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The chapter reprint attached with this page is a true copy of the following:

USP28-NF23, Official Monographs/Dextran, pp 601-602

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ionization detector and contains a suitable column, 1.8 m × 2.0 mm, packed with 5% liquid phase G2 on support S1A. The column and injection port are maintained isothermally at 170° and 180°, respectively. Using a suitable carrier gas, adjust the flow rate so that the derivatized pantolactone elutes in about 4 minutes. Chromatograph five injections of the *Standard solution 2,* and record the peak responses as directed under *Procedure:* the relative standard deviation of the peak response ratios (R_S) of the five injections is not more than 2.0%. The retention time of the derivatized pantolactone is about 0.75 relative to that of the derivatized internal standard. In a suitable chromatogram, the resolution factor between the two peaks is not less than 2.0.

Procedure—Inject about 0.5 µL of *Standard solution 2* into the gas chromatograph, record the chromatogram to obtain not less than 40% of maximum recorder response, and measure the peak responses of the derivatized pantolactone and the derivatized internal standard. Similarly, inject about 0.5 µL of the *Test solution,* record the chromatogram, and measure the peak responses of the corresponding components. Calculate the quantity, in mg, of pantolactone in the portion of Preparation taken by the formula:

$$0.4C_S(R_U/R_S),$$

in which C_S is the concentration, in mg per mL, of USP Pantolactone RS in *Standard solution 1,* and R_U and R_S are the ratios of the peak response due to the pantolactone to that due to the internal standard obtained from the *Test solution* and the *Standard solution 2,* respectively.

Other requirements—It meets the requirements for *Refractive index, Water, Residue on ignition, Limit of aminopropanol,* and *Assay* under *Dexpanthenol.*

Dextran 1

» Dextran 1 is a low molecular weight fraction of dextran, consisting of a mixture of isomaltooligosaccharides. It is obtained by controlled hydrolysis and fractionation of dextrans produced by fermentation of *Leuconostoc mesenteroides* (strain NRRL B-512; CIP 78.59, or its sub-strains, for example *L. mesenteroides* B-512F; NCTC, 10817), in the presence of sucrose. It is a glucose polymer in which the linkages between glucose units are almost exclusively α-1,6. Its weight-average molecular weight is about 1000.

Packaging and storage—Store in well-closed containers at a temperature between 4° and 30°.

Labeling—Where it is intended for use in preparing injectable dosage forms, the label states that it is sterile or must be subjected to further processing during the preparation of injectable dosage forms.

USP Reference standards ⟨11⟩—*USP Dextran 1 RS. USP Endotoxin RS.*

Identification—

A: *Infrared Absorption* ⟨197K⟩—To 1 to 2 mg each of USP Dextran 1 RS and the sample add one to two drops of water, grind in an agate mortar for 1 to 2 minutes, add about 300 mg of potassium bromide, and mix to a slurry. [NOTE—Do not grind.] Dry under vacuum at 40° for 15 minutes, and if it is not dry, continue drying for another 15 minutes. Crush the residue, prepare a disk, and run the IR spectrum with a blank potassium bromide disk in the reference beam.

B: It meets the requirements of the test for *Molecular weight distribution and average molecular weight.*

Absorbance ⟨851⟩—The absorbance of a 15% solution in water at 375 nm is not more than 0.12, water being used as the blank.

Specific rotation ⟨781S⟩: between +148° and +164° at 20°, for a solution in water, on the dried basis (dry at 70° under vacuum to constant weight), and corrected for the content of sodium chloride.

Microbial limits ⟨61⟩—The total aerobic microbial count does not exceed 10^2 cfu per g, determined by plate-count; and the total

Bacterial endotoxins ⟨85⟩ (where it is labeled as intended for use in the preparation of injectables): not more than 25.0 USP Endotoxin Units per g.

pH ⟨791⟩: between 4.5 and 7.0, in a 15% solution in water.

Loss on drying ⟨731⟩—Dry it at 100° to 105° for 5 hours: it loses not more than 5.0% of its weight.

Heavy metals, *Method II* ⟨231⟩: not more than 5 µg per g.

Limit of alcohol and related impurities—

Test solution—Proceed as directed for *Test solution* in the test for *Limit of alcohol and related impurities* under *Dextran 40,* except to use 5.0 g of Dextran 1.

Standard solution, Chromatographic system, and *Procedure*—Proceed as directed in the test for *Limit of alcohol and related impurities* under *Dextran 40.* The total area of peaks from impurities in the *Test solution* does not exceed the area of the *n*-propyl alcohol solution peak.

Limit of sodium chloride—Dissolve 5 g of Dextran 1, accurately weighed, in 100 mL of water. Add 0.2 mL of potassium chromate TS, and titrate with 0.1 N silver nitrate VS (see *Titrimetry* ⟨541⟩). Each mL of 0.1 N silver nitrate is equivalent to 5.844 mg of sodium chloride: not more than 1.5% of sodium chloride is found.

