MSD® Active GLP-1 (ver. 2) Assay Kit

DATA SHEET

For the quantitative determination of Active GLP-1 in human, mouse, rat serum and plasma

Alzheimer's Disease
BioProcess
Cardiac
Cell Signaling
Clinical Immunology
Cytokines
Hypoxia
Immunogenicity
Inflammation
Metabolic
Oncology
Toxicology
Vascular

Catalog Numbers

Active GLP-1 (ver. 2) Kit	
Kit size	
1 plate	K150JWC-1
5 plates	K150JWC-2
25 plates	K150JWC-4

Ordering information:

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MSD Technology Overview

MESO SCALE DISCOVERY'S MULTI-ARRAY® Technology is a multiplex immunoassay system that enables the measurement of biomarkers utilizing the next generation of electrochemiluminescent detection. In an MSD assay, specific capture antibodies for the analytes are coated in arrays in each well of a 96-well carbon electrode plate surface. The detection system uses patented SULFO-TAG™ labels that emit light upon electrochemical stimulation initiated at the electrode surfaces of the MULTI-ARRAY and MULTI-SPOT® plates. MSD assays have low background, are highly stable and are non-radioactive.

MSD assays require minimal sample volume compared to traditional ELISA. With an MSD assay, ten different biomarkers can be analyzed simultaneously with typical sample volumes less than 25 µL. These assays have high sensitivity, up to five logs of linear dynamic range, and excellent performance in complex biological matrices. Combined, these advantages enable the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions.

This datasheet outlines the performance of MSD Active GLP-1 (ver. 2) assay.

Glucagon-like peptide-1 (GLP-1), a post-translational product of preproglucagon, is a 3.5 kD protein hormone produced in intestinal L cells and plays a key role in the promotion of glucose-dependent insulin secretion and insulin biosynthesis. In addition, GLP-1 works in concert with insulin to inhibit glucose secretion and thus lower overall blood glucose levels. Through the activation of different physiological systems, it plays roles in gastric emptying upon nutrient intake and in the regulation of short-term feeding behavior. Upon release, its action is mediated through a single G-protein-coupled receptor. The cleaved peptides, commonly referred to as GLP-1 (7-36) amide and GLP-1 (7-37) are the biologically active forms of GLP-1. In vivo, these active isoforms are rapidly cleaved by dipeptidyl peptidase IV (DPP IV). The primary amino acid sequence for GLP-1 is conserved among mammalian species, i.e. human, mouse, rat, monkey, canine, etc. MSD offers a comprehensive array of GLP-1 assays that measure active, total and amidated isoforms of the GLP-1 protein using detection antibodies that recognize the amino acids in the C-terminus region of the peptide.

Assay Sensitivity

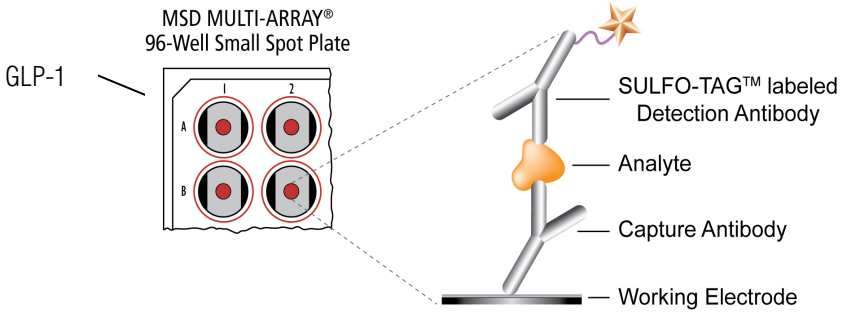
Note: 1 pmol/L = 3.297pg/mL

	Active GLP-1
LLOD (pg/mL)	0.12

The lower limit of detection (LLOD) is the calculated concentration of the signal that is 2.5 standard deviations over the zero Calibrator.

The MSD Active GLP-1 (ver. 2) assay is available in the singleplex format on MSD 96-well Small Spot plate. This assay detects GLP-1 (7-36) amide and GLP-1 (7-37) isoforms using 25 µL sample volume.

For a complete list of products, please check our website at www.mesoscale.com.

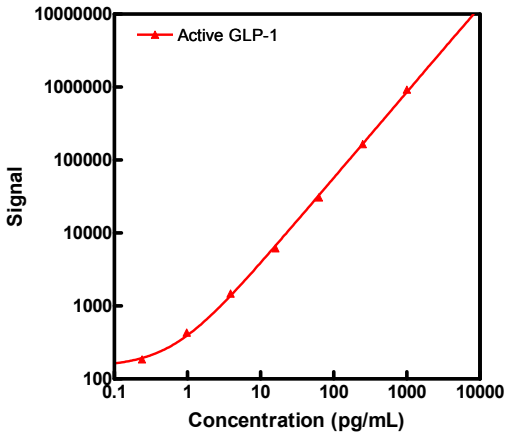


Example Protocol

1. Add Blocking solution, incubate for 1 hour at RT.
2. Wash. Add Assay Diluent + Calibrator / Sample, incubate for 2 hours at RT.
3. Wash. Add Detection Antibody, incubate for 1 hour at RT.
4. Wash. Add Read Buffer T, read.

Typical Standard Curve:

The following standard curves demonstrate the wide dynamic range (3-4 logs) of the MSD Active GLP-1 (ver. 2) assay. This allows for accurate quantification of many biological samples without the need for dilution.



