beled units of Insulin Zinc Injection (Aqueous Suspension) by proceeding as directed in the Assay (iv) under Insulin Injection. Divide healthy rabbits weighing more than 1.8 kg, fasted for not less than 14 hours before injection, into 2 equal groups of not less than 3. Inject subcutaneously an amount of the standard solution or the sample solution equivalent to 0.3 units per kg of body mass to the animals of each group. Collect blood before and 1 hour and 2.5 hours after injection, then proceed as directed in the Assay (viii) under Insulin Injection, and calculate the ratio of the average blood sugar level of 1 hour and 2.5 hours after to that of before injection of each animal: the mean value for the group injected the sample solution is not less than that for the group injected the standard solution.

Nitrogen content Perform the test as directed under the Nitrogen Determination: not less than 0.50 mg and not more than 0.64 mg of nitrogen (N: 14.01) is found for each labeled 100 Units.

Assay **(1)** Insulin—Proceed as directed in the Assay under Insulin Injection with the clear liquid obtained from Crystalline Insulin Zinc Injection (Aqueous Suspension) by adjusting the pH to about 2.5 with diluted hydrochloric acid (1 in 100).

(2) Zinc—Measure exactly a volume of Crystalline Insulin Zinc Injection (Aqueous Suspension), equivalent to about 200 Units according to the labeled units, add 1 mL of 0.1 mol/L hydrochloric acid TS and sufficient water to make exactly 200 mL, then dilute with water to contain 0.6 to 1.0 μg of zinc (Zn: 65.39) per mL, and use this solution as the sample solution. Separately, pipet a volume of Standard Zinc Solution for atomic absorption spectrophotometry, dilute with water to contain 0.4 to 1.2 μg of zinc (Zn: 65.39) per mL, and use this solution as the standard solution. Perform the test with the sample solution and the standard solution as directed under the Atomic Absorption Spectrophotometry according to the following conditions, and determine the amount of zinc in the sample solution using the calibration curve obtained from the absorbance of the standard solution.

Gas: Combustible gas—Acetylene gas
Supporting gas—Air
Lamp: Zinc hollow-cathode lamp
Wavelength: 213.9 nm

(3) Crystalline insulin—Measure accurately a volume of Crystalline Insulin Zinc Injection (Aqueous Suspension), equivalent to about 400 Units according to the labeled Units, centrifuge, discard the supernatant liquid, suspend the residue in 5 mL of water, add 10 mL of sodium acetate-acetone TS, shake for 3 minutes, and centrifuge. Discard the supernatant liquid, and repeat the above treatment on the residue. Wash down the residue into a Kjeldahl flask with 15 mL of sulfuric acid, and perform the test as directed under the Nitrogen Determination: the amount of nitrogen (N: 14.01) is not less than 85% of the total nitrogen content. Calculate the total nitrogen content for insulin Units of a sample from the values of nitrogen obtained in the Nitrogen content.

Containers and storage Containers—Hermetic containers.
Storage—In a cold place, and avoid freezing.

Expiration date 24 months after preparation.

Insulin Zinc Protamine Injection (Aqueous Suspension)

プロタミンインスリン亜鉛水性懸濁注射液

Insulin Zinc Protamine Injection (Aqueous Suspension) is an aqueous suspension for injection. It contains not less than 90% and not more than 110% of the labeled Insulin Units, and not less than 0.12 mg and not more than 0.30 mg of zinc (Zn: 65.39) for each labeled 100 Units.

Method of preparation Prepare as directed under Injections, with Insulin, Protamine Sulfate and Zinc Chloride. It contains 0.38 to 0.63 g of Dibasic Sodium Phosphate, 1.4 to 1.8 g of Concentrated Glycerin, and 0.18 to 0.22 g of Cresol or 0.22 to 0.28 g of Phenol for each 100 mL of Insulin Zinc Protamine Injection (Aqueous Suspension).