Limit of nitrogenous impurities ⟨461⟩ *(where it is labeled as intended for use in the preparation of injectables)*—

Sulfate solution—To 1000 mL sulfuric acid add 5 g of anhydrous cupric sulfate and 500 g of potassium sulfate. Dissolve by heating, and store at 60°. [NOTE—If storage at 60° is not possible, prepare a smaller quantity of *Sulfate solution* on the day of use, adjusting proportions accordingly.]

Indicator—Dilute a mixture of 20 mL of a 0.1% solution of bromocresol green in alcohol and 4 mL of methyl red TS with water to 100 mL.

Procedure—Transfer 0.2 g Dextran 1, accurately weighed, to a micro-Kjeldahl flask. Add 4 mL of *Sulfate solution.* Heat until the solution exhibits a clear green color and the sides of the flask are free from carbonaceous material. Cool, cautiously add 30 mL of water, mix, and transfer the solution to a steam distillation unit. Rinse the Kjeldahl flask with three 5-mL portions of water, adding the washings to the solution. Add 15 mL of 45% sodium hydroxide solution, immediately close the distillation apparatus, and start steam distillation immediately. Receive the distillate in 1 mL of *Indicator* and sufficient water to cover the end of the condensing tube. Upon completion of the distillation, remove the receiving flask, and rinse the end of the condensing tube with a small quantity of water, adding the rinse to the distillate. Titrate the distillate with 0.010 N hydrochloric acid until the color changes from blue to reddish violet. Perform a blank determination, and make any necessary correction. The corrected volume of 0.010 N hydrochloric acid required to change the color does not exceed 0.15 mL (110 ppm of nitrogen).

Molecular weight distribution and average molecular weight—

Mobile phase—Prepare a filtered and degassed solution of sodium chloride containing 2.9 g per liter.

Calibration solution—Prepare a solution containing about 0.45 mg of isomaltotriose (3 glucose units) and 0.60 mg of sodium chloride per mL.

Reference solution—Prepare a solution of USP Dextran 1 RS in *Mobile phase* containing 6.0 to 6.5 mg per mL.

Test solution—Prepare a solution of Dextran 1 in *Mobile phase* containing 6.0 to 6.5 mg per mL.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a differential refractive index detector and two 10-mm × 30-cm columns in series that contain packing L54 and are maintained at 20–25°. The flow rate is 0.07 to 0.08 mL per minute, maintained constant to ±1%.

Procedure—Inject about 100 µL of the *Calibration solution,* record the chromatogram, and note the retention times of the peaks. Separately inject equal volumes (about 100 µL) of the *Reference solution* and *Test solution,* and record the chromatograms. Using the retention times in the chromatogram of *Calibration solution,* identify the peaks due to isomaltotriose and sodium chloride in the chromatograms of *Reference solution* and *Test solution.* Disregard the peak due to sodium chloride in *Reference solution* and *Test*

solution. Calculate the weight-average molecular weight, M_w, by the formula:

$$\Sigma W_i M_i,$$

in which W_i is the weight proportion of oligosaccharide *i;* and M_i is the molecular weight of oligosaccharide *i*. Use the following molecular weight values for calculation:

Glucose	180
Isomaltose	342
Isomaltotriose	504
Isomaltotetraose	666
Isomaltopentaose	828
Isomaltohexaose	990
Isomaltoheptaose	1152
Isomaltooctaose	1314
Isomaltononaose	1476
Isomaltodecaose	1638
Isomaltoundecaose	1800
Isomaltododecaose	1962
Isomaltotridecaose	2124
Isomaltotetradecaose	2286
Isomaltopentadecaose	2448
Isomaltohexadecaose	2610
Isomaltoheptadecaose	2772
Isomaltooctadecaose	2934
Isomaltononadecaose	3096

Calculate the amounts of the fractions with fewer than 3 and with more than 9 glucose units for the *Reference solution* and the *Test solution:* the M_w and amounts of the fractions obtained for the *Reference solution* are within the values stated in the data sheet that accompanies USP Dextran 1 RS. The M_w of Dextran 1 is between 850 and 1150. The fraction with fewer than 3 units of glucose is less than 15%, and the fraction with more than 9 units of glucose is less than 20%.

Dextran 40

Dextrans.
Dextrans [*9004-54-0*].

» Dextran 40 is derived by controlled hydrolysis and fractionation of polysaccharides elaborated by the fermentative action of certain strains of *Leuconostoc mesenteroides* (NRRL, B.512 F; NCTC, 10817) on a sucrose substrate. It is a glucose polymer in which the linkages between glucose units are almost entirely of the α-1 : 6 type. Its weight average molecular weight is in the 35,000 to 45,000 range.

Packaging and storage—Preserve in well-closed containers. Store at 25°, excursions permitted between 15° and 30°.

Labeling—Where it is intended for use in preparing injectable dosage forms, the label states that it is sterile or must be subjected to further processing during the preparation of injectable dosage forms.