Active GLP-1		
Conc. (pg/mL)	**Average Signal**	**%CV**
0	92	8.0
0.24	185	3.6
0.98	430	6.4
3.9	1486	4.5
16	6184	1.9
63	30816	0.7
250	164880	1.1
1000	925335	0.5

Spike Recovery:

Serum, EDTA plasma, and heparin plasma samples from human, mouse and rat were spiked with the Calibrators at multiple values throughout the range of the assay. Measured analyte represents average spike recovery in multiple pooled serum and plasma samples. Results of spike-recovery may vary based on the individual samples.
% Recovery = measured /expected x 100

	Human			
Sample	**Spike Conc. (pg/mL)**	**Measured Conc. (pg/mL)**	**Measured Conc. %CV**	**% Recovery**
Serum	0	0.5	10.3	
	10	11	1.3	103
	100	94	2.6	94
	800	936	4.0	117
EDTA Plasma	0	0.4	23.5	
	10	11	6.6	105
	100	92	4.5	91
	800	909	5.3	114
Heparin Plasma	0	0.5	7.2	
	10	9	4.5	90
	100	85	3.5	84
	800	819	2.6	102

Sample	Mouse				Rat			
	Spike Conc. (pg/mL)	Measured Conc. (pg/mL)	Measured Conc. %CV	% Recovery	Spike Conc. (pg/mL)	Measured Conc. (pg/mL)	Measured Conc. %CV	% Recovery
Serum	0	0.6	4.3		0	0.9	11.9	
	10	11	0.9	105	10	12	4.5	114
	100	89	0.4	88	100	116	1.6	115
	800	858	1.3	107	800	938	3.1	117
EDTA Plasma	0	0.7	8.7		0	0.4	2.8	
	10	9	1.8	85	10	11	2.9	104
	100	68	5.8	68	100	83	1.0	82
	800	631	6.5	79	800	833	1.0	104
Heparin Plasma	0	2.2	5.7		0	0.4	22.2	
	10	15	7.9	120	10	12	0.0	112
	100	112	6.7	110	100	81	0.3	81
	800	965	6.0	120	800	900	0.9	113

Linearity:

Linearity was measured by spiking Calibrator levels in pooled serum, EDTA and heparin plasma samples from human, mouse and rat followed by subsequent dilution. Percent recovery is calculated as the measured concentration divided by the concentration of the previous dilution (expected).

% Recovery = measured x dilution factor / expected x 100.

Sample	Human			
	Fold Dilution	Conc. (pg/mL)	Conc. %CV	% Recovery
Serum	1	96	4.6	
	2	48	3.1	99
	4	23	1.6	97
	8	13	1.6	109
EDTA Plasma	1	87	0.5	
	2	43	0.9	98
	4	22	3.4	104
	8	11	11.2	101
Heparin Plasma	1	85	3.5	
	2	43	4.0	101
	4	22	2.3	103
	8	12	7.8	105

Sample	Fold Dilution	Mouse Conc. (unit)	Mouse Conc. %CV	Mouse % Recovery	Rat Conc. (unit)	Rat Conc. %CV	Rat % Recovery
Serum	1	49	2.7		45	4.5	
	2	26	0.6	107	25	4.9	111
	4	13	2.7	98	12	0.1	95
	8	7	4.9	111	7	0.1	112
EDTA Plasma	1	40	3.7		44	1.7	
	2	23	2.4	114	27	3.0	120
	4	12	5.8	102	13	4.6	98
	8	7	0.1	114	7	1.1	105
Heparin Plasma	1	44	0.8		39	5.9	
	2	23	0.8	106	23	1.9	116
	4	12	2.7	101	13	3.2	116
	8	7	2.8	111	7	3.6	104

Cross Reactivity:

The cross-reactivity shown below is calculated based on signal generated using different GLP-1 isoforms.

Active GLP-1	
Form	Cross-Reactivity
GLP-1 (7-36) amide	100%
GLP-1 (9-36) amide	< 0.1%
GLP-1 (1-36) amide	< 0.1%
GLP-1 (7-37)	31%
GLP-1 (1-37)	< 0.1%

References using MSD platform for the measurement of GLP-1:

1. Read, P.A., Khan, F.Z., Heck, P.M., Hoole, S.P., Dutka, D.P. (2010) ***DPP-4 Inhibition by Sitagliptin Improves the Myocardial Response to Dobutamine Stress and Mitigates Stunning in a Pilot Study of Patients with Coronary Artery Disease.*** Circ Cardiovasc Imaging. 2010 Jan 14 [Epub ahead of print]
2. Sauve, M., Ban, K., Momen, M.A., Zhou, Y.Q., Henkelman, R.M., Husain, M., Drucker, D.J. (2010) ***Genetic deletion or pharmacological inhibition of dipeptidyl peptidase-4 improves cardiovascular outcomes following myocardial infarction in mice.*** Diabetes. Vol. 59(4):1063-73
3. Fujita, Y., Wideman, R.D., Speck, M., Asadi, A., King, D.S., Webber, T.D., Haneda, M., Kieffer, T.J. (2009) ***Incretin release from gut is acutely enhanced by sugar but not by sweeteners in vivo.*** Am J Physiol Endocrinol Metab. Vol. 296(3):E473-9
4. Lauffer, L.M., Iakoubov, R., Brubaker, P.L. (2009) ***GPR119 is essential for oleoylethanolamide-induced glucagon-like peptide-1 secretion from the intestinal enteroendocrine L-cell.*** Diabetes. Vol. 58(5):1058-66

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