

(ii) *Assay procedures.* Use any of the following methods; however, the results obtained from the microbiological agar diffusion assay shall be conclusive.

(a) *Microbiological agar diffusion assay.* Proceed as directed in § 436.105 of this chapter, diluting an aliquot of the stock solution with solution 1 to the reference concentration of 1.0 microgram of cephaloridine per milliliter (estimated).

(b) *Iodometric assay.* Proceed as directed in § 436.204 of this chapter. If it is packaged for dispensing, dilute an aliquot of the stock solution with distilled water to the prescribed concentration.

NOTE: The 10 milliliters of 0.01*N* iodine must be added within 20 seconds after the addition of the 2.0 milliliters of 1.2*N* HCl, and the assay should be completed within 1 hour after the sample and standard are first put into solution. The working standard should be dried as described in § 436.200(a) of this chapter.

(c) *Hydroxylamine colorimetric assay.* Proceed as directed in § 436.205 of this chapter.

(2) *Sterility.* Proceed as directed in § 436.20 of this chapter, using the method described in paragraph (e)(1) of that section.

(3) *Pyrogens.* Proceed as directed in § 436.32(b) of this chapter, using a solution containing 50 milligrams of cephaloridine per milliliter.

(4) [Reserved]

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

(6) *pH.* Proceed as directed in § 436.202 of this chapter, using an aqueous solution containing 250 milligrams of cephaloridine per milliliter. If it is packaged for dispensing, however, use the solution obtained after reconstituting the drug as directed in the labeling.

(7) *Specific rotation.* Dilute an accurately weighed sample with sufficient distilled water to give a concentration of approximately 10 milligrams of cephaloridine per milliliter. Proceed as directed in § 436.210 of this chapter using a 2.0-decimeter polarimeter tube.

(8) *Crystallinity.* Proceed as directed in § 436.203(a) of this chapter.

(9) *Identity.* Using a 0.0025-percent solution of the sample in water and a suitable spectrophotometer, record the

ultraviolet absorption spectrum from 220 to 310 nanometers. The spectrum compares qualitatively to that of the cephaloridine working standard similarly tested.

[39 FR 19040, May 30, 1974, as amended at 43 FR 9800, Mar. 10, 1978; 50 FR 19919, May 13, 1985]

§ 442.25a Sterile cephalothin sodium.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Sterile cephalothin sodium is the sodium salt of the compound formed by reaction of thiophene-2-acetic acid with 7-amino-cephalosporanic acid. The 7-amino-cephalosporanic acid is obtained from a kind of cephalosporin. It is so purified and dried that:

(i) Its potency is not less than 850 micrograms of cephalothin per milligram on an anhydrous basis. If it is packaged for dispensing, its potency is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of cephalothin that it is represented to contain.

(ii) It is sterile.

(iii) It is nonpyrogenic.

(iv) [Reserved]

(v) Its loss on drying is not more than 1.5 percent.

(vi) Its pH in an aqueous solution is not less than 4.5 and not more than 7.0.

(vii) The specific rotation in an aqueous solution containing 50 milligrams of cephalothin sodium per milliliter at 25° C is +129°±5°.

(viii) It gives a positive identity test.

(ix) It is crystalline.

(2) *Packaging.* In addition to the requirements of § 432.1 of this chapter, if it is packaged for dispensing and is intended for both intravenous and intramuscular use, each vial shall contain the equivalent of 1 gram of cephalothin; except that if it is packaged for dispensing and is intended solely for intravenous use, each vial shall contain the equivalent of 4 grams of cephalothin.

(3) *Labeling.* In addition to the labeling requirements prescribed by § 432.5 of this chapter, if it is packaged for dispensing, each package shall bear on its label and labeling, the following statement: "After reconstitution, store in a

refrigerator and use within 48 hours. If kept at room temperature, use within 6 hours.”

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

(i) Results of test and assay on the batch for potency, sterility, pyrogens, loss on drying, pH, specific rotation, crystallinity, and identity.

(ii) Samples of the batch:

(a) If the batch is packaged for re-packing or for use as an ingredient in the manufacture of another drug:

(1) For all tests except sterility: 10 packages, each containing equal portions of approximately 500 milligrams.

(2) For sterility testing: 20 packages, each containing equal portions of approximately 300 milligrams.

(b) If the batch is packaged for dispensing:

(1) For all tests except sterility: A minimum of 10 immediate containers of the batch.

(2) For sterility testing: 20 immediate containers collected at regular intervals throughout each filling operation.

(b) *Tests and methods of assay*—(1) *Potency*—(i) *Sample preparation.* Dissolve an accurately weighed sample in sufficient 1.0 percent potassium phosphate buffer, pH 6.0 (solution 1), for the microbiological agar diffusion assay, distilled water for the iodometric assay or hydroxylamine colorimetric assay, to give a stock solution of convenient concentration; also if it is packaged for dispensing, reconstitute as directed in the labeling. Then, using a suitable hypodermic needle and syringe, remove all of the withdrawable contents if it is represented as a single-dose container; or, if the labeling specifies the amount of potency in a given volume of the resultant preparation, remove an accurately measured representative portion from each container. Dilute with either solution 1 or distilled water as specified above to give a stock solution of convenient concentration.