Description Insulin Zinc Protamine Injection (Aqueous Suspension) is a white suspension. When allowed to stand, it separates into a white precipitate and a colorless, supernatant liquid, and it readily becomes suspension again on gentle shaking.

When it is examined microscopically, no large particles are seen.

Identification Adjust the pH of Insulin Zinc Protamine Injection (Aqueous Suspension) to between 2.5 and 3.5 with dilute hydrochloric acid: the particles dissolve, and the solution is clear and colorless.

pH 7.0 - 7.4

Purity **(1)** Protein—Perform the test as directed under the Nitrogen Determination: not exceeding 1.25 mg of nitrogen (N: 14.01) is found for each labeled 100 Units.

(2) Dissolved insulin—Perform the following test with the clear liquid obtained by centrifuging Insulin Zinc Protamine Injection (Aqueous Suspension): not more than 2.5% of the labeled Units is found.

Use a clear liquid of Insulin Zinc Protamine Injection (Aqueous Suspension) as the sample solution, and prepare the standard solution by proceeding as directed in the Assay (iv) under Insulin Injection to adjust its concentration to 2.5% of the labeled units of Insulin Zinc Protamine Injection (Aqueous Suspension). Divide the healthy rabbits weighing not less than 1.8 kg, fasted for not less than 14 hours before injection, into 2 equal groups of not less than 3. Inject subcutaneously an amount of the standard solution or the sample solution equivalent to 0.3 units per kg of body mass. Collect blood before and 1 hour and 2.5 hours after injection, proceed as directed in the Assay (viii) under Insulin Injection, and calculate the ratios of the average blood sugar content in each rabbit measured 1 hour and 2.5 hours after injection to the content before injection: the mean value for the group injected with the sample solution is not less than the mean value for the group injected with the standard solution.

Assay **(1)** Insulin—Proceed as directed in the Assay under Injection with the clear liquid obtained by adjusting the pH to 2.5 with diluted hydrochloric acid (1 in 100).

(2) Zinc—Measure accurately a volume of Insulin Zinc

Protamine Injection (Aqueous Suspension), equivalent to about 200 Units according to the labeled units, add 1 mL of 0.1 mol/L hydrochloric acid TS and sufficient water to make exactly 200 mL, dilute with water to contain 0.6 to 1.0 μg of zinc (Zn: 65.39) in 1 mL, and use this solution as the sample solution. Separately, pipet a volume of Standard Zinc Solution for the Atomic Absorption Spectrophotometry, dilute with water to contain 0.4 to 1.2 μg of zinc (Zn: 65.39) per ml, and use this solution as the standard solution. Perform the test with the sample solution and the standard solution according to the Atomic Absorption Spectrophotometry under the following conditions, and determine the amount of zinc in the sample solution using the analytical curve obtained from the absorbance of the standard solution.

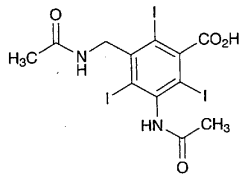
Gas: Combustible gas—Acetylene gas
Supporting gas—Air
Lamp: Zinc hollow-cathode lamp
Wavelength: 213.9 nm

Containers and storage Containers—Hermetic containers.
Storage—In a cold place, and avoid freezing.

Expiration date 24 months after preparation.

Iodamide

ヨーダミド



$C_{12}H_{11}I_3N_2O_4$: 627.94
3-Acetylamino-5-acetylaminomethyl-2,4,6-triiodobenzoic acid [*440-58-4*]

Iodamide, calculated on the dried basis, contains not less than 98.5% of $C_{12}H_{11}I_3N_2O_4$.

Description Iodamide occurs as a white, crystalline powder. It is odorless.

It is slightly soluble in water and in ethanol (95), and practically insoluble in diethyl ether.

It dissolves in sodium hydroxide TS and in sodium carbonate TS.

It gradually changes in color by light.

