abcam

Product datasheet

Anti-STAT5a antibody [E289] ab32043

Recombinant RabMAb

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Overview

Product name	Anti-STAT5a antibody [E289]
Description	Rabbit monoclonal [E289] to STAT5a
Host species	Rabbit
Specificity	The antibody recognises Stat5a. It does not cross-react with other Stat family members. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF, Flow Cyt, IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	within Human STAT5a aa 750 to the C-terminus (C terminal). The exact sequence is proprietary. Database link: P42229
Positive control	WB: A431 cell lysate. IHC-P: Human squamous lung carcinoma. ICC/IF: Jurkat cells. Flow Cyt: Jurkat cells.
General notes	A trial size is available to purchase for this antibody. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.** This product is a recombinant rabbit monoclonal antibody.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity	Protein A purified
Clonality	Monoclonal
Clone number	E289
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab32043** in the following tested applications.

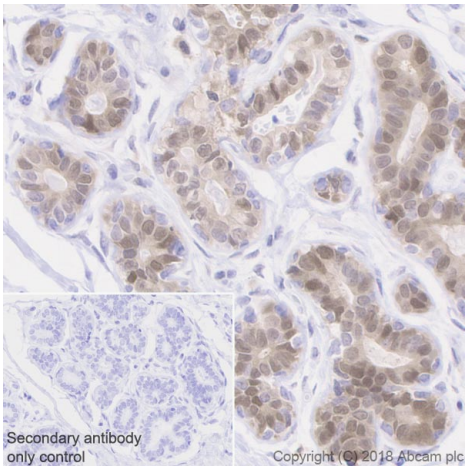
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★	1/1000. Detects a band of approximately 92 kDa (predicted molecular weight: 91 kDa).
IHC-P		1/1000. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat. See IHC antigen retrieval protocols
ICC/IF		1/100 - 1/500.
Flow Cyt		1/100. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. **For unpurified use at 1/10.**
IP		1/20. **For unpurified use at 1/80.**

Target

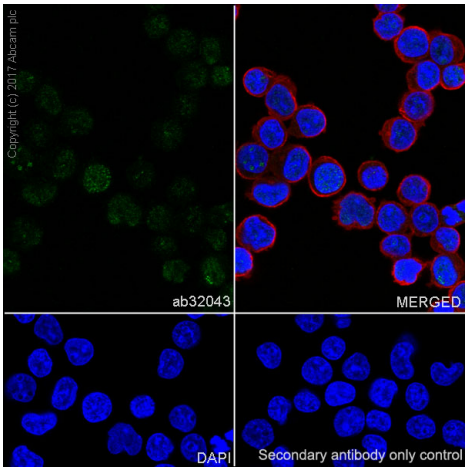
Function	Carries out a dual function: signal transduction and activation of transcription. Binds to the GAS element and activates PRL-induced transcription.
Sequence similarities	Belongs to the transcription factor STAT family. Contains 1 SH2 domain.
Post-translational modifications	Tyrosine phosphorylated in response to IL-2, IL-3, IL-7, IL-15, GM-CSF, growth hormone, prolactin, erythropoietin and thrombopoietin. Tyrosine phosphorylation is required for DNA-binding activity and dimerization. Serine phosphorylation is also required for maximal transcriptional activity.
Cellular localization	Cytoplasm. Nucleus. Translocated into the nucleus in response to phosphorylation.

Images



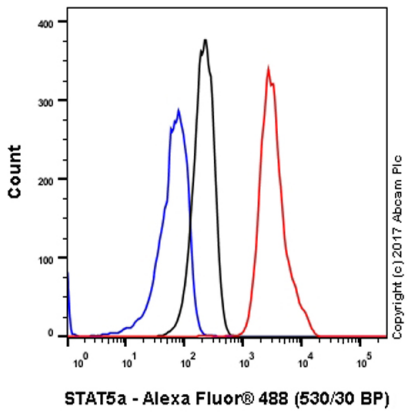
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-STAT5a antibody [E289] (ab32043)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast tissue sections labeling STAT5a with purified ab32043 at 1:1000 dilution (0.12 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use). PBS instead of the primary antibody was used as the negative control.



