Magnetic Luminex® Performance Assay Human CCL3/MIP-1α Kit

Catalog Number: LUHM270
Pack Size: 100 Tests

SPECIFICATIONS AND USE

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

Microparticle Region
- Region-34

Components
- Microparticle Concentrate (Part 894445) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 892632) 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).

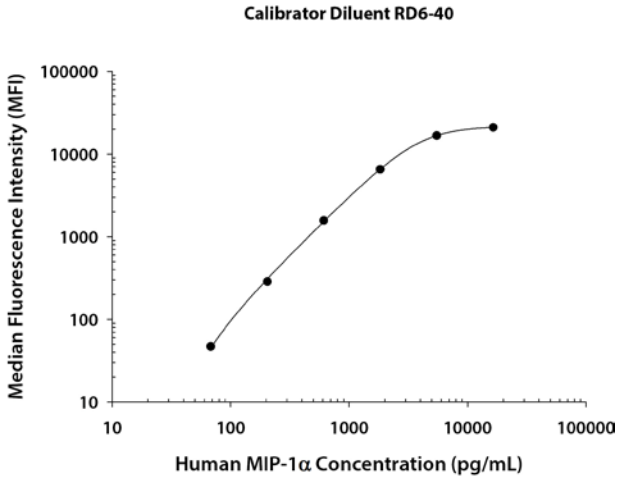
Storage
- Store the unopened kit at 2-8 °C. Do not use past the expiration date on the label.
- **Avoid freezing microparticles.**
- **Protect microparticles from light.**

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

TYPICAL DATA

This human MIP-1α 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.

Note: *When assaying cell culture supernate samples using Calibrator Diluent RD5K, a six-point standard curve (23-5500 pg/mL) is recommended. When assaying serum/plasma samples using Calibrator Diluent RD6-40, a six-point standard curve (67.9-16,500 pg/mL) is recommended.*



Standard	pg/mL	MFI	Average	Corrected
Blank	0	74 76	75	____
1	16,500	21,019 21,063	21,041	20,966
2	5500	16,739 16,900	16,820	16,745
3	1833	6566 6615	6591	6516
4	611	1634 1660	1647	1572
5	204	356 367	361	286
6	68	121 122	122	47

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.

Ten assays were evaluated, and the MDD of human MIP-1α ranged from 1.25-28.3 pg/mL. The mean MDD was 8.11 pg/mL.

R&D Systems, Inc.
1-800-343-7475

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.

	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	11	11	10
Mean (pg/mL)	212	1689	4791	181	1765	6563
Standard Deviation	22.8	192	730	30	84	924
% CV	10.8	11.4	15.2	16.5	4.8	14.1

Recovery and Linearity – Samples containing and/or spiked with high concentrations of MIP-1α were evaluated for recovery and were serially diluted to evaluate assay linearity.

Recovery		
Sample Type	Average % Recovery	Range (%)
Cell culture supernates	95	84-104
Serum	106	95-114
EDTA plasma	100	77-111
Heparin plasma	112	104-129

Linearity					
		Cell culture supernates	Serum	EDTA Plasma	Heparin Plasma
1:2	Average % of Expected	103	102	98	94
	Range (%)	93-122	93-108	80-112	79-104
1:4	Average % of Expected	103	95	99	94
	Range (%)	83-124	83-101	81-116	84-109
1:8	Average % of Expected	106	96	97	96
	Range (%)	88-121	83-103	76-116	83-111

Specificity - This assay recognizes natural and recombinant human MIP-1α. 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-1β
FGF-6	IL-5 Rα	MIP-3β	IL-2	Tpo	IL-6	IL-8	IL-1β	RANTES
FGF-9	IL-6 R	MCP-2	IL-4	TNF-α	IL-10	IL-10	IL-1ra	Tpo
FGF-10	IL-10 R	MCP-3	IL-5	VEGF	TNF-α	Leptin	IL-2	TNF-α
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.