

(7) *pH*. Proceed as directed in § 436.202 of this chapter, using an aqueous solution containing 10 milligrams per milliliter.

(8) *Absorptivity*. Dissolve approximately 40 milligrams of the sample, accurately weighed, in approximately 150 milliliters of distilled water by mixing thoroughly. Dilute to 250 milliliters with distilled water and mix thoroughly. Transfer a 10.0-milliliter aliquot of this solution to a 100-milliliter volumetric flask, add approximately 75 milliliters of distilled water and 5.0

milliliters of 5*N* NaOH, dilute to volume with water, and mix thoroughly. Treat a sample of the tetracycline hydrochloride working standard in the same manner. Exactly 6 minutes after the addition of the NaOH, determine the absorbance of each solution at 380 nanometers, using a suitable spectrophotometer and distilled water as the blank. Determine the percent absorptivity of the sample relative to the absorptivity of the standard using the following calculation:

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

where: *m* = Percent moisture in the sample.

(9) *4-Epianhydrotetracycline*. Proceed as directed in § 436.309 of this chapter.

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

(11) *Identity*. Proceed as directed in § 436.308 of this chapter.

[43 FR 11160, Mar. 17, 1978; 43 FR 34456, Aug. 4, 1978, as amended at 44 FR 31636, June 1, 1979; 46 FR 60568, Dec. 11, 1981; 50 FR 19920, May 13, 1985]

§ 446.82 Tetracycline phosphate complex.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity*. Tetracycline phosphate complex is [4S-(4 α ,4 α ,5 α ,6 β , 12 α)] - 4 - (dimethylamino) - 1,4,4a,5,5a,6,11,12a - octahydro - 3,6,10,12,12a - pentahydroxy - 6 - methyl - 1,11 - dioxo - 2 - naphthacenicarboxamide phosphate complex. It is so purified and dried that:

(i) Its potency is not less than 750 micrograms per milligram on the anhydrous basis.

(ii) [Reserved]

(iii) Its moisture content is not more than 9 percent.

(iv) Its pH in an aqueous suspension containing 10 milligrams per milliliter is not less than 2.0 and not more than 4.0.

(v) When calculated on the anhydrous basis, its absorptivity at 380 nanometers relative to that of the tet-

racycline hydrochloride working standard similarly treated is 82.0 \pm 4.9 percent.

(vi) Its 4-epianhydrotetracycline content is not more than 2.0 percent.

(vii) It passes the identity test, showing a presence of phosphate, a content of not more than 0.2 percent chloride, and a content of not more than 1 percent tetracycline base.

(viii) It is crystalline.

(2) *Labeling*. In addition to the requirements of § 432.5 of this chapter, each such package shall bear on its label or labeling the statement "For use only in the manufacture of non-parenteral drugs".

(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, 4-epianhydro tetracycline content, identity, and crystallinity.

(ii) Samples required: 10 packages, each containing approximately 60 milligrams.

(b) *Tests and methods of assay—(1) Potency*. Proceed as directed in § 436.106 of this chapter, preparing the sample for assay as follows: Dissolve an accurately weighed sample in sufficient 0.1*N* hydrochloric acid to obtain a concentration of 1,000 micrograms of tetracycline hydrochloride per milliliter (estimated). Further dilute an aliquot of

the stock solution with sterile distilled water to the reference concentration of 0.24 microgram of tetracycline hydrochloride per milliliter (estimated).

(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 a suspension containing 10 milligrams of the sample per milliliter.

(5) *Absorptivity*. Dissolve approximately 40 milligrams of the sample, accurately weighed, in 2.0 milliliters of 0.1N HCl and dilute to 250 milliliters with distilled water. Transfer a 10.0

milliliter aliquot of this solution to a 100-milliliter volumetric flask, add about 75 milliliters of distilled water and 5.0 milliliters of 5N NaOH, dilute to volume with water, and mix thoroughly. Treat a sample of the tetracycline hydrochloride working standard in the same manner. Exactly 6 minutes after the addition of NaOH, determine the absorbance of each solution at 380 nanometers, using a suitable spectrophotometer and distilled water as the blank. Determine the percent absorptivity of the sample relative to the absorptivity of the standard using the following calculations:

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

where: *m* = Percent moisture in the sample.

(6) *4-Epianhydrotetracycline*. Proceed as directed in § 436.309 of this chapter.

(7) *Identity*—(i) *Presence of phosphate*. Prepare a filtrate as follows: Suspend 100 milligrams of the sample in 10 milliliters of distilled water and filter a small portion by gravity. Transfer 1.0 milliliter of the filtrate to a 100-milliliter glass-stoppered cylinder, add 10.0 milliliters of distilled water, 2.0 milliliters of ammonium molybdate test solution, 1.0 milliliter of stannous chloride test solution, and 10.0 milliliters of isobutyl alcohol-benzene mixture (1:1 ratio), all in the order named. Shake vigorously for 1 minute, allow the layers to separate, and examine the top organic layer. In the presence of phosphate, the top layer turns blue.

