Magnetic Luminex® Performance Assay Human VEGF Kit

Catalog Number: LUHM293
Pack Size: 100 Tests

SPECIFICATIONS AND USE

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

Microparticle Region
- Region-39

Components
- Microparticle Concentrate (Part 894451) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Biotin-Antibody Concentrate (Part 892638) 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) or Magnetic Luminex Performance Assay Human Angiogenesis Base Kit A (Catalog Number LANM000).

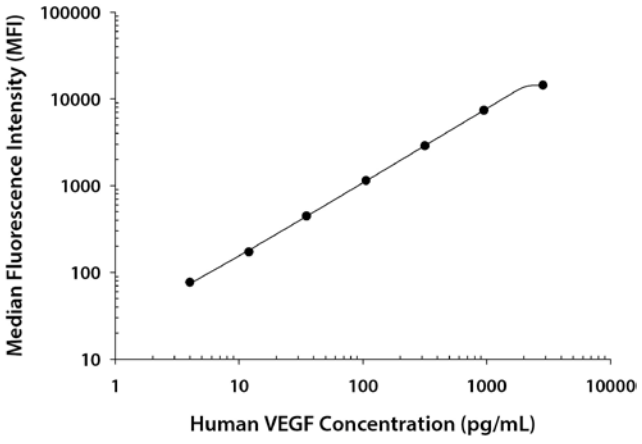
Storage
- Store the unopened kit at 2-8 °C. Do not use past the expiration date on the label.
- **Avoid freezing microparticles.**
- **Protect microparticles from light.**

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

TYPICAL DATA

This human VEGF 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: *This kit, when used with Human Base Kit A, utilizes a 3-fold dilution series.*
This kit, when used with the Human Angiogenesis Base Kit A, utilizes a 4-fold dilution series.



Standard	pg/mL	MFI	Average	Corrected
Blank	0	52 53	53	——
1	2850	14,427 14,428	14,427	14,374
2	950	7452 7455	7454	7401
3	317	2928 2934	2931	2878
4	106	1190 1204	1197	1144
5	35	494 501	497	444
6	12	224 226	225	172
7	4	129 130	130	77

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.

Forty-three assays were evaluated, and the MDD of human VEGF ranged from 0.24-1.84 pg/mL. The mean MDD was 0.81 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)	21	71	527	100	920	1015
Standard Deviation	1.4	5.0	19.5	7.8	87	87
% CV	6.7	7.0	3.7	7.8	9.5	8.6

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

Recovery		
Sample Type	Average % Recovery	Range (%)
Cell culture supernates	98	86-120
Serum	109	95-131
EDTA plasma	91	59-118
Heparin plasma	101	93-120

Linearity					
		Cell culture supernates	Serum	EDTA Plasma	Heparin Plasma
1:2	Average % of Expected	93	103	111	98
	Range (%)	84-103	88-116	93-132	88-112
1:4	Average % of Expected	92	101	113	102
	Range (%)	78-101	77-120	90-147	94-111
1:8	Average % of Expected	94	99	114	111
	Range (%)	76-130	80-122	88-142	90-133

Specificity - This assay recognizes natural and recombinant human VEGF. 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 Angiogenesis Panel:
6Ckine	IL-2 Rα	LIF	G-CSF	GM-CSF	GM-CSF	ENA-78	IL-8	Angiogenin
CNTF	IL-2 Rβ	LIF R	GM-CSF	IFN-γ	IL-1α	G-CSF	IL-10	Angiopoietin-1
β-ECGF	IL-2 Rγ	MIP-1α	IFN-γ	IL-1α	IL-1β	GM-CSF	IL-17	Endostatin
FGF acidic	IL-3 Rα	MIP-3α	IL-1α	IL-1β	IL-2	IFN-γ	MCP-1	FGF acidic
FGF-4	IL-4 R	MIP-3β	IL-1ra	IL-2	IL-4	IL-1α	MIP-1α	FGF basic
FGF-5	IL-5 Rα	MCP-2	IL-1	IL-4	IL-6	IL-1β	MIP-1β	PlGF
FGF-6	IL-6 R	MCP-3	IL-2	IL-6	IL-8	IL-1ra	RANTES	PDGF-AA
FGF-9	IL-10 R	MCP-4	IL-4	IL-10	IL-10	IL-2	Tpo	PDGF-BB
FGF-10	IL-3	M-CSF	IL-5	TNF-α	Leptin	IL-4	TNF-α	Thrombospondin-2
FGF-18	IL-7	TNF RI	IL-6		TNF-α	IL-5	VEGF	VEGF-D
GCP-2	IL-9	TNF-α	IL-8			IL-6		
GROα	IL-11	VEGF-D	IL-10					
GROβ	IL-12 p40		IL-17					
GROγ	IL-12 p70		MIP-1α					
I-309	IL-13		MIP-1β					
IGF-I	IL-15		RANTES					
IGF-II	IL-16		Tpo					
IL-1 RI	IL-17		TNF-α					
IL-1 RII	IL-18		VEGF					

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.