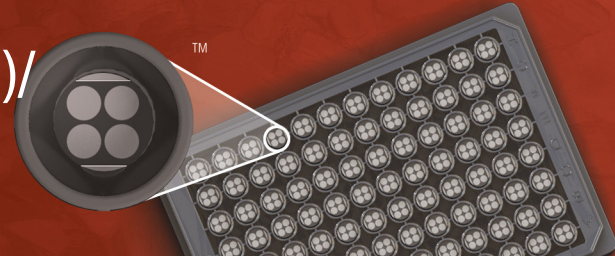


# MSD® Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 Assay: Whole Cell Lysate Kit

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(Thr202/Tyr204;  
Thr185/Tyr187)/Total ERK1/2  
Assay: Whole Cell Lysate Kit

Kit size

1 plate	K15107D-1
5 plates	K15107D-2
20 plates	K15107D-3

Phospho-ERK1/2 Whole Cell  
Lysate Set

200 µg	C11CM-1
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## Ordering information

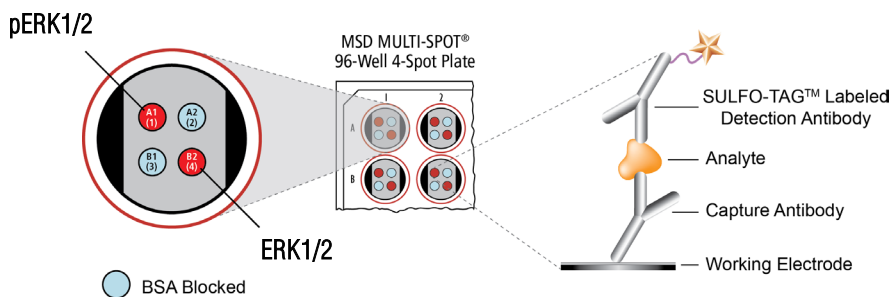
MSD Customer Service  
Phone: 1-301-947-2085  
Fax: 1-301-990-2776  
Email: CustomerService@mesoscale.com

## Company Address

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Gaithersburg, MD 20877 USA

www.mesoscale.com®

For Research Use Only.  
Not for use in diagnostic  
procedures.



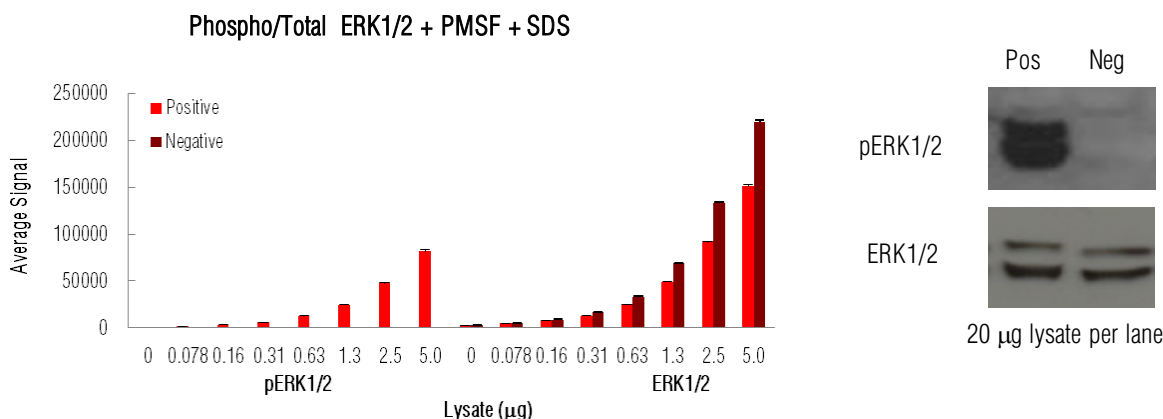
**ERK** (Extracellular signal Regulated Kinases) 1 and 2, are proline-directed serine/threonine protein kinases, also known as p44 MAPK (Mitogen-Activated Protein Kinase) and p42 MAPK, respectively. These closely-related kinase isoforms are activated by various extracellular signals including growth factors, cytokines, hormones, and neurotransmitters. The activation occurs through the phosphorylation of threonine 202 and tyrosine 204 of ERK1 and threonine 185 and tyrosine 187 of ERK2 by the upstream kinases MEK1 and MEK2. Activated ERK1/2 phosphorylates targets in both the cytosol and the nucleus. Cytoplasmic substrates for ERK include SOS, MNK1/2, and the 90 kDa ribosomal protein S6 kinases (RSKs). Nuclear translocation of activated ERK affects gene expression and DNA replication by the phosphorylation of MSK 1 and 2 and the transcription factors Elk-1, Sap1, and Sap2.