(ii) *Chloride content*. To 1.0 milliliter of the filtrate prepared as directed in the first sentence of paragraph (b)(7)(i) of this section, add 1 drop of silver nitrate test solution and 1 drop of nitric acid. Any turbidity produced is not greater than that obtained by similarly treating 1.0 milliliter of 0.057N hydrochloric acid.

(iii) *Determination of percent tetracycline base*. This test is used to determine the quantity of tetracycline present as base in mixtures with phosphate salts.

(a) *Reagents*—(1) 1,4-Dioxane.

(2) Purified dioxane: Pass the dioxane through a column of Amberlite IRA 400 (OH-) resin or equivalent.

(3) Perchloric acid, 0.01N: Dilute 0.84 milliliter of 70 percent perchloric acid to 1,000 milliliters with purified dioxane; standardize at least once every 2 days, as follows: Weigh accurately about 70 milligrams of diphenylguanidine, and dissolve in 50 milliliters of ethyl alcohol in a 250-milliliter flask. Add two drops of methyl red, and titrate with the perchloric acid solution until the yellow color changes to orange. Deduct the volume of the perchloric acid consumed by 50 milliliters of the ethyl alcohol, and calculate the normality. Each 2.113 milligrams of diphenylguanidine is equivalent to 1 milliliter of 0.01N perchloric acid.

(4) Methyl red indicator: Dissolve 100 milligrams of methyl red in 100 milliliters of methyl alcohol.

(b) *Procedure*. Place an accurately weighed 1-gram sample into a 50-milliliter Erlenmeyer flask, add 10.0 milliliters of purified dioxane and shake the mixture manually for about 2 minutes. Allow to settle, decant all the supernatant liquid into a 50-milliliter polyethylene centrifuge tube, cover with Parafilm (or equivalent), and centrifuge until clear (about 3 minutes).

Pipette 5.0 milliliters of the clear, supernatant solution into a 50-milliliter beaker, stir magnetically, and titrate with 0.01*N* perchloric acid, using meth-

yl red as the indicator. The endpoint is the last color change to orange when a drop of titrant is added. Calculate the percent tetracycline base as follows:

$$\text{Percent tetracycline base} = \frac{\text{Milliliters of acid used} \times \text{Normality} \times 0.4445 \times 200}{\text{Weight of sample}}$$

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

[43 FR 11161, Mar. 17, 1978; 43 FR 34456, Aug. 4, 1978, as amended at 50 FR 19920, May 13, 1985]

Subpart B—Oral Dosage Forms

§ 446.110 Chlortetracycline hydrochloride capsules.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity*. Chlortetracycline hydrochloride capsules are composed of chlortetracycline hydrochloride and one or more suitable and harmless diluents, lubricants, and fillers. Each capsule contains 50, 100, or 250 milligrams of chlortetracycline hydrochloride. The potency is satisfactory if it is not less than 90 percent and not more than 120 percent of the number of milligrams of chlortetracycline hydrochloride that it is represented to contain. The loss on drying is not more than 1 percent. The chlortetracycline hydrochloride used conforms to the standards prescribed by § 446.10(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 complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on:

(a) The chlortetracycline hydrochloride used in making the batch for potency, loss on drying, pH, crystallinity, and identity.

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

(ii) Samples required:

(a) The chlortetracycline hydrochloride used in making the batch: 10 packages, each containing approximately 300 milligrams.

(b) The batch: A minimum of 36 capsules.

(b) *Test 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 0.01*N* hydrochloric acid to give a stock solution of convenient concentration. Blend for 3 to 5 minutes. Remove an aliquot of the stock solution and further dilute with sterile distilled water to the reference concentration of 0.06 microgram of chlortetracycline hydrochloride per milliliter (estimated).

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

[43 FR 11162, Mar. 17, 1978; 43 FR 34456, Aug. 4, 1978, as amended at 50 FR 19920, May 13, 1985]

§ 446.115 Demeclocycline oral dosage forms.

§ 446.115a Demeclocycline oral suspension.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity*. Demeclocycline oral suspension is composed of demeclocycline with or without one or more suitable and harmless buffer substances, suspending and stabilizing agents, and preservatives suspended in a suitable and harmless vehicle. Each milliliter contains demeclocycline equivalent to 15 milligrams of demeclocycline hydrochloride. Its potency is satisfactory if it is not less than 90 percent and not more than 125 percent of the number of milligrams of demeclocycline hydrochloride equivalent that it is represented to contain. The pH is not less than 4 and not more than 5.8. The demeclocycline used conforms to the standards prescribed by § 446.15(a)(1).