

Technical Data Sheet

Human TNF Standard

Product Information

Material Number:	558472
Component:	51-9003509
Description:	Human TNF Standard
Size:	1 Vial(s) (1 ea)
Storage Buffer:	Lyophilized in an aqueous buffered solution containing BSA and ProClin™ 150.

Description

This product contains lyophilized recombinant human tumor necrosis factor (TNF). When reconstituted, this product can be used as a standard control with the BD™ CBA Human TNF Flex Set (Cat. No. 560112 or 558273) or with the BD™ CBA Human TNF Enhanced Sensitivity Flex Set (Cat. No. 561516). Please refer to the technical data sheet included with the BD™ CBA Human TNF Flex Set or the BD™ CBA Human TNF Enhanced Sensitivity Flex Set for further information.

Preparation and Storage

Store the lyophilized standard at 4°C prior to reconstitution.

Lyosphere(s) from 1 vial of Human TNF Standard, when reconstituted in 4.0 mL Assay Diluent, approximates the CBA activity of Human TNF at a concentration of 2500 pg/mL. Following reconstitution, maintain the Human TNF at 4°C and use within 12 hours. Any unused reconstituted standard must be discarded after this period (do not store or reuse).

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

The Human TNF Standard has been tested to assure to function as a standard when used with the BD™ Cytometric Bead Array (CBA). Investigators are encouraged to refer to the technical data sheet for the BD™ CBA Human TNF Flex Set (Cat. No. 560112 or 558273) or the BD™ CBA Human TNF Enhanced Sensitivity Flex Set (Cat. No. 561516). The Human TNF Standard is lyophilized and should be reconstituted and serially diluted before mixing with CBA capture beads and PE detection reagents. An abbreviated instruction has been provided below (Preparation of Human TNF Standard Protocol). The Human TNF Standard should be reconstituted using Assay Diluent, which is provided with the BD™ CBA Human Soluble Protein Master Buffer Kit (Cat. No. 558264 or 558265), the BD™ CBA Human Enhanced Sensitivity Master Buffer Kit (Cat. No. 561521 or 561523) or it may also be purchased separately (Cat. No. 560104).

Preparation of Human TNF Standard for use with the BD™ CBA Human TNF Flex Set (Cat. No. 560112 or 558273):

1. Open one vial of the lyophilized Human TNF Standard. Transfer the lyosphere(s) to a polypropylene tube (BD Falcon™, Cat. No. 352097). Label the tube "Top Standard".
2. Reconstitute the standard with 4.0 mL of Assay Diluent. Allow the reconstituted standard to equilibrate for at least 15 minutes before making dilutions. **Mix the reconstituted protein by pipette only. Do not vortex or mix vigorously.**
3. Label additional 12 x 75 mm tubes (BD Falcon™, Cat. No. 352008) and arrange them in the following order: Top Standard, 1:2, 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, and 1:256.
4. Add 500 µl of Assay Diluent to each of the dilution tubes.
5. Perform a serial dilution by transferring 500 µl from the Top Standard to the 1:2 dilution tube and mix thoroughly. **Do not vortex, mix by pipet only.** Continue making serial dilutions by transferring 500 µl from the 1:2 tube to the 1:4 tube and so on to the 1:256 tube and mix thoroughly. Prepare one tube containing only Assay Diluent to serve as the 0 pg/mL negative control.

Preparation of Human TNF Standard for use with the BD™ CBA Human TNF Enhanced Sensitivity Flex Set (Cat. No. 561516):

1. Open one vial of the lyophilized Human TNF Standard. Transfer the lyosphere(s) to a polypropylene tube (BD Falcon™, Cat. No. 352097).
2. Reconstitute the standard with 4.0 mL of Assay Diluent. Allow the reconstituted standard to equilibrate for at least 15 minutes before making dilutions. **Mix the reconstituted protein by pipette only. Do not vortex or mix vigorously.**
3. Label additional 12 x 75 mm tubes (BD Falcon™, Cat. No. 352008) and arrange them in the following order: Top Standard, 1:3, 1:9, 1:27, 1:81, 1:243, and 1:729.
4. Add 460 µl of Assay Diluent into the Top Standard tube.
5. Add 400 µl of Assay Diluent to each of the remaining dilution tubes.
6. Transfer 40 µl of the reconstituted standard into the Top Standard tube. Mix by pipette or gently vortexing for no longer than 3 seconds.

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7. Perform a serial dilution by transferring 200 µl from the Top Standard to the 1:3 dilution tube and mix thoroughly. **Do not vortex, mix by pipet only.** Continue making serial dilutions by transferring 200 µl from the 1:3 tube to the 1:9 tube and so on to the 1:729 tube and mix thoroughly. Prepare one tube containing only Assay Diluent to serve as the 0 fg/mL negative control.

Suggested Companion Products

Catalog Number	Name	Size	Clone
558273	Human TNF Flex Set	1 Vial(s)	(none)
558264	Human Soluble Protein Master Buffer Kit	100 Tests	(none)
558265	Human Soluble Protein Master Buffer Kit	500 Tests	(none)
560104	Assay Diluent	30 mL	(none)
352097	BD Falcon™ Conical Centrifuge Tubes, 15 ML		(none)
352008	BD Falcon™ Round Bottom Tubes, 6 ML		(none)
561516	Human TNF Enhanced Sensitivity Flex Set	100 Tests	(none)
561521	Human Enhanced Sensitivity Master Buffer Kit	100 Tests	(none)
561523	Human Enhanced Sensitivity Master Buffer Kit	500 Tests	(none)
560112	Human TNF Flex Set	100 Tests	(none)

Product Notices

- ProClin is a trademark of Rohm and Haas Company.
- Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- Please refer to www.bdbiosciences.com/pharminggen/protocols for technical protocols.
- Warning: CBA lyophilized standard contains 0.02% (w/w) of a CMIT/MIT mixture (3:1), which is a mixture of: 5-chloro-2-methyl-4-isothiazolin-3-one [EC No 247-500-7] and 2-methyl-4-isothiazolin-3-one [EC No 220-239-6] (3:1).
Hazard statement: May cause an allergic skin reaction.
Precautionary statements: Wear protective gloves/eye protection. Wear protective clothing. Avoid breathing mist/vapours/spray. If skin irritation or rash occurs: Get medical advice/attention. IF ON SKIN: Wash with plenty of water. Dispose of contents/container in accordance with local/regional/national/international regulations.

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