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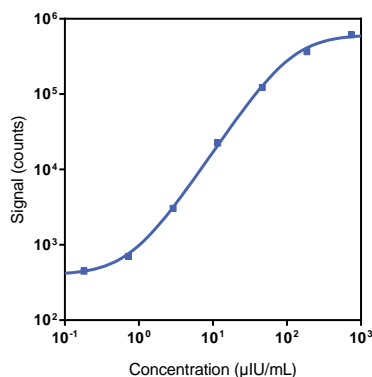
Company Address

MESO SCALE DISCOVERY[®]
 A division of
 Meso Scale Diagnostics, LLC.
 1601 Research Boulevard
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Product Options	Catalog Number	Description
Multiplex	K151ACM, K251ACM	U-PLEX Metabolic Group 1 (hu)
	K151AEM, K251AEM	U-PLEX Immuno-Oncology Group 1 (hu)
Singleplex	K1516HK-1/-2/-4	U-PLEX Human Insulin Assay with SECTOR [™] plates
	K1516HK-21/-22/-24	U-PLEX Human Insulin Assay with QuickPlex [®] plates
	K2516HK-2/-4	U-PLEX Human Insulin Assay with 384-well plates
Antibody Set	B216H-2/-3	U-PLEX Human Insulin Antibody Set
Protocol	U-PLEX Product Inserts are available at www.mesoscale.com	

The 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 Insulin 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 (µIU/mL)	LLOD Range (µIU/mL)
Insulin	0.32	0.31-0.36

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. (µIU/mL)	Average Intra-run Conc. (%CV)	Inter-run Conc. (%CV)
High	347	3.4	16.8
Mid	89	2.6	15.3
Low	15	2.7	20.8

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 Insulin

Tested Samples

Sample Type	Serum (N=12)	EDTA Plasma (N=12)	P800 Plasma (N=8)
Median (µU/mL)	5.9	5.9	5.7
Range (µU/mL)	1.2-21	1.1-22	1.6-33
% Detected	100	100	100

Normal serum, EDTA plasma, and P800 plasma samples were diluted 4-fold prior to the assay.

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	83	76-88	2	85	80-90	2	82	72-99	2	82	71-90
8	110	107-112	8	107	105-112	8	110	101-117	8	112	108-117
16	106	103-110	16	105	101-110	16	113	101-126	16	113	103-121

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.

$$\% \text{ Recovery} = (\text{measured concentration} / \text{expected concentration}) \times 100$$

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	72-77	83	79-90	80	73-89	82	73-91
Mid	84	83-88	91	85-96	89	80-100	91	88-93
Low	92	88-97	100	95-105	97	91-103	100	95-105

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.

$$\% \text{ Recovery} = (\text{measured concentration} / \text{expected concentration}) \times 100$$

Specificity

To assess specificity, the Insulin 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-α, HAVCR2/TIM-3, HVEM/TNFRSF14, ICOS, ICOS-L/B7-H2, I-309, IFN-α2a, IFN-β, IFN-γ, IL-1α, IL-1β, IL-1RA, IL-2, IL-2Rα, 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-λ1, IL-31, IL-33, Insulin, IP-10, LAG3, Leptin, LH, LIGHT/TNFSF14, MCP-1, MCP-2, MCP-4, M-CSF, MDC, MIF, MIG, MIP-1α, MIP-1β, 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-1α, Tie-2, TIGIT, TLR1, TNF-α, TNF-β, TNF-RI, TNF-RII, TPO, TRAIL, TSLP, VEGF-A, VEGF-D, VEGFR-1/Flt-1, YKL-40, and β-NGF). Nonspecific binding was less than 2.0%.

$$\% \text{ Nonspecificity} = (\text{nonspecific signal} / \text{specific signal}) \times 100$$

The Insulin assay will cross-react with the Proinsulin assay. We do not recommend multiplexing the Insulin assay with the Proinsulin assay 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 µM 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: Insulin is included in Calibrator 15. The human Insulin Calibrator is a full-length recombinant protein expressed in *E. coli*.

Antibodies: The U-PLEX Human Insulin 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.

