abcam

Product datasheet

Anti-PTP1B antibody [EPR22474] ab244207

敲除验证 重组 RabMAb

7 References 14 图像

概述

产品名称	Anti-PTP1B抗体[EPR22474]
描述	兔单克隆抗体[EPR22474] to PTP1B
宿主	Rabbit
特异性	IHC is recommended for human only.
经测试应用	**适用于:** Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP
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: HAP1, HeLa, HepG2, A549, MCF7, SW480, Jurkat, RAW 264.7, PC-12, NIH/3T3 and HCT 116 whole cell lysates. IHC-P: Human breast cancer and colon cancer tissue. ICC/IF: HeLa, wild type HAP1 and HCT 116 cells. Flow Cyt (intra): HeLa and HCT 116 cells. IP: PTP1B IP in HCT 116 whole cell lysate.
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: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
纯度	Protein A purified

克隆	单克隆
克隆编号	EPR22474
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab244207于以下的经测试应用

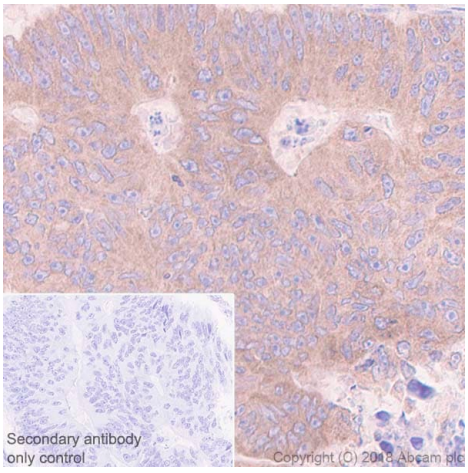
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/50.
WB		1/1000. Predicted molecular weight: 50 kDa.
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. IHC is recommended for human only.
ICC/IF		1/50.
IP		1/30.

靶标

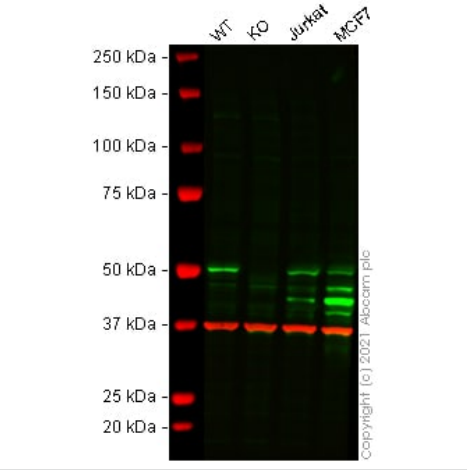
功能	May play an important role in CKII- and p60c-src-induced signal transduction cascades.
序列相似性	Belongs to the protein-tyrosine phosphatase family. Non-receptor class 1 subfamily. Contains 1 tyrosine-protein phosphatase domain.
翻译后修饰	Oxidized on Cys-215; the Cys-SOH formed in response to redox signaling reacts with the alpha-amido of the following residue to form a 4-amino-3-isothiazolidinone serine cross-link, triggering a conformational change that inhibits substrate binding and activity. The active site can be restored by reduction.
细胞定位	Endoplasmic reticulum membrane.

图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PTP1B antibody [EPR22474] (ab244207)

Immunohistochemical analysis of paraffin-embedded human colon cancer tissue labeling PTP1B with ab244207 at 1/1000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP) secondary antibody. Cytoplasmic staining in human colon cancer (PMID:27752061) is observed. Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).
Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).



Western blot - Anti-PTP1B antibody [EPR22474] (ab244207)

All lanes : Anti-PTP1B antibody [EPR22474] (ab244207) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : PTPN1 knockout HeLa cell lysate
Lane 3 : Jurkat cell lysate
Lane 4 : MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

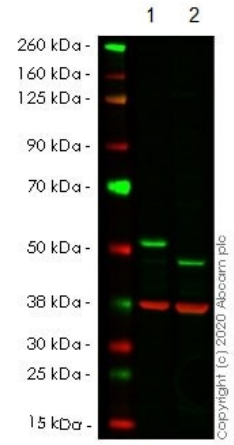
Performed under reducing conditions.

Predicted band size: 50 kDa
Observed band size: 51 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab244207 observed at 51 kDa. Red - loading control **ab8245** (Mouse anti-

GAPDH antibody [6C5]) observed at 37 kDa.

ab244207 was shown to react with PTP1B in wild-type HeLa cells in Western blot with loss of signal observed in PTPN1 knockout cell line **ab265014** (PTPN1 knockout cell lysate **ab257617**). Wild-type HeLa and PTPN1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5 % milk in TBS-T (0.1 % Tween®) before incubation with ab244207 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated 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 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-PTP1B antibody [EPR22474] (ab244207)

All lanes : Anti-PTP1B antibody [EPR22474] (ab244207) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : PTPN1 CRISPR/Cas9 edited HeLa cell lysate

Lysates/proteins at 20 µg per lane.

