abcam

Product datasheet

Anti-DUSP6 antibody [EPR129Y] ab76310

敲除验证 重组 RabMAb

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概述

产品名称	Anti-DUSP6抗体[EPR129Y]
描述	兔单克隆抗体[EPR129Y] to DUSP6
宿主	Rabbit
经测试应用	**适用于:** IHC-P, Flow Cyt (Intra), WB, IP **不适用于:** ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: 3T3, and A431 cell lysates; IHC-P: Human gastric carcinoma and human pancreas tissues; IHC-Fr: Mouse brain tissue; Flow Cyt (intra): NIH-3T3 and HeLa cells; IP: NIH-3T3 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. Avoid freeze / thaw cycle.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR129Y

同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab76310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。

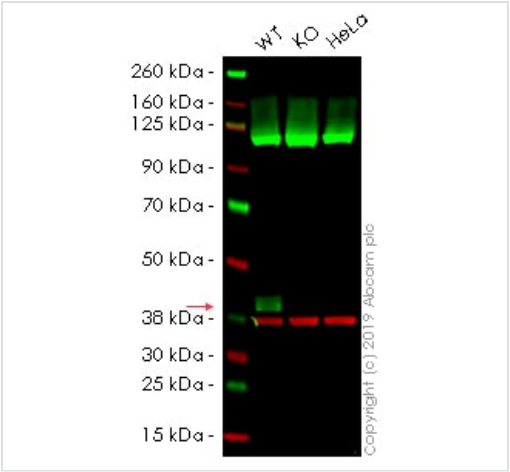
应用	Ab评论	说明
IHC-P	★★★★☆ (1)	1/50 - 1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
Flow Cyt (Intra)		1/20 - 1/200. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB	★★★★★ (8)	1/500 - 1/1000. Detects a band of approximately 42,44 kDa (predicted molecular weight: 42 kDa).
IP	★★★★★ (2)	1/40 - 1/50.

应用说明	Is unsuitable for ICC/IF.

靶标

功能	Inactivates MAP kinases. Has a specificity for the ERK family.
序列相似性	Belongs to the protein-tyrosine phosphatase family. Non-receptor class dual specificity subfamily. Contains 1 rhodanese domain. Contains 1 tyrosine-protein phosphatase domain.
细胞定位	Cytoplasm.

图片



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

All lanes : Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/500 dilution

Lane 1 : Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 2 : DUSP6 knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

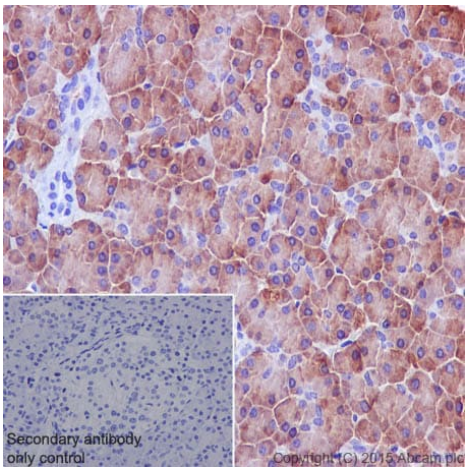
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 42 kDa

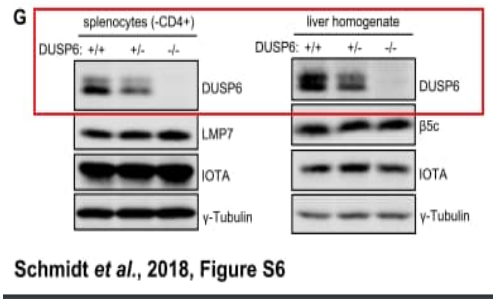
Lanes 1 - 3: Merged signal (red and green). Green - ab76310 observed at 42 kDa. Red - loading control, **ab130007**, observed at 130 kDa.

ab76310 was shown to recognize in wild-type A431 cells as signal was lost at the expected MW in DUSP6 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and DUSP6 knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% Milk. Ab76310 and **ab130007** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/500 dilution and 1/20000 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 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-DUSP6 antibody [EPR129Y] (ab76310)

