abcam

Product datasheet

Anti-JAK2 antibody [EPR108(2)] ab108596

KO VALIDATED Recombinant RabMAb

3 Abreviews 130 References 8 Images

Overview

Product name	Anti-JAK2 antibody [EPR108(2)]
Description	Rabbit monoclonal [EPR108(2)] to JAK2
Host species	Rabbit
Tested applications	**Suitable for:** ICC/IF, WB, IP **Unsuitable for:** IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: A549, TF-1, K562, THP-1, Jurkat, NIH 3T3, Ramos, C6 and IM-9 cell lysates. ICC/IF: K562, Jurkat and Ramos cells.
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**.

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. Stable for 12 months at -20°C.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 59% PBS, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR108(2)

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab108596 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
ICC/IF	★★★★☆ (1)	1/150. **For unpurified use at 1/100 - 1/500.**
WB	★★★★☆ (2)	1/5000. Detects a band of approximately 130 kDa (predicted molecular weight: 131 kDa). **For unpurified use at 1/1000.**
IP		1/80.

Application notes Is unsuitable for IHC-P.

Target

Function Non-receptor tyrosine kinase involved in various processes such as cell cycle progression, apoptosis, mitotic recombination, genetic instability and histone modifications. In the cytoplasm, plays a pivotal role in signal transduction via its association with cytokine receptors, which constitutes an initiating step in signaling for many members of the cytokine receptor superfamily including the receptors for growth hormone (GHR), prolactin (PRLR), leptin (LEPR), erythropoietin (EPOR), granulocyte-macrophage colony-stimulating factor (CSF2), thrombopoietin (THPO) and multiple interleukins. Following stimulation with erythropoietin (EPO) during erythropoiesis, it is autophosphorylated and activated, leading to its association with erythropoietin receptor (EPOR) and tyrosine phosphorylation of residues in the EPOR cytoplasmic domain. Also involved in promoting the localization of EPOR to the plasma membrane. Also acts downstream of some G-protein coupled receptors. Plays a role in the control of body weight (By similarity). Mediates angiotensin-2-induced ARHGEF1 phosphorylation. In the nucleus, plays a key role in chromatin by specifically mediating phosphorylation of 'Tyr-41' of histone H3 (H3Y41ph), a specific tag that promotes exclusion of CBX5 (HP1 alpha) from chromatin.

Tissue specificity Expressed in blood, bone marrow and lymph node.

Involvement in disease Note=Chromosomal aberrations involving JAK2 are found in both chronic and acute forms of eosinophilic, lymphoblastic and myeloid leukemia. Translocation t(8;9)(p22;p24) with PCM1 links the protein kinase domain of JAK2 to the major portion of PCM1. Translocation t(9;12)(p24;p13) with ETV6.
Defects in JAK2 are a cause of susceptibility to Budd-Chiari syndrome (BCS) [MIM:600880]. It is a syndrome caused by obstruction of hepatic venous outflow involving either the hepatic veins or the terminal segment of the inferior vena cava. Obstructions are generally caused by thrombosis and lead to hepatic congestion and ischemic necrosis. Clinical manifestations observed in the majority of patients include hepatomegaly, right upper quadrant pain and abdominal ascites. Budd-Chiari syndrome is associated with a combination of disease states including primary myeloproliferative syndromes and thrombophilia due to factor V Leiden, protein C deficiency and antithrombin III deficiency. Budd-Chiari syndrome is a rare but typical complication in patients with polycythemia vera.
Defects in JAK2 are a cause of polycythemia vera (PV) [MIM:263300]. A myeloproliferative

disorder characterized by abnormal proliferation of all hematopoietic bone marrow elements, erythroid hyperplasia, an absolute increase in total blood volume, but also by myeloid leukocytosis, thrombocytosis and splenomegaly.
Defects in JAK2 gene may be a cause of essential thrombocythemia (ET) [MIM:187950]. ET is characterized by elevated platelet levels due to sustained proliferation of megakaryocytes, and frequently lead to thrombotic and haemorrhagic complications.
Defects in JAK2 are a cause of myelofibrosis (MYELOF) [MIM:254450]. Myelofibrosis is a disorder characterized by replacement of the bone marrow by fibrous tissue, occurring in association with a myeloproliferative disorder. Clinical manifestations may include anemia, pallor, splenomegaly, hypermetabolic state, petechiae, ecchymosis, bleeding, lymphadenopathy, hepatomegaly, portal hypertension.
Defects in JAK2 are a cause of acute myelogenous leukemia (AML) [MIM:601626]. AML is a malignant disease in which hematopoietic precursors are arrested in an early stage of development.

Sequence similarities

Belongs 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 2 protein kinase domains. The second one probably contains the catalytic domain, while the presence of slight differences suggest a different role for protein kinase 1.

