Technical Data Sheet

Purified Mouse Anti-Stat3 (pS727)

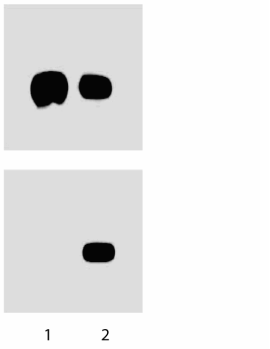
Product Information

Material Number:	**612542**
Size:	50 µg
Concentration:	250 µg/ml
Clone:	49/p-Stat3
Immunogen:	Phosphorylated Human Stat3
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human Tested in Development: Mouse, Rat
Target MW:	92 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

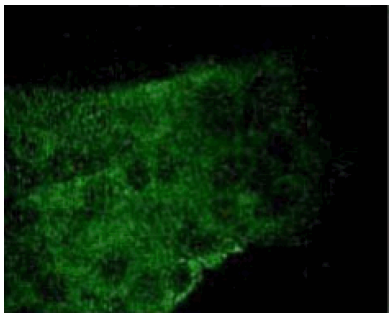
Description

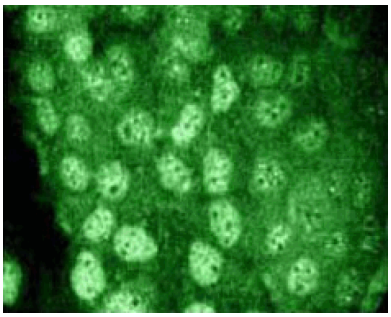
The Stat proteins function both as cytoplasmic signal transducers and as activators of transcription. Seven mammalian Stat proteins have been identified: Stat1-4, Stat5a, 5b, and Stat6. Stat3 is a 92-kDa protein that is activated as a DNA binding protein through cytokines, such as IL-6, and growth factors, such as EGF. Stat3 is phophorylated at serine 727 (S727) via the MAPK pathway. The S727 residue is located at a conserved Pro-X-Ser-Pro sequence, which is recognized by the protein kinase ERK. Activation through the S727 residue is thought to lead to initiation of transcription. Upon activation, Stat3 dimerizes, translocates to the nucleus, and binds DNA response elements thereby regulating gene expression. It appears that Stat3 binds to DNA as a homodimer, but it is also capable of binding as a heterodimer with Stat1. In addition to serine phosphorylation, Stat3 is also phosphorylated at tyrosine 705 by JAK1 in response to cytokine stimulation. Stat3 is widely expressed and can bind to the sis-inducible element (SIE) site from the c-fos promoter. This site is similar to the GAS element that is present in IFN-γ-induced genes. Thus, phosphorylation of S727 in Stat3 occurs in response to growth factors and cytokines, and is essential for normal transcription activity.

The 49/p-Stat3 monoclonal antibody recognizes the S727-phosphorylated form of Stat3 (isoform 1). The fluorochrome-conjugated formats have been evaluated using a human model system. However, the unconjugated form of this antibody (Cat. no. 612542 or 612543) is also effective for western blot analysis of human, mouse, and rat tissue.



A431 cells were either left untreated (left lane) or treated (right lane) with 100 ng/ml EGF for 5 minutes at 37°C. *The top panel was probed with Stat3 (Cat. No. 610189) and the bottom was probed with Stat3 (pS727) (Cat. No. 612542). Lane 1: 1:250, lane 2: 1:500 dilution of the mouse anti-Stat3 antibody.*





A431 cells were serum starved and treated with EGF (100 ng/ml) for 5 minutes, then fixed in 1:1 methanol-acetone for 10 minutes at -20°C. *Immunofluorescent staining was performed using Stat3 (pS727) (Cat. No. 612542).*

Preparation and Storage

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

Application Notes

Application

Western blot	Routinely Tested

BD Biosciences

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United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	888.259.0187	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

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Immunofluorescence	Tested During Development

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

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)

Kanai M, Konda Y, Nakajima T, et al. Differentiation-inducing factor-1 (DIF-1) inhibits STAT3 activity involved in gastric cancer cell proliferation via MEK-ERK-dependent pathway. *Oncogene.* 2003; 22(22):548-554.(Clone-specific: Western blot)

Schuringa JJ, Dekker LV, Vellenga E, Kruijer W. Sequential activation of Rac-1, SEK-1/MKK-4, and protein kinase Cδ is required for interleukin-6-induced STAT3 Ser-727 phosphorylation and transactivation. *J Biol Chem.* 2001; 276:27709-27715.(Biology)

Smith PD, Crompton MR. Expression of v-src in mammary epithelial cells induces transcription via STAT3. *Biochem J.* 1998; 15:331-381.(Biology)