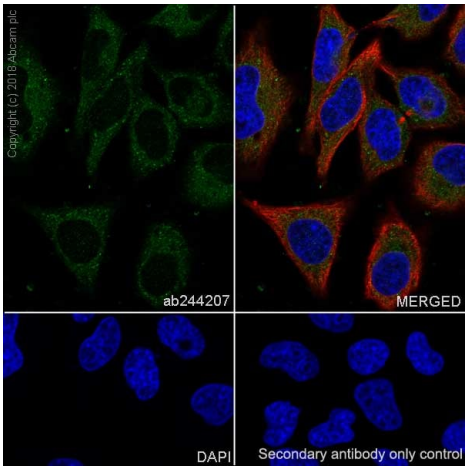
Performed under reducing conditions.

Predicted band size: 50 kDa
Observed band size: 50 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab244207 observed at 50 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

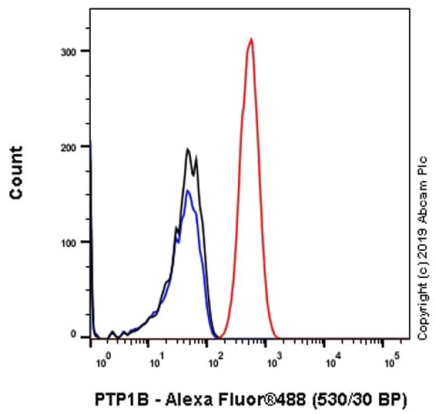
ab244207 was shown to react with PTP1B in wild-type HeLa cells in western blot. The band observed in CRISPR/Cas9 edited cell line **ab265014** (CRISPR/Cas9 edited cell lysate **ab257617**) lane below 50kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type HeLa and PTPN1 CRISPR/Cas9 edited 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. ab244207 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) 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.



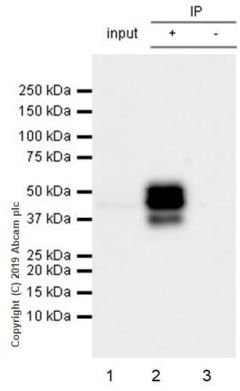
Immunocytochemistry/ Immunofluorescence - Anti-PTP1B antibody [EPR22474] (ab244207)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cells labeling PTP1B with ab244207 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in HeLa cells is observed. The nuclear counterstain is DAPI (blue). Counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red). The negative control is the secondary antibody only.



Flow Cytometry (Intracellular) - Anti-PTP1B antibody [EPR22474] (ab244207)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cell line labeling PTP1B with ab244207 at 1/500 (red) compared with a Rabbit monoclonal IgG (**ab172730**)isotype control (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**), at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-PTP1B antibody [EPR22474] (ab244207)

PTP1B was immunoprecipitated from 0.35 mg HCT 116 (human colorectal carcinoma epithelial cell) whole cell lysate with ab244207 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab244207 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/5000 dilution.

Lane 1: HCT 116 whole cell lysate 10 µg (Input).

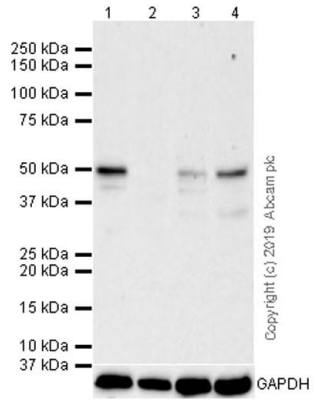
Lane 2: ab244207 IP in HCT 116 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab244207 in HCT 116 whole cell lysate.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 30 seconds.

The observed MW is consistent with what described in the literatures. (PMID: 18253097; PMID: 11895943; PMID: 19797268).



Western blot - Anti-PTP1B antibody [EPR22474] (ab244207)

All lanes : Anti-PTP1B antibody [EPR22474] (ab244207) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : PTP1B knockout HAP1 whole cell lysate

Lane 3 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 4 : HCT 116 (human colorectal carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 50 kDa

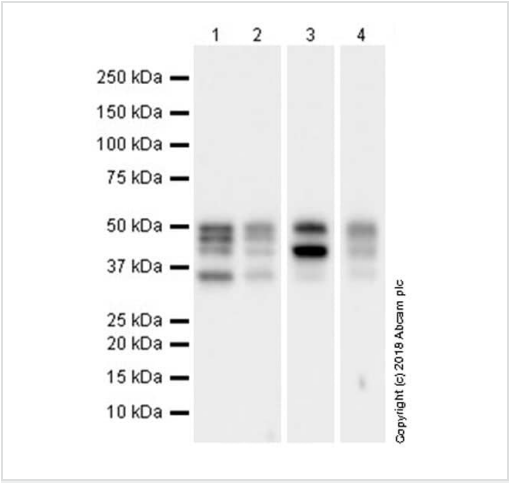
ab244207 was shown to specifically react with PTP1B in wild-type HAP1 cells as signal was lost in PTP1B knockout cells. Wild-type and PTP1B knockout samples were subjected to SDS-PAGE. ab244207 and **ab181602** (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-

RAD® ChemiDoc™ MP instrument using the ECL technique.

The expression profile observed is consistent with what has been described in the literature (PMID: 18253097; PMID: 11895943; PMID: 19797268). The bands below 50 kDa may represent truncated forms and cleaved fragments.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



Western blot - Anti-PTP1B antibody [EPR22474] (ab244207)

All lanes : Anti