Technical Data Sheet

Purified Mouse Anti-Stat1 (pY701)

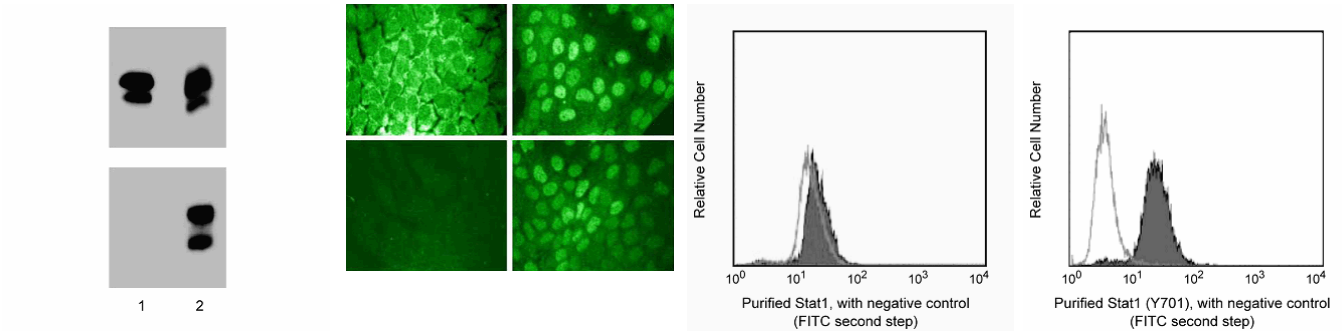
Product Information

Material Number:	**612133**
Size:	150 µg
Concentration:	250 µg/ml
Clone:	14/P-STAT1
Immunogen:	Phosphorylated Human Stat1 (pY701) Peptide
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human Tested in Development: Mouse
Target MW:	91/84 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

Description

Stat (**S**ignal **t**ransducer and **a**ctivators of **t**ranscription) proteins are critical mediators of the biologic activity of cytokines, including interleukins, interferons, erythropoietin, and growth factors. Ligand-receptor interaction leads to activation of constitutively associated JAK family kinases and subsequent recruitment/activation of Stat proteins by tyrosine phosphorylation. Active Stat proteins then move to the nucleus to promote transcription of cytokine-inducible genes. Seven Stat proteins have been cloned, each of which is differentially expressed and/or activated in a cytokine-specific and cell type-specific manner. Stat1 and Stat2 are components of the ISGF3 (**I**nterferon-**S**timulated **G**ene **F**actor **3**) complex, which is the primary transcription activator induced by the binding of the interferon to a specific cell-surface receptor. Stat1 has two alternatively spliced isoforms, 91-kDa Stat1α and 84-kDa Stat1β; Stat1α has 38 additional C-terminal amino acids. In response to the binding of IFNα, IFNγ, EGF, PDGF, or CSF-1 to their respective receptors, the Stat1 subunits become tyrosine-phosphorylated at Y701, and the complex is translocated to the nucleus. This results in the formation of an active complex that includes the DNA-binding p48 subunit. This complex is responsible for modulating the transcription of the interferon-stimulated genes (ISGs).

The 14/P-STAT1monoclonal antibody recognizes the phosphorylated Y701 in Stat1α and Stat1β.



Western blot analysis for Stat1 (pY701) (far left figure). *A431 cells (Human epithelial carcinoma; ATCC CRL-1555) were either left untreated (lane 1) or treated with 100 ng/ml EGF for 5 minutes at 37°C (lane 2). The top panel was probed with a mouse anti-Stat1 antibody (Cat. No. 610115) while the bottom panel was probed with the mouse anti-Stat1 (pY701) antibody at a 1:1000 dilution.*

Immunofluorescence staining for Stat1 (pY701) (middle left figure). *A431 cells (Human epithelial carcinoma; ATCC CRL-1555) were either untreated (top left and bottom left quadrants) or were serum starved and then treated with 100 ng/ml EGF for 5 minutes, then fixed in 3.75% paraformaldehyde with 0.2% Trtion-X 100 (top right and bottom right quandrants). Immunofluorescent staining was performed with a mouse anti-Stat1 antibody (Cat. No. 610115) (top left and top right quadrants) and the mouse anti-Stat1 (pY701) antibody (bottom left and bottom right quadrants).*

Flow cytometric staining for Stat1 (pY701) (middle right and far right figures). *U-937 cells (Human histiocytic lymphoma; ATCC CRL-1593.2) were either untreated (unshaded histograms) or serum starved overnight and treated with 1000 units/mL of IFN-γ for 15 min (shaded histograms). Cells were fixed with 1% formaldehyde, followed by 80% ethanol and BD Cytofix/Cytoperm™ (Cat. No. 554714). Cells were then stained with a mouse anti-Stat1 antibody (Cat. No. 610185) (middle right figure) or the mouse anti-Stat1 (pY701) antibody (far right figure).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at -20°C.

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Application Notes

Application

Western blot	Routinely Tested
Intracellular staining (flow cytometry)	Tested During Development
Immunofluorescence	Tested During Development
Immunoprecipitation	Tested During Development

Recommended Assay Procedure:

Western blot: Please refer to http://www.bdbiosciences.com/pharmingen/protocols/Western_Blotting.shtml.

Suggested Companion Products

Catalog Number	Name	Size	Clone
611447	A431 Cell Lysate	500 µg	(none)
611448	A431 + EGF Cell Lysate	500 µg	(none)
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
554001	FITC Goat Anti-Mouse Ig	0.5 mg	Polyclonal
610115	Purified Mouse Anti-Stat1	50 µg	1/Stat1
610185	Purified Mouse Anti-Stat1	50 µg	42/Stat1
554714	BD Cytofix/Cytoperm™ Fixation/Permeablization Kit	250 tests	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Darnell JE Jr. STATs and gene regulation. *Science.* 1997; 277(5332):1630-1635.(Biology)

Fu XY, Zhang JJ. Transcription factor p91 interacts with the epidermal growth factor receptor and mediates activation of the c-fos gene promoter. *Cell.* 1993; 74(6):1135-1145.(Biology)