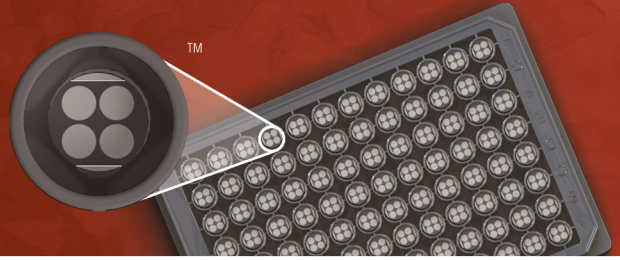
MSD® Total Met Assay Whole Cell Lysate Kit

For quantitative determination in human whole cell lysate samples



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

Catalog Numbers

Total Met Assay: Whole Cell Lysate Kit	
Kit size	
1 plate	K151DMD-1
5 plates	K151DMD-2
20 plates	K151DMD-3

Phospho-Met (Tyr1349) Whole Cell Lysate Set	
200 µg	C11DL-1

Ordering information

MSD Customer Service
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Company Address

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Gaithersburg, MD 20877 USA

www.mesoscale.com®

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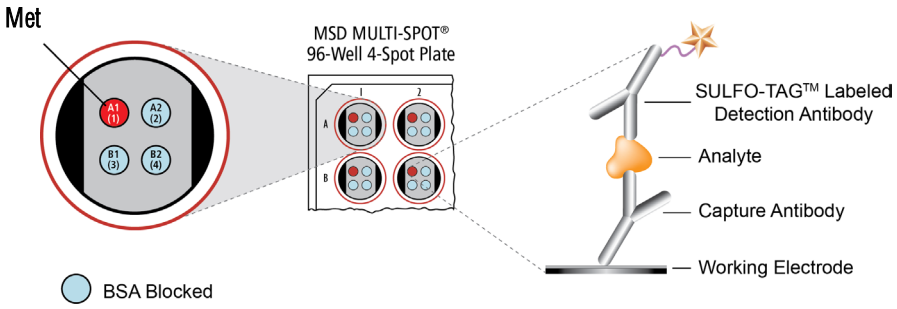

c-Met, also known as the hepatocyte growth factor receptor, is a proto-oncogene with tyrosine kinase activity. Hepatocyte growth factor (HGF) is the only identified ligand for Met and upon ligand binding, the Met receptor dimerizes, autophosphorylates its catalytic residues, and prepares to bind adaptor proteins to continue signaling through downstream mediators.[1] HGF activation of Met induces growth, proliferation, cell survival, motility, and angiogenesis.[2] Met has a tyrosine kinase domain, containing phosphorylated residues Tyr1234 and Tyr1235, and a multi-substrate docking site, containing phosphorylated residues Tyr1349 and Tyr1356.[3] Phosphorylation of these residues are key to the direct biological effects of Met activation as well as the ability of Met to signal through other downstream signaling cascades such as the PI3K signaling cascade, SRC, STAT, and Ras-Raf-Mek-Erk cascades.[4]
Met mutations and receptor upregulation have been identified in many different types of cancer, such as gastric, renal, thyroid, ovarian, pancreatic, prostatic, and breast cancers, colorectal carcinomas, and medulloblastoma.[4] Due to the involvement of Met in so many different types of cancers, there has been a lot of research and drug development directed towards disruption and modulation of the HGF-Met interactions and the downstream signaling cascades controlled through Met-HGF binding.

The MSD Total Met Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Total Met Assay are illustrated below. The signal and ratio values provided below are example data; individual results may vary depending upon the samples tested. Western blot analyses of each lysate type were performed with phospho-Met (Tyr1349) and total Met antibodies and are shown below for comparison.
Growing HeLa cells (negative) were treated with sodium vanadate (1 mM; 4 hours) and HGF (200 ng/mL; 5 minutes) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-total Met antibody on one of the four spatially distinct electrodes per well. Total Met was detected with anti-total Met antibody conjugated with MSD SULFO-TAG™ reagent.

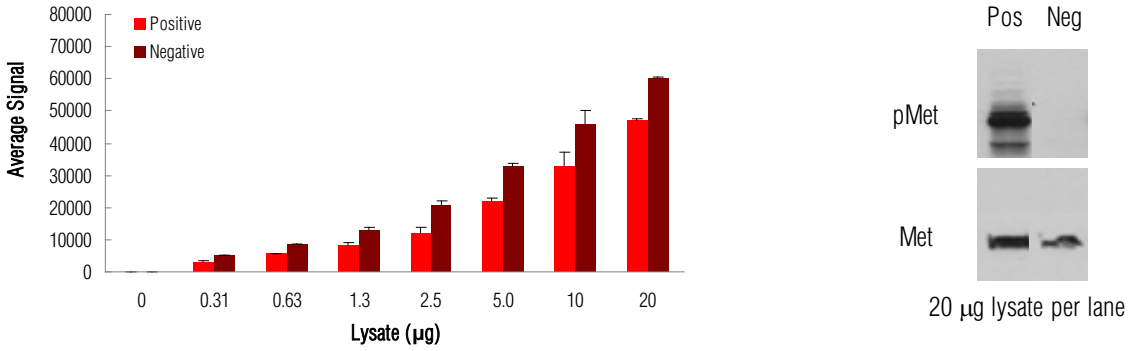

Fig. 1: Sample data generated with the MULTI-ARRAY® Total Met Assay. Increased signal is observed with the titration of both pMet positive and negative cell lysates. The Total Met Assay provides a quantitative measure of the data obtained with the traditional Western blot.

Lysate Titration

Data for pMet positive and negative HeLa cell lysates using the MULTI-ARRAY Total Met Assay are presented below.

Lysate (µg)	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	26	3	11	47	5	11	
0.31	3166	100	3	5177	143	3	0.6
0.63	5517	129	2	8573	406	5	0.6
1.3	8220	820	10	13140	746	6	0.6
2.5	12259	1554	13	20612	848	4	0.6
5.0	22149	966	4	32759	2236	7	0.7
10	32927	4255	13	46027	1848	4	0.7
20	47263	419	1	59943	2291	4	0.8

MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample amounts of 25 µg/well or less without compromising speed or performance
- **Large dynamic range**: Linear range of up to five logs enables the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions
- **Minimal background:** The stimulation mechanism (electricity) is decoupled from the signal (light)
- **Simple protocols:** Only labels near the electrode surface are detected, enabling no-wash assays
- **Flexibility:** Labels are stable, non-radioactive, and conveniently conjugated to biological molecules
- **High sensitivity and precision:** Multiple excitation cycles of each label enhance light levels and improve sensitivity

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

References

1. Yap TA, de Bono JS. Targeting the HGF/c-Met Axis: State of Play. Mol Cancer Ther. 2010 May;9(5):1077-9.
2. Eder JP, Vande Woude GF, Boerner SA, LoRusso PM. Novel therapeutic inhibitors of the c-Met signaling pathway in cancer. Clin Cancer Res. 2009 Apr 1;15(7):2207-14.
3. Birchmeier C, Birchmeier W, Gherardi E, Vande Woude GF. Met, metastasis, motility and more. Nat Rev Mol Cell Biol. 2003 Dec;4(12):915-25.
4. Comoglio PM, Giordano S, Trusolino L. Drug development of MET inhibitors: targeting oncogene addiction and expedience. Nat Rev Drug Discov. 2008 Jun;7(6):504-16.