate RS.

B: To the filtrate saved from *Identification* test *A*, add 6 N acetic acid dropwise until the pH is between 4 and 5. Allow crystals of sulfacetamide to develop. Filter the crystals, wash with a small amount of water, and dry at 105° for 2 hours: the IR absorption spectrum of a potassium bromide dispersion of the crystals so obtained exhibits maxima only at the same wavelengths as a preparation of USP Sulfacetamide Sodium RS, similarly treated.

Sterility ⟨71⟩: meets the requirements.

pH ⟨791⟩: between 6.0 and 7.4.

Assay for sulfacetamide sodium—

Mobile phase—Prepare a filtered and degassed mixture of water, methanol, and glacial acetic acid (890:100:10). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Sulfacetamide Sodium RS in a mixture of water and methanol (4:1), and dilute quantitatively, and stepwise if necessary, with the same solvent mixture to obtain a solution having a known concentration of about 30 µg per mL.

Assay preparation—Transfer an accurately measured volume of Ophthalmic Suspension, freshly mixed and free from air bubbles, equivalent to about 100 mg of sulfacetamide sodium, to a 100-mL volumetric flask, dilute with a mixture of water and methanol (4:1) to volume, and mix. Dilute 3.0 mL of this solution with the same solvent mixture to 100.0 mL, and mix.

System suitability preparation—Dissolve about 3 mg of sulfanilamide in 100 mL of the *Standard preparation*, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4.6-mm × 25-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. Chromatograph the *Standard preparation* and the *System suitability preparation*, and record the peak responses as directed for *Procedure:* the column efficiency determined for the analyte peak is not less than 1500 theoretical plates; the resolution, *R*, between the sulfacetamide and sulfanilamide peaks is not less than 3; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 90 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 sulfacetamide sodium ($C_8H_9N_2NaO_3S.H_2O$) in each mL of the Ophthalmic Suspension taken by the formula:

$$3.33(254.24 / 236.23)C(r_U / r_S)$$

in which 254.24 and 236.23 are the molecular weights of sulfacetamide sodium monohydrate and anhydrous sulfacetamide sodium, respectively; *C* is the concentration, in µg per mL, calculated on the anhydrous basis, of USP Sulfacetamide Sodium 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.

Assay for prednisolone acetate—

Mobile phase—Prepare a filtered and degassed mixture of water and acetonitrile (60:40). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Prednisolone Acetate RS in methanol to obtain a solution containing about 2 mg per mL. Transfer 2.0 mL of this solution to a 100-mL volumetric flask, and dilute with a solvent mixture prepared by dissolving 2.72 g of monobasic potassium phosphate in 300 mL of water and 700 mL of methanol. The *Standard preparation* has a known concentration of about 0.04 mg per mL.

Assay preparation—Using a "To contain" pipet, transfer an accurately measured volume of Ophthalmic Suspension, freshly mixed and free from air bubbles, equivalent to about 10 mg of prednisolone acetate, to a 250-mL volumetric flask. Rinse the pipet with the solvent mixture described under *Standard preparation*, collecting the rinsings in the flask, dilute with the same solvent mixture to volume, and mix.

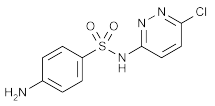
Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4.0-mm × 30-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. Chromatograph the *Standard preparation*, and record the peak responses as directed for *Procedure:* the column efficiency determined from the analyte peak is not less than 3000 theoretical plates; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 30 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 prednisolone acetate ($C_{23}H_{30}O_6$) in each mL of the Ophthalmic Suspension taken by the formula:

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

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

Sulfachlorpyridazine



$C_{10}H_9ClN_4O_2S$ 284.72

N[1]-(6-Chloro-3-pyridazinyl)sulfanilamide [80-32-0].

» Sulfachlorpyridazine contains not less than 97.0 percent and not more than 103.0 percent of $C_{10}H_9ClN_4O_2S$, calculated on the dried basis.

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

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

USP Reference standards ⟨11⟩**—**
USP Sulfachlorpyridazine RS

Identification—

A: *Infrared Absorption* ⟨197M⟩.

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

Clarity and color of solution—Dissolve 1.0 g of it in 50 mL of 0.1 N methanolic hydrochloric acid prepared by diluting 8.6 mL of hydrochloric acid with methanol to obtain 1000 mL of solvent: a clear solution is produced that is not deeper in color than pale yellow.

