abcam

Product datasheet

Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free ab218121

敲除验证 重组 RabMAb

4 References 12 图像

概述

产品名称	Anti-PFKFB3抗体[EPR12594] - BSA and Azide free
描述	兔单克隆抗体[EPR12594] to PFKFB3 - BSA and Azide free
宿主	Rabbit
经测试应用	**适用于:** Flow Cyt (Intra), IP, ICC/IF, WB, IHC-P
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Jurkat and HeLa whole cell lysate (**ab150035**); Human melanoma tissue; HeLa and A431 cells, Mouse skin tissue lysate, Rat breast tissue lysate, AR42 and L6 whole cell lysates, HAP1 whole cell lysate, AR42J (rat pancreatic tumor epithelial cell) whole cell lysate, IP: Mouse skin tissue lysate, AR42J, whole cell lysate ICC: HeLa, A431 cells IHC: human melanoma tissue Flow: A431 (human epidermoid carcinoma) cells,
常规说明	ab218121 is the carrier-free version of **ab181861**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. 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. Do Not Freeze.
存储溶液	pH: 7.2 Constituent: PBS
无载体	是
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR12594
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab218121于以下的经测试应用**

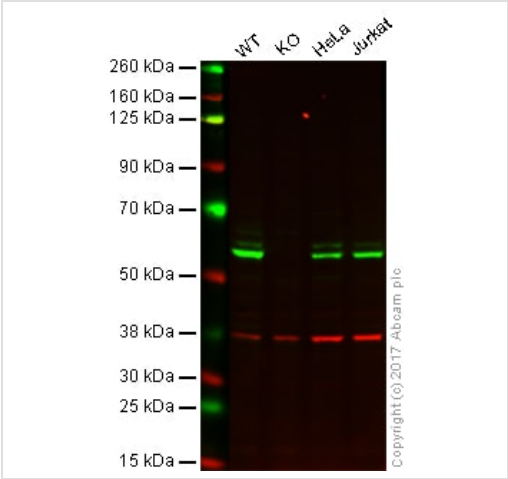
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 58 kDa (predicted molecular weight: 60 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

靶标

功能	Synthesis and degradation of fructose 2,6-bisphosphate.
组织特异性	Ubiquitous.
序列相似性	In the C-terminal section; belongs to the phosphoglycerate mutase family.
翻译后修饰	Phosphorylation by AMPK stimulates activity.

图片



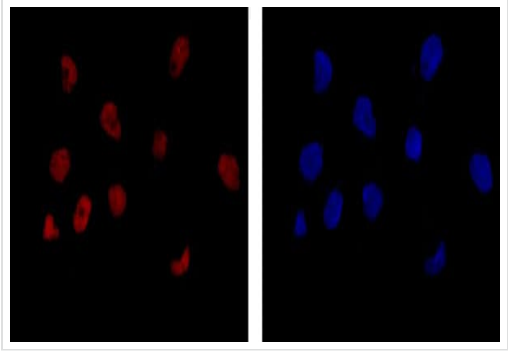
Western blot - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

This WB data was generated using the same anti-PFKFB3 antibody clone, EPR12594, in a different buffer formulation (cat# **ab181861**).

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: PFKFB3 knockout HAP1 whole cell lysate (20 µg)
Lane 3: HeLa whole cell lysate (20 µg)
Lane 4: Jurkat whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - **ab181861** observed at 60 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

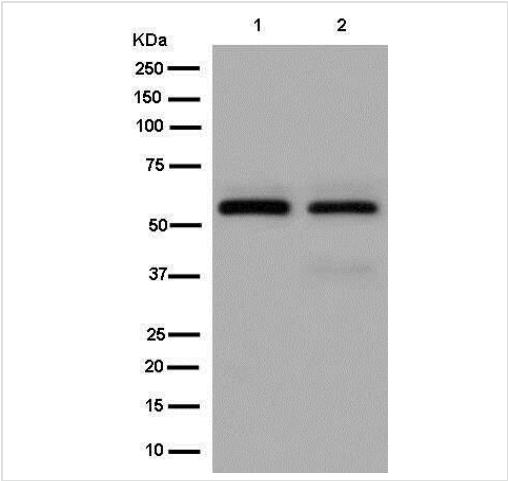
ab181861 was shown to specifically react with PFKFB3 in wild-type HAP1 cells as signal was lost in PFKFB3 knockout cells. Wild-type and PFKFB3 knockout samples were subjected to SDS-PAGE. Ab181861 and **ab9484** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 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.



