

Qty: 50 µg/100 µL

Mouse anti-phospho-STAT1

Catalog No. 33-3400

Lot No. See product label

## Mouse anti-phospho-STAT1

### FORM

This monoclonal antibody is supplied as a 100 µL aliquot at a concentration of 0.5 mg/mL in phosphate buffered saline (pH 7.4) containing 0.1% sodium azide. This antibody was purified from mouse ascites using various methods.

**CLONE:** ST1P-11A5

**ISOTYPE:** IgG<sub>2a-kappa</sub>

### IMMUNOGEN

Synthetic tyrosine-phosphorylated peptide encompassing the conserved C-terminal tyrosine phosphorylation site (Y701) of murine STAT1 protein. This peptide differs from the corresponding human sequence by one amino acid.

### SPECIFICITY

This antibody reacts specifically with the tyrosine-701 phosphorylated form of STAT1 and does not exhibit appreciable cross-reactivity with corresponding tyrosine phosphorylated forms of other STAT proteins or with other endogenous phosphotyrosine-containing proteins. To test and confirm the exclusive recognition of tyrosine phosphorylated STAT1, Western blots were carried out on lysates of 293 cells transfected with a STAT1 expression vector together with a wild type or kinase "dead" JAK1 expression vector. In addition, recognition of endogenous tyrosine phosphorylated STAT1 was confirmed on Western blots with cell lysates derived from serum starved or EGF stimulated A431 cells and on Western blots with IFN $\alpha$ -stimulated mouse embryo fibroblast lysates.

### REACTIVITY

This antibody is reactive with human and mouse STAT proteins. Reactivity with other species has not been tested.

### USAGE

The dilutions listed below are recommended starting points. Optimal dilutions for this antibody should be determined by the investigator for each application.














<b>Western Blotting:</b>	1-2 µg/mL
<b>ELISA:</b>	0.1-1.0 µg/mL
<b>Immunohistochemistry (FFPE)*:</b>	5 µg/mL

\* For best results in immunohistochemistry with formalin-fixed, paraffin-embedded tissues, heat induced epitope retrieval (HIER) with citrate buffer, pH 6.0, is required prior to staining.

### STORAGE

Store at 2-8°C for up to one month. Aliquot and store at -20°C for long-term storage. Avoid repeated freezing and thawing.

Explanation of symbols

Symbol	Description	Symbol	Description
	Catalogue Number		Batch code
	Research Use Only		In vitro diagnostic medical device
	Use by		Temperature limitation
	Manufacturer		European Community authorised representative
	Without, does not contain		With, contains
	Protect from light		Consult accompanying documents
	Directs the user to consult instructions for use (IFU), accompanying the product.		

(cont'd)

**For Research Use Only. CAUTION: Not for human or animal therapeutic or diagnostic use.**

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PI333400

(Rev 06/10) DCC-10-1790

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**BACKGROUND<sup>(1-10)</sup>**

In unstimulated cells, STAT proteins exist largely in the cytoplasm as latent transcription factors. In response to treatment of target cells with cytokines or in some cases growth factors, STATs undergo tyrosine phosphorylation, homo- or heterodimerization, nuclear translocation, and DNA binding, which results in transcriptional activation of target genes. Phosphorylation of a conserved tyrosine residue located near the C-terminus of all STAT proteins is required for both dimerization and DNA binding. Tyrosine phosphorylation is therefore a useful marker for STAT activation. At least one and oftentimes several STAT proteins are activated in response to cytokines that utilize receptors from the cytokine receptor superfamily. Nevertheless, a striking specificity of specific STAT activation is seen in response to individual cytokines. Stimulation of responsive cells with IFN $\alpha/\beta$  induces the formation of a transcription complex known as ISGF3. This complex binds to the interferon-stimulated response element (ISRE), activating the transcription of responsive genes. The ISGF3 complex consists of tyrosine phosphorylated-STAT1 $\alpha$ , STAT1 $\beta$  (p84), STAT2 and p48, a 48 kDa DNA-binding protein that is specific for the IFN-stimulated response element. Formation of this complex and its migration into the nucleus is dependent upon tyrosine phosphorylation of STAT1 $\alpha/\beta$  and STAT2. Stimulation of cells with IFN- $\gamma$  results in tyrosine phosphorylation of STAT1 $\alpha$  (p91), but not of STAT2. Phosphorylation of STAT1 $\alpha$  results in its homodimerization and migration into the nucleus where it binds to the IFN- $\gamma$  activated site (GAS). The STAT1 $\alpha$  and STAT1 $\beta$  isoforms arise by alternative splicing of a single gene. The only difference between the two proteins is that STAT1 $\beta$  lacks 38 C-terminal amino acids. Contained within these 38 terminal amino acids of STAT1 $\alpha$  is a critical serine residue (Ser727) whose phosphorylation is required for maximal IFN- $\gamma$  induced transcription.

**REFERENCES**

1. Levy DE, et al. *Genes Dev* 2:383-393 (1988)
2. Schindler C, et al. *Science* 257:809-815 (1992).
3. Schindler C, et al. *PNAS* 89:7836-7839 (1992).
4. Qureski SA, et al. *PNAS* 92:3829-3833 (1995).
5. Fu X-Y, et al. *PNAS* 87:8555-8559 (1990).
6. Veals SA, et al. *Mol Cell Biol* 12: 3315-3324 (1992).
7. Shuai K, et al. *Science* 259:1808-1812 (1992).
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10. Ihle JN, et al. *Trends Biochem Sci* 19:222-227 (1994).

**RELATED PRODUCTS**

<b>Product</b>	<b>Clone/PAD*</b>	<b>Cat. No.</b>
STAT Sampler Pack	(see www.invitrogen.com)	90-0700
Ms x phospho-STAT1 (Y701)	ST1P-11A5	33-3400
Rb x STAT1 $\alpha$	Z-341	71-4300
Ms x STAT1 $\alpha$	ST1-3D4	33-1400
Ms x STAT2	1D4D8	32-2300
Ms x STAT3	ST3-5G7	13-7000
Rb x phospho-STAT4	ST4P	71-7900
Rb x STAT4	Z-17S	71-4500
Ms x STAT4	ST4-5D6	33-2300
Rb x Phospho-STAT5 (Y694)	ZyAL	71-6900
Ms x phospho-STAT5 (Y694)	ST5P-4A9	33-6000
Rb x STAT5a	Z-82	71-2400
Ms x STAT5a	ST5a-2H2	13-3600
Rb x STAT5b	Z-61	71-2500
Ms x STAT5b	ST5b-10G1	13-5300
Ms x STAT5 (pan)	ST5-8F7	33-5900
Ms x STAT6	ST6-3E4	13-5400

\*PAD: Polyclonal Antibody Designation

<b>Product</b>	<b>Conjugate</b>	<b>Cat. No.</b>
Protein A	Sepharose <sup>®</sup> 4B	10-1041
rec-Protein G	Sepharose <sup>®</sup> 4B	10-1241

<b>Conjugate</b>	<b>ZyMAX<sup>™</sup> Goat x Rabbit IgG (H+L)</b>	<b>ZyMAX<sup>™</sup> Goat x Mouse IgG (H+L)</b>
Purified	81-6100	81-6500
FITC	81-6111	81-6511
TRITC	81-6114	81-6514
Cy <sup>™</sup> 3	81-6115	81-6515
Cy <sup>™</sup> 5	81-6116	81-6516
HRP	81-6120	81-6520
AP	81-6122	81-6522
Biotin	81-6140	81-6540

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