The MSD Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

## Typical Data

Representative results for the Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 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-ERK1/2 (Thr202/Tyr204; Thr185/Tyr187) and total ERK1/2 antibodies and are shown below for comparison.

Logarithmically growing Jurkat cells were treated with PMA (200 nM; 15 minutes) (positive) or LY294002 (50 µM; 2.5 hours) (negative). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-phospho-ERK1/2 (Thr202/Tyr204; Thr185/Tyr187) antibody and anti-total ERK1/2 antibody on spatially distinct electrodes within a well. Phosphorylated and total ERK1/2 were detected with anti-total ERK1/2 antibody conjugated with MSD SULFO-TAG™ reagent.



**Fig. 1:** Sample data generated with the MULTI-SPOT Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 Assay. Increased signal for phosphorylated ERK1/2 was observed with only pERK1/2 positive cell lysate. Total ERK1/2 signal increased throughout the titration of both pERK1/2 positive and negative cell lysates. The Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 Assay provides a quantitative measure of the data obtained with the traditional Western blot.

# MSD Phosphoprotein Assays

## Lysate Titration

Data for pERK1/2 positive and negative Jurkat cell lysates using the MULTI-SPOT Phospho(Thr202/Tyr204; Thr185/Tyr187)/Total ERK1/2 Assay are presented below.

	Lysate (µg)	Positive			Negative			P/N
		Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
pERK1/2	0	142	11	7.7	154	11	7.4	
	0.078	1528	46	3.0	117	17	14.1	13
	0.16	3039	78	2.6	135	11	8.2	22
	0.31	5798	456	7.9	120	7	5.5	48
	0.63	12529	270	2.2	133	5	3.4	94
	1.3	24736	590	2.4	141	14	10.2	175
	2.5	47530	1744	3.7	168	11	6.4	282
	5.0	81470	6167	7.6	169	5	3.1	482
ERK1/2	0	2478	202	8.1	2608	25	1.0	
	0.078	4769	115	2.4	5490	149	2.7	0.9
	0.16	7436	134	1.8	8888	524	5.9	0.8
	0.31	12866	994	7.7	16681	360	2.2	0.8
	0.63	25172	680	2.7	33392	1190	3.6	0.8
	1.3	49017	1656	3.4	68684	2086	3.0	0.7
	2.5	91404	4035	4.4	133622	4238	3.2	0.7
	5.0	150915	6931	4.6	219290	6149	2.8	0.7

## 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. Carnahan J, Beltran PJ, Babij C, Le Q, Rose MJ, Vonderfecht S, Kim JL, Smith AL, Nagapudi K, Broome MA, Fernando M, Kha H, Belmontes B, Radinsky R, Kendall R, Burgess TL. Selective and potent Raf inhibitors paradoxically stimulate normal cell proliferation and tumor growth. *Mol Cancer Ther.* 2010 Aug;9(8):2399-410. Epub 2010 Jul 27.
2. Brake R, Starnes C, Lu J, Chen D, Yang S, Radinsky R, Borges L. Effects of palifermin on antitumor activity of chemotherapeutic and biological agents in human head and neck and colorectal carcinoma xenograft models. *Mol Cancer Res.* 2008 Aug;6(8):1337-46.
3. 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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