abcam

Product datasheet

Anti-JAK3 antibody [EP909Y] ab45141

Recombinant RabMAb

★★★★★ 1 Abreviews 10 References 9 Images

Overview

Product name Anti-JAK3 antibody [EP909Y]

Description Rabbit monoclonal [EP909Y] to JAK3

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, IHC-P, ICC/IF

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human JAK3 aa 1100 to the C-terminus (C terminal). The exact

> sequence is proprietary. Database link: P52333

Positive control Flow Cyt (intra): Jurkat cells. WB: TF-1 cell lysate, HEL, KARPAS-299, HaCaT. IHC-P: Human NK

cell lymphoma, Human large B cell lymphoma ICC/IF: KARPAS-299IP: TF-1 whole cell lysate

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

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: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

1

Purity Tissue culture supernatant

Clonality Monoclonal
Clone number EP909Y

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab45141 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
Flow Cyt (Intra)		1/100. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/2000. Predicted molecular weight: 125 kDa.
IP		1/60.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	**** <u>(1)</u>	Use at an assay dependent concentration.

Target	•
--------	---

Function Tyrosine kinase of the non-receptor type, involved in the interleukin-2 and interleukin-4 signaling

pathway. Phosphorylates STAT6, IRS1, IRS2 and PI3K.

Tissue specificity In NK cells and an NK-like cell line but not in resting T-cells or in other tissues. The S-form is more

commonly seen in hematopoietic lines, whereas the B-form is detected in cells both of

hematopoietic and epithelial origins.

Involvement in disease Defects in JAK3 are a cause of severe combined immunodeficiency autosomal recessive T-cell-

negative/B-cell-positive/NK-cell-negative (T(-)B(+)NK(-) SCID) [MIM:600802]. A form of severe combined immunodeficiency (SCID), a genetically and clinically heterogeneous group of rare congenital disorders characterized by impairment of both humoral and cell-mediated immunity, leukopenia, and low or absent antibody levels. Patients present in infancy recurrent, persistent infections by opportunistic organisms. The common characteristic of all types of SCID is absence

of T-cell-mediated cellular immunity due to a defect in T-cell development.

Sequence similaritiesBelongs to the protein kinase superfamily. Tyr protein kinase family. JAK subfamily.

Contains 1 FERM domain.

Contains 1 protein kinase domain.

Contains 1 SH2 domain.

Domain Possesses two phosphotransferase domains. The second one probably contains the catalytic

domain (By similarity), while the presence of slight differences suggest a different role for domain

1.

Post-translational modifications

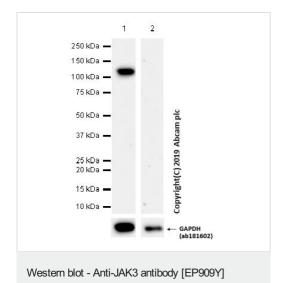
Tyrosine phosphorylated in response to IL-2 and IL-4.

Cellular localization

Endomembrane system. Wholly intracellular, possibly membrane associated.

Images

(ab45141)



All lanes : Anti-JAK3 antibody [EP909Y] (ab45141) at 1/1000 dilution

Lane 1 : TF-1 (Human Erythroleukemia erythroblast) whole cell lysate

Lane 2: HaCaT (Human skin keratinocyte) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 125 kDa Observed band size: 125 kDa

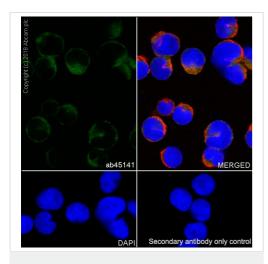
Exposure time: 3 minutes

Blocking/Diluting buffer and concentration 5% NFDM/TBST

The expression profile observed in HaCaT is consistent with the

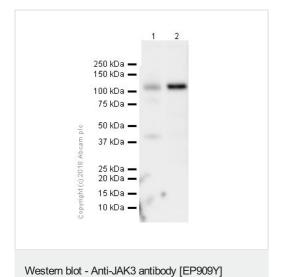
literature (PMID: 11709700).

Negative control: HaCaT (PMID: 11709700)



Immunocytochemistry/ Immunofluorescence - Anti-JAK3 antibody [EP909Y] (ab45141)

Immunocytochemistry/Immunofluorescence analysis of KARPAS-299 (human anaplastic large cell lymphoma) labelling JAK3 with ab45141 at a dilution of 1:100 dilution (12 μ g/ml). Cells were fixed with 100% Methanol. Goat anti rabbit lgG (Alexa Fluor[®] 488, **ab150077**) (1:1000 dilution (2 μ g/ml)) was used as the secondary antibody. The cells were co-stained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) 1:200 (2.5 μ g/ml). Nuclei counterstained with DAPI (blue). Control: PBS instead of the primary antibody.