Acidity—Prepare a suspension of 3.0 g of it in 150.0 mL of carbon dioxide-free water, and heat at 70° for 5 minutes, maintaining the suspension. Cool rapidly in an ice bath to

$20 \pm 0.5°$, stirring by mechanical means. Filter the suspension using vacuum, and collect the filtrate. Titrate 25.0 mL of the clear filtrate with 0.1 N sodium hydroxide VS, using 2 drops of thymolphthalein TS as the indicator. Transfer a second 25.0-mL portion of the clear filtrate to a 250-mL conical flask, add 10 mL of hydrochloric acid, and cool in an ice bath to 15°. Add about 25 g of crushed ice, prepared from frozen purified water, and titrate with 0.1 M sodium nitrite VS, stirring vigorously, until the titrated solution produces an immediate, stable, blue color on starch-iodide paper. The volume of 0.1 N sodium hydroxide consumed in the titration of the first 25.0-mL portion of the filtrate does not exceed the volume of 0.1 M sodium nitrite consumed in the titration of the second 25.0-mL portion of the filtrate by more than 0.5 mL.

Loss on drying ⟨731⟩: Dry it at 105° 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⟩: 0.002%.

Assay—

pH 2.5 phosphate buffer—Dissolve 14 g of monobasic potassium phosphate in 1600 mL of water, adjust with phosphoric acid to a pH of 2.5 ± 0.1, dilute with water to 2000 mL, and mix.

Mobile phase—Prepare a filtered and degassed mixture of *pH 2.5 phosphate buffer* and methanol (700:300). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Prepare a stock solution of USP Sulfachlorpyridazine RS in methanol having a known concentration of about 0.5 mg per mL. Transfer 3.0 mL of this stock solution to a 100-mL volumetric flask, dilute with *Mobile phase* to volume, and mix. Filter this solution through a nylon filter having a porosity of 0.5 µm or finer, and use the filtrate as the *Standard preparation.* The *Standard preparation* contains about 15 µg of USP Sulfachlorpyridazine RS per mL.

Assay preparation—Transfer about 50 mg of Sulfachlorpyridazine, accurately weighed, to a 100-mL volumetric flask. Dissolve in and dilute with methanol to volume, and mix. Transfer 3.0 mL of this solution to a second 100-mL volumetric flask, dilute with *Mobile phase* to volume, and mix. Filter this solution through a filter having a porosity of 0.5 µm or finer, and use the filtrate as the *Assay preparation*.

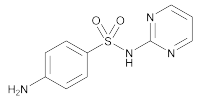
Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 265-nm detector, a 4.6-mm × 25-cm analytical column containing 5-µm packing L1, and a guard column containing 5-µm packing L1, and is maintained at about 40°. The flow rate is about 1 mL per minute. Chromatograph 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 responses for the major peaks. Calculate the quantity, in mg, of $C_{10}H_9ClN_4O_2S$ in the portion of Sulfachlorpyridazine taken by the formula:

$$(10 / 3)(C)(r_U / r_S)$$

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

Sulfadiazine



$C_{10}H_{10}N_4O_2S$ 250.28

Benzenesulfonamide, 4-amino-*N*-2-pyrimidinyl-; *N*[1]-2-Pyrimidinylsulfanilamide [68-35-9].

DEFINITION

Sulfadiazine contains NLT 98.0% and NMT 102.0% of $C_{10}H_{10}N_4O_2S$, calculated on the dried basis.

IDENTIFICATION

- **A. Infrared Absorption** ⟨197K⟩
- **B.**

Sample: 50 mg

Analysis: Carefully melt the *Sample* in a small test tube.

Acceptance criteria: A reddish brown color develops upon melting, and the fumes evolved during decomposition do not discolor moistened lead acetate test paper (distinction from sulfathiazole).

- **C.**

Sample: 1 g

Analysis 1: Gently heat the *Sample* in a small test tube until a sublimate is formed. Collect a few mg of the sublimate with a glass rod, and mix in a test tube with 1 mL of a solution (1 in 20) of resorcinol in alcohol. Add 1 mL of sulfuric acid, and mix by shaking.

Acceptance criteria 1: A deep red color appears at once.

Analysis 2: Cautiously dilute the mixture obtained in *Analysis 1* with 25 mL of ice-cold water, and add an excess of 6 N ammonium hydroxide.

Acceptance criteria 2: A blue or reddish blue color is produced.

ASSAY

- **Procedure**

Mobile phase: Acetonitrile, water, and glacial acetic acid (12:87:1)

Standard solution: 1 mg/mL of USP Sulfadiazine RS in 0.025 N sodium hydroxide

Sample solution: 1 mg/mL of Sulfadiazine in 0.025 N sodium hydroxide

Chromatographic system

(See *Chromatography* ⟨621⟩*, System Suitability*.)

Mode: LC

Detector: UV 254 nm

Column: 4-mm × 30-cm; packing L1

Flow rate: 2 mL/min

Injection volume: 10 µL

System suitability

Sample: *Standard solution*

Suitability requirements

Tailing factor: NMT 1.5

Relative standard deviation: NMT 2.0%, five injections

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of sulfadiazine ($C_{10}H_{10}N_4O_2S$) in the portion of Sulfadiazine taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times 100$$

r_U = peak response of sulfadiazine from the *Sample solution*

r_S = peak response of sulfadiazine from the *Standard solution*

C_S = concentration of USP Sulfadiazine RS in the *Standard solution* (mg/mL)