Luminex® Performance Assay Human IL-6 Kit

Catalog Number: LUH206
**Pack Size:** 100 Tests

SPECIFICATIONS AND USE

Recommended Sample Types	• Cell culture supernates, serum, EDTA plasma, and heparin plasma.
Microparticle Region	• Region-32
Components	• Microparticle Concentrate (Part 891062) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
	• Biotin-Antibody Concentrate (Part 892622) is supplied as a 100X concentrated stock solution (0.075 mL) with preservatives.
Other Supplies Required	• Luminex Performance Assay Human Base Kit A (Catalog Number LUH000) or Luminex Performance Assay Human Obesity Base Kit (Catalog Number LOB000).
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 Luminex Performance Assay procedure.

TYPICAL DATA

This human IL-6 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.
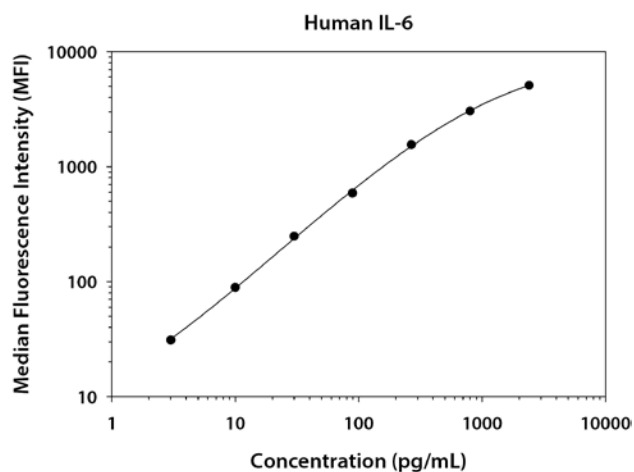
Standard	pg/mL	MFI	Average	Corrected
Blank	0	18 20	19	——
1	2400	4875 5348	5112	5094
2	800	2910 3216	3063	3045
3	267	1540 1599	1570	1552
4	89	575 636	606	588
5	30	256 275	266	248
6	10	104 109	107	89
7	3	48 50	49	31

PERFORMANCE CHARACTERISTICS

All data were collected with assays run as a multiplex.

Sensitivity - The Minimum Detectable Dose (MDD) was determined by adding two standard deviations to the mean MFI of twenty zero standard replicates and calculating the corresponding concentration.
Forty-three assays were evaluated, and the MDD of human IL-6 ranged from 0.10-1.11 pg/mL. The mean MDD was 0.36 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)	30.2	180	903	34	189	900
Standard Deviation	1.43	7.78	41.9	3.00	13.0	53.0
% CV	4.7	4.3	4.6	8.8	6.9	5.9

Recovery and Linearity - Samples containing and/or spiked with high concentrations of IL-6 were evaluated for recovery and were serially diluted to evaluate assay linearity.

Recovery				Linearity					
Sample Type	Average % Recovery	Range (%)				Cell culture supernates	Serum	EDTA Plasma	Heparin Plasma
Cell culture supernates	96	91-100		1:2	Average % of Expected	101	106	103	99
					Range (%)	87-117	104-109	82-124	86-110
Serum	108	95-118		1:4	Average % of Expected	103	112	122	101
					Range (%)	86-127	103-117	116-129	85-114
EDTA plasma	102	95-116		1:8	Average % of Expected	102	115	126	100
					Range (%)	82-130	97-126	121-131	87-110
Heparin plasma	109	96-129							

Specificity - This assay recognizes natural and recombinant human IL-6. 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 Panel A:	Recombinant human multiplex partners Obesity Panel:
6Ckine	IL-3 Rα	MCP-3	G-CSF	GM-CSF	GM-CSF	ENA-78	Adiponectin
CNTF	IL-4 R	MCP-4	GM-CSF	IFN-γ	IL-1α	FGF basic	CRP
β-ECGF	IL-5 Rα	M-CSF	IFN-γ	IL-1α	IL-1β	G-CSF	Factor D
FGF acidic	IL-6 R	TNF RI	IL-1α	IL-1β	IL-2	GM-CSF	IL-10
FGF-4	IL-10 R	TNF-β	IL-1β	IL-2	IL-4	IFN-γ	Leptin
FGF-5	IL-3	VEGF$_{121}$	IL-1ra	IL-4	IL-6	IL-1α	MCP-1
FGF-6	IL-7	VEGF$_{165}$	IL-2	IL-6	IL-8	IL-1β	Resistin
FGF-9	IL-9	VEGF-D	IL-4	IL-10	IL-10	IL-1ra	Serpin E1
FGF-10	IL-11		IL-5	TNF-α		IL-2	TNF-α
FGF-18	IL-12/IL-23 p40		IL-6			IL-4	
GCP-2	IL-12 p70		IL-10			IL-5	
GROα	IL-13		IL-17			IL-8	
GROβ	IL-15		MIP-1α			IL-10	
GROγ	IL-16		MIP-1β			IL-17	
I-309	IL-17		RANTES			MCP-1	
IGF-I	IL-18		TNF-α			MIP-1α	
IGF-II	LIF		Tpo			MIP-1β	
IL-1 RI	LIF R					RANTES	
IL-1 RII	MIP-1δ					TNF-α	
IL-2 Rα	MIP-3α					Tpo	
IL-2 Rβ	MIP-3β					VEGF	
IL-2 Rγ	MCP-2						

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 vacuum manifold device made to accommodate a microplate is necessary for washing. Adjust the vacuum to between 15 and 40 cm Hg.
- Discrepancies may exist in values obtained for the same analyte utilizing different technologies.

Luminex Performance Assays afford the user the benefit of multi-analyte 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.

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