

Magnetic Luminex $^{\circ}$ Performance Assay Human TNF- α High Sensitivity Kit

Catalog Number: LHSCM210
Pack Size: 100 Tests

SPECIFICATIONS AND USE

Recommended Sample Types

- Human HS Cytokine Base Kit A: Serum, EDTA plasma, and heparin plasma.
- **Human HS Cytokine Base Kit B:** Cell culture supernates, serum, EDTA plasma, and heparin plasma.

Microparticle Region Components

- Region-12
- Microparticle Concentrate (Part 894500) is supplied as a 50X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 894057) is supplied as a 100X concentrated stock solution (0.075 mL) with preservatives.

Other Supplies Required

 Magnetic Luminex Performance Assay Human High Sensitivity Cytokine Base Kit A (Catalog Number LHSCM000) or Magnetic Luminex Performance Assay Human High Sensitivity Cytokine Base Kit B (Catalog Number LBHS000).

Storage

- Store the unopened kit at 2-8 °C. Do not use past the expiration date on the label.
- Avoid freezing microparticles.
- Protect microparticles from light.

Instructions for Use

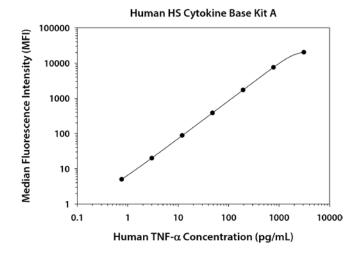
 Refer to the appropriate Base Kit insert for the Magnetic Luminex Performance Assay procedure.

TYPICAL DATA

This human TNF- α standard curve is provided only for demonstration. A standard curve must be generated each time an assay is run, utilizing values from the Standard Value Card included in the Base Kit.

Human HS Cytokine Base Kit A: When using Calibrator Diluent RD6-40, a seven point standard curve (0.76-3100 pg/mL) is recommended.

Human HS Cytokine Base Kit B: When running cell culture supernate samples using Calibrator Diluent RD5K, a six-point standard curve (1.20-1230 pg/mL) is recommended. When running serum/plasma samples using Calibrator Diluent RD6-65, a seven-point standard curve (1.20-4920 pg/mL) is recommended.



Standard	pg/mL	MFI	Average	Corrected
Blank	0	15 16	16	
1	3100	20,284 20,416	20,350	20,334
2	775	7375 7725	7550	7534
3	194	1722 1756	1739	1723
4	48	397 400	399	383
5	12	102 106	104	88
6	3	35 36	36	20
7	0.76	20 21	21	5

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PRECISION

Intra-assay Precision (precision within an assay)

Three samples of known concentration were tested twenty times on one plate to assess precision within an assay.

Inter-assay Precision (precision between assays)

Three samples of known concentration were tested in separate assays to assess precision between assays.

	Int	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3	
n	20	20	20	60	60	60	
Mean (pg/mL)	8.1	65	1533	7.5	61	1654	
Standard Deviation	0.4	2.7	105	0.8	4.9	169	
% CV	4.9	4.1	6.8	10.7	8.0	10.2	

RECOVERY & LINEARITY

Samples were spiked with human TNF- α and evaluated for recovery and were serially diluted to evaluate assay linearity.

Recovery				
Sample Type	Average % Recovery	Range (%)		
Cell culture supernates*	105	99-111		
Serum	86	73-101		
EDTA plasma	79	73-87		
Heparin plasma	79	70-94		

Linearity					
		Cell culture supernates*	Serum	EDTA Plasma	Heparin Plasma
1:2	Average % of Expected	98	108	111	112
	Range (%)	95-101	98-116	107-115	110-113
1:4	Average % of Expected	94	112	115	111
	Range (%)	93-96	105-125	97-128	107-117
1:8	Average % of Expected	93	116	114	116
	Range (%)	90-97	105-139	99-129	110-123

^{*}Cell culture supernates are valid samples in Human HS Cytokine Base Kit B only.

SENSITIVITY

All data were collected with assays run as a multiplex.

Data obtained with polystyrene and magnetic beads were equivalent.

Twenty-eight assays were evaluated, and the minimum detectable dose (MDD) of human TNF- α ranged from 0.13-0.54 pg/mL. The mean MDD was 0.29 pg/mL.

The MDD was determined by adding two standard deviations to the MFI of twenty zero standard replicates and calculating the corresponding concentration.

CORRELATION

This assay has been correlated to the Quantikine® ELISA Kit with a slope of 0.9-1.1 and an R² value greater than 0.9.

SPECIFICITY

Note: *Refer to the base kit insert for a complete list of analytes tested for cross-reactivity and interference.*

This assay recognizes natural and recombinant human TNF- α .

Recombinant human TNF RI interferes at concentrations > 25.0 ng/mL in this assay.

TECHNICAL HINTS

- Protect the microparticles and streptavidin-PE from light at all times.
- Refer to the appropriate Base Kit Standard Value Card for reconstitution volume and values of the reconstituted standard.
- Diluted microparticles cannot be stored. Make a fresh dilution of microparticles each time the assay is run.
- The use of a magnetic device made to accommodate a microplate is necessary for washing.
- Discrepancies may exist in values obtained for the same analyte utilizing different technologies.

Magnetic Luminex Performance Assays afford the user the benefit of multianalyte analysis of cytokines in a complex sample. A single, multipurpose diluent for each sample type is used to optimize recovery, linearity, and reproducibility. Such a multipurpose, single diluent may not optimize any single analyte to the same degree that a unique diluent selected for analysis of that analyte can optimize conditions. Therefore, some performance characteristics may be more variable than those for assays designed specifically for single analyte analysis.

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