Magnetic Luminex® Performance Assay Human CXCL5/ENA-78 Kit

Catalog Number: LUHM254
Pack Size: 100 Tests

SPECIFICATIONS AND USE

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

Microparticle Region
- Region-12

Components
- Microparticle Concentrate (Part 894440) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 892627) is supplied as a 100X concentrated stock solution (0.075 mL) with preservatives.

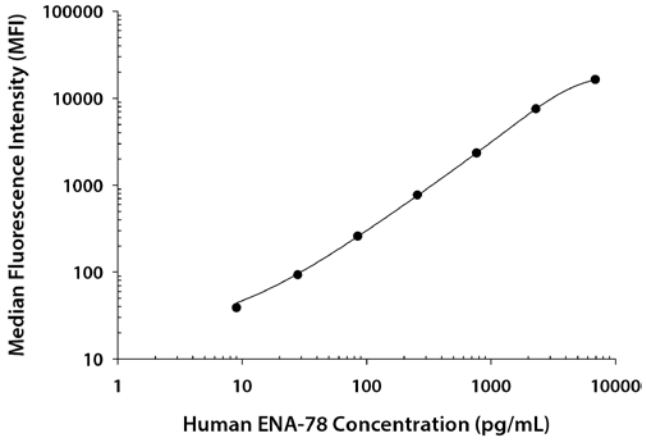
Other Supplies Required
- Magnetic Luminex Performance Assay Human Base Kit A (Catalog Number LUHM000).

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 ENA-78 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	16 16	16	—
1	6900	16,337 16,437	16,387	16,371
2	2300	7517 7582	7549	7533
3	767	2354 2363	2358	2342
4	256	787 789	788	772
5	85	274 276	275	259
6	28	108 110	109	93
7	9	55 55	55	39

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.

Twenty-two assays were evaluated, and the MDD of human ENA-78 ranged from 1.44-4.14 pg/mL. The mean MDD was 2.71 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 separate assays to assess precision between assays.

	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	296	682	1446	310	682	1390
Standard Deviation	14	28	44.2	33	69	127
% CV	4.7	4.1	3.1	10.6	10.1	9.1

Recovery and Linearity – Samples containing and/or spiked with high concentrations of ENA-78 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	105	92-118	1:2	Average % of Expected	95	100	100	99
				Range (%)	86-119	87-106	87-139	85-107
Serum	97	79-122	1:4	Average % of Expected	89	100	95	97
				Range (%)	85-95	90-107	49-170	79-110
EDTA plasma	102	86-121	1:8	Average % of Expected	88	99	97	91
				Range (%)	83-96	86-115	67-188	71-104
Heparin plasma	108	96-126						

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