

## DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human STAT5b in Western blots.
<b>Source</b>	Monoclonal Mouse IgG <sub>2B</sub> Clone # 389215
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	Human STAT5b synthetic peptide MDSQWIPHAQS Accession # NP_036580
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

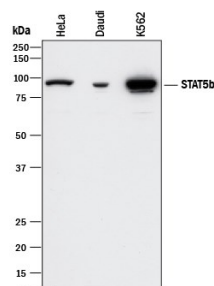
## APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Western Blot</b>	1 µg/mL	See Below
<b>Immunoprecipitation</b>	2 µg/1 mg cell lysate	Cell lysate of HAP1 human near-haploid cell line
<b>Intracellular Staining by Flow Cytometry</b>	2.5 µg/10 <sup>6</sup> cells	See Below
<b>Simple Western</b>	20 µg/mL	HeLa human cervical epithelial carcinoma cell line
<b>CyTOF-ready</b>	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	
<b>Knockout Validated</b>	STAT5b is specifically detected in HeLa human cervical epithelial carcinoma parental cell line but is not detectable in STAT5b knockout HeLa cell line.	

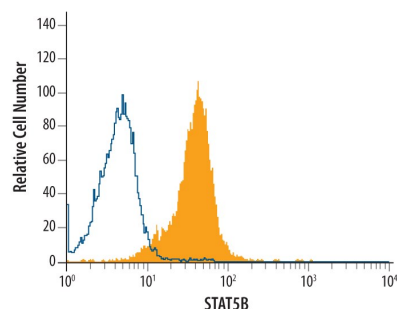
## DATA

### Western Blot



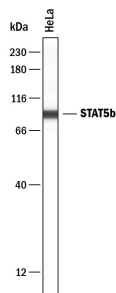
**Detection of Human STAT5b by Western Blot.** Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line, Daudi human Burkitt's lymphoma cell line, and K562 human chronic myelogenous leukemia cell line. PVDF membrane was probed with 1 µg/mL of Mouse Anti-Human STAT5b Monoclonal Antibody (Catalog # MAB1584) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for STAT5b at approximately 90 kDa (as indicated). This experiment was conducted under reducing conditions and using Immoblot Buffer Group 1.

### Intracellular Staining by Flow Cytometry



**Detection of STAT5b in Jurkat Human Cell Line by Flow Cytometry.** Jurkat human acute T cell leukemia cell line was stained with Mouse Anti-Human STAT5b Monoclonal Antibody (Catalog # MAB1584, filled histogram) or isotype control antibody (Catalog # MAB0041, open histogram), followed by Allophycocyanin-conjugated Anti-Mouse IgG F(ab')<sub>2</sub> Secondary Antibody (Catalog # F0101B).

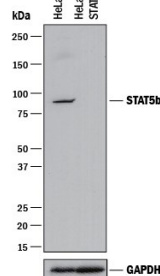
## Simple Western



**Detection of Human STAT5b by Simple Western™.** Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for STAT5b at approximately 91 kDa (as indicated) using 20 µg/mL of Mouse Anti-Human STAT5b Monoclonal Antibody (Catalog # MAB1584). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

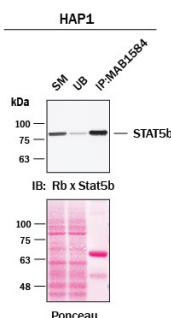


## Knockout Validated



**Western Blot Shows Human STAT5b Specificity by Using Knockout Cell Line.** Western blot shows lysates of HeLa human cervical epithelial carcinoma parental cell line and STAT5b knockout HeLa cell line (KO). PVDF membrane was probed with 1 µg/mL of Mouse Anti-Human STAT5b Monoclonal Antibody (Catalog # MAB1584) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for STAT5b at approximately 90 kDa (as indicated) in the parental HeLa cell line, but is not detectable in knockout HeLa cell line. GAPDH (Catalog # Catalog # MAB5718) is shown as a loading control. This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

## Immunoprecipitation



**Detection of Stat5b by Immunoprecipitation.** Immunoprecipitation was performed on cell lysate of HAP1 human near-haploid cell line using 2.0 µg of Mouse Anti-Human Stat5b Monoclonal Antibody (Catalog # MAB1584) pre-coupled to protein G or protein A beads. Immunoprecipitated Stat5b was detected with a Rabbit Anti-Stat5b antibody. The Ponceau stained transfers of each blot are shown. SM=10% starting material; UB=10% unbound fraction; IP=immunoprecipitated. Image, protocol, and testing courtesy of YCharOS Inc. (ycharos.com).

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

## BACKGROUND

Signal Transduction and Activator of Transcription 5B (STAT5b) is a member of the Jak/STAT signal transduction pathway and is activated by a variety of cytokines (IL-22, IL-6, IFN $\alpha$ ). Upon cytokine activation, STAT5b is phosphorylated and translocates to the nucleus where it binds to the DNA consensus site TTCN<sub>3</sub>GAA and activates transcription of specific genes. STAT5b shares 93% amino acid identity with STAT5a. STAT5b is involved in a wide array of biological processes ranging from regulating apoptosis to adult mammary gland proliferation, differentiation and survival.