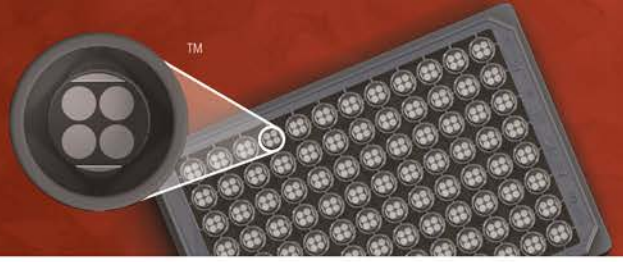
MSD® Phospho-p53 (Ser15) Assay Whole Cell Lysate Kit

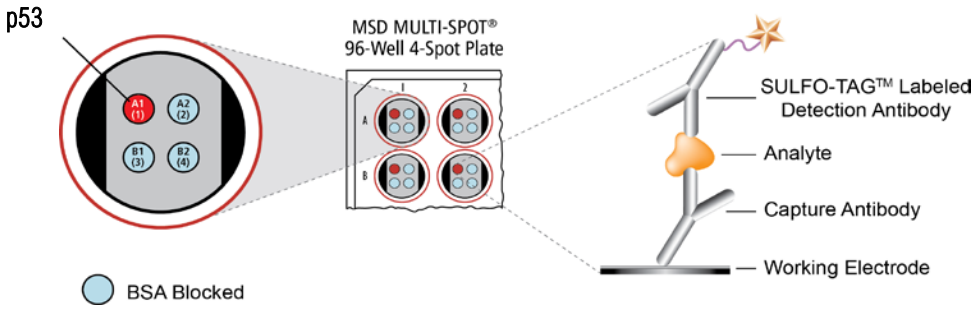
For quantitative determination in human whole cell lysate samples



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

Catalog Numbers

Phospho-p53 (Ser15) Assay: Whole Cell Lysate Kit	
Kit size	
1 plate	K151DAD-1
5 plates	K151DAD-2
20 plates	K151DAD-3



p53 (protein 53) is a transcription factor and tumor suppressor protein with an apparent molecular weight of 53 kDa that plays a critical role in cell cycle regulation, progression, and apoptosis.[1] MDM2 is a potent negative regulator of p53 through its binding and subsequent polyubiquitination of p53, resulting in proteasome dependent degradation.[2] This negative regulation can be relieved both through phosphorylation of p53, resulting in destabilization of the MDM2-p53 interaction,[3] and through phosphorylation and ubiquitination of MDM2.[1] p53 is the most commonly mutated gene in cancer, and a functional copy of p53 is required to maintain a non-tumorigenic phenotype.[4] When cell repair is possible, p53 activates genes that pause the cell cycle allowing time for DNA repair, but when damage is extensive, p53 activates the BCL-2 family of proteins leading to apoptosis.[5] p53's role as a transcription factor and the negative regulation of the protein by MDM2 mediated polyubiquitination has been extensively researched due to its crucial role in cancer prevention and cell cycle control.

The MSD Phospho-p53 (Ser15) Assay is available on 96-well 4-spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Phospho-p53 (Ser15) Assay are illustrated below. The signal and ratio values provided are example data; individual results may vary depending upon the samples tested. Western blot analyses of each lysate type were performed with phospho-p53 (Ser15) and total p53 antibodies and are shown for comparison.
Growing HT29 cells (negative) were harvested 1 hour after UV radiation (40 mJ/cm^2) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-phospho-p53 antibody on one of the four spatially distinct electrodes per well. Phosphorylated p53 was detected with anti-total p53 antibody conjugated with MSD SULFO-TAG™.

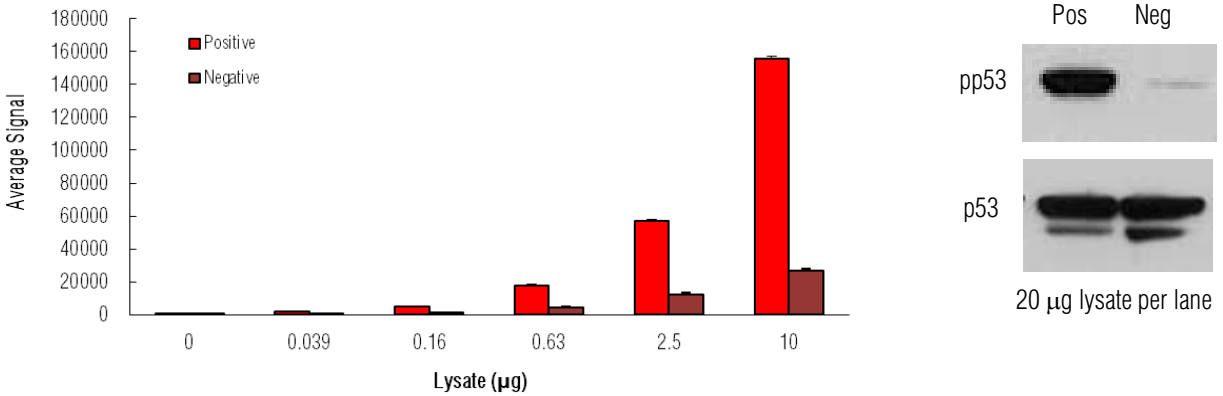


Fig. 1: Sample data generated with the MULTI-ARRAY® Phospho-p53 (Ser15) Assay. Increased signal is observed with the titration of both pp53 positive and negative cell lysates. Signal for pp53 negative cell lysate remains low throughout the titration.The Phospho-p53 (Ser15) Assay provides a quantitative measure of the data obtained with the traditional Western blot.

Ordering information

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

Company Address

MESO SCALE DISCOVERY®
A division of
Meso Scale Diagnostics, LLC.
9238 Gaither Road
Gaithersburg, MD 20877 USA

www.mesoscale.com®

Lysate Titration

Data for pp53 positive and negative HT29 cell lysates using the MULTI-ARRAY Phospho-p53 (Ser15) Assay are presented below.

Lysate (µg)	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	57	8	14.0	41	3	7.3	
0.039	1627	14	0.8	463	1	0.2	3.5
0.16	5241	148	2.8	1383	25	1.8	3.8
0.63	17832	728	4.1	4415	8	0.2	4.0
2.5	56714	1356	2.4	11920	335	2.8	4.8
10	155763	1512	1.0	26580	553	2.1	5.9

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. Inuzuka H, Fukushima H, Shaik S, Wei W. Novel insights into the molecular mechanisms governing Mdm2 ubiquitination and destruction. Oncotarget. 2010 Nov;1(17):685-90.
2. Midgley CA, Lane DP. p53 protein stability in tumour cells is not determined by mutation but is dependent on Mdm2 binding. Oncogene. 1997 Sep 4;15(10):1179-89.
3. Hirao A, Kong YY, Matsuoka S, Wakeham A, Ruland J, Yoshida H, Liu D, Elledge SJ, Mak TW. DNA damage-induced activation of p53 by the checkpoint kinase Chk2. Science. 2000 Mar 10;287(5459):1824-7.
4. Kenzelmann Broz D, Attardi LD. In vivo analysis of p53 tumor suppressor function using genetically engineered mouse models. Carcinogenesis. 2010 Aug;31(8):1311-8.
5. Brady CA, Attardi LD. p53 at a glance. J Cell Sci. 2010 Aug 1;123(Pt 15): 2527-32.