(ii) *Assay procedures.* Use any of the following methods; however, the results obtained from the microbiological agar diffusion assay shall be conclusive.

(a) *Microbiological agar diffusion assay.* Proceed as directed in § 436.105 of

this chapter, diluting an aliquot of the stock solution with solution 1 to the reference concentration of 1.0 microgram of cephalothin per milliliter (estimated).

(b) *Iodometric assay.* Proceed as directed in §436.204 of this chapter. If it is packaged for dispensing, dilute an aliquot of the stock solution with distilled water to the prescribed concentration.

NOTE: The 10 milliliters of 0.01*N* iodine must be added within 20 seconds after the addition of the 2.0 milliliters of 1.2*N* HCl, and the assay should be completed within 1 hour after the sample and standard are first put into solution. The working standard should be dried as described in §436.200(a) of this chapter.

(c) *Hydroxylamine colorimetric assay.* Proceed as directed in §436.205 of this chapter.

(2) *Sterility.* Proceed as directed in §436.20 of this chapter, using the method described in paragraph (e)(1) of that section.

(3) *Pyrogens.* Proceed as directed in §436.32(b) of this chapter, using a solution containing 50 milligrams of cephalothin per milliliter.

(4) [Reserved]

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

(6) *pH.* Proceed as directed in §436.202 of this chapter, using an aqueous solution containing 250 milligrams per milliliter; however, if it is packaged for dispensing, use the solution obtained after reconstituting the drug as directed in the labeling.

(7) *Specific rotation.* Dilute an accurately weighed sample with sufficient distilled water to give a concentration of approximately 50 milligrams per milliliter. Proceed as directed in §436.210 of this chapter, using a 1.0-decimeter polarimeter tube and calculate the specific rotation on an anhydrous basis.

(8) *Crystallinity.* Proceed as directed in §436.203(a) of this chapter.

(9) *Identity.* Using a 0.0025-percent solution of the sample in water and a suitable spectrophotometer, record the ultraviolet absorption spectrum from 220 to 310 nanometers. The spectrum compares qualitatively to that of the

cephalothin working standard similarly tested.

[39 FR 19040, May 30, 1974, as amended at 46 FR 46312, Sept. 18, 1981; 48 FR 11427, Mar. 18, 1983; 50 FR 19919, May 13, 1985]

§ 442.27 Cephalixin monohydrate.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity.* Cephalixin monohydrate is the monohydrate form of 7-(D-*alpha*-amino-*alpha*-phenylacetamido)-3-methyl-3-cephem-4-carboxylic acid. It is so purified and dried that:

- (i) Its potency is not less than 900 micrograms of cephalixin per milligram on an anhydrous basis.
- (ii) [Reserved]
- (iii) Its moisture content is not less than 4.0 nor more than 8.0 percent.
- (iv) Its pH in an aqueous solution containing 50 milligrams per milliliter is not less than 3.0 nor more than 5.5.
- (v) When calculated on an anhydrous basis, its absorptivity at 262 nanometers is not less than 95 percent and not more than 104 percent of that of the cephalixin standard similarly treated and corrected for potency.
- (vi) It gives a positive identity test.
- (vii) It is crystalline.

(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, moisture, pH, absorptivity, identity, and crystallinity.
- (ii) Samples required: 10 packages, each containing approximately 300 milligrams.

(b) *Tests and methods of assay—(1) Potency.* Use either of the following methods; however, the results obtained from

the microbiological agar diffusion assay shall be conclusive.

(i) *Microbiological agar diffusion assay.* Proceed as directed in § 436.105 of this chapter, preparing the sample for assay as follows: Dissolve an accurately weighed sample in sufficient 1 percent potassium phosphate buffer, pH 6.0 (solution 1), to give a stock solution containing 1.0 milligram per milliliter (estimated). Further dilute an aliquot of the stock solution with solution 1 to the reference concentration of 20 micrograms of cephalixin per milliliter (estimated).

(ii) *Iodometric assay.* Proceed as directed in § 436.204 of this chapter.

NOTE: The 10 milliliters of 0.01N iodine must be added within 20 seconds after the addition of the 2.0 milliliters of 1.2N hydrochloric acid, and the assay should be completed within 1 hour after the sample and standard are first put into solution.

(2) [Reserved]

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

(4) *pH.* Proceed as directed in § 436.202 of this chapter, using an aqueous suspension containing 50 milligrams per milliliter.

(5) *Absorptivity.* Determine the absorbance of the sample and standard solutions in the following manner: Dissolve accurately weighed portions of approximately 50 milligrams each of the sample and standard in 250 milliliters of distilled water. Transfer a 10-milliliter aliquot to a 100-milliliter volumetric flask and dilute to volume with distilled water. Using a suitable spectrophotometer and distilled water as the blank, determine the absorbance of each solution at 262 nanometers. Determine the percent absorptivity of the sample relative to the absorptivity of the standard using the following calculations:

$$\text{Percent relative absorptivity} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \times \frac{\text{Milligrams of standard}}{\text{Milligrams of sample}} \times \frac{\text{Potency of standard in micrograms per milligram}}{100 - m} \times \frac{10}{100 - m}$$

where:
m=percent moisture in the sample.

(6) *Identity.* Proceed as directed in § 436.211 of this chapter, using the 0.5