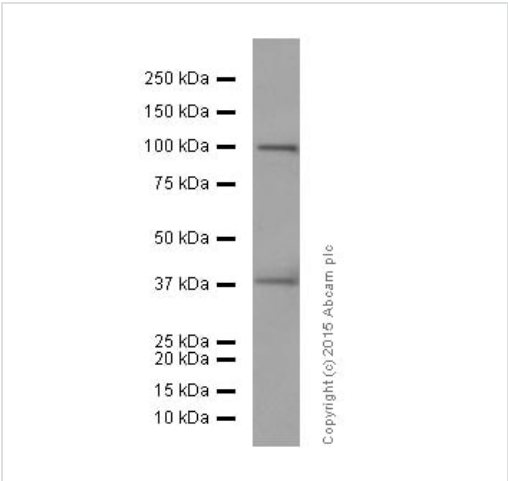
Immunohistochemical staining of paraffin embedded human pancreas with purified ab76310 at a working dilution of 1/100. The secondary antibody used is HRP goat anti-rabbit IgG H&L (**ab97051**) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Immunoblot analysis of indicated proteins in splenocytes after CD4+ sort, liver homogenate or purified CD4+ T cells from WT, DUSP6+/- or DUSP6-/- mice to confirm knockout or heterozygosity for DUSP6 at protein level.

This image was generated using **ab220811**, the same antibody but in a PBS-only buffer formulation.



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/5000 dilution (purified) + HepG2 cell lysate at 20 µg

Secondary

HRP goat anti-rabbit IgG (H+L) at 1/50000 dilution

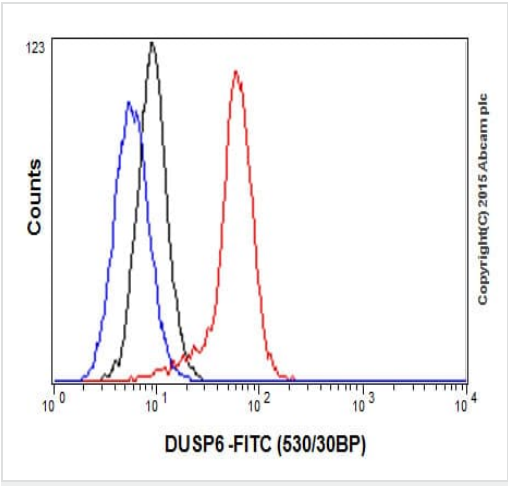
Predicted band size: 42 kDa

Observed band size: 42,44 kDa

Blocking buffer: 5% NFDM/TBST

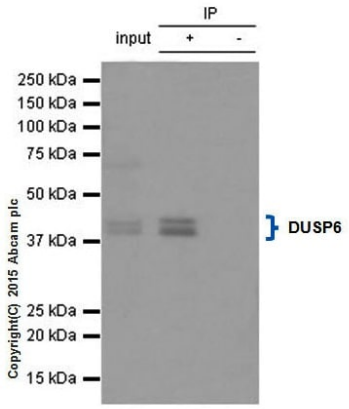
Dilution buffer: 5% NFDM/TBST

ab76310 detects an unspecific band around 100 kDa in human materials.



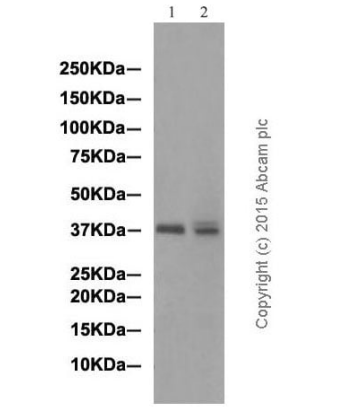
Flow Cytometry (Intracellular) - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Overlay histogram showing NIH-3T3 cells fixed in 4% PFA and stained with purified ab76310 at a dilution of 1 in 200 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 500. Rabbit monoclonal IgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line).



