Human Granzyme A

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Scientific Support

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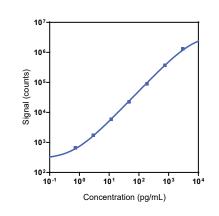
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www.mesoscale.com®	Product Options	Catalog Number	Description
	Multiplex	K151AEM, K251AEM	U-PLEX Immuno-Oncology Group 1 (human)
Ordering Information MSD Customer Service Phone: 1-240-314-2795 Fax: 1-301-990-2776 Email: CustomerService@ mesoscale.com	Singleplex	K151G8K-1/-2/-4	U-PLEX Human Granzyme A Assay with SECTOR [™] plates
		K151G8K-21/-22/-24	U-PLEX Human Granzyme A Assay with QuickPlex Ultra $^{\rm M}$ plates
		K251G8K-2/-4	U-PLEX Human Granzyme A Assay with 384-well plates
	Antibody Set	B22G8-2/-3	U-PLEX Human Granzyme A Antibody Set
	Protocol	U-PLEX Product Inserts are available at <u>www.mesoscale.com</u>	

The MESO SCALE DISCOVERY® U-PLEX platform was designed to provide ultimate flexibility for detection of biomarkers in a wide variety of sample types. This datasheet provides the representative performance of the U-PLEX® Human Granzyme A Assay tested on U-PLEX 96-well SECTOR plates run as a multiplex. The data do not represent the product specifications. Under your experimental conditions, the assay may perform differently from the representative data. U-PLEX assays are offered in either singleplex or multiplex; both are available on 96- or 384-well plates. See a U-PLEX product insert for instrument compatibility.

Representative Calibration Curve and Sensitivity



Assay	Median LLOD (pg/mL)	LLOD Range (pg/mL)	
Granzyme A	0.49	0.35-0.61	

The Calibrator curve was fitted with a 4-parameter logistic model with a 1/Y² weighting. The lower limit of detection (LLOD) is a calculated concentration corresponding to 2.5 standard deviations above the background (zero Calibrator).

Precision

Control	Average Conc. (pg/mL)	Average Intra-run Conc. (%CV)	Inter-run Conc. (%CV)
High	792	3.8	3.9
Mid	142	2.7	6.6
Low	23	2.9	8.1

Controls were made by spiking Calibrator into assay diluent at 3 levels within the quantitative range of the assay. Average intra-run concentration %CV is the average %CV of the control replicates within an individual run. Inter-run concentration %CV is the variability of controls across multiple runs.

For Research Use Only. Not for use in diagnostic procedures.





MSD® U-PLEX Human Granzyme A

Tested Samples

Sample Type	Serum (N=10)	EDTA Plasma (N=10)	Normal Lysate (N=5)	Tumor Lysate (N=5)
Median (pg/mL)	17	31	246	705
Range (pg/mL)	13-23	16-81	48-898	16-11,000
% Detected	100	100	100	100

Normal serum and plasma samples were diluted 4-fold prior to the assay. Lysates were tested at a protein concentration of 0.5 mg/mL.

Dilution Linearity

Serum			EDTA Plasma		
Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range
2	92	84 - 105	2	94	82 - 104
8	105	98 - 111	8	102	97 - 108
16	111	96 - 127	16	109	101 - 120

Normal human serum and EDTA plasma were spiked with Calibrator and tested at different dilutions. Percent recovery at each dilution level was normalized to the dilutionadjusted, 4-fold concentration. Samples may benefit from additional dilution with assay diluent to reduce matrix effects.

% Recovery = (measured concentration / expected concentration) x 100

Spike Recovery

	Ser	um	EDTA Plasma	
Spike Level	Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range
High	73	60 - 86	107	102 - 117
Mid	82	65 - 93	108	100 - 119
Low	82	65 - 93	112	106 - 116

Normal serum and plasma were spiked with Calibrator at 3 levels. Spiked samples were diluted 4-fold to determine the expected concentration of the analyte. Samples may benefit from additional dilution with assay diluent to reduce matrix effects.

% Recovery = (measured concentration / expected concentration) x 100

Specificity

To assess specificity, the Granzyme A Antibody Set was tested individually against a larger panel of analytes for nonspecific binding: APRIL/TNFSF13, BAFF R/TNFRSF13C, BCMA/TNFRSF17, CD20, CD27, CD276/B7-H3, CD28, CD40L (soluble), CTACK, CTLA-4, ENA-78, Eotaxin, Eotaxin-2, Eotaxin-3, EPO, E-Selectin, FGF (basic), FLT3L, Fractalkine, G-CSF, Galectin-9, GITR/TNFRSF18, GITRL/TNFSF18, GM-CSF, gp130 (soluble), Granzyme A, Granzyme B, GRO- α , HAVCR2/TIM-3, HVEM/TNFRSF14, I-309, ICOS, ICOSL/B7-H2, IFN- α 2a, IFN- β , IFN- γ , IL-1 α , IL-1 β , IL-1RA, IL-10, IL-12/IL-23p40, IL-12p70, IL-13, IL-15, IL-16, IL-17A, IL-17A/F, IL 17C, IL-17D, IL-17E/IL 25, IL-17F, IL-18, IL-21, IL-22, IL 23, IL-27, IL-29/IFN- λ 1, IL-2R α , IL-3, IL-31, IL-33, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IP-10, I-TAC, LAG 3, LIGHT/TNFSF14, MCP-1, MCP-2, MCP-4, M CSF, MDC, MIF, MIG, MIP-1 α , MIP-5, MMP-1, MMP-2, MMP-7, MMP-9, Nectin-4, OX40/TNFRSF4, PD1, PD-L1, PD-L2, Pentraxin 3, Perforin, PIGF, P Selectin, RAGE (soluble), RANKL/TNFSF11, RANTES, S100A12, TARC, Tie-2, TIGIT, TLR-1, TNF-RI, TNF-RII, TNF- α , TNF β , TPO, TRAIL, TSLP, VEGF-A, VEGF-D, VEGFR-1/FIt-1 and YKL-40. Nonspecific binding was less than 2.0%.

% Nonspecificity = (nonspecific signal / specific signal) x 100

Diluent Compatibility

Diluents 58 and 3 are provided with this assay. MSD offers a range of assay and antibody diluents for separate purchase. Depending on your assay needs, other diluents may be tested.

Assay Components

Calibrator: Granzyme A is included in Calibrator 23. The human Granzyme A Calibrator is Granzyme A (26–262) expressed in a mouse cell line. **Antibodies:** The U-PLEX Human Granzyme A Assay uses a mouse monoclonal antibody for capture and a mouse monoclonal antibody for detection. **Assay generation:** A

Note: This datasheet contains representative assay performance data. In custom multiplex formats, the assay may perform differently from the representative data shown.

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