Immunocytochemistry/ Immunofluorescence - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

Immunofluorescent analysis of 4% paraformaldehyde-fixed A431 cells labeling PFKFB3 with **ab181861** at 1/100 dilution, followed by Goat anti rabbit IgG (Alexa Fluor®555) at 1/200 dilution. Counter stained with Dapi (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).



Western blot - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

All lanes : Anti-PFKFB3 antibody [EPR12594] (**ab181861**) at 1/20000 dilution

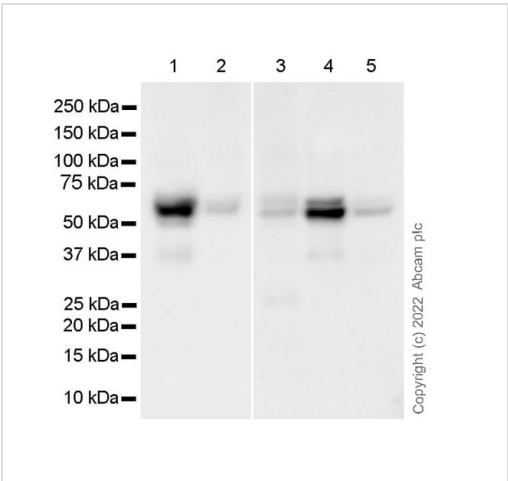
Lane 1 : Jurkat cell lysate
Lane 2 : HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugate at 1/1000 dilution

Predicted band size: 60 kDa
Observed band size: 58 kDa

Blocking buffer: 5% NFDM/TBST



Western blot - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

All lanes : Anti-PFKFB3 antibody [EPR12594] (**ab181861**) at 1/1000 dilution

Lane 1 : A431 (human epidermoid carcinoma epithelial cell) whole cell lysate
Lane 2 : Mouse skin tissue lysate
Lane 3 : Rat breast tissue lysate
Lane 4 : AR42J (rat pancreatic tumor epithelial cell) whole cell lysate
Lane 5 : L6 (rat skeletal muscle myoblast) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 60 kDa
Observed band size: 60 kDa

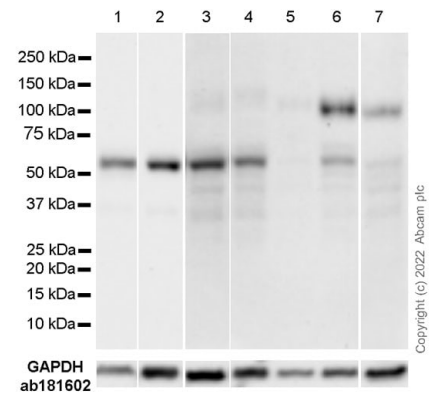
Exposure time: 15 seconds

Blocking buffer: 5% NFDM/TBST

Exposure time: 15 seconds

This blot was developed using a high sensitivity ECL substrate.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).



Western blot - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

All lanes : Anti-PFKFB3 antibody [EPR12594] (**ab181861**) at 1/1000 dilution

Lane 1 : A431 (human epidermoid carcinoma epithelial cell), whole cell lysate
Lane 2 : bEnd.3 (mouse brain endothelial cell), whole cell lysate
Lane 3 : NIH/3T3 (mouse embryonic fibroblast), whole cell lysate
Lane 4 : 4T1 (mouse mammary gland carcinoma epithelial cell), whole cell lysate
Lane 5 : Undifferentiated 3T3-L1 (mouse embryonic fibroblast), whole cell lysate
Lane 6 : 3T3-L1 (mouse embryonic fibroblast) differentiated into adipocyte-like cells, whole cell lysate
Lane 7 : C2C12 (mouse myoblast), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 60 kDa

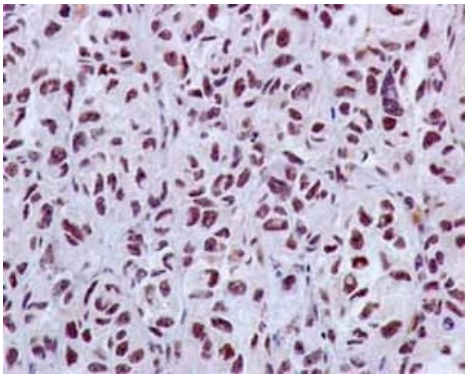
Observed band size: 60 kDa

Exposure time: Lane 1-2: 26 seconds, Lane 3-7: 48 seconds

Blocking buffer and concentration: 5% NFDM/TBST

Lane 3-7 were developed using a high sensitivity ECL substrate.
The expression level of PFKFB3 is upregulated during 3T3-L1 differentiation (PMID: 16306349).
The band at approximately 110 kDa is likely to be PFKFB3 dimer (PMID: 31889092).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).