Immunoprecipitation - Anti-DUSP6 antibody [EPR129Y] (ab76310)

ab76310 (purified) at 1/20 immunoprecipitating DUSP6 in 10 µg NIH-3T3 (Lanes 1 and 2, observed at 42 and 44 kDa). Lane 3 - PBS. For western blotting, a HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500). Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

All lanes : Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/1000 dilution (purified)

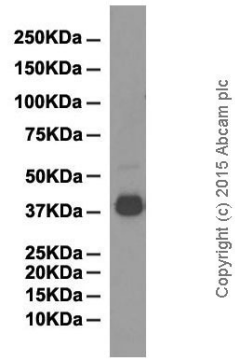
Lane 1 : rat brain lysate
Lane 2 : NIH/3T3 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : HRP goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 42 kDa
Observed band size: 42,44 kDa

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/1000 dilution (purified) + mouse brain at 10 µg

Secondary

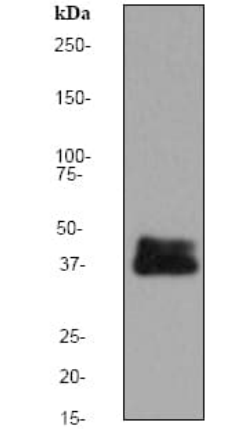
HRP goat anti-rabbit IgG (H+L) at 1/50000 dilution

Predicted band size: 42 kDa

Observed band size: 42,44 kDa

Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

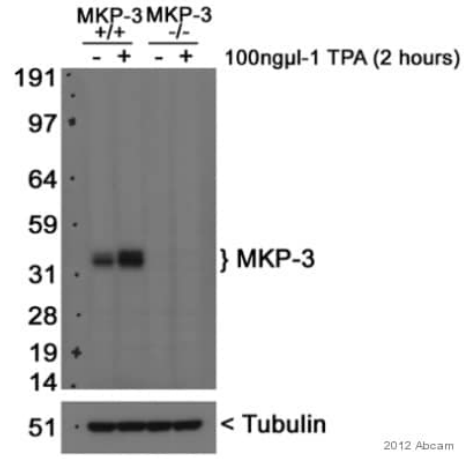
Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/500 dilution (unpurified) + 3T3 cell lysate at 10 µg

Secondary

HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 42 kDa

Observed band size: 42,44 kDa



Western blot - Anti-DUSP6 antibody [EPR129Y] (ab76310)

This image was taken from an abreview by Graeme Stewart.

All lanes : Anti-DUSP6 antibody [EPR129Y] (ab76310) at 1/4000 dilution (unpurified)

Lane 1 : Marker
Lane 2 : Lysate from wild type primary murine embryonic fibroblasts (MEFs) untreated at 10 µg
Lane 3 : Lysate from wild type primary murine embryonic fibroblasts (MEFs) treated with 100ngµl-1, 12-O-tetradecanoylphorbol-13-acetate (TPA) for 2 hours at 10 µg
Lane 4 : Lysate from MKP-3 null primary murine embryonic fibroblasts (MEFs) untreated at 10 µg
Lane 5 : Lysate from MKP-3 null primary murine embryonic fibroblasts (MEFs) treated with 100ngµl-1, 12-O-tetradecanoylphorbol-13-acetate (TPA) for 2 hours at 10 µg

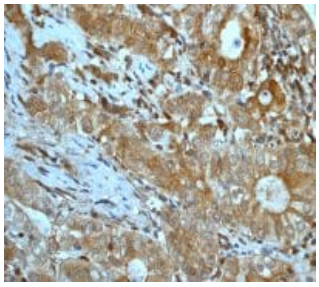
Secondary
All lanes : Goat anti Rabbit HRP conjugate at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 42 kDa
Observed band size: 42 kDa

Exposure time: 2 minutes



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Immunohistochemical staining of paraffin-embedded human gastric carcinoma using unpurified ab76310 at 1/50 dilution.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Flow Cytometry (Intracellular) - Anti-DUSP6 antibody [EPR129Y] (ab76310)

Overlay histogram showing HeLa cells stained with unpurifiedab76310 (red line). The cells were fixed with methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions

followed by the antibody (ab76310, 1/20 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit monoclonal IgG (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde/permeabilized with 0.1% PBS-Tween 20 used under the same conditions.