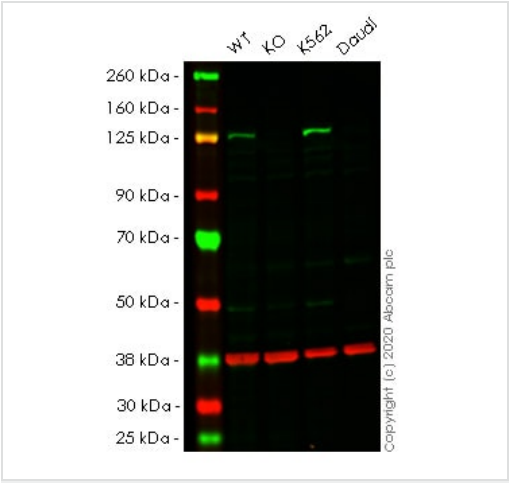
Post-translational modifications

Autophosphorylated, leading to regulate its activity. Leptin promotes phosphorylation on tyrosine residues, including phosphorylation on Tyr-813. Autophosphorylation on Tyr-119 in response to EPO down-regulates its kinase activity. Autophosphorylation on Tyr-868, Tyr-966 and Tyr-972 in response to growth hormone (GH) are required for maximal kinase activity.

Cellular localization

Endomembrane system. Nucleus.

Images



Western blot - Anti-JAK2 antibody [EPR108(2)] (ab108596)

All lanes : Anti-JAK2 antibody [EPR108(2)] (ab108596) at 1/1000 dilution

Lane 1 : Wild-type A549 cell lysate
Lane 2 : JAK2 knockout A549 cell lysate
Lane 3 : K562 cell lysate
Lane 4 : Daudi cell lysate

Lysates/proteins at 20 µg per lane.

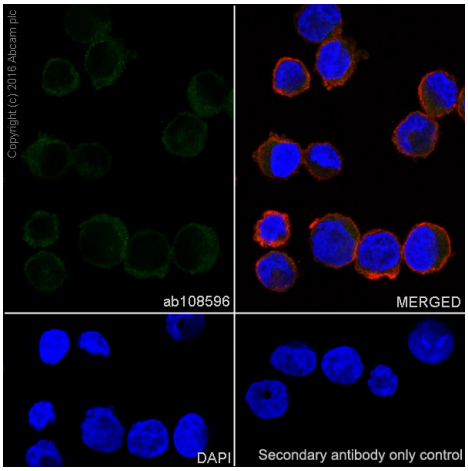
Performed under reducing conditions.

Predicted band size: 131 kDa
Observed band size: 131 kDa

Lanes 1- 4: Merged signal (red and green). Green - ab108596

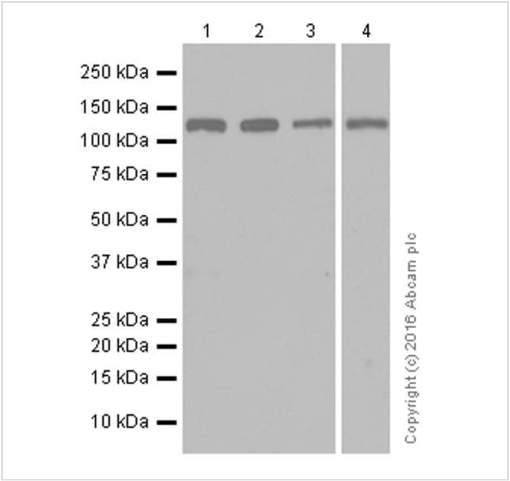
observed at 131 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab108596 was shown to react with JAK2 in Wild-Type A549 cells in western blot. Loss of signal was observed when knockout cell line **ab267113** (knockout cell lysate **ab256963**) was used. Wild-Type A549 and JAK2 knockout A549 cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab108596 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-JAK2 antibody [EPR108(2)] (ab108596)

Immunocytochemistry/ Immunofluorescence analysis of K562 (Human chronic myelogenous leukemia cell line from bone marrow) cells labeling JAK2 with purified ab108596 at 1/150 dilution (8.5µg/ml). Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, a goat anti-rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1/1000 dilution. Ab195889, anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used as the counter stain at 1/200 (2.5 µg/ml). PBS instead of the primary antibody was the negative control. DAPI was used as a nuclear counterstain.



Western blot - Anti-JAK2 antibody [EPR108(2)] (ab108596)

All lanes : Anti-JAK2 antibody [EPR108(2)] (ab108596) at 1/5000 dilution (purified)

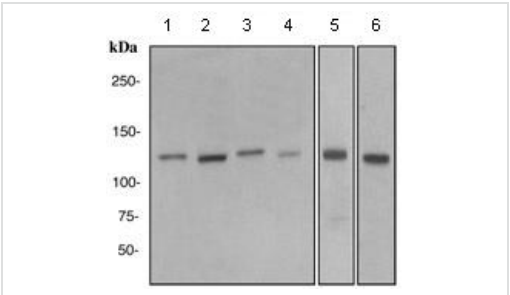
Lane 1 : TF-1 (Human bone marrow erythroleukemia cell line) whole cell lysate
Lane 2 : K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate
Lane 3 : C6 (Rat glial tumour cell line) whole cell lysate
Lane 4 : NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**)

Predicted band size: 131 kDa
Observed band size: 130 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST.