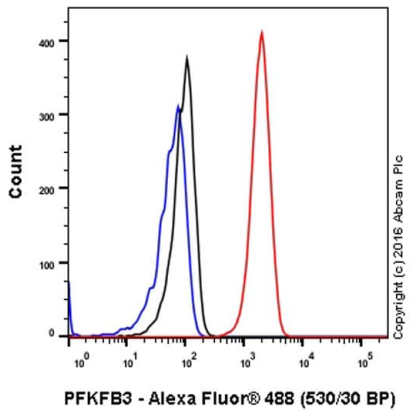


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

This IHC data was generated using the same anti-PFKFB3 antibody clone, EPR12594, in a different buffer formulation (cat# **ab181861**).

Immunohistochemical analysis of paraffin-embedded human melanoma tissue labeling PFKFB3 with **ab181861** at 1/50 dilution followed by prediluted HRP Polymer for Rabbit IgG. Counter stained with Hematoxylin.

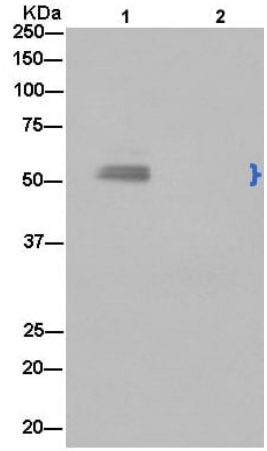
Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

Intracellular Flow Cytometry analysis of A431 (human epidermoid carcinoma) cells labeling PFKFB3 with purified **ab181861** at 1/210 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).

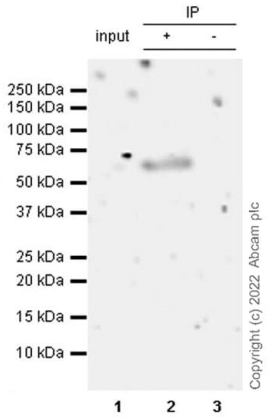


Immunoprecipitation - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

Western blot analysis of PFKFB3 in HeLa cell lysate immunoprecipitated using **ab181861** at 1/50 dilution (Lane 1). Lane 2: Negative control.

Secondary antibody: Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).



Immunoprecipitation - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

PFKFB3 was immunoprecipitated from 0.35 mg of Mouse skin tissue lysate with **ab181861** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab181861** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/5000 dilution.

Lane 1: Mouse skin tissue lysate 10 µg (Input).

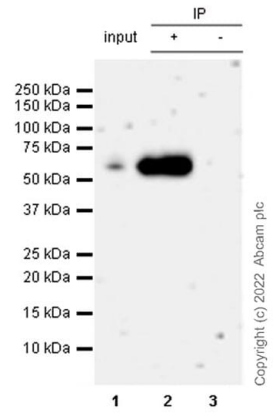
Lane 2: **ab181861** IP in Mouse skin tissue lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab181861** in Mouse skin tissue lysate

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

Exposure time: 180 seconds.

This blot was developed using a high sensitivity ECL substrate.



Immunoprecipitation - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

PFKFB3 was immunoprecipitated from 0.35 mg of AR42J (rat pancreatic tumor epithelial cell) whole cell lysate with **ab181861** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab181861** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/5000 dilution.

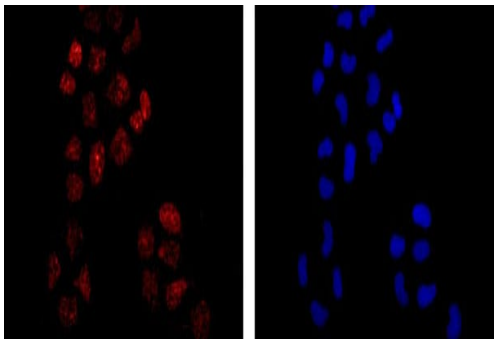
Lane 1: AR42J (rat pancreatic tumor epithelial cell) whole cell lysate 10 µg (Input).

Lane 2: **ab181861** IP in AR42J whole cell lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab181861** in AR42J whole cell lysate

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

Exposure time: 180 seconds.



Immunocytochemistry/ Immunofluorescence - Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

Immunofluorescent analysis of acetone-fixed HeLa cells labeling PFKFB3 with **ab181861** at 1/100 dilution, followed by Goat anti rabbit IgG (Alexa Fluor®555) at 1/200 dilution. Counter stained with Dapi (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab181861**).

Anti-PFKFB3 antibody [EPR12594] - BSA and Azide free (ab218121)

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