

(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:

(A) The azithromycin used in making the batch for potency, moisture, pH, residue on ignition, heavy metals, specific rotation, crystallinity, and identity.

(B) The batch for content, moisture, pH, and identity.

(ii) Samples, if required by the Director, Center for Drug Evaluation and Research:

(A) The azithromycin used in making the batch: 10 packages, each containing approximately 1,000 milligrams.

(B) The batch: A minimum of 30 packages.

(b) *Tests and methods of assay—(1) Azithromycin content.* Proceed as directed in § 452.60(b)(1), preparing the dissolving solvent and sample solution and calculating the azithromycin content as follows:

(i) *Dissolving solvent.* Dissolve 2.2 grams of potassium phosphate monobasic in 1,590 milliliters of ultrapure deionized or high-performance liquid chromatographic-grade water. Add 600 milliliters of 2-propanol, 480 milliliters of ethanol, and 330 milliliters of acetonitrile, adjust to pH 8.4 with 10M potassium hydroxide and shake on a reciprocating shaker for 30 minutes. The dissolving solvent is 0.01M monobasic potassium phosphate:2-propanol:ethanol:acetonitrile (53:20:16:11, by volume).

(ii) *Preparation of sample solution.* Quantitatively transfer the contents of one package into a 500-milliliter volumetric flask. Add about 350 milliliters of dissolving solvent and shake on a reciprocating shaker for 30 minutes. Dilute to volume with dissolving solvent, stopper the flask, and mix well. Place 40 milliliters of the resulting suspension into a suitably sized centrifuge tube. Stopper the tube and centrifuge the suspension (about 20 minutes at 1,000 revolutions per minute). Pipet 10.0 milliliters of the diluted solution into a 50-milliliter volumetric flask and dilute to volume with mobile phase (de-

scribed in § 452.60(b)(1)(i)). Pipet 2.0 milliliters of the diluted solution into a 50-milliliter volumetric flask and dilute to volume with mobile phase. The final dilution of the sample and standard must be identical. The final concentration of azithromycin in the sample solution is 0.003 milligram per milliliter (estimated).

(iii) *Calculations.* Calculate the azithromycin content as follows:

$$\text{Milligrams of azithromycin per package} = \frac{A_U \times P_s \times d}{A_s \times 1,000}$$

where:

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

$A_s$  = Area of the azithromycin peak in the chromatogram of the azithromycin working standard;

$P_s$  = Azithromycin activity in the azithromycin working standard solution in micrograms per milliliter; and

$d$  = Dilution factor of the sample =  $500 \times 50 / 10 \times 50 / 10 \times 50 / 2$ .

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

(3) *pH.* Proceed as directed in § 436.202 of this chapter, using the drug constituted as directed in the labeling. Allow the constituted suspension to sit for 10 minutes undisturbed before making the measurement.

(4) *Identity.* Using the high-performance liquid chromatographic procedure described in paragraph (b)(1) of this section, the retention time for the peak of the active ingredient must be within 2 percent of the retention time for the peak of the corresponding reference standard.

[59 FR 52078, Oct. 14, 1994]

#### § 452.175 Troleandomycin oral dosage forms.

##### § 452.175a Troleandomycin capsules.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Troleandomycin capsules are capsules composed of troleandomycin and one or more suitable buffers, diluents, binders, lubricants, and colorings. Each capsule contains 125 milligrams or 250 milligrams

of troleandomycin. Its potency is satisfactory if it is not less than 90 percent and not more than 120 percent of the number of milligrams of troleandomycin that it is represented to contain. The loss on drying is not more than 5 percent. The troleandomycin used conforms to the standards prescribed by § 452.75(a)(1).

(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 the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on:

(a) The troleandomycin used in making the batch for potency, loss on drying, pH, residue on ignition, identity,  $R_f$  value, acetyl value (only if more than one spot is present in the determination of  $R_f$  value), and crystallinity.

(b) The batch for potency and loss on drying.

(ii) Samples required:

(a) The troleandomycin used in making the batch: 10 packages, nine containing approximately equal portions of not less than 500 milligrams and one containing not less than 2 grams.

(b) The batch: A minimum of 30 capsules.

(b) *Tests and methods of assay—(1) Potency.* Proceed as directed in § 436.106 of this chapter, preparing the sample for assay as follows: Place a representative number of capsules into a high-speed glass blender jar containing sufficient 80 percent isopropyl alcohol solution (solution 15) to obtain a stock solution containing 1,000 micrograms of troleandomycin per milliliter (estimated). Blend for 3 to 5 minutes. Further dilute an aliquot of the stock solution with distilled water to the reference concentration of 25 micrograms of troleandomycin per milliliter (estimated).

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

[39 FR 19149, May 30, 1974, as amended at 48 FR 3960, Jan. 28, 1983; 50 FR 19921, May 13, 1985]

#### § 452.175b Troleandomycin oral suspension.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Troleandomycin oral suspension is troleandomycin and one or more suitable buffers, dispersants, flavorings, colorings, and preservatives suspended in a suitable and harmless vehicle. Each milliliter contains 25 milligrams of troleandomycin. Its potency is satisfactory if it is not less than 90 percent and not more than 125 percent of the number of milligrams of troleandomycin that it is represented to contain. Its pH is not less than 5.0 and not more than 8.0. The troleandomycin used conforms to the standards prescribed by § 452.75(a)(1).

(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 the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on:

(a) The troleandomycin used in making the batch for potency, loss on drying, pH, residue on ignition, identity,  $R_f$  value, acetyl value (only if more than one spot is present in the determination of  $R_f$  value), and crystallinity.

(b) The batch for potency and pH.

(ii) Samples required:

(a) The troleandomycin used in making the batch: 10 packages, nine containing approximately equal portions of not less than 500 milligrams and one containing not less than 2 grams.

(b) The batch: A minimum of five immediate containers.

(b) *Tests and methods of assay—(1) Potency.* Proceed as directed in § 436.106 of this chapter, preparing the sample for assay as follows: Dilute an accurately measured representative portion of the sample with 80 percent isopropyl alcohol solution (solution 15) to obtain a stock solution containing 1,000 micrograms of troleandomycin per milliliter (estimated). Further dilute an aliquot of the stock solution with distilled water to the reference concentration of 25 micrograms of troleandomycin per milliliter (estimated).