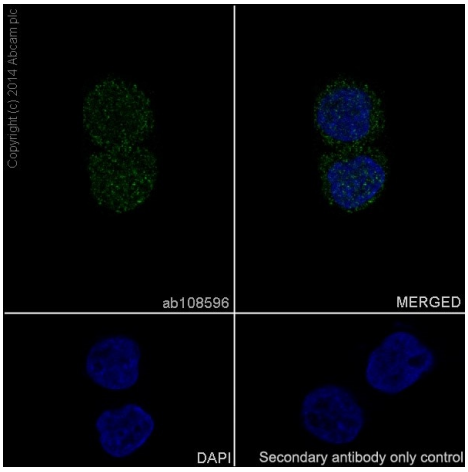
Western blot - Anti-JAK2 antibody [EPR108(2)] (ab108596)

All lanes : Anti-JAK2 antibody [EPR108(2)] (ab108596) at 1/1000 dilution (unpurified)

Lane 1 : TF-1 cell lysate
Lane 2 : K562 cell lysate
Lane 3 : THP-1 cell lysate
Lane 4 : Jurkat cell lysate
Lane 5 : NIH3T3 cell lysate
Lane 6 : IM-9 cell lysate

Lysates/proteins at 10 µg per lane.

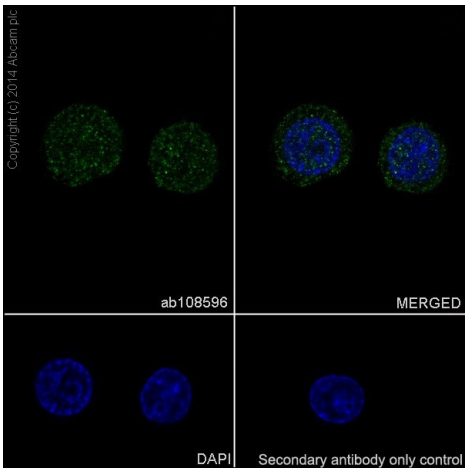
Predicted band size: 131 kDa
Observed band size: 130 kDa



Immunocytochemistry/ Immunofluorescence - Anti-JAK2 antibody [EPR108(2)] (ab108596)

Immunocytochemistry/Immunofluorescence analysis of Jurkat (Human T cell leukemia cells from peripheral blood) cells labelling JAK2 with unpurified ab108596 at 1/300 (7.0 µg/mL). Cells were fixed with 4% Paraformaldehydeand permeabilized with 0.1% Triton X-100. **ab150077**, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000, 2 µg/mL) was used as the secondary antibody. Nuclei were stained with DAPI (blue).

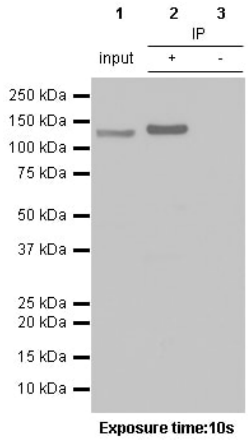
Confocal image showing nuclear and cytoplasmic staining on Jurkat cell line.



Immunocytochemistry/ Immunofluorescence - Anti-JAK2 antibody [EPR108(2)] (ab108596)

Immunocytochemistry/Immunofluorescence analysis of Ramos (Human Burkitt's lymphoma) cells labelling JAK2 with unpurified 108596 at 1/300 (7.0 µg/mL). Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000, 2 µg/mL) was used as the secondary antibody. Nuclei were stained with DAPI (blue).

Confocal image showing nuclear and cytoplasmic staining on Ramos cell line.



Immunoprecipitation - Anti-JAK2 antibody [EPR108(2)] (ab108596)

ab108596 at 1/80 dilution (20 µg/mL) immunoprecipitating JAK2 in K562 (Human chronic myelogenous leukemia lymphoblast) cell lysate.

Lane 1 (input): K562(Human chronic myelogenous leukemia lymphoblast) whole cell lysate 10µg

Lane 2 (+): K562 whole cell lysate, 350µg + ab108596, 2µg

Lane 3 (-): K562 cell lysate, 350µg + rabbit IgG (**ab172730**), 2µg

For western blotting, ab108596 at 1/500 dilution (3.0 µg/mL) and **ab131366** VeriBlot for IP (HRP) was used at 1/1000.

Blocking and dilution buffer: 5% NFDM/TBST.

Anti-JAK2 antibody [EPR108(2)] (ab108596)

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