abcam

Product datasheet

Anti-USP9x antibody [EPR13809(B)] - N-terminal ab180191





重组 RabMAb

★★★★★ 1 Abreviews 4 References 10 图像

概述

产品名称 Anti-USP9x抗体[EPR13809(B)] - N-terminal

描述 兔单克隆抗体[EPR13809(B)] to USP9x - N-terminal

宿主 Rabbit

经测试应用 适用于: Flow Cyt (Intra), IHC-P, WB, ICC/IF, IP

种属反应性 与反应: Mouse, Human

预测可用于: Rat 📤

免疫原 Synthetic peptide within Human USP9x aa 1-100 (Cysteine residue). The exact sequence is

proprietary.

Database link: Q93008

阳性对照 WB: HeLa, Hap1, T84, NIH3T3, Jurkat and HepG2 cell lysates. IHC-P: Human kidney and testis

tissues. IP: HeLa and NIH3T3 cell lysates. Flow Cyt (intra): Jurkat cells. ICC/IF: HeLa cells.

常规说明 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**.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

纯度 Protein A purified

克隆 单克隆

克隆编号 EPR13809(B)

同种型 lgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab180191于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/10 - 1/100. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Antigen retrieval is recommended.
WB	**** (1)	1/1000 - 1/10000. Predicted molecular weight: 292 kDa.
ICC/IF		1/100 - 1/250.
IP		1/20.

靶标

功能 Deubiquitinase involved both in the processing of ubiquitin precursors and of ubiquitinated

proteins. May therefore play an important role regulatory role at the level of protein turnover by preventing degradation of proteins through the removal of conjugated ubiquitin. Essential component of TGF-beta/BMP signaling cascade. Regulates chromosome alignment and segregation in mitosis by regulating the localization of BIRC5/survivin to mitotic centromeres. Specifically hydrolyzes both 'Lys-29'- and 'Lys-33'-linked polyubiquitins chains. Specifically deubiquitinates monoubiquitinated SMAD4, opposing the activity of E3 ubiquitin-protein ligase

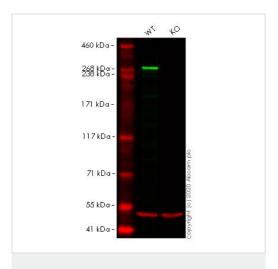
TRIM33.

组织**特异性** Widely expressed in embryonic and adult tissues.

序列相似性 Belongs to the peptidase C19 family.

细胞定位 Cytoplasm.

图片



Western blot - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

All lanes : Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: USP9X knockout HeLa cell lysate

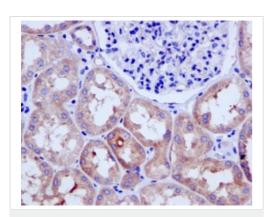
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 292 kDa **Observed band size:** 290 kDa

Lanes 1-2: Merged signal (red and green). Green - ab180191 observed at 290 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab180191 was shown to react with USP9x in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265665 (knockout cell lysate ab257790) was used. Wild-type HeLa and USP9X knockout HeLa 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. ab180191 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated 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.

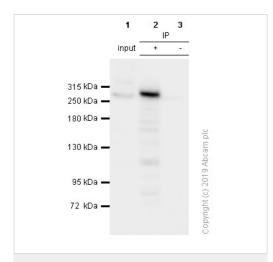


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-USP9x antibody

[EPR13809(B)] - N-terminal (ab180191)

Immunohistochemical analysis of paraffin embedded Human kidney tissue labeling USP9x with ab180191 at 1/50.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

ab180191 (purified) at 1/20 dilution immunoprecipitating ETS2 in HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 μg .

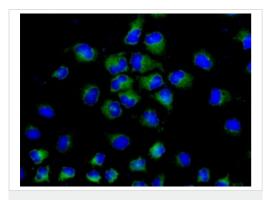
Lane 1 (input): HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 μg

Lane 2 (+): ab180191 & HeLa whole cell lysate

Lane 3 (-): Rabbit monoclonal $\lg G$ (ab172730) instead of ab180191 in HeLa whole cell lysate

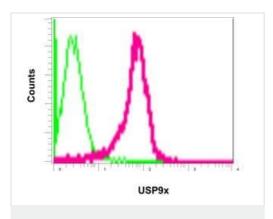
For western blotting, ab180191 at 1/500 dilution (0.23 μ g/mL) and veriBlot for IP secondary antibody (HRP) (<u>ab131366</u>) at 1/1000 dilution was used.

Blocking and diluting buffer: 5% NFDM /TBST.



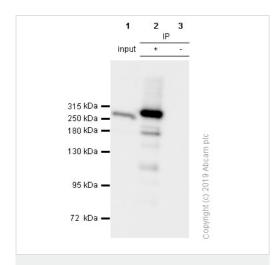
Immunocytochemistry/ Immunofluorescence - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

Immunofluorescent analysis of HeLa cells labeling USP9x with ab180191 at 1/100 (green) and DAPI staining (blue).



Flow Cytometry (Intracellular) - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

Intracellular flow cytometrical analysis of permeabilized Jurkat cells labeling USP9x with ab180191 at 1/10 (red) or a rabbit lgG negative (green).



Immunoprecipitation - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

ab180191 (purified) at 1/20 dilution immunoprecipitating ETS2 in NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate 10 μg .

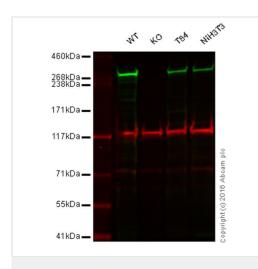
Lane 1 (input): NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate 10 µg.

Lane 2 (+): ab180191 & NIH/3T3 whole cell lysate

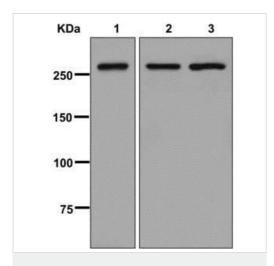
Lane 3 (-): Rabbit monoclonal lgG (ab172730) instead of
ab180191 in NIH/3T3 whole cell lysate

For western blotting, ab180191 at 1/500 dilution (0.23 μ g/mL) and veriBlot for IP secondary antibody (HRP) (**ab131366**) at 1/1000 dilution was used.

Blocking and diluting buffer: 5% NFDM /TBST.



Western blot - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)



Western blot - Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191)

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

Lane 2: USP9x knockout HAP1 cell lysate (20 μg)

Lane 3: T84 cell lysate (20 µg)

Lane 4: NIH3T3 cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab180191 observed at 270 kDa. Red - loading control, <u>ab18058</u>, observed at 117 kDa.

ab180191 was shown to specifically react with USP9x when USP9x knockout samples were used. Wild-type and USP9x knockout samples were subjected to SDS-PAGE. ab180191 and ab18058 (loading control to Vinculin) were diluted at 1 µg/ml and 1/10 000 respectively and incubated overnight at 4°C. 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 1/10 000 dilution for 1 h at room temperature before imaging.

All lanes : Anti-USP9x antibody [EPR13809(B)] - N-terminal (ab180191) at 1/1000 dilution

Lane 1: Jurkat cell lysate

Lane 2: HeLa cell lysate

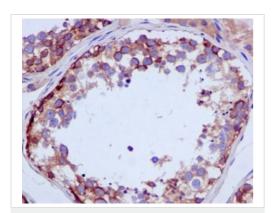
Lane 3: HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 292 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-USP9x antibody

[EPR13809(B)] - N-terminal (ab180191)

Immunohistochemical analysis of paraffin embedded Human testis tissue labeling USP9x with ab180191 at 1/50.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



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