

(iv) Its heavy metals content is not more than 30 parts per million.

(v) It gives a positive identity test for vancomycin.

(2) *Labeling.* It shall be labeled in accordance with the requirements of § 432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for potency, chromatographic purity, moisture, heavy metals, and identity.

(ii) Samples required: 12 packages, each containing approximately 500 milligrams.

(b) *Tests and methods of assay—(1) Potency.* Proceed as directed in § 436.105 of this chapter, preparing the sample for assay as follows: Place an accurately weighed sample of approximately 100 milligrams in a 100-milliliter volumetric flask and dissolve in approximately 50 milliliters of distilled water and 1.0 milliliter of 0.1N hydrochloric acid. Swirl or sonicate to dissolve the sample and bring to volume with distilled water. Further dilute an aliquot of this solution with 0.1M potassium phosphate buffer, pH 4.5 (solution 4), to the reference concentration of 10 micrograms of vancomycin per milliliter (estimated).

(2) *Chromatographic purity.* Proceed as directed in § 436.366 of this chapter. The relative amount of vancomycin B is not less than 92 percent, and the relative amount of any related substance is not more than 3 percent.

(3) *Moisture.* Proceed as directed in § 436.201 of this chapter.

(4) *Heavy metals.* Proceed as directed in § 436.208 of this chapter.

(5) *Identity.* Proceed as directed in § 436.211 of this chapter, using the 0.5 percent potassium bromide disc preparation as described in § 436.211(b)(1).

[59 FR 8400, Feb. 22, 1994]

§ 455.88 Rifabutin.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Rifabutin is an amorphous red-violet powder. It is (9S, 12E, 14S, 15R, 16S, 17R, 18R, 19R, 20S, 21S, 22E, 24Z)-6,16,18, 20-tetrahydroxy-1'-isobutyl-14-methoxy-7, 9,15,17,19,21, 25-

heptamethylspiro[9,4-(epoxypentadeca[1,11,13]trienimino)-2H-furo[2', 3':7,8] naphth[1,2-d]imidazole-2, 4'-piperidine]-5,10, 26-(3H, 9H)-trione-16-acetate. It is very slightly soluble in water, sparingly soluble in ethanol, and soluble in chloroform and methanol. It is so purified and dried that:

(i) Its potency is not less than 950 micrograms and not more than 1,020 micrograms of rifabutin activity per milligram on an anhydrous basis.

(ii) Its content for the four major related substances detected by high-performance liquid chromatography (HPLC) is not more than 1.0 percent each. All other unknown related substances are not more than 0.5 percent. The total of all related substances is not more than 3.0 percent.

(iii) Its moisture content is not more than 2.5 percent.

(iv) Its *N*-isobutylpiperidone content is not more than 0.5 percent.

(v) It gives a positive identity test.

(2) *Labeling.* It shall be labeled in accordance with the requirements of § 432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for rifabutin potency, related substances, moisture, *N*-isobutylpiperidone, and identity.

(ii) Samples, if required by the Director, Center for Drug Evaluation and Research: 10 packages each containing approximately 300 milligrams.

(b) *Tests and methods of assay—(1) Potency.* Proceed as directed in § 436.216 of this chapter, using ambient temperature, an ultraviolet detection system operating at a wavelength of 254 ± 1 nanometers, an 11 centimeters X 4.7 millimeters (i.d.) column packed with microparticulate (5 to 7 micrometers in diameter) packing material such as octylsilane chemically bonded to porous silica (U.S. Pharmacopeia designation L7), a flow rate of about 1.0 milliliter per minute, and a manual or automatic injector capable of injecting 10 microliters. The retention time for rifabutin is between 9 and 11 minutes. Reagents; working standard, sample,

and resolution solutions; system suitability requirements; and calculations are as follows:

(i) *Reagents*—(A) *Hydrochloric acid, 2N*. Dilute 85 milliliters of hydrochloric acid (37 percent) with distilled water to 500 milliliters.

(B) *Potassium dihydrogen phosphate, 0.1M*. Prepare a solution containing 15.4 grams of potassium dihydrogen phosphate monohydrate (potassium phosphate monobasic) per liter of distilled water.

(C) *Sodium hydroxide, 2N*. Dissolve 8 grams of sodium hydroxide pellets in 100 milliliters of distilled water.

(D) *Mobile phase*. Acetonitrile:phosphate buffer, pH 6.5, 50:50. Mix equal quantities of acetonitrile and 0.1M potassium dihydrogen phosphate and adjust to an apparent pH of 6.5 ± 0.1 by dropwise addition of 2N sodium hydroxide. Filter through a suitable filter capable of removing particulate matter 0.5 micron in diameter and degas it just prior to its introduction into the chromatograph. Slight adjustments of the mobile phase components ratio may be made in order to meet the system suitability requirements described in the system suitability tests in paragraph (b)(1)(iii) of this section.

(ii) *Preparation of working standard, sample, and resolution test solution*—(A) *Working standard solution*. Accurately weigh approximately 25 milligrams of the rifabutin working reference standard into a 50-milliliter volumetric flask. Add 5 milliliters of acetonitrile. Dissolve and dilute to volume with mobile phase and mix to obtain a solution having a known concentration of about 0.5 milligram of rifabutin per milliliter.

(B) *Sample solution*. Accurately weigh approximately 25 milligrams of sample into a 50-milliliter volumetric flask. Add 5 milliliters of acetonitrile. Dissolve and dilute to volume with mobile phase and mix to obtain a solution containing 0.5 milligram of rifabutin per milliliter (estimated).

