AKRON BIO

FREQUENTLY ASKED QUESTIONS

Recombinant Human Interleukin-2 (rHu IL-2) Closed System Solutions™ (CSS)

Bags (1 MIU) Cat. # AR1045-0010 | Bags (2 MIU) Cat. # AR1045-0020 | Protein Concentration (10 µg/mL)
Bags (7.5 MIU) Cat. # AR1050-0010 | Bags (15 MIU) Cat. # AR1050-0020 | Protein Concentration (75 µg/mL)

1. Why use Akron's Recombinant Human Interleukin-2 (rHu IL-2) Closed System Solutions™ (CSS)?

Akron's novel rHu IL-2 CSS liquid formulation and sterile bag packaging increase safety and ease of use by eliminating the reconstitution step during manufacture, allowing for the direct introduction of cytokine material into culture media in a fully contained manner.

Akron's Recombinant Human Interleukin-2 (rHu IL-2) is manufactured following all relevant cGMP guidelines for ancillary materials and is supported by a Type II Master File (MF) with the FDA and a MF Type 1 with Health Canada. It is purified in a pharmaceutical facility without the use of histidine tags and nickel columns. Sterile filtration with aseptic filling are followed by Endotoxin and Sterility testing performed per USP/EP on the final product. Our rHu IL-2 amino acid sequence is identical to Proleukin® (aldesleukin), and its functional similarity in T cell expansion has been evaluated and confirmed (see Product Page).

2. What are the recommended storage conditions for Akron's rHu IL-2 CSS?

We recommend storing this product at 2-8 °C.

3. What is the white precipitate inside the bag that is observed while storing under recommended conditions?

During storage at 2-8 °C, a white precipitate can appear inside the bag as a result of a reversible interaction between the rHu IL-2 protein and the excipients at this temperature. The white precipitate disappears after a few minutes at room temperature and has no relevant impact on the strength, purity, or biological activity of Akron's rHu IL-2. Once the precipitate has reached dissolution, a colorless, clear liquid should be observed within the bag, ensuring that it is ready to use.

Note: Do not use the bag until the precipitate completely dissolves and is no longer visible.

4. What are the shipping conditions for Akron's rHu IL-2 CSS?

This product ships with cold packs. rHu IL-2 CSS units are packed in storage cassettes and shipped in validated insulated CSafe Parcel shippers.

5. What is the shelf-life for Akron's rHu IL-2 CSS?

This product is currently under a long-term stability program.

6. What organism is used to express Akron's rHu IL-2?

This product is expressed in *E. coli*.

7. How is Akron's rHu IL-2 different than native IL-2?

Akron's rHu IL-2 is not glycosylated because it is derived from *E. coli* and has an identical amino acid sequence to Proleukin® (aldesleukin), which differs from the native IL-2 amino sequence in the following ways: Akron's rHu IL-2 does not have an N-terminal alanine, and it has a serine substituted in place of a cysteine at position 125. It is likely that the aggregation state is also different than native IL-2.

8. What is the molecular weight of Akron's rHu IL-2?

Akron's rHu IL-2 was characterized by liquid chromatography-electrospray ionization-tandem mass spectrometry analysis and was found to have an observed molecular mass of 15,319 Da. Every batch of product is released using a reducing SDS-PAGE molecular weight assay to directly compare against a reference standard.

9. Do you have a Master File (MF) available for this product?

The active substance in this product, Akron's rHu IL-2, has a Type II eCTD MF (#026152) on file with the FDA and an MF Type I (#e250089) on file with Health Canada. We can provide you a Letter of Authorization that will permit these agencies to refer to the information on file in support of your submissions.

10. Is a TSE/BSE statement available for this product?

Yes, a TSE/BSE statement is available upon request.

11. Is virus and pathogen inactivation included in the manufacturing process?

No, because viruses that infect bacteria (bacteriophages) do not pose a known threat to human cells. Virus reduction manufacturing steps are usually not included when purifying material from a bacterial host, as is the case with Akron's rHu IL-2.

12. What safety testing is done on this product?

Every lot of final product is tested with methods per USP/EP, for both Endotoxin (USP <85> / EP 2.6.14) and Sterility (USP <71> / EP 2.6.1).

13. Which cell types are suitable?

Akron's cGMP-compliant rHu IL-2 can be used to promote the activation and proliferation of numerous immune cell types, including CAR-T cells, TCR-T cells, Tregs, TILs, NK cells, CIK cells, B cells, monocytes, and macrophages.

14. Do you have an SDS for this product?

Yes, an SDS is available upon request for this product.

15. How does Akron measure activity for rHu IL-2?

Akron uses a validated leukocyte proliferation assay to report activity. The rHu IL-2 induced proliferation of T lymphocyte HT-2 cells is measured against the World Health Organization (WHO) 2nd international standard for Interleukin-2, held by the National Institute for Biological Standards and Control (NIBSC 86/500). Akron uses a parallel-line concentration-response model to calculate a relative potency, per guidelines set forth in USP <1032> and USP <1034> (see Technical Overview).

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16. Is the performance comparable to competitor rHu IL-2 products?

The specific activity (MIU/mg) of Akron's rHu IL-2 batches have historically been comparable to the specific activity of the current international standard (NIBSC 86/500) for IL-2, known to be approximately 13.73 MIU/mg. Akron's rHu IL-2 amino acid sequence is identical to Proleukin® (aldesleukin), and its functional similarity in T Cells has been evaluated and confirmed (see Product Page Data).

17. What is the intended use for the product?

For research use or further manufacturing use in *ex vivo* cell therapy applications. This product is not intended for direct *in vivo* use or for direct clinical use as a drug, therapeutic, biologic, or medical device.

18. What packaging options are available for rHu IL-2 CSS?

Akron's rHu IL-2 CSS comes packaged in a sterile 25 mL fluoropolymer bag chamber. The outlet tube is made from two different weldable materials to choose from. The top portion is made from TPE AdvantaFlex® (1/8" ID x 1/4" OD) and the bottom portion is made from standard weldable PVC (3/32" ID x 5/32" OD). These packaging materials are extensively validated, controlled, and qualified to ensure a consistent experience.

19. What closed-connection options are available to remove the material from the bag?

There are two different tubing materials available to make weldable connections to your closed-system process line (for full instructions, see "Methods of Use" document).