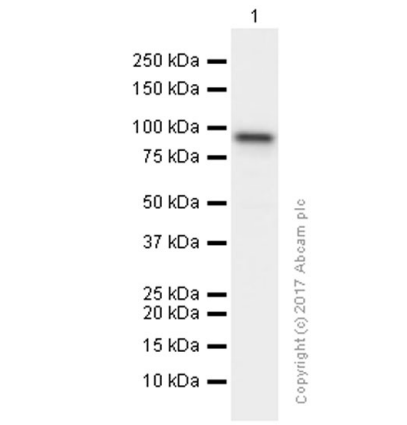
Immunocytochemistry/ Immunofluorescence - Anti-STAT5a antibody [E289] (ab32043)

Immunocytochemistry/ Immunofluorescence analysis of Jurkat (human T cell leukemia T lymphocyte) cells labeling STAT5a with purified ab32043 at 1:100 (1.2 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry - Anti-STAT5a antibody [E289] (ab32043)

Flow Cytometry analysis of Jurkat (human acute T cell leukemia) cells labeling STAT5a with purified ab32043 at 1:100 dilution (1 µg/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1:2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



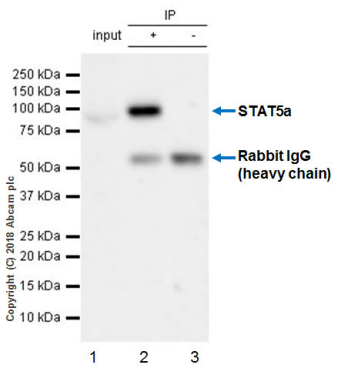
Western blot - Anti-STAT5a antibody [E289] (ab32043)

Anti-STAT5a antibody [E289] (ab32043) at 1/20000 dilution (purified) + K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysates at 20 µg

Secondary

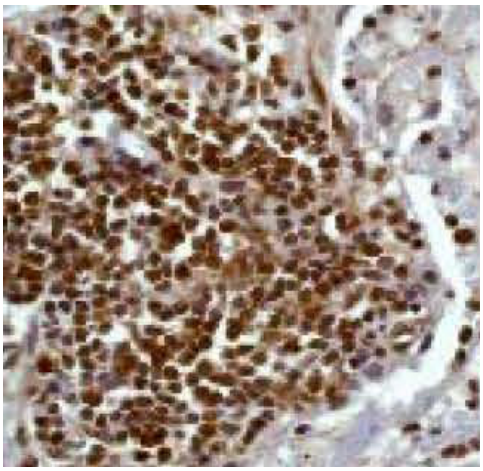
Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 91 kDa
Observed band size: 92 kDa



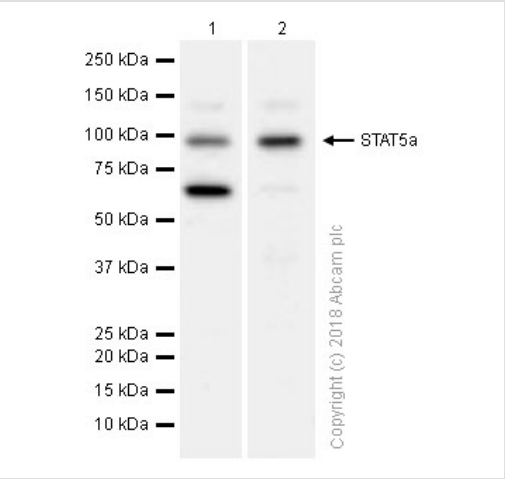
Immunoprecipitation - Anti-STAT5a antibody [E289] (ab32043)

ab32043 (purified) at 1:20 dilution (0.6ug) immunoprecipitating in TF-1 whole cell lysate. TF-1 (Human Erythroleukemia erythroblast) whole cell lysate 10ug
Lane 2 (+): ab32043 & TF-1 whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab32043 in TF-1 whole cell lysate
For western blotting, VeriBlot for IP secondary antibody (HRP) (ab131366) was used as the secondary antibody at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-STAT5a antibody [E289] (ab32043)

Unpurified ab32043 at 1/250 dilution, staining human squamous lung carcinoma by Immunohistochemistry, paraffin-embedded tissue.



