Anti-JAK2 Antibody [6-D3]

M1501-8



Product Type:	Mouse monoclonal IgG2b, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 130 kDa
Clone number:	6-D3
Description:	Janus kinase 2 (commonly called JAK2) is a non-receptor tyrosine kinase. It is a member of the Janus kinase family and has been implicated in signaling by members of the type II cytokine receptor family (e.g. interferon receptors), the GM-CSF receptor family (IL-3R, IL-5R and GM-CSF-R), the gp130 receptor family (e.g., IL-6R), and the single chain receptors (e.g. Epo-R, Tpo-R, GH-R, PRL-R). JAK2 signaling is activated downstream from the prolactin receptor. Jak - 2 kinase mutations were found to have a high correlation with abnormal heart defects in those of Southeast Asian descent carrying the PYFA gene. Mutations in JAK2 have been implicated in polycythemia vera, essential thrombocythemia, and myelofibrosis as well as other myeloproliferative disorders.
Immunogen:	Recombinant protein within Human JAK2 aa 883-1,132 / 1,132.
Positive control:	A549 cell lysate, K-562 cell lysate, TF-1 cell lysate, Jurkat cell lysate, RAW264.7 cell lysate, C6 cell lysate, K-562, RAW264.7, human kidney tissue, human lung carcinoma tissue, mouse kidney tissue, rat kidney tissue.
Subcellular location:	Cytopasm, nucleus
Database links:	SwissProt: O60674 Human
Recommended Dilutions: WB IF-Cell IHC-P	1:2,000 1:50-1:100 1:200
Storage Buffer:	1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!C$ after thawing. Aliquot store at -20 $^\circ\!C$ or -80 $^\circ\!C$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

5 Service mail:support@huabio.cn



Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

M1501-8 - Page 2

Images



Lane 1: A549-si NT cell lysate Lane 2: A549-si JAK2 cell lysate

Lysates/proteins at 15 µg/Lane.

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

Exposure time: 45 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (M1501-8) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/100,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of JAK2 on different lysates with Mouse anti-JAK2 antibody (M1501-8) at 1/1,000 dilution.

Lane 1: K-562 cell lysate Lane 2: TF-1 cell lysate Lane 3: Jurkat cell lysate Lane 4: RAW264.7 cell lysate Lane 5: C6 cell lysate

Lysates/proteins at 20 µg/Lane.

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

Exposure time: 40 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (M1501-8) at 1/1,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.



Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn



Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation



kDa <u>ko</u>y 250-150-

> 72-55-42-

35 25 JAK2 ~130kDa

GAPDH



Fig3: Immunocytochemistry analysis of K-562 cells labeling JAK2 with Mouse anti-JAK2 antibody (M1501-8) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-JAK2 antibody (M1501-8) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor[™] 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

JAK2 with Mouse anti-JAK2 antibody (M1501-8) at 1/100 dilution.

Fig4: Immunocytochemistry analysis of RAW264.7 cells labeling

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-JAK2 antibody (M1501-8) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Mouse IgG H&L (iFluor[™] 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn



Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation



Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Mouse anti-JAK2 antibody (M1501-8) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1501-8) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig6: Immunohistochemical analysis of paraffin-embedded human lung carcinoma tissue with Mouse anti-JAK2 antibody (M1501-8) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1501-8) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig7: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Mouse anti-JAK2 antibody (M1501-8) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1501-8) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

 が よ い www.huabio.cn

Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation



Fig8: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Mouse anti-JAK2 antibody (M1501-8) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1501-8) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. "JAK2 phosphorylates histone H3Y41 and excludes HP1alpha from chromatin." Dawson M.A., Bannister A.J., Gottgens B., Foster S.D., Bartke T., Green A.R., Kouzarides T. Nature 461:819-822(2009)
- "Heme controls the regulation of protein tyrosine kinases Jak2 and Src." Yao X., Balamurugan P., Arvey A., Leslie C., Zhang L. Biochem. Biophys. Res. Commun. 403:30-35(2010)
- "Phosphorylation of p27Kip1 by JAK2 directly links cytokine receptor signaling to cell cycle control." Jakel H., Weinl C., Hengst L. Oncogene 30:3502-3512(2011)

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880



Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Celt=Immunofluorescence (Celt) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Service mail:support@huabio.cn

Technical:0086-571-89986345