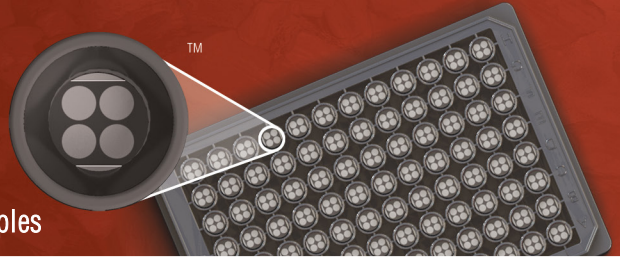
MSD® Total S6RP Assay Whole Cell Lysate Kit

For quantitative determination in human, monkey, 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

Total S6RP Assay: Whole Cell Lysate Kit	
Kit size	
1 plate	K150DHD-1
5 plates	K150DHD-2
20 plates	K150DHD-3

Phospho-S6RP Whole Cell Lysate Set	
200 µg	C10DF-1

Ordering information

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

Company Address

MESO SCALE DISCOVERY®
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Meso Scale Diagnostics, LLC.
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Gaithersburg, MD 20877 USA

www.mesoscale.com®

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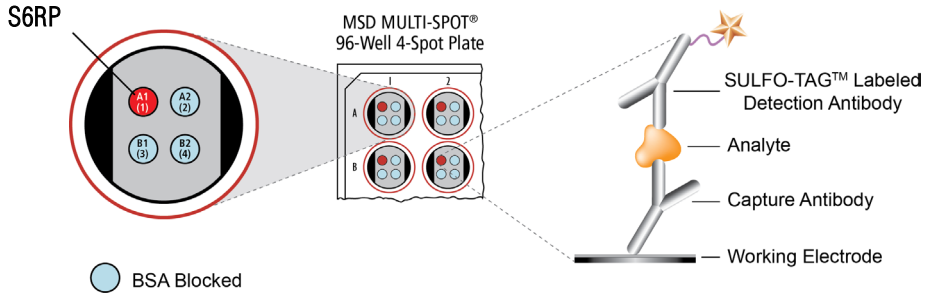

S6 Ribosomal Protein (S6RP) is the S6 subunit of the 40S ribosome and it functions to increase translation of mRNA containing a 5′-terminal oligopyramidine tract (5′-TOP).[1] mRNA with a 5'-TOP generally encode proteins involved in the translational machinery, such as proteins involved in ribosome formation.[2] S6RP functions to control translation of proteins which are constituents of the ribosome, and therefore helps to control overall levels of protein translation. The function of S6RP is phosphorylation dependent, and S6RP is phosphorylated by P70S6K in a mitogen dependent fashion.[3] Residues Ser235, Ser236, Ser240, and Ser244, located within the C-terminus of S6RP, are phosphorylated and important for activation of S6RP.[4]

The MSD Total S6RP Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Total S6RP 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-S6RP and total S6RP antibodies and are shown below for comparison.
Growing low density Jurkat cells were treated with LY294002 (50 µM, 2.5 hours) (negative) or PMA (200 nM, 15 minutes) (positive). Whole cell lysates were added to MSD MULTI-SPOT® 4-Spot plates coated with anti-total S6RP antibody on one of the four spatially distinct electrodes per well. Total S6RP was detected with anti-total S6RP antibody conjugated with MSD SULFO-TAG™ reagent.

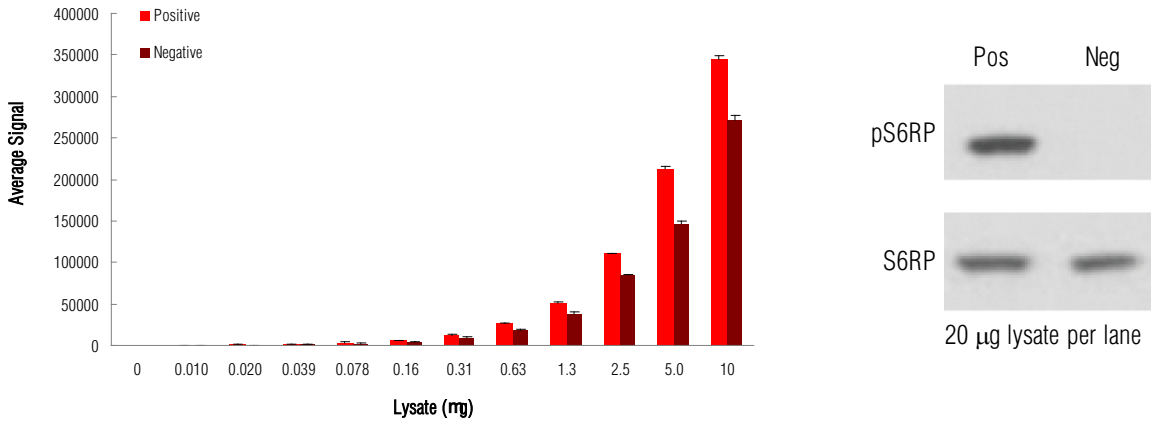

Fig. 1: Sample data generated with the MULTI-ARRAY® Total S6RP Assay. Increased signal is observed with the titration of both pS6RP positive and negative cell lysates. The Total S6RP Assay provides a quantitative measure of the data obtained with the traditional Western blot.

Lysate Titration

Data for pS6RP positive and negative Jurkat cell lysates using the MULTI-ARRAY Total S6RP Assay are presented below.

Lysate (µg)	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	36	7	18.2	36	7	18.2	
0.010	531	59	11.1	345	12	3.4	1.5
0.020	949	52	5.5	640	71	11.1	1.5
0.039	1687	351	20.8	996	73	7.3	1.7
0.078	3119	750	24.1	1892	158	8.4	1.6
0.16	5620	120	2.1	3797	280	7.4	1.5
0.31	12067	729	6.0	9117	339	3.7	1.3
0.63	26919	375	1.4	18406	2314	12.6	1.5
1.3	50707	1980	3.9	37853	3219	8.5	1.3
2.5	110913	198	0.2	85055	1693	2.0	1.3
5.0	212449	2635	1.2	147117	23681	16.1	1.4
10	344779	5018	1.5	271824	5059	1.9	1.3

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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References

1. Avruch J, Belham C, Weng Q, Hara K, Yonezawa K. The p70 S6 kinase integrates nutrient and growth signals to control translational capacity. Prog Mol Subcell Biol. 2001;26:115-54.
2. Avni D, Biberman Y, Meyuhas O. The 5′ terminal oligopyrimidine tract confers translational control on TOP mRNAs in a cell type- and sequence context-dependent manner. Nucleic Acids Res. 1997 Mar 1;25(5):995-1001.
3. Pende M, Um SH, Mieulet V, Sticker M, Goss VL, Mestan J, Mueller M, Fumagalli S, Kozma SC, Thomas G. S6K1(−/−)/S6K2(−/−) mice exhibit perinatal lethality and rapamycin-sensitive 5′-terminal oligopyrimidine mRNA translation and reveal a mitogen-activated protein kinase-dependent S6 kinase pathway. Mol Cell Biol. 2004 Apr;24(8):3112-24.
4. Flotow H, Thomas G. Substrate recognition determinants of the mitogen-activated 70K S6 kinase from rat liver. J Biol Chem. 1992 Feb 15;267(5):3074-8.