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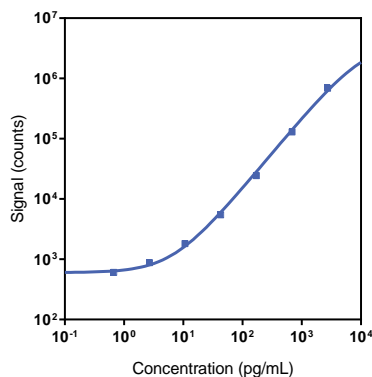
## Company Address

MESO SCALE DISCOVERY<sup>®</sup>  
 A division of  
 Meso Scale Diagnostics, LLC.  
 1601 Research Boulevard  
 Rockville, MD 20850-3173 USA

Product Options	Catalog Number	Description
Multiplex	K151ACM, K251ACM	U-PLEX Metabolic Group 1 (hu)
	K151AEM, K251AEM	U-PLEX Immuno-Oncology Group 1 (hu)
Singleplex	1515XK-1/-2/-4	U-PLEX Human Ghrelin (total) Assay with SECTOR <sup>™</sup> plates
	K1515XK-21/-22/-24	U-PLEX Human Ghrelin (total) Assay with QuickPlex <sup>®</sup> plates
	K2515XK-2/-4	U-PLEX Human Ghrelin (total) Assay with 384-well plates
Antibody Set	B215X-2/-3	U-PLEX Human Ghrelin (total) Antibody Set
Protocol	U-PLEX Product Inserts are available at <a href="http://www.mesoscale.com">www.mesoscale.com</a>	

The U-PLEX<sup>®</sup> 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 Ghrelin (total) Assay tested on U-PLEX 96-well 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)
Ghrelin (total)	1.7	1.4-1.9

The Calibrator curve was fitted with a 4-parameter logistic model with a  $1/Y^2$  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	1,770	3.2	10.8
Mid	687	3.0	11.6
Low	280	3.0	11.8

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 Ghrelin (total)

## Tested Samples

Sample Type	Serum (N=12)	EDTA Plasma (N=12)	P800 Plasma (N=8)
Median (pg/mL)	2.2	15	57
Range (pg/mL)	ND-4.3	ND-112	10-88
% Detected	33	58	100

Normal serum, EDTA plasma, and P800 plasma samples were diluted 4-fold prior to the assay. ND = non-detectable (<LLOD)

## Dilution Linearity

Serum			EDTA Plasma			P800 Plasma			Cell Culture Media		
Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range
2	81	72-87	2	90	87-93	2	88	83-92	2	94	86-100
8	112	106-119	8	104	100-108	8	103	99-106	8	106	102-110
16	126	116-135	16	107	99-113	16	110	102-117	16	112	106-117

Normal human serum, EDTA plasma, P800 plasma, and cell culture media were spiked with Calibrator and tested at different dilutions. Percent recovery at each dilution level was normalized to the dilution-adjusted, 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

Spike Level	Serum		EDTA Plasma		P800 Plasma		Cell Culture Media	
	Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range
High	75	44-88	95	84-108	92	84-98	89	82-95
Mid	79	52-92	97	90-109	95	90-101	87	81-91
Low	79	44-94	99	87-109	95	92-101	89	83-93

Normal serum, EDTA plasma, P800 plasma, and cell culture media 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 Ghrelin (total) Antibody Set was tested individually against a larger panel of analytes for nonspecific binding (APRIL/TNFSF13, BAFF, BAFF-R/TNFRSF13C, BCMA/TNFRSF17, BDNF, C-Peptide, CD20, CD27, CD28, CD40L (soluble), CD276/B7-H3, CTACK, CTLA-4, Desghrelin, ENA-78, Eotaxin, Eotaxin-2, Eotaxin-3, EPO, E-Selectin, FGF (basic), FGF-21, FGF-23, FLT3L, Fractalkine, FSH, Galectin-9, G-CSF, GITRL/TNFSF18, GTR/TNFRSF18, Ghrelin (Ser3-octanoylated), gp130 (soluble), GIP (1-42), GIP (3-42), GLP-1 (7-36), GLP-1 (9-36), Glucagon, GM-CSF, Granzyme A, Granzyme B, GRO- $\alpha$ , HAVCR2/TIM-3, HVEM/TNFRSF14, ICOS, ICOS-L/B7-H2, I-309, IFN- $\alpha$ 2a, IFN- $\beta$ , IFN- $\gamma$ , IL-1 $\alpha$ , IL-1 $\beta$ , IL-1RA, IL-2, IL-2R $\alpha$ , IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, 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- $\lambda$ 1, IL-31, IL-33, Insulin, IP-10, LAG3, Leptin, LH, LIGHT/TNFSF14, MCP-1, MCP-2, MCP-4, M-CSF, MDC, MIF, MIG, MIP-1 $\alpha$ , MIP-1 $\beta$ , MIP-5, MMP-1, MMP-2, MMP-7, Nectin-4, OX40/TNFRSF4, PD1, PD-L1, PD-L2, Pentraxin 3, Perforin, PIGF, PP, Proinsulin, proMMP-9, P-Selectin, PYY (3-36), RAGE (soluble), RANKL/TNFSF11, RANTES, S100A12, SDF-1 $\alpha$ , Tie-2, TIGIT, TLR1, TNF- $\alpha$ , TNF- $\beta$ , TNF-RI, TNF-RII, TPO, TRAIL, TSLP, VEGF-A, VEGF-D, VEGFR-1/Flt-1, YKL-40, and  $\beta$ -NGF). Nonspecific binding was less than 2.0%.

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

Octanoylated Ghrelin (Calibrator for Ghrelin [active]) cross-reacts with the Ghrelin (total) assay as expected. We do not recommend multiplexing the Ghrelin (total) and Ghrelin (active) assays on the same plate.

## Diluent Compatibility

The data included in this document were collected with Assay Diluent 13 (supplemented with 1,000 kIU/mL Aprotinin [provided] and 100  $\mu$ M diprotin A [not provided]) and Antibody Diluent 11. MSD offers a range of assay and antibody diluents for separate purchase. Depending on your assay needs, other diluents may be tested. Diprotin A should be purchased separately.

## Assay Components

**Calibrator:** Ghrelin (total) is included in Calibrator 13. The human Ghrelin (total) Calibrator is a synthetic peptide.

**Antibodies:** The U-PLEX Human Ghrelin (total) Assay uses a mouse monoclonal antibody for capture and a rabbit polyclonal 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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