(C) *Resolution test solution*. Dissolve approximately 10 milligrams of rifabutin in 2 milliliters of methanol and add 1 milliliter of 2N sodium hydroxide. Allow to stand for 3 to 4 minutes and then add 1 milliliter of 2N hydrochloric acid. Mix and dilute to 50

milliliters with mobile phase. Store aliquots of this solution in the frozen state for future use.

(iii) *System suitability requirements*. Using the apparatus and conditions described in this section, test the chromatographic system by injecting the resolution test solution. The chromatogram shows one major degradation peak and two minor degradation peaks eluting at relative retention times (RRT) of 0.5–0.6, 0.65–0.75, and 0.8–0.9, respectively, followed by the rifabutin peak.

(A) *Asymmetry factor*. The asymmetry factor (A_s) is satisfactory if it is not less than 1.0 and not more than 4.0 further if a butin peak.

(B) *Efficiency of the column*. The absolute efficiency (h) is satisfactory if it is not more than 11 for the rifabutin peak, equivalent to 2,000 theoretical plates for a 11-centimeter column of 5-micrometer particles.

(C) *Resolution factor*. The resolution factor (R) between the peak for rifabutin and its closest eluting degradation product (generated in situ as described in paragraph (b)(1)(iii) of this section and eluting at RRT of 0.8–0.9) is satisfactory if it is not less than 1.3.

(D) *Coefficient of variation (relative standard deviation)*. The coefficient of variation (S_R in percent of 5 replicate injections of the rifabutin working standard solution) is satisfactory if it is not more than 2.0 percent. If the system suitability parameters have been met, then proceed as described in § 436.216(b) of this chapter.

(iv) *Calculations*. Calculate the micrograms of rifabutin per milligram of sample on an anhydrous basis as follows:

$$\frac{\text{Micrograms of rifabutin}}{\text{per milligram}} = \frac{A_U \times P_s \times 100}{A_s \times C_U \times (100 - m)}$$

where:

A_U = Area of the rifabutin peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_s = Area of the rifabutin peak in the chromatogram of the rifabutin working standard;

P_s = Rifabutin activity in the rifabutin working standard solution in micrograms per milliliter;

C_U = Milligrams of sample per milliliter of

sample solution; and
m = Percent moisture content of the sample.

(2) *Related substances.* Proceed as directed in paragraph (b)(1) of this section for potency using the sample prepared as described in paragraph (b)(1)(ii)(B) of this section and calculating the amounts of related substances as follows.

(i) *Calculations.* Calculate the percentage of related substances as follows:

$$\text{Percent individual HPLC-related substance} = \frac{A_i \times 100}{A_t}$$

$$\text{Percent total HPLC-related substances} = \frac{A \times 100}{A_t}$$

where:

A_i = Area of the individual related substance peak;

A = The sum of areas of all peaks minus the area due to the rifabutin peak and solvent front peak; and

A_t = The sum of areas of all peaks in the chromatogram excluding the solvent peak.

(ii) [Reserved]

(3) *Moisture.* Proceed as directed in § 436.201 of this chapter.

(4) *N-Isobutylpiperidone.* Proceed as directed in § 436.369 of this chapter.

(5) *Identity.* (i) Proceed as directed in § 436.211 of this chapter, using the sample preparation method described in paragraph (b)(1) of that section using a 1 to 2 percent mixture in potassium bromide.

(ii) The identity of rifabutin is confirmed by the qualitative comparison of the HPLC of the sample to the rifabutin working standard as directed in paragraph (b)(1) of this section.

[59 FR 40807, Aug. 10, 1994; 59 FR 46479, Sept. 8, 1994]

§ 455.90a Sterile vidarabine monohydrate.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Vidarabine monohydrate is the monohydrate form of 9-β - D - arabinofuranosyl - 9H - purin - 6-amine. It is a white to off-white powder. It is so purified and dried that:

(i) Its vidarabine content is not less than 845 micrograms and not more than 985 micrograms of vidarabine per milligram.

(ii) It is sterile.

(iii) [Reserved]

(iv) Its loss on drying is not less than 5 percent and not more than 7 percent.

(v) Its specific rotation in dimethylformamide at 25° C is -60.5°±4.5°.

(vi) It passes the identity test for vidarabine.

(2) *Labeling.* In addition to the labeling requirements prescribed by § 432.5(b) of this chapter, this drug shall be labeled "vidarabine".

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for vidarabine content, sterility, loss on drying, specific rotation, and identity.

(ii) Samples required:

(a) For all tests except sterility: 10 packages, each containing approximately 500 milligrams.

(b) For sterility testing: 20 packages, each containing approximately 200 milligrams.

(b) *Tests and methods of assay—(1) Vidarabine content.* Proceed as directed in § 436.325 of this chapter.

(2) *Sterility.* Proceed as directed in § 436.20 of this chapter, using the method described in paragraph (e)(2) of that section, except use 100 milligrams in lieu of 300 milligrams.

(3) [Reserved]

(4) *Loss on drying.* Proceed as directed in § 436.200(e) of this chapter.

(5) *Specific rotation.* Using a solution containing 10 milligrams of vidarabine per milliliter in dimethylformamide and a polarimeter tube 1.0 decimeter in length, proceed as directed in § 436.210 of this chapter, except determine the specific rotation at 365 nanometers.

(6) *Identity.* Proceed as directed in § 436.211 of this chapter, using the 0.5 percent potassium bromide disc prepared as described in paragraph (b)(1) of that section.

[42 FR 44224, Sept. 2, 1977; 43 FR 9802, Mar. 10, 1978, as amended at 44 FR 30334, May 25, 1979; 50 FR 19921, May 13, 1985]