abcam

Product datasheet

Anti-KDM5C / Jarid1C / SMCX antibody ab34718





★★★★★ <u>5 Abreviews</u> <u>12 References</u> 6 Images

Overview

Product name Anti-KDM5C / Jarid1C / SMCX antibody

Description Rabbit polyclonal to KDM5C / Jarid1C / SMCX

Host species Rabbit

Tested applications Suitable for: WB, ICC/IF

Species reactivity Reacts with: Human

Predicted to work with: Pig

Immunogen Synthetic peptide conjugated to KLH derived from within residues 1500 to the C-terminus of

Human Jarid1C/ SMCX.Read Abcam's proprietary immunogen policy(Peptide available as

ab35501.)

Positive control WB: HEK-293T, HAP1 and Y79 cell lysates. ICC/IF: HeLa cells.

General notes The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

> Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any guestions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -Storage instructions

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

1

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

Applications

Target

The Abpromise guarantee Our Abpromise guarantee covers the use of ab34718 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	★★★★ (3)	1/250. Detects a band of approximately 180 kDa (predicted molecular weight: 176 kDa). Abcam recommends using milk as the blocking agent.
ICC/IF	*** <u>*</u>	Use a concentration of 1 µg/ml.

Function	Histone demethylase that specifically demethylates 'Lys-4' of histone

Histone demethylase that specifically demethylates 'Lys-4' of histone H3, thereby playing a central role in histone code. Does not demethylate histone H3 'Lys-9', H3 'Lys-27', H3 'Lys-36', H3 'Lys-79' or H4 'Lys-20'. Demethylates trimethylated and dimethylated but not monomethylated H3 'Lys-4'. Participates in transcriptional repression of neuronal genes by recruiting histone deacetylases

and REST at neuron-restrictive silencer elements.

Tissue specificity Expressed in all tissues examined. Highest levels found in brain and skeletal muscle.

Involvement in disease Defects in KDM5C are the cause of mental retardation syndromic X-linked JARID1C-related

(MRXSJ) [MIM:300534]. MRXSJ is characterized by significantly sub-average general intellectual functioning associated with impairments in adaptative behavior and manifested during the developmental period. MRXSJ patients manifest mental retardation associated with variable

features such as slowly progressive spastic paraplegia, seizures, facial dysmorphism.

Sequence similarities Belongs to the JARID1 histone demethylase family.

Contains 1 ARID domain. Contains 1 JmjC domain. Contains 1 JmjN domain.

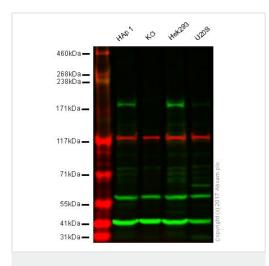
Contains 2 PHD-type zinc fingers.

Domain The first PHD-type zinc finger domain recognizes and binds H3-K9Me3.

Both the JmjC domain and the JmjN domain are required for enzymatic activity.

Cellular localization Nucleus.

Images



Western blot - Anti-KDM5C / Jarid1C / SMCX antibody (ab34718)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

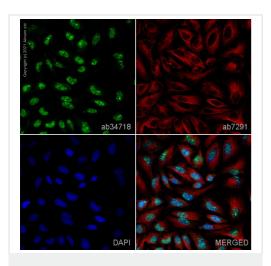
Lane 2: KDM5C knockout HAP1 whole cell lysate (20 µg)

Lane 3: HEK293 whole cell lysate (20 µg)

Lane 4: U2OS whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab34718 observed at 175 kDa. Red - loading control, <u>ab18058</u>, observed at 120 kDa.

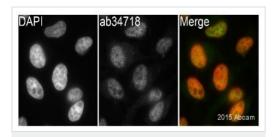
ab34718 was shown to specifically recognize KDM5C in wild-type HAP1 cells as signal was lost at the expected MW in KDM5C knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and KDM5C knockout samples were subjected to SDS-PAGE. Ab34718 and ab18058 (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 1/250 dilution and 1/20,000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1/20,000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-KDM5C / Jarid1C / SMCX antibody (ab34718)

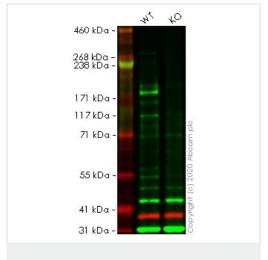
ab34718 staining KDM5C / Jarid1C / SMCX in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab34718 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor[®] 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor[®] 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Immunocytochemistry/ Immunofluorescence - Anti-KDM5C / Jarid1C / SMCX antibody (ab34718)

PFA-fixed, 0.5% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells stained for KDM5C / Jarid1C / SMCX (green) using ab34718 at 1/200 dilution in ICC/IF. Counter-stained with DAPI in order to highlight the nucleus (red). Please refer to abreview for further experimental details.



Western blot - Anti-KDM5C / Jarid1C / SMCX antibody (ab34718)

All lanes : Anti-KDM5C / Jarid1C / SMCX antibody (ab34718) at 1/250 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: KDM5C knockout HEK-293T cell lysate

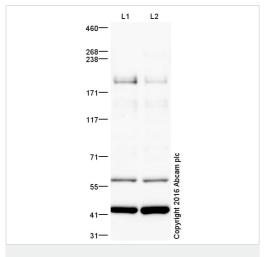
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 176 kDa **Observed band size:** 175 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab34718 observed at 175 kDa. Red - loading control <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab34718 was shown to react with KDM5C / Jarid1C / SMCX in wild-type HEK-293T cells in western blot with loss of signal observed in KDM5C knockout cell line ab266251 (KDM5C knockout cell lysate ab257494). Wild-type and KDM5C knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab34718 and ab7291 (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 250 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-KDM5C / Jarid1C / SMCX antibody (ab34718)

All lanes: Anti-KDM5C / Jarid1C / SMCX antibody (ab34718) at 1 µg/ml

Lane 1: HEK293 (Human) Whole Cell Lysate

Lane 2: Y79 (Human retinoblastoma cell line) Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) preadsorbed at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

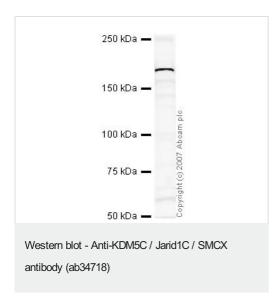
Predicted band size: 176 kDa **Observed band size:** 180 kDa

Additional bands at: 45 kDa, 58 kDa. We are unsure as to the

identity of these extra bands.

Exposure time: 20 minutes

This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% Milk before being incubated with ab34718 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution ab133406.



Anti-KDM5C / Jarid1C / SMCX antibody (ab34718) at 1/250 dilution + HEK-293 whole cell lysate (**ab7902**) at 20 µg

Secondary

IRDye 680 Conjugated Goat Anti-Rabbit IgG (H+L) at 1/10000 dilution

Performed under reducing conditions.

Predicted band size: 176 kDa **Observed band size:** 176 kDa

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