Magnetic Luminex® Performance Assay Human TNF-α Kit

Catalog Number: LUHM210
Pack Size: 100 Tests

SPECIFICATIONS AND USE

Recommended Sample Types
- Cell culture supernates, serum, EDTA plasma, and heparin plasma.

Microparticle Region
- Region-37

Components
- Microparticle Concentrate (Part 894449) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 892637) is supplied as a 100X concentrated stock solution (0.075 mL) with preservatives.

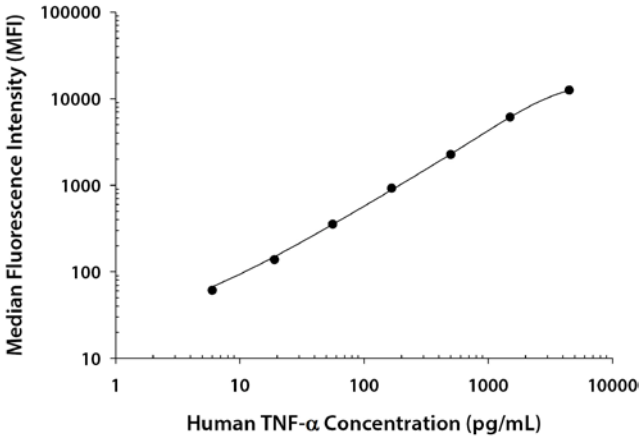
Other Supplies Required
- Magnetic Luminex Performance Assay Human Base Kit A (Catalog Number LUHM000).

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 Base Kit insert for the 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.



Standard	pg/mL	MFI	Average	Corrected
Blank	0	31 31	31	——
1	4500	12,438 12,572	12,505	12,474
2	1500	6121 6139	6130	6099
3	500	2292 2301	2297	2266
4	167	945 957	951	920
5	56	383 387	385	354
6	19	168 169	169	138
7	6	90 93	92	61

PERFORMANCE CHARACTERISTICS

All data were collected with assays run as a multiplex.
Data obtained with polystyrene and magnetic beads were equivalent.

Sensitivity - The Minimum Detectable Dose (MDD) was determined by adding two standard deviations to the MFI of twenty zero standard replicates and calculating the corresponding concentration.

Fourteen assays were evaluated, and the MDD of human TNF-α ranged from 0.10-1.50 pg/mL. The mean MDD was 0.60 pg/mL.

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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 twenty-five separate assays to assess precision between assays.

	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	25	25	25
Mean (pg/mL)	39.2	232	1326	44	234	1237
Standard Deviation	1.66	8.56	63.9	3.0	15	87
% CV	4.2	3.7	4.8	7.3	6.2	7.0

Recovery and Linearity – Samples containing and/or spiked with high concentrations of TNF-α were evaluated for recovery and were serially diluted to evaluate assay linearity.

Recovery		
Sample Type	Average % Recovery	Range (%)
Cell culture supernates	93	80-108
Serum	108	97-114
EDTA plasma	99	78-128
Heparin plasma	105	95-115

Linearity					
		Cell culture supernates	Serum	EDTA Plasma	Heparin Plasma
1:2	Average % of Expected	108	104	107	101
	Range (%)	96-117	96-116	95-122	91-108
1:4	Average % of Expected	108	107	110	98
	Range (%)	90-121	96-123	90-135	94-105
1:8	Average % of Expected	108	110	111	96
	Range (%)	87-120	102-141	67-145	96-98

Specificity - This assay recognizes natural and recombinant human TNF-α. The assay was tested for cross-reactivity and interference with the following factors. Less than 0.5% cross-reactivity and interference was observed.

Recombinant human:			Recombinant mouse:		Recombinant rat:	Recombinant porcine:	Recombinant human multiplex partners:	
6Ckine	IL-1 RII	IL-17	G-CSF	IL-8	GM-CSF	GM-CSF	ENA-78	IL-6
CNTF	IL-2 Rα	IL-18	GM-CSF	IL-10	IFN-γ	IL-1α	FGF basic	IL-8
β-ECGF	IL-2 Rβ	LIF	IFN-γ	IL-17	IL-1α	IL-1β	G-CSF	IL-10
FGF acidic	IL-2 Rγ	LIF R	IL-1α	MIP-1α	IL-1β	IL-2	GM-CSF	IL-17
FGF-4	IL-3 Rα	MIP-1α	IL-1ra	MIP-1β	IL-2	IL-4	IFN-γ	MCP-1
FGF-5	IL-4 R	MIP-3α	IL-1	RANTES	IL-4	IL-6	IL-1α	MIP-1α
FGF-6	IL-5 Rα	MIP-3β	IL-2	Tpo	IL-6	IL-8	IL-1β	MIP-1β
FGF-9	IL-6 R	MCP-2	IL-4	TNF-α	IL-10	IL-10	IL-1ra	RANTES
FGF-10	IL-10 R	MCP-3	IL-5	VEGF	TNF-α	Leptin	IL-2	Tpo
FGF-18	IL-3	MCP-4	IL-6			TNF-α	IL-4	VEGF
GCP-2	IL-7	M-CSF					IL-5	
GROα	IL-9	TNF RI						
GROβ	IL-11	TNF-α						
GROγ	IL-12 p40	$VEGF_{121}$						
I-309	IL-12 p70	$VEGF_{165}$						
IGF-I	IL-13	VEGF-D						
IGF-II	IL-15							
IL-1 RI	IL-16							

TECHNICAL HINTS

- Protect the microparticles and streptavidin-PE from light at all times.
- Refer to the 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.

Luminex Performance Assays afford the user the benefit of multianalyte analysis of biomarkers in a complex sample. For each sample type, a single, multipurpose diluent is used to optimize recovery, linearity, and reproducibility. Such a multipurpose 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.