(ab45141)

All lanes: Anti-JAK3 antibody [EP909Y] (ab45141) at 1.21 µg/ml

Lane 1: KARPAS-299 (Human anaplastic large cell lymphoma) whole cell lysate

Lane 2 : HEL (Human Erythroleukemia erythroblast) whole cell lysate

Lysates/proteins at 20 µg per lane.

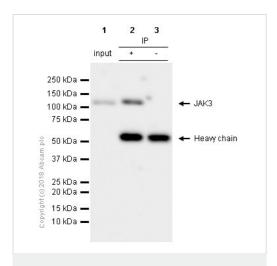
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 125 kDa Observed band size: 125 kDa

Exposure time: 26 seconds

Blocking and diluting buffer: 5% NFDM/TBST.



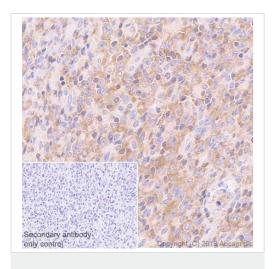
Immunoprecipitation - Anti-JAK3 antibody [EP909Y] (ab45141)

Lane 1 (input): TF-1 (Human Erythroleukemia erythroblast) whole cell lysate, 10µg

Lane 2 (+): TF-1 whole cell lysate

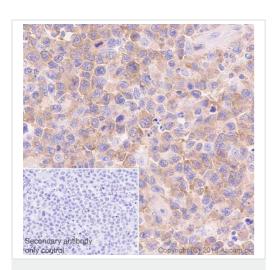
Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab45141 in TF-1 whole cell lysate.

ab45141 at 1/60 immunoprecipitating JAK3 in TF-1 whole cell lysate. For western blotting, ab45141 was used as a primary antibody at 1/500 dilution (2.42 µg/ml). **ab131366**, VeriBlot for IP Detection Reagent was used for detection at 1/1000 dilution. Blocking and diluting buffer used was 5% NFDM/TBST.



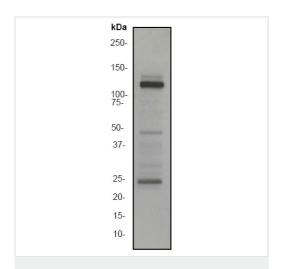
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-JAK3 antibody [EP909Y] (ab45141)

Immunohistochemistry (Formalin/PFA-fixed paraffin embedded sections) analysis of Human NK cell lymphoma tissue sections labeling JAK3 using purified ab45141. Samples were incubated the primary antibody at 1:2000 dilution (0.60 µg/ml). Hematoxylin was used as a counterstain. PBS instead of primary antibody was used for negative control. A ready to use ImmunoHistoProbe one step HRP Polymer at 1:0 dilution was used as the secondary antibody. Heatm mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-JAK3 antibody [EP909Y] (ab45141)

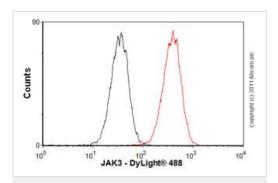
Immunohistochemistry (Formalin/PFA-fixed paraffin embedded sections) analysis of Human large B cell lymphoma tissue sections labeling JAK3 using purified ab45141. Samples were incubated the primary antibody at 1:2000 dilution (0.60 µg/ml). Hematoxylin was used as a counterstain. PBS instead of primary anitbody was used for negative control. A ready to use ImmunoHistoProbe one step HRP Polymer at 1:0 dilution was used as the secondary antibody. Heatm mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0).



Western blot - Anti-JAK3 antibody [EP909Y] (ab45141)

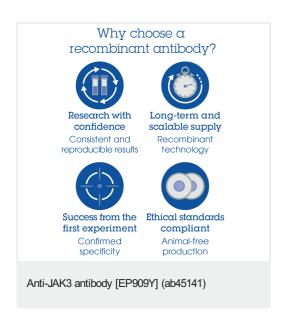
Anti-JAK3 antibody [EP909Y] (ab45141) at 1/2000 dilution + TF-1 (Human bone marrow erythroleukemia cell line) cell lysate

Predicted band size: 125 kDa Observed band size: 125 kDa



Flow Cytometry (Intracellular) - Anti-JAK3 antibody [EP909Y] (ab45141)

Overlay histogram showing Jurkat (Human T cell leukemia cell line from peripheral blood) cells stained with ab45141 (red line). The cells were fixed with 80% methanol (5 minutes) and then permeabilized with 0.1% PBS-Tween for 20 minutes. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab45141, 1/100 dilution) for 30 minutes at 22°C. The secondary antibody used was DyLight[®] 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 minutes at 22°C. Isotype control antibody (black line) was rabbit lgG (1µg/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in Jurkat cells fixed with 4% paraformaldehyde (10 minutes)/permeabilized in 0.1% PBS-Tween used under the same conditions.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit https://www.abcam.com/abpromise or contact our technical team.

Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors