Technical Data Sheet

Purified Mouse Anti-Human Stat5 (pY694)

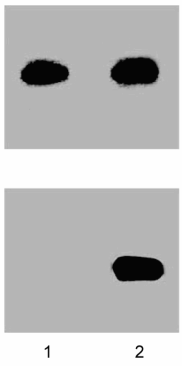
Product Information

Material Number:	**611965**
Size:	150 µg
Concentration:	250 µg/ml
Clone:	47/Stat5(pY694)
Immunogen:	Phosphorylated Human Phosphorylated Stat5 Peptide
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Target MW:	92 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

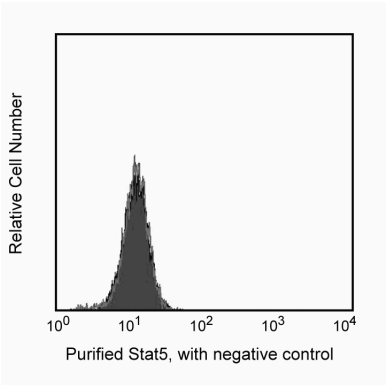
Description

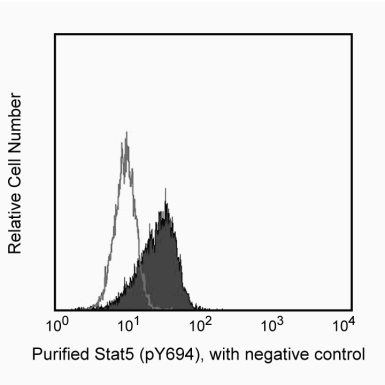
Stat (**S**ignal **t**ransducer and **a**ctivators of **t**ranscription) proteins are critical mediators of the biologic activity of cytokines, including interleukins, interferons, erythropoietin, and growth factors. Ligand-receptor interaction leads to activation of constitutively associated JAK family 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. Stat5 has been characterized and shown to be encoded by two separate genes, Stat5a and Stat5b that share over 90% identity at the amino acid level. Stat5a has been shown to be involved in lactogenesis and mammary development, while Stat5b has been shown to be involved in growth hormone signaling and to play a role in liver gene expression. Both Stat5a and Stat5b share similarities, both are involved in IL-2 induced peripheral T cell proliferation. The peptide hormone, prolactin, binds to the prolactin receptor (PRLR) to initiate the lactogenic response. There are at least three forms of PRLR; however, only the long form is able to activate the 92-kDa Stat5 protein by inducing phosphorylation at Y694. Once phosphorylated, Stat5 becomes an essential transcription factor which binds to the β-casein gene promoter. The presence of an SH2 domain within Stat5 suggests that it may directly interact with protein tyrosine kinases (PTKs) such as JAK2.

The 47 monoclonal antibody recognizes the phosphorylated Y694 of Stat5a. The homologous phosphorylation site in Stat5b is Y699.



A431 cells were either left untreated (-) or treated (+) with 100 ng/ml EGF for 5 minutes at 37°C. *The top panel was probed with Stat5 (Cat. No. 610191) and the bottom was probed with Stat5 (pY694) (Cat. No. 611965).*





U937 cells were serum starved overnight and treated with GMCSF (15 ng/ml) for 20 minutes. *Cells were then fixed with 1% formaldehyde, followed by 80% EtOH, then with BD Cytofix/Cytoperm™ Fixation/Permeabilization Solution Kit (Cat. No. 554714). Cells were then stained with Stat5 (Cat. No. 610191) and Stat5 (pY694) (Cat. No. 611964).*

Preparation and Storage

Store undiluted at -20°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Application Notes

Application

Western blot	Routinely Tested

BD Biosciences

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Suggested Companion Products

Catalog Number	Name	Size	Clone
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
610191	Purified Mouse Anti-Stat5	50 µg	89/Stat5

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
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.
5. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Gouilleux F, Wakao H, Mundt M, Groner B. Prolactin induces phosphorylation of Tyr694 of Stat5 (MGF), a prerequisite for DNA binding and induction of transcription. *EMBO J.* 1994; 13(18):4361-4369. (Biology)

Park DS, Lee H, Frank PG, et al. Caveolin-1-deficient mice show accelerated mammary gland development during pregnancy, premature lactation, and hyperactivation of the Jak-2/STAT5a signaling cascade. *Mol Biol Cell.* 2002; 13(10):3416-3430. (Clone-specific: Western blot)

Wakao H, Gouilleux F, Groner B. Mammary gland factor (MGF) is a novel member of the cytokine regulated transcription factor gene family and confers the prolactin response. *EMBO J.* 1994; 13(9):2182-2191. (Biology)

Williams TM, Cheung MW, Park DS, et al. Loss of caveolin-1 gene expression accelerates the development of dysplastic mammary lesions in tumor-prone transgenic mice. *Mol Biol Cell.* 2003; 14(3):1027-1042. (Clone-specific: Western blot)

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