

Technical Data Sheet

Human IL-6 Flex Set

Product Information

Material Number:558276Size:100 testsBead Position:A7

Reactivity: QC Testing: Human

Testing in Development: Non-human Primate

Assay Range: 10 - 2,500 pg/mL

Component Description: Human IL-6 Standard
Component Mat. No: 51-9003499

Component Storage Buffer: Lyophilized in an aqueous buffered solution containing BSA

and ProClinTM 150.

Component Description: Human IL-6 PE Detection Reagent

Component Mat. No: 51-9004046

Component Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09%

sodium azide.

Component Description: Human IL-6 Capture Bead A7

Component Mat. No: 51-9005287

Component Storage Buffer: Aqueous buffered solution containing fetal bovine serum and

≤0.09% sodium azide.

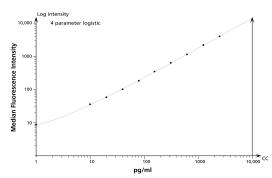


Figure 1. Example BD CBA Human IL-6 Flex Set standard curve. Data acquired on a BD FACSArray bioanalyzer and analyzed using FCAP Array Software

Description

The BDTM CBA Human IL-6 Flex Set is a bead-based immunoassay capable of measuring human interleukin-6 (IL-6) in serum, plasma, and cell culture supernatant samples. Human and non-human primate reactivity was determined by testing samples with the BD CBA Human IL-6 Flex Set. The biology and function of IL-6 has been extensively reviewed in the literature. For more information on bead-based immunoassays, refer to the product insert for the BD CBA Human Soluble Protein Master Buffer Kit (Cat. No. 558264 or 558265).

Preparation and Storage

This BDTM CBA Flex Set contains one vial each of Capture Bead and PE Detection Reagent and two vials of Standard. The Capture Bead and PE Detection Reagent components of this flex set have been formulated to a 50x concentration to ensure product performance when multiplexed. The Standard component is lyophilized and should be transferred to a 15 mL polypropylene tube for reconstitution. When reconstituted in 4.0 mL Assay Diluent, the standard has a protein concentration of 2,500 pg/mL. Discard unused reconstituted standard, do not store or reuse. Store lyophilized standard and other components at 4°C. Protect Capture Beads and the PE Detection Reagent from prolonged exposure to light.

Application Notes

Recommended Assay Procedure: The BD CBA Human IL-6 Flex Set must be used in conjunction with a BD CBA Human Soluble Protein Master Buffer Kit (Cat. No. 558264, 100 tests, or 558265, 500 tests), a flow cytometer, and FCAP Array™ Software. Detailed instructions on the use of this product can be found in the manual for the BD CBA Human Soluble Protein Master Buffer Kit. When following the directions in the Master Buffer Kit, the top standard point for the BD CBA Human IL-6 Flex Set will be 2,500 pg/mL. An example standard curve is shown in Figure 1.

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When multiplexing the BD CBA Human IL-6 Flex Set assay with the BD CBA Human LT-α Flex Set assay (Cat. No. 560083), significantly higher background will be seen in the human IL-6 assay. This increased background, while reducing the sensitivity of the human IL-6 assay, will not effect the quantitation of human IL-6 or LT-α in any other way.

The BD CBA Human IL-6 Flex Set should not be used in the same assay well with any non-BD CBA Human Soluble Protein Flex Set reagents (such as BD CBA Mouse Soluble Protein or Cell Signaling Flex Sets). For an updated assay compatibility chart for the BD CBA Human Soluble Protein Flex Sets, please refer to the BD CBA Flex Set System homepage at http://www.bdbiosciences.com/cbasetup.

Performance

Limit of Detection: The theoretical limit of detection is 1.6 pg/mL and was determined by evaluating the estimated result of the average MFI of the negative control (0 pg/mL, n=30) + 2 standard deviations.

		Inter-Assay Reproducibility			Intra-Assay Reproducibility			
Specifici	ty	Mean (pg/ml)	Standard Deviation	%CV	Mean Standard (pg/ml) Deviation		%CV	
Human IL-6	Sample 1	38.8	2.4	6%	35.6	1.2	3%	
	Sample 2	144.7	9.2	6%	145.9	4.6	3%	
	Sample 3	609.0	47.6	8%	645.1	10.9	2%	

Reproducibility: The inter-assay and intra-assay reproducibility were determined for the BD CBA Human IL-6 Flex Set by evaluating ten replicates of three different sample levels (inter-assay) and two replicates of three different sample levels from four separate experiments (intra-assay) respectively.

Specificity	Cell Culture Supernatant		Ser	um	Plasma		
	Average % Recovery	Range	Average % Recovery	Range	Average % Recovery	Range	
Human IL-6	86%	76 - 96%	83%	75 - 90%	77%	72 - 81%	

Recovery: Cell culture supernatant, serum, or EDTA-treated plasma were spiked with three different levels of protein. The spiked samples were assayed and the results were compared with expected values. Serum and plasma samples were diluted 1:4 before the protein was spiked into each. Serum is a pool of 800 - 1000 donors and the plasma was pooled from at least 20 donors.

Sample Dilution	Cell Culture Supernatant		Ser	um	Plasma		
	Detected (pg/ml)	% of Expected	Detected (pg/ml)	% of Expected	Detected (pg/ml)	% of Expected	
Spiked sample	601.5	100%	1,206.5	100%	1,198.2	100%	
1:2	289.1	96%	587.8	97%	483.8	81%	
1:4	146.3	97%	301.0	100%	298.2	100%	

Linearity: Cell culture supernatant, 1:4 diluted serum, or 1:4 diluted EDTA-treated plasma were spiked with protein and serially diluted. The diluted samples were assayed and the results were compared with the original spiked sample.

Product Notices

- 1. ProClin is a trademark of Rohm and Haas Company.
- Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

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