USP Reference standards ⟨11⟩—*USP Dextran 40 RS. USP Dextran 4 Calibration RS. USP Dextran 10 Calibration RS. USP Dextran 40 Calibration RS. USP Dextran 70 Calibration RS. USP Dextran 250 Calibration RS. USP Dextran V_o Marker RS. USP Dextran 40 System Suitability RS. USP Endotoxin RS.*

Color of solution—The absorbance of a solution in water (1 in 10), measured in a 4-cm cell determined at 375 nm against a water blank, is not greater than 0.20.

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: Prepare four *Test solutions* of Dextran 40 in water, in such a manner that the concentrations are accurately known and approximately evenly distributed in the range of 2% to 0.5%. Using a capillary tube viscosimeter having dimensions such that the flow time and of the *Test solution* at 20°. Calculate the viscosity numbers of each of the *Test solutions* by the formula:

$$\{\ln[(R_D)(t / t_0)]\} / C,$$

in which R_D is the ratio of the density of the individual *Test solution* to that of water; t and t_0 are the flow times for the *Test solution* and water, respectively; and C is the concentration, in g per mL, of Dextran 40 in the *Test solution.* Plot the viscosity numbers of each of the *Test solutions* against their respective concentrations, and draw the straight line of best fit through the points and extrapolate to zero concentration: the value of the intercept is between 18 and 23 mL per g.

Specific rotation ⟨781S⟩**:** between +195° and +203°.

Test solution: 20 mg per mL, heated, if necessary, on a water bath to dissolve.

Bacterial endotoxins ⟨85⟩ *(where it is labeled as intended for use in the preparation of injectables)*—When tested in Sodium Chloride Injection (1 in 10), it contains not more than 1.0 USP Endotoxin Unit per mL.

Safety—Inject intravenously 1.0 mL of a sterile 1 in 10 solution of 10% Dextran 40 in saline TS into each of five mice weighing 18 to 20 g. The injection period is not less than 10 seconds and not greater than 15 seconds. If there are no deaths within 72 hours, it meets the requirements of the test. If 1 or more animals die, continue the test using 10 mice weighing 20 ± 0.5 g. If all animals survive for 72 hours, the requirements of the test are met.

pH ⟨791⟩**:** between 4.5 and 7.0, in a solution (1 in 10).

Loss on drying ⟨731⟩—Dry it at 105° for 5 hours: it loses not more than 7.0% of its weight.

Sulfate ⟨221⟩—A 1.5-g portion shows no more sulfate than corresponds to 0.45 mL of 0.020 N sulfuric acid (0.03%).

Heavy metals, *Method II* ⟨231⟩**:** 5 µg per g.

Limit of nitrogenous impurities *(where it is labeled as intended for use in the preparation of injectables)*—

Sulfate solution—To 1000 mL of sulfuric acid add 5 g of anhydrous cupric sulfate and 500 g of potassium sulfate. Dissolve by heating, and store at 60°. [NOTE—If storage at 60° is not possible, prepare a smaller quantity of *Sulfate solution* on the day of use, adjusting the proportions accordingly.]

Indicator—Dilute a mixture of 20 mL of a 0.1% solution of bromocresol green in alcohol and 4 mL of methyl red TS with water to 100 mL.

Procedure—Transfer 0.2 g, accurately weighed, to a micro-Kjeldahl flask. Add 4 mL of *Sulfate solution.* Heat until the solution exhibits a clear green color and the sides of the flask are free from carbonaceous material. Cool, and transfer the solution to a steam distillation unit. Rinse the Kjeldahl flask three times with 5 mL of water, adding the washings to the solution. Add 15 mL of 45% sodium hydroxide solution, immediately close the distillation apparatus, and commence steam distillation without delay. Receive the distillate in 1 mL of *Indicator* in a 100-mL flask, keeping the end of the condensing tube below the liquid surface for 5 minutes and above the liquid surface for 1 minute. Upon completion of the distillation, remove the receiving flask, and rinse the end of the condensing tube with a small quantity of water, adding the rinse to the distillate. Titrate the distillate with 0.010 N hydrochloric acid until the color changes from blue to reddish violet. Perform a blank determination, and make any necessary correction. The corrected volume of 0.010 N hydrochloric acid titrated does not exceed 0.14 mL (0.01%, as N).

Limit of alcohol and related impurities—

Test solution—Dissolve without heating 5.0 g in 100 mL of water, and distill the solution, collecting the first 45 mL of the distillate. Dilute the distillate with water to 50.0 mL, and mix.

Standard solution—To 25.0 mL of the *Test solution* add 0.5 mL of a 2.5% (w/v) solution of *n*-propyl alcohol.

Chromatographic system—The gas chromatograph is equipped with a flame-ionization detector and contains a 2-mm × 1.8-m column packed with support S3. The column temperature is maintained at about 160°, the injection port temperature is maintained at about 240°, and the detector is maintained at about 210°. The carrier gas is nitrogen, flowing at a rate of about 25 mL per minute. [NOTE—Injector seals may deteriorate after multiple injections of the *Standard* and *Test solutions*. Inspect the seals before making a series of injections.]