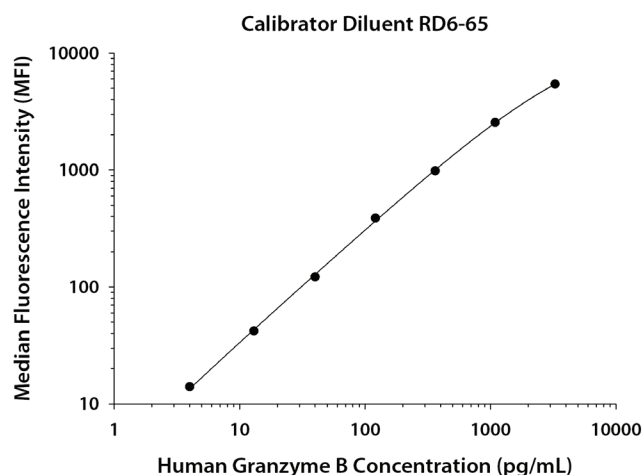


- Recommended Sample Types** • Cell culture supernates, serum, EDTA plasma, and heparin plasma
- Microparticle Region** • Region-38
- Components** • Human Granzyme B Magnetic Microparticles (Part 898851) is supplied as a 100X concentrated stock (0.075 mL) with preservatives.
- Other Supplies Required** • Magnetic Luminex® Performance Assay Human XL Discovery Base Kit (R&D Systems®, Catalog # LUXLM000).
- 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 Magnetic Luminex® Performance Assay procedure.

TYPICAL DATA

This human Granzyme B 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	26 27	26	—
1	3270	5447 5461	5454	5428
2	1090	2552 2599	2576	2550
3	363	972 1048	1010	984
4	121	411 416	414	388
5	40	146 150	148	122
6	13	68 68	68	42
7	4	40 41	40	14

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.

Six assays were evaluated, and the MDD of human Granzyme B ranged from 0.463-3.64 pg/mL. The mean MDD was 1.04 pg/mL.

PRECISION

Intra-assay Precision (precision within an assay) - Two samples of known concentration were tested twenty times on one plate to assess intra-assay precision.

Inter-assay Precision (precision between assays) - Two samples of known concentration were tested in twenty-five separate assays to assess inter-assay precision.

Sample	Intra-Assay Precision		Inter-Assay Precision	
	1	2	1	2
n	20	20	25	25
Mean (pg/mL)	32.4	1207	31.3	1306
Standard deviation	0.717	17.7	4.88	158
CV (%)	2.2	1.5	15.6	12.1

RECOVERY

Samples containing and/or spiked with human Granzyme B were evaluated for recovery.

Sample Type	Average % Recovery	Range
Cell culture supernates	106	91-120%
Serum	118	91-155%
EDTA plasma	122	95-145%
Heparin plasma	118	94-164%

LINEARITY

Samples containing and/or spiked with human Granzyme B were serially diluted to evaluate assay linearity.

		Cell culture supernates	Serum	EDTA plasma	Heparin plasma
1:2	Average % of Expected	102	107	95	99
	Range (%)	98-105	106-109	94-96	93-102
1:4	Average % of Expected	103	98	97	112
	Range (%)	96-107	94-102	95-99	107-119
1:8	Average % of Expected	108	96	95	111
	Range (%)	94-115	91-99	92-99	104-119

SPECIFICITY

Note: Refer to the base kit insert for a complete list of analytes tested for cross-reactivity and interference.

This assay recognizes natural and recombinant human Granzyme B.

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.

Magnetic 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.