

$$R_u = \frac{\text{Area of the daunorubicin sample peak}}{\text{Area of the internal standard peak}}$$

$$R_s = \frac{\text{Area of the daunorubicin standard peak}}{\text{Area of the internal standard peak}}$$

W_s = Weight of the daunorubicin working standard in milligrams;

V = Volume in milliliters of the internal standard solution added to the vials;

P = Potency of the daunorubicin working standard in micrograms per milligram.

(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(a) of this chapter, using a solution containing 2.25 milligrams of daunorubicin per milliliter.

(4) *Depressor substances*. Proceed as directed in § 436.35 of this chapter.

(5) *Moisture*. Proceed as directed in § 436.201 of this chapter, using the sample preparation method described in paragraph (d)(4) of that section.

(6) *pH*. Proceed as directed in § 436.202 of this chapter, using the sample obtained after reconstituting the drug as directed in the labeling.

(7) *Identity*. The high-pressure liquid chromatogram of the sample determined as directed in paragraph (b)(1) of this section compares qualitatively to that of the daunorubicin working standard.

[45 FR 75198, Nov. 14, 1980, as amended at 50 FR 47214, Nov. 15, 1985]

§ 450.224 Doxorubicin hydrochloride injectable dosage forms.

§ 450.224a Doxorubicin hydrochloride for injection.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity*. Doxorubicin hydrochloride for injection is a freeze-dried powder whose components are doxorubicin hydrochloride and lactose. It may also contain methylparaben. Its doxorubicin hydrochloride content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams of doxorubicin hydrochloride that it is

represented to contain. It is sterile. It contains not more than 2.2 U.S.P. endotoxin units per milligram of doxorubicin hydrochloride. Its moisture content is not more than 4.0 percent. When reconstituted as directed in the labeling, its pH is not less than 4.5 and not more than 6.5. It passes the identity test. The doxorubicin hydrochloride used conforms to the standards prescribed by § 450.24(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 doxorubicin hydrochloride used in making the batch for doxorubicin hydrochloride content, residue solvents, depressor substances, moisture, pH, crystallinity, identity, and total related impurities.

(b) The batch for doxorubicin hydrochloride content, sterility, bacterial endotoxins, moisture, pH, and identity.

(ii) Samples required:

(a) The doxorubicin hydrochloride used in making the batch: 14 packages, each containing approximately 40 milligrams.

(b) The batch:

(1) For all tests except sterility: A minimum of 34 immediate containers.

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

(b) *Tests and methods of assay*. Doxorubicin hydrochloride is toxic. It must be handled with care in the laboratory. Solutions should not be pipetted by mouth. Transfer all dry powders in a suitable hood while wearing rubber gloves. If the substance contacts the skin, wash with soap and water. Dispose of all waste material by dilution with large volumes of sodium hypochlorite (bleach) solution.

(1) *Doxorubicin hydrochloride content (high-performance liquid chromatography)*. Proceed as directed in § 450.24(b)(1), preparing the sample solution and calculating the doxorubicin hydrochloride content as follows:

(i) *Sample solution*. Prepare the sample solution by rinsing the contents of the vial into an appropriate sized volumetric flask with sufficient mobile phase to obtain a concentration of 0.1 milligram of doxorubicin hydrochloride per milliliter (estimated).

(ii) *Calculations*. Calculate the doxorubicin hydrochloride content per vial as follows:

$$\text{Milligrams of doxorubicin hydrochloride per vial} = \frac{A_U \times P_S \times d}{A_S \times 1,000}$$

where:

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

A_S = Area of the doxorubicin hydrochloride peak in the chromatogram of the doxorubicin hydrochloride working standard;

P_S = Doxorubicin hydrochloride activity in the doxorubicin hydrochloride working standard solution in micrograms per milliliter; and

d = Dilution factor of the sample.

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

(3) *Bacterial endotoxins*. Proceed as directed in the United States Pharmacopeia (U.S.P.) Bacterial Endotoxin Test, using a solution of doxorubicin hydrochloride for injection containing 1.1 milligrams of doxorubicin hydrochloride per milliliter. The specimen under test contains not more than 2.2 U.S.P. endotoxin units per milligram of doxorubicin hydrochloride.

(4) [Reserved]

(5) *Moisture*. Proceed as directed in § 436.201 of this chapter, using the sample preparation method described in paragraph (d)(4) of that section.

(6) *pH*. Proceed as directed in § 436.202 of this chapter, using the sample obtained after reconstituting the drug as directed in the labeling, except in lieu of saline use distilled water.

(7) *Identity*. The high-pressure liquid chromatogram of the sample deter-

mined as directed in paragraph (b)(1) of this section, compares qualitatively to that of the doxorubicin hydrochloride working standard.

[41 FR 14185, Apr. 2, 1976, as amended at 43 FR 44837, Sept. 29, 1978; 46 FR 60568, Dec. 11, 1981; 50 FR 19676, May 10, 1985. Redesignated at 53 FR 37292, Sept. 26, 1988; 59 FR 9641, Mar. 1, 1994]

§ 450.224b Doxorubicin hydrochloride injection.

(a) *Requirements for certification—(1) Standards of identity, strength, quality, and purity*. Doxorubicin hydrochloride injection is an aqueous solution of doxorubicin hydrochloride in an isosmotic diluent. Each milliliter contains doxorubicin hydrochloride equivalent to 2 milligrams of doxorubicin hydrochloride. Its doxorubicin hydrochloride content is satisfactory if it is not less than 90 percent and not more than 115 percent of the number of milligrams it is represented to contain. It is sterile. It contains not more than 2.2 U.S.P. endotoxin units per milligram of doxorubicin hydrochloride. Its pH is not less than 2.5 and not more than 3.5. It passes the identity test. The doxorubicin hydrochloride used conforms to the standards prescribed by § 450.24(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 doxorubicin hydrochloride used in making the batch for doxorubicin hydrochloride content, residue solvents, depressor substances, moisture, pH, crystallinity, identity, and total related impurities.

(B) The batch for doxorubicin hydrochloride content, sterility, bacterial endotoxins, pH, and identity.

(ii) Samples required:

(A) The doxorubicin hydrochloride used in making the batch: 14 packages, each containing approximately 40 milligrams.

(B) The batch:

(1) For all tests except sterility: A minimum of 34 immediate containers.