Western blot - Anti-STAT5a antibody [E289] (ab32043)

All lanes : Anti-STAT5a antibody [E289] (ab32043) at 1/1000 dilution (Purified)

Lane 1 : Mouse brain lysates

Lane 2 : Rat brain lysates

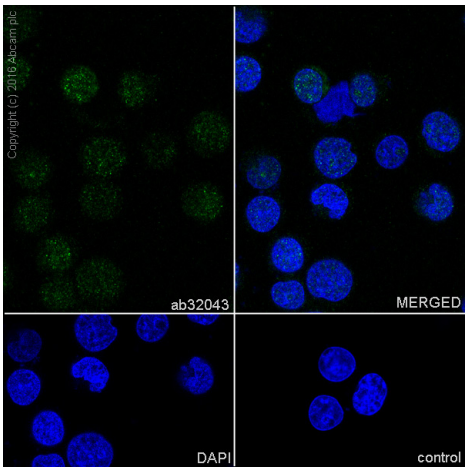
Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

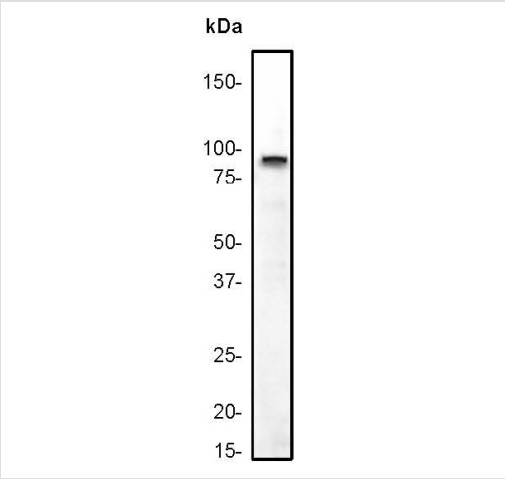
Predicted band size: 91 kDa

Observed band size: 92 kDa



Immunocytochemistry/ Immunofluorescence - Anti-STAT5a antibody [E289] (ab32043)

Immunocytochemistry/Immunofluorescence analysis of Jurkat cells labelling STAT5a with unpurified ab32043 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody.
Control: PBS only.
Nuclear counter stain: DAPI.



Western blot - STAT5a antibody [E289] (ab32043)

Anti-STAT5a antibody [E289] (ab32043) at 1/1000 dilution (unpurified) + A431 cell lysate

Predicted band size: 91 kDa
Observed band size: 92 kDa

Western blot

Anti-STAT5a antibody [E289] (ab32043) at 1/1000 dilution (unpurified) + Recombinant Human STAT5a protein (ab84627) at 0.01 µg

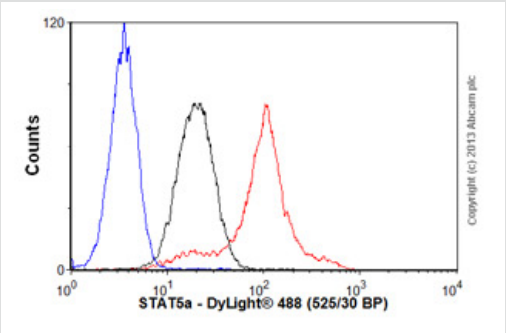
Secondary
Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 91 kDa

Exposure time: 20 seconds



Flow Cytometry-Anti-STAT5a antibody [E289] (ab32043)

Overlay histogram showing Jurkat cells stained with unpurified ab32043 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32043, 1/100 dilution) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line). Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in Jurkat cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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