

Magnetic Luminex® Performance Assay Human IFN- γ Kit

Catalog Number: LUHM285 Pack Size: 100 Tests

SPECIFICATIONS AND USE

Recommended Sample Types	Cell culture supernates, serum, EDTA plasma, and heparin plasma.
Microparticle Region	• Region-18
Components	• Microparticle Concentrate (Part 894444) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
	• Biotin-Antibody Concentrate (Part 892631) 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 IFN-γ 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	20 20	20	
1	3250	13,539 13,637	13,588	13,568
2	1083	7081 7167	7124	7104
3	361	2890 2896	2893	2873
4	120	1243 1258	1250	1230
5	40	492 504	498	478
6	13	206 211	209	189
7	4	99 101	100	80

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-two assays were evaluated, and the MDD of human IFN- γ ranged from 0.09-1.27 pg/mL. The mean MDD was 0.31 pg/mL.

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.

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.

	Int	ra-assay Precisi	on	Inter-assay Precision			
Sample	1	2	3	1	2	3	
n	20	20	20	20	20	20	
Mean (pg/mL)	10	28	196	29	270	445	
Standard Deviation	0.76	1.86	6.04	3.6	14.0	23.2	
% CV	7.6	6.6	3.0	12.4	5.2	5.2	

Recovery and Linearity – Samples containing and/or spiked with high concentrations of IFN- γ 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	100	05 107		1.2	Average % of Expected	100	100	104	83	
supernates	102	93-107		1:2	Range (%)	83-132	96-105	100-108	66-98	
Serum	103	92-119		1:4	Average % of Expected	99	101	107	82	
					Range (%)	79-126	93-106	105-109	71-96	
EDTA plasma 99	00	55-114		1:8	Average % of Expected	86	104	103	85	
	99				Range (%)	75-122	93-112	100-109	72-97	
Heparin plasma	99	86-111								

Specificity - This assay recognizes natural and recombinant human IFN-γ. 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-8
CNTF	IL-2 Ra	IL-18	GM-CSF	IL-10	IFN-7	IL-1α	FGF basic	IL-10
β-ECGF	IL-2 Rβ	LIF	IFN-γ	IL-17	IL-1α	IL-1β	G-CSF	IL-17
FGF acidic	IL-2 Rγ	LIF R	IL-1α	MIP-1a	IL-1β	IL-2	GM-CSF	MCP-1
FGF-4	IL-3 Ra	MIP-1a	IL-1ra	MIP-1β	IL-2	IL-4	IL-1α	MIP-1a
FGF-5	IL-4 R	MIP-3a	IL-1	RANTES	IL-4	IL-6	IL-1β	MIP-1β
FGF-6	IL-5 Ra	MIP-3β	IL-2	Тро	IL-6	IL-8	IL-1ra	RANTES
FGF-9	IL-6 R	MCP-2	IL-4	TNF- α	IL-10	IL-10	IL-2	Тро
FGF-10	IL-10 R	MCP-3	IL-5	VEGF	TNF-α	Leptin	IL-4	TNF-a
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						
GR0β	IL-11	TNF-α						
GR0γ	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.