

## Technical Data Sheet

## Mouse Th1/Th2/Th17 Cytokine Standards

## Product Information

Material Number:	561665
Size:	1 Vial(s)
Storage Buffer:	Lyophilized in an aqueous buffered solution containing BSA and ProClin™ 150.
Component:	51-9006250
Description:	C) Mouse Th1/Th2/Th17 Cytokine Standards

## Description

The Mouse Th1/Th2/Th17 Cytokine Standards contains a mixture of individually lyophilized recombinant proteins made up of recombinant Mouse Interleukin (IL)-2, IL-4, IL-6, IL-10, IL-17A, Interferon- $\gamma$  (IFN- $\gamma$ ), and Tumor Necrosis Factor (TNF). The reconstituted Mouse Th1/Th2/Th17 Cytokine Standards are designed for use as standard controls with the BD™ Cytometric Bead Array (CBA) Mouse Th1/Th2/Th17 Cytokine Kit (Cat. No. 560485). Please refer to the manual included with the BD™ Cytometric Bead Array (CBA) Mouse Th1/Th2/Th17 Cytokine Kit for further information.

## Preparation and Storage

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

Lyospheres from 1 vial of the Mouse Th1/Th2/Th17 Cytokine Standard, when reconstituted in 2.0 mL Assay Diluent, approximates the CBA activity of Mouse IL-2, IL-4, IL-6, IL-10, IL-17A, Interferon- $\gamma$  (IFN- $\gamma$ ), and Tumor Necrosis Factor (TNF) at a concentration of 5000 pg/mL. Following reconstitution, maintain the Mouse Th1/Th2/Th17 Cytokine Standard 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/immunoassay	Routinely Tested
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## Recommended Assay Procedure:

The Mouse Th1/Th2/Th17 Cytokine Standard has been tested with BD™ Cytometric Bead Array (CBA) Mouse Th1/Th2/Th17 Cytokine Kit (Cat. No. 560485) to assure function as a standard in the CBA assay. Investigators are encouraged to refer to the CBA kit manual or to the brief instructions below (Preparation of Mouse Th1/Th2/Th17 Cytokine Standard Protocol). The Mouse Th1/Th2/Th17 Cytokine Standard should be reconstituted using Assay Diluent supplied with the aforementioned CBA kit (component no. 51-2432KC) or it may be purchased separately (Cat. No. 560104).

**Preparation of Mouse Th1/Th2/Th17 Cytokine Standard:** The Mouse Th1/Th2/Th17 Cytokine Standard is lyophilized and should be reconstituted and serially diluted before mixing with CBA capture beads and PE detection reagents.

1. Open one vial of the lyophilized Mouse Th1/Th2/Th17 Cytokine Standard. Transfer all the lyospheres to a 15 mL polypropylene tube (BD Falcon™, Cat. No. 352097). Label the tube "Top Standard".
2. Reconstitute the standard with 2.0 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 300  $\mu$ L of Assay Diluent to each of the dilution tubes.
5. Perform a serial dilution by transferring 300  $\mu$ L 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 300  $\mu$ L 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.

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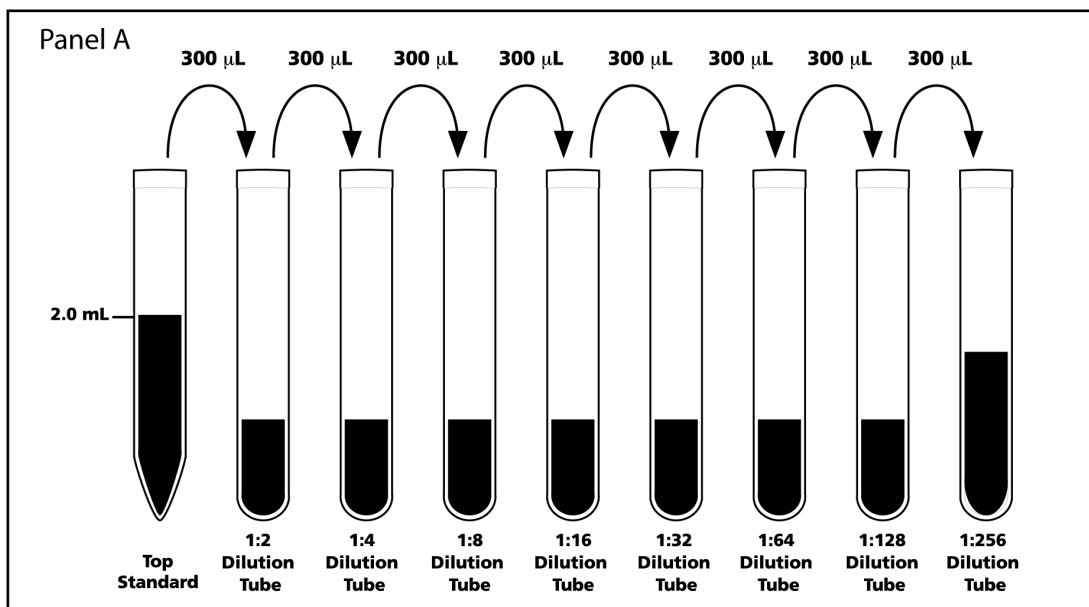
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**Panel B**

Protein (pg/ml)	Top Standard	1:2 Dilution Tube	1:4 Dilution Tube	1:8 Dilution Tube	1:16 Dilution Tube	1:32 Dilution Tube	1:64 Dilution Tube	1:128 Dilution Tube	1:256 Dilution Tube
Mouse IL-2	5000	2500	1250	625	312.5	156	80	40	20
Mouse IL-4	5000	2500	1250	625	312.5	156	80	40	20
Mouse IL-6	5000	2500	1250	625	312.5	156	80	40	20
Mouse IL-10	5000	2500	1250	625	312.5	156	80	40	20
Mouse IL-17A	5000	2500	1250	625	312.5	156	80	40	20
Mouse IFN- $\gamma$	5000	2500	1250	625	312.5	156	80	40	20
Mouse TNF	5000	2500	1250	625	312.5	156	80	40	20

**Preparation of Mouse Th1/Th2/Th17 Cytokine Standard Dilutions (Panel A) and Mouse Th1/Th2/Th17 Cytokine Standard Concentrations after Dilutions (Panel B).**

## Product Notices

- Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
- Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- ProClin is a trademark of Rohm and Haas Company.
- 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.

## References

Curran MA, Montalvo W, Yagita H, Allison JP. PD-1 and CTLA-4 combination blockade expands infiltrating T cells and reduces regulatory T and myeloid cells within B16 melanoma tumors. *Proc Natl Acad Sci U S A*. 2010; 107(9):4275-4280. (Biology: Cytometric Bead Array)  
Matarollo SR, Yong M, Tan L, Frazer IH, Leggatt GR. Secretion of IFN- $\gamma$  but not IL-17 by CD1d-restricted NKT cells enhances rejection of skin grafts expressing epithelial cell-derived antigen. *J Immunol*. 2010; 184(10):5663-5669. (Biology: Cytometric Bead Array)  
Silverman JM, Clos J, Horakova E, et al. Leishmania exosomes modulate innate and adaptive immune responses through effects on monocytes and dendritic cells. *J Immunol*. 2010; 185(9):5011-5022. (Biology: Cytometric Bead Array)

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