Status: Currently Official on 15-Feb-2025
Official Date: Official as of 01-May-2018
Document Type: USP Monographs
DocId: GUID-73C27AB1-B876-46A5-BDD7-81D84331D940_3_en-US
DOI: https://doi.org/10.31003/USPNF_M40605_03_01
DOI Ref: mk0sz

Insulin Human Injection

DEFINITION

Insulin Human Injection is an isotonic, sterile solution of Insulin Human in Water for Injection. It has a potency of NLT 95.0% and NMT 105.0% of the potency stated on the label, expressed in USP Insulin Human Units/mL.

IDENTIFICATION

• **A.** The retention time of the major peak of *Sample solution A* or *Sample solution B* corresponds to that of the *Standard solution*, as obtained in the *Assay*.

ASSAY

• **Procedure**

Solution A: Dissolve 28.4 g of anhydrous sodium sulfate in 1000 mL of water. Pipet 2.7 mL of phosphoric acid into the solution, and adjust with ethanolamine to a pH of 2.3, if necessary.

Mobile phase: Acetonitrile and *Solution A* (26:74). [Note—The acetonitrile is warmed to NLT 20° to avoid precipitation.]

System suitability solution: 1.5 mg/mL of insulin human in 0.01 N hydrochloric acid. Allow to stand at room temperature for NLT 3 days to obtain a solution containing NLT 5% of A-21 desamido insulin human.

Standard solution: 1.5 mg/mL of USP Insulin Human RS in 0.01 N hydrochloric acid

Sample solution A (for Injection labeled as containing 40 USP Insulin Human Units/mL)**:** Add 2.5 µL of 9.6 N hydrochloric acid for each mL of an accurately measured volume of Injection. Allow the suspension, if present, to clarify, and mix.

Sample solution B (for Injection labeled as containing 100 USP Insulin Human Units/mL)**:** Add 2.5 µL of 9.6 N hydrochloric acid for each mL of an accurately measured volume of Injection. Allow the suspension, if present, to clarify, and mix. [Note—Pooling several package units may be necessary to obtain sufficient volume of the sample.] Pipet 2 mL of this solution into a 5-mL volumetric flask, dilute with 0.01 N hydrochloric acid to volume, and mix.

Chromatographic system

(See *Chromatography (621), System Suitability*.)

Mode: LC

Detector: UV 214 nm

Column: 4.6-mm × 15-cm; packing L1

Column temperature: 40°

Flow rate: 1 mL/min

Injection volume: 20 µL

System suitability

Samples: *System suitability solution* and *Standard solution*

Suitability requirements

Resolution: NLT 2.0 between insulin human and A-21 desamido insulin human, *System suitability solution*

Tailing factor: NMT 1.8 for the insulin human peak, *System suitability solution*

Relative standard deviation: NMT 1.6%, *Standard solution*

Analysis

Samples: *Standard solution* and either *Sample solution A* or *Sample solution B*

Measure the peak responses for insulin human and A-21 desamido insulin human. Calculate the potency, in USP Insulin Human Units/mL, of the Injection taken:

$$\text{Result} = (\Sigma r_U / \Sigma r_S) \times C_S \times D$$

Σr_U = sum of the peak responses of insulin human and A-21 desamido insulin human from the *Sample solution*

Σr_S = sum of the peak responses of insulin human and A-21 desamido insulin human from the *Standard solution*

C_S = concentration of USP Insulin Human RS in the *Standard solution* (USP Insulin Human Units/mL)

D = dilution factor used to prepare the *Sample solution*

Acceptance criteria: 95.0%–105.0% of the potency stated on the label, expressed in USP Insulin Human Units/mL

OTHER COMPONENTS

• **Zinc Determination ⟨591⟩:** 10–40 µg for every 100 USP Insulin Human Units

PRODUCT-RELATED SUBSTANCES AND IMPURITIES

• **Physicochemical Analytical Procedures for Insulins,** *Limit of High Molecular Weight Proteins* ⟨121.1⟩

Proceed as directed in *Limit of High Molecular Weight Proteins*, except prepare the following *Sample solution*. It meets the requirements.

Sample solution: Quantitatively add 4 µL of 6 N hydrochloric acid to each mL of an accurately measured volume of Injection, and mix.

Acceptance criteria: NMT 1.7%

SPECIFIC TESTS

• **pH ⟨791⟩:** 7.0–7.8

• **Particulate Matter in Injections ⟨788⟩:** Meets the requirements for small-volume injections

• **Bacterial Endotoxins Test ⟨85⟩:** NMT 80 USP Endotoxin Units/100 USP Insulin Human Units

• **Sterility Tests ⟨71⟩:** Meets the requirements when tested as directed in *Test for Sterility of the Product to Be Examined, Membrane Filtration*

• **Injections and Implanted Drug Products ⟨1⟩:** Meets the requirements

ADDITIONAL REQUIREMENTS

• **Packaging and Storage:** Preserve and dispense in the unopened, multiple-dose container provided by the manufacturer. Store in a refrigerator, protect from sunlight, and avoid freezing.

• **Labeling:** Label it to indicate that it has been prepared with Insulin Human produced by methods based on recombinant DNA technology or that it is derived by enzymatic modification of insulin from porcine pancreas. Label it to state that it is to be stored in a refrigerator and that freezing is to be avoided. The label states the potency in USP Insulin Human Units/mL.

• **USP Reference Standards ⟨11⟩**

USP Insulin Human RS

Auxiliary Information - Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
INSULIN HUMAN INJECTION	Jennifer Tong Sun Senior Scientist II	BIO2 Biologics Monographs 2 - Proteins

Chromatographic Database Information: Chromatographic Database

Most Recently Appeared In:
Pharmacopeial Forum: Volume No. PF 40(5)

Current DocID: GUID-73C27AB1-B876-46A5-BDD7-81D84331D940_3_en-US
Previous DocID: GUID-73C27AB1-B876-46A5-BDD7-81D84331D940_1_en-US
DOI: https://doi.org/10.31003/USPNF_M40605_03_01
DOI ref: mk0sz