Identification **(1)** To 0.01 g of Iodamide add 5 mL of hydrochloric acid, and heat in a water bath for 5 minutes: the solution responds to the Qualitative Tests for primary aromatic amines.

(2) Heat 0.1 g of Iodamide over a flame: a purple gas evolves.

(3) Determine the infrared absorption spectrum of Iodamide, previously dried, as directed in the potassium bromide disk method under the Infrared Spectrophotometry, and compare the spectrum with the Reference Spectrum: both spectra exhibit similar intensities of absorption at the same wave numbers. If any difference appears between the spectra, dissolve 1 g of Iodamide in 100 mL of water by heating, and concentrate the solution to about 30 mL by gentle boiling. After cooling, collect the formed crystals by filtration, dry, and repeat the test on the dried crystals.

Purity **(1)** Clarity and color of solution—Dissolve 1.0 g of Iodamide in 10 mL of diluted sodium hydroxide TS (1 in 5): the solution is clear and colorless.

(2) Primary aromatic amines—Dissolve 0.20 g of Iodamide in 5 mL of water and 1 mL of sodium hydroxide TS, add 4 mL of a solution of sodium nitrite (1 in 100) and 10 mL of 1 mol/L hydrochloric acid TS, shake well, and allow to stand for 2 minutes. To this solution add 5 mL of ammonium amidosulfate TS, shake thoroughly, allow to stand for 1 minute, add 0.4 mL of a solution of 1-naphthol in ethanol (95) (1 in 10), 15 mL of sodium hydroxide TS and water to make exactly 50 mL, and determine the absorbance at 485 nm as directed under the Ultraviolet-visible Spectrophotometry, using a solution, prepared in the same manner, as the blank: the absorbance of the solution is not more than 0.12.

(3) Soluble halide—Dissolve 2.5 g of Iodamide in 20 mL of water and 2.5 mL of ammonia TS, then add 20 mL of dilute nitric acid and water to make 100 mL. Allow to stand for 15 minutes with occasional shaking, and filter. Discard the first 10 mL of the filtrate, transfer 25 mL of the subsequent filtrate to a Nessler tube, and add ethanol (95) to make 50 mL. Use this solution as the test solution, and proceed as directed under the Chloride Limit Test. Prepare the control solution with 0.10 mL of 0.01 mol/L hydrochloric acid VS and 6 mL of dilute nitric acid, and dilute with water to 25 mL, then with ethanol (95) to 50 mL.

(4) Iodine—Dissolve 0.20 g of Iodamide in 2 mL of sodium hydroxide TS, add 2.5 mL of 0.5 mol/L sulfuric acid TS, allow to stand for 10 minutes with occasional shaking, then add 5 mL of chloroform, shake vigorously and allow to stand: the chloroform layer remains colorless.

(5) Heavy metals—Proceed with 2.0 g of Iodamide according to Method 4, and perform the test. Prepare the control solution with 2.0 mL of Standard Lead Solution (not more than 10 ppm).

(6) Arsenic—Prepare the test solution with 1.0 g of Iodamide according to Method 3, and perform the test using Apparatus B (not more than 2 ppm).

Loss on drying Not more than 3.0% (1 g, 105°C, 4 hours).

Residue on ignition Not more than 0.10% (1 g).

Assay Weigh accurately about 0.5 g of Iodamide in a saponification flask, dissolve in 40 mL of sodium hydroxide TS, add 1 g of zinc powder, connect the flask with a reflux condenser, boil for 30 minutes, cool, and filter. Wash the flask and filter paper with 50 mL of water, and combine the washings with the filtrate. Add 5 mL of acetic acid (100), and titrate with 0.1 mol/L silver nitrate VS until the color of the precipitate changes from yellow to green (indicator: 1 mL of tetrabromophenolphthalein ethyl ester TS).

Each mL of 0.1 mol/L silver nitrate VS
= 20.931 mg of $C_{12}H_{11}I_3N_2O_4$

Containers and storage Containers—Tight containers.
Storage—Light-resistant.