

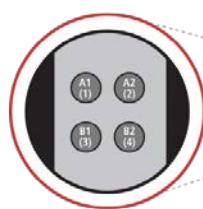
# MSD® Total STAT4 Kit

For quantitative determination in human whole cell lysate samples

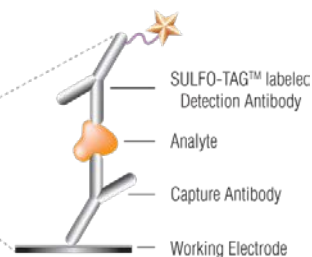
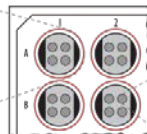


Alzheimer's Disease  
BioProcess  
Cardiac  
**Cell Signaling**  
Clinical Immunology  
Cytokines  
Growth Factors  
Hypoxia  
Immunogenicity  
Inflammation  
Metabolic  
Oncology  
Toxicology  
Vascular

1. STAT4
2. BSA blocked
3. BSA blocked
4. BSA blocked



MSD MULTI-SPOT®  
96-Well 4-Spot Plate



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.<sup>1,2</sup> 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.<sup>3</sup> Functional STAT4 dimers translocate into the nucleus and activate cytokine responsive gene transcription, leading to Th1 cell differentiation, monocyte activation, and interferon-gamma production.<sup>1,2</sup> STAT4 contributes to autoimmune disorder pathogenesis and anti-viral immune responses.<sup>4,5</sup> The MSD Total STAT4 assay is available on 96-well, 4-spot plates. This datasheet outlines the performance of the assay.

## Catalog Numbers

Total STAT4 Kit	
Kit Size	Catalog #
1 plate	K1500VD-1
5 plates	K1500VD-2
25 plates	K1500VD-4

## Typical Data

Representative results for the Total STAT4 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-total STAT4 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.

## Ordering Information

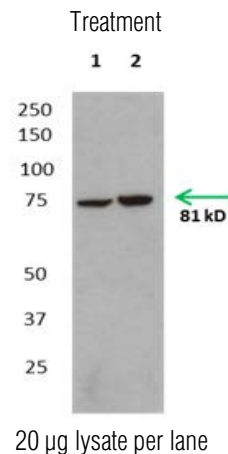
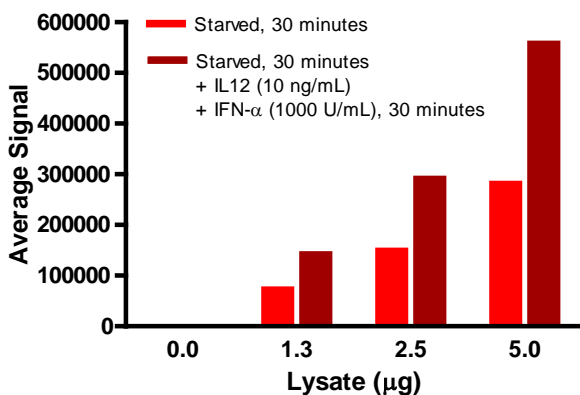
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**Figure 1:** Sample data generated with Total STAT4 assay. Increased signal is observed with the titration of lysates starved for 30 minutes (treatment 1) and with lysates starved for 30 minutes then co-incubated with IL-12 (10 ng/mL) and interferon-alpha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

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

# MSD Phosphoprotein Assays

## Lysate Titration

Data for positive and negative cell lysates using the Total STAT4 Kit are presented below.

Lysate ( $\mu\text{g}/\text{well}$ )	Treatment 1			Treatment 2		
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV
0	85	7	8.2	100	11	10.7
1.3	83 467	1586	1.9	152 976	1682	1.1
2.5	159 608	2713	1.7	301 771	1811	0.6
5.0	291 583	6706	2.3	568 487	7959	1.4

For a complete list of products, please visit our website at [www.mesoscale.com](http://www.mesoscale.com).

## The MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample volumes of 25  $\mu\text{L}$  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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