

MSD® Phospho-STAT4 (Tyr693) Kit

For quantitative determination in human whole cell lysate samples



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

Catalog Numbers

Phospho-STAT4 (Tyr693) Kit	
Kit Size	Catalog #
1 plate	K150PAD-1
5 plates	K150PAD-2
25 plates	K150PAD-4

Ordering Information

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

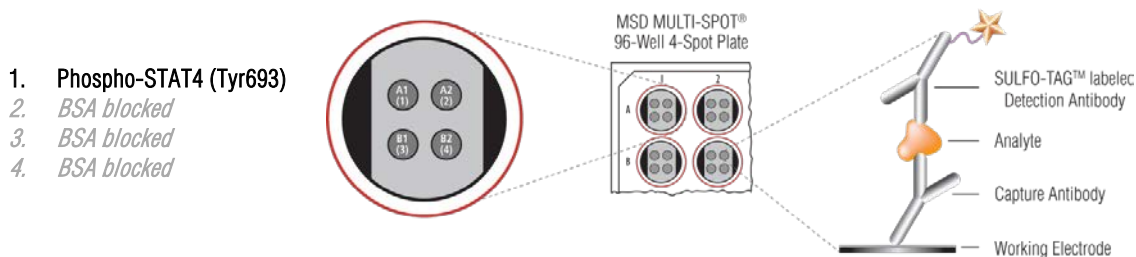
Scientific Support

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Company Address

MESO SCALE DISCOVERY®
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Not for use in
diagnostic procedures.



Signal transducer and activator of transcription 4 (STAT4) is a transcription factor that transduces interleukin-12, interleukin-23, and type-1 interferon cytokine signals in T-cells and monocytes.^{1,2} Following exposure to cytokines, the cytokine receptor-associated Janus kinases (JAK) phosphorylate tyrosine residues present on cytoplasmic STAT4 proteins. STAT4 phosphorylation at tyrosine residue 693 allows homodimerization through src homology 2 domains.³ Functional STAT4 dimers translocate into the nucleus and activate cytokine responsive gene transcription, leading to Th1 cell differentiation, monocyte activation, and interferon-gamma production.^{1,2} STAT4 contributes to autoimmune disorder pathogenesis and anti-viral immune responses.^{4,5} The MSD Phospho-STAT4 (Tyr693) assay is available on 96-well, 4-spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Phospho-STAT4 (Tyr693) Kit are illustrated below. The signal and ratio values provided are examples; individual results will vary depending upon the samples tested. Western blot analyses of each lysate type are shown for comparison.

Growing human T cells were starved for 30 minutes (treatment 1) or starved for 30 minutes then co-incubated with IL-12 (10 ng/mL) and interferon-alpha (1000 U/mL) for 30 minutes (treatment 2). Whole cell lysates were added to MSD MULTI-SPOT®, 4-spot plates coated with anti-Phospho-STAT4 (Tyr693) antibody on one of the four spatially distinct electrodes in each well. Phospho-STAT4 (Tyr693) was detected with anti-total STAT4 antibody conjugated with MSD SULFO-TAG™ reagent.

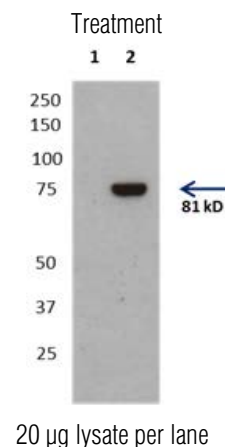
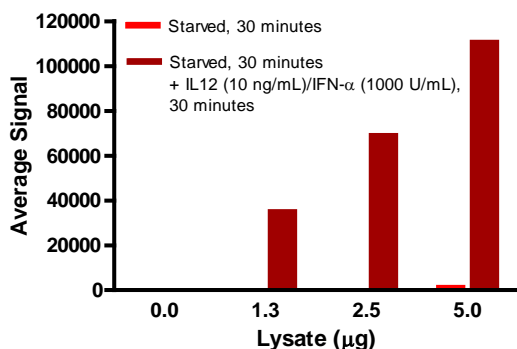


Figure 1: Sample data generated with Phospho-STAT4 (Tyr693) assay. Increased signal is observed with the titration of lysates from treatment 2 [starved for 30 minutes and then co-incubated with IL-12 (10 ng/mL) and interferon-alpha (1000 U/mL) for 30 minutes]. Signal for lysates starved for 30 minutes (treatment 1) remains low throughout the titration. The Phospho-STAT4 (Tyr693) assay provides a quantitative measure of the data obtained with the traditional Western blot.

MSD Phosphoprotein Assays

Lysate Titration

Data for positive and negative cell lysates using the Phospho-STAT4 (Tyr693) Kit are presented below.

Lysate ($\mu\text{g}/\text{well}$)	Treatment 1			Treatment 2		
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV
0	63	17	27.4	63	19	30.0
1.3	862	23	2.7	37 080	1706	4.6
2.5	1783	307	12.6	71 064	4264	6.0
5.0	3291	415	17.2	112 659	4619	4.1

For a complete list of products, please visit our website at www.mesoscale.com.

The MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample volumes of 25 μL 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 response (light signal), minimizing matrix interference
- **Simple protocols:** Only labels bound near the electrode surface are excited, enabling assays with fewer washes
- **Flexibility:** Labels are stable, non-radioactive, and conveniently conjugated to biological molecules

References

1. Wurster AL, et al. The biology of STAT4 and STAT6. *Oncogene*. 2000;19:2577-2584.
2. Korman BD, et al. STAT4: genetics, mechanisms, and implications for autoimmunity. *Curr Allergy Asthma Rep*. 2008 Sep;8(5):398-403.
3. Visconti R, et al. Importance of the MKK6/p38 pathway for interleukin-12-induced STAT4 serine phosphorylation and transcriptional activity. *Blood*. 2000 Sep 1;96(5):1844-52.
4. Svensson A, et al. STAT4 regulates anti-viral gamma interferon responses and recurrent disease during herpes simplex virus 2 infection. *J Virol*. 2012 Sep;86(17):9409-15.
5. Cheng X, et al. Adiponectin induces pro-inflammatory programs in human macrophages and CD4+ T Cells. *J Biol Chem*. 2012 Oct 26;287(44):36896-904.

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