

# MSD® Phospho-JNK (Thr183/Tyr185) Assay Whole Cell Lysate Kit

For quantitative determination in human, mouse, and rat whole cell lysate samples



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

## Catalog Numbers

Phospho-JNK (Thr183/Tyr185) Assay: Whole Cell Lysate Kit	
Kit size	
1 plate	K150CUD-1
5 plates	K150CUD-2
20 plates	K150CUD-3

Phospho-JNK Whole Cell Lysate Set	
200 µg	C11CU-1

## Ordering information

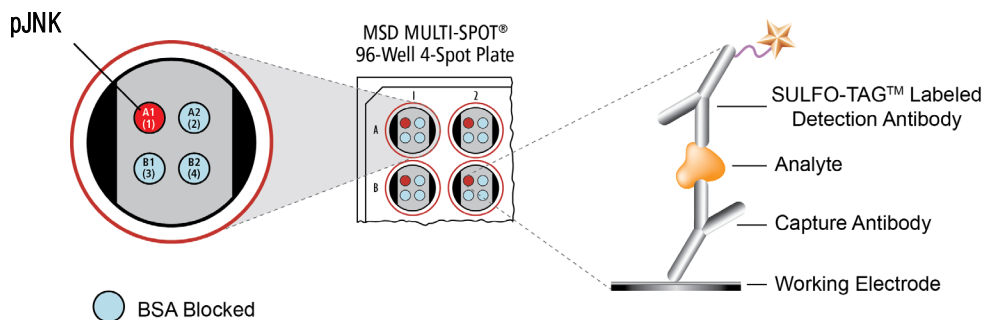
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## Company Address

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Not for use in diagnostic procedures.



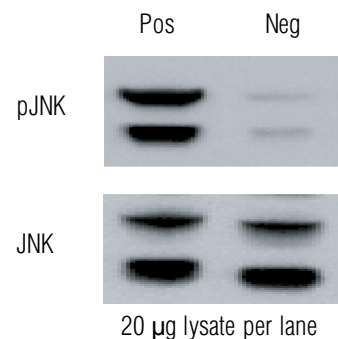
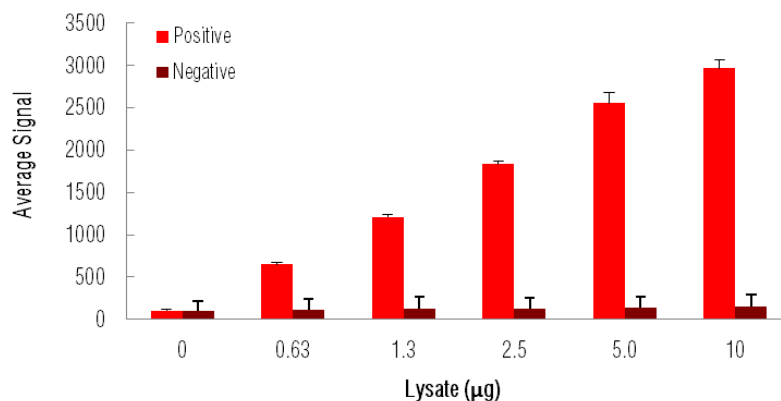
The **c-Jun N-terminal kinase (JNK)** family of kinases, also known as SAPK (stress-activated protein kinase) and MAPK8 (mitogen-activated protein kinase-8), are encoded by three genes (JNK 1-3) that produce more than 10 isoforms. Inflammatory cytokines and cellular stresses such as UV light and osmotic shock lead to JNK activation through the intercellular signaling cascade including Ras and Rac, MEKK1, and MKK4 or MKK7. JNK is fully activated by phosphorylation on threonine 183 and tyrosine 185 by MKK4 and MKK7. Activated JNK in turn phosphorylates the transcription factors c-Jun, ATF-2, and Elk-1, thereby regulating gene transcription. JNK is involved with the function of the mitochondrial apoptotic pathway, mediating the release of cytochrome c and the phosphorylation of Bim and Bmf. JNK also affects expression of many cytokine genes, as well as playing a role in tumor development, embryonic morphogenesis, and cell survival.

The MSD Phospho-JNK (Thr183/Tyr185) Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

## Typical Data

Representative results for the Phospho-JNK (Thr183/Tyr185) 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-JNK (Thr183/Tyr185) and total JNK antibodies and are shown below for comparison.

Growing HEK293 cells were treated with rapamycin (1 µM; 3 hours) (negative). Serum deprived HEK293 cells were harvested 30 minutes after UV irradiation (40 mJ/cm<sup>2</sup>) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-total JNK antibody on one of the four spatially distinct electrodes per well. Phosphorylated JNK was detected with anti-phospho-JNK antibody labeled with MSD SULFO-TAG™ reagent.



**Fig. 1:** Sample data generated with the MULTI-ARRAY® Phospho-JNK (Thr183/Tyr185) Assay. Increased signal is observed with the titration of pJNK positive cell lysate. Signal for negative lysate remains low throughout the titration. The Phospho-JNK (Thr183/Tyr185) Assay provides a quantitative measure of the data obtained with the traditional Western blot.

# MSD Phosphoprotein Assays

## Lysate Titration

Data for pJNK positive and negative HEK293 cell lysates using the MULTI-ARRAY Phospho-JNK (Thr183/Tyr185) Assay are presented below.

Lysate (µg)	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	106	9	8.6	106	9	8.6	
0.63	654	20	3.0	119	22	18.5	5.5
1.3	1208	33	2.8	133	1	1.1	9.1
2.5	1835	33	1.8	124	10	8.0	15
5.0	2552	124	4.9	135	4	2.6	19
10	2969	95	3.2	148	7	4.8	20

## 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](http://www.mesoscale.com)

## References using MSD's platform for the measurement of phosphoproteins

1. Hua F, Henstock PV, Tang B. ERK activation by GM-CSF reduces effectiveness of p38 inhibitor on inhibiting TNFalpha release. *Int Immunopharmacol.* 2010 Jul;10(7):730-7.
2. Skepner JE, Shelley LD, Ji C, Reidich B, Luo Y. Chronic treatment with epoxyeicosatrienoic acids modulates insulin signaling and prevents insulin resistance in hepatocytes. *Prostaglandins & Other Lipid Mediators.* 2011 Feb;94(1-2):3-8.
3. Engstrom L, Pinzon-Ortiz MC, Li Y, Chen SC, Kinsley D, Nelissen R, Fine JS, Mihara K, Manfra D. Characterization of a murine keyhole limpet hemocyanin (KLH)-delayed-type hypersensitivity (DTH) model: role for p38 kinase. *Int Immunopharmacol.* 2009 Sep; 9(10):1218-27.
4. Gowan SM, Hardcastle A, Hallsworth AE, Valenti MR, Hunter LJ, de Haven Brandon AK, Garrett MD, Raynaud F, Workman P, Aherne W, Eccles SA. Application of meso scale technology for the measurement of phosphoproteins in human tumor xenografts. *Assay Drug Dev Technol.* 2007 Jun;5(3):391-401.

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