

SPECIFICATIONS AND USE

Recommended Sample Types

- Cell culture supernates, serum, EDTA plasma, and heparin plasma.

Microparticle Region

- Region-17

Components

- Microparticle Concentrate (Part 891058) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 892619) 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).

Storage

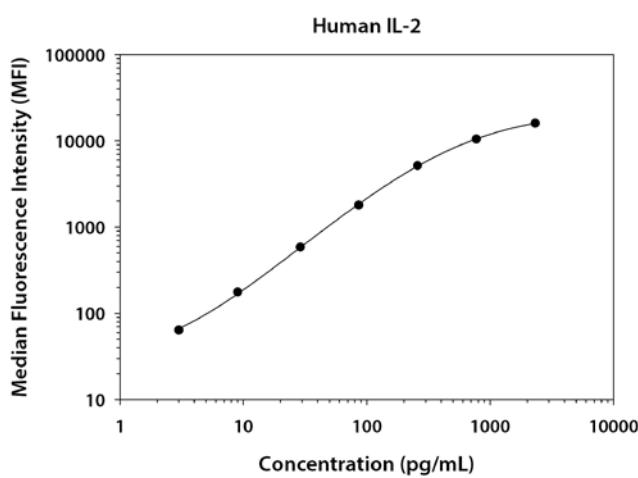
- Store the unopened kit at 2-8 °C. Do not use past the expiration date on the label.
- Avoid freezing microparticles.**
- Protect microparticles from light.**

Instructions for Use

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

TYPICAL DATA

This human IL-2 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	75 87	81	—
1	2320	15,942 16,306	16,124	16,043
2	773	10,557 10,583	10,570	10,489
3	258	5151 5368	5260	5179
4	86	1788 1984	1886	1805
5	29	660 675	668	587
6	9	257 260	258	177
7	3	140 150	145	64

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.

Thirty-six assays were evaluated, and the MDD of human IL-2 ranged from 0.31-2.23 pg/mL. The mean MDD was 0.89 pg/mL.

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 separate assays to assess precision between assays.

Sample	Intra-assay Precision			Inter-assay Precision		
	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	10	23	203	28	265	543
Standard Deviation	0.52	1.19	6.39	3.7	34	52.5
% CV	5.2	5.2	3.1	13.2	12.8	9.7

Recovery and Linearity - Samples containing and/or spiked with high concentrations of IL-2 were evaluated for recovery and were serially diluted to evaluate assay linearity.

Sample Type	Recovery		Linearity					
	Average % Recovery	Range (%)		Cell culture supernates	Serum	EDTA Plasma	Heparin Plasma	
Cell culture supernates	94	89-98	1:2	Average % of Expected	101	103	104	100
				Range (%)	89-116	99-106	97-109	88-118
Serum	109	98-119	1:4	Average % of Expected	102	99	100	93
				Range (%)	86-124	88-109	91-104	80-114
EDTA plasma	100	88-111	1:8	Average % of Expected	102	95	96	90
				Range (%)	84-121	80-112	89-102	81-105
Heparin plasma	103	90-123						

Specificity - This assay recognizes natural and recombinant human IL-2. 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-6	GM-CSF	ENA-78	IL-8	
CNTF	IL-2 R α	IL-18	GM-CSF	IL-10	IFN- γ	IL-1 α	FGF basic	IL-10
β -ECGF	IL-2 R β	LIF	IFN- γ	IL-17	IL-1 α	IL-1 β	G-CSF	IL-17
FGF acidic	IL-2 R γ	LIF R	IL-1 α	MIP-1 α	IL-1 β	IL-2	GM-CSF	MCP-1
FGF-4	IL-3 R α	MIP-1 δ	IL-1 β	MIP-1 β	IL-2	IL-4	IFN- γ	MIP-1 α
FGF-5	IL-4 R	MIP-3 α	IL-1ra	RANTES	IL-4	IL-6	IL-1 α	MIP-1 β
FGF-6	IL-5 R α	MIP-3 β	IL-2	TNF- α	IL-6	IL-8	IL-1 β	RANTES
FGF-9	IL-6 R	MCP-2	IL-4	Tpo	IL-10	IL-10	IL-1ra	TNF- α
FGF-10	IL-10 R	MCP-3	IL-5	VEGF	TNF- α		IL-4	Tpo
FGF-18	IL-3	MCP-4				IL-5		VEGF
GCP-2	IL-7	M-CSF				IL-6		
GRO α	IL-9	TNF RI						
GRO β	IL-11	TNF- β						
GRO γ	IL-12/IL-23 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 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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