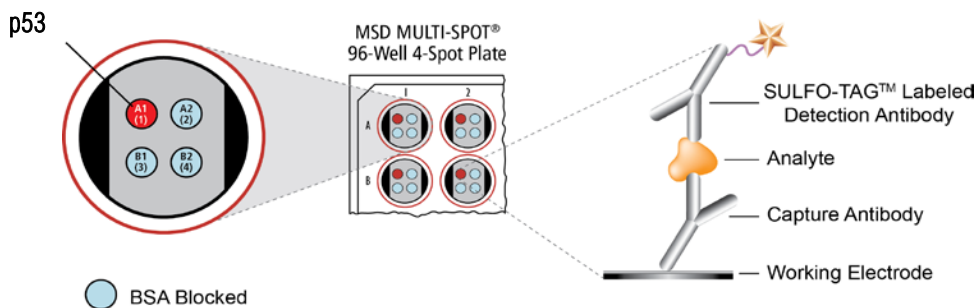


MSD® Total p53 Assay Whole Cell Lysate Kit

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



p53 (protein 53) is a transcription factor and tumor suppressor protein with an apparent molecular weight of 53 kDa, which 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 which 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 Total p53 Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Total p53 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-p53 (Ser15) and total p53 antibodies and are shown below for comparison.

Growing HT29 cells (negative) were harvested 1 hour after UV radiation (40 mJ/cm²) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-total p53 antibody on one of the four spatially distinct electrodes per well. Total p53 was detected with anti-total p53 antibody conjugated with MSD SULFO-TAG™ reagent.

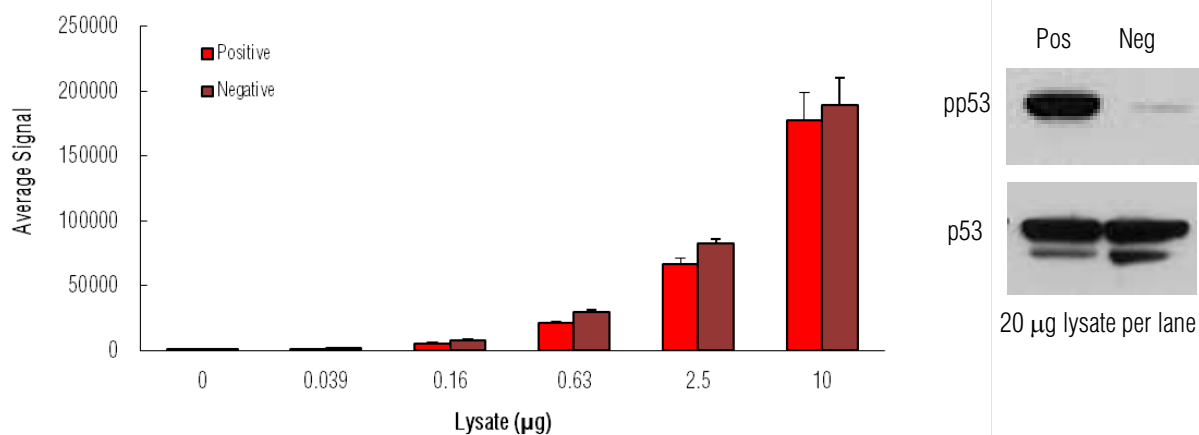


Fig. 1: Sample data generated with the MULTI-ARRAY® Total p53 Assay. Increased signal for total p53 was observed with both pp53 positive and negative cell lysates. The Total p53 Assay provides a quantitative measure of the data obtained with the traditional Western blot.

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

Catalog Numbers

Total p53 Assay: Whole Cell Lysate Kit

Kit size

1 plate	K150DBD-1
5 plates	K150DBD-2
20 plates	K150DBD-3

Ordering information

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

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procedures.

MSD Phosphoprotein Assays

Lysate Titration

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

Lysate (µg)	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	58	27	46.6	29	22	75.8	
0.039	1367	131	9.6	1756	120	6.9	0.8
0.16	5227	929	17.8	7626	423	5.5	0.7
0.63	21451	962	4.5	29932	1177	3.9	0.7
2.5	66969	3914	5.8	82259	1901	2.3	0.8
10	177763	21584	12.1	188994	1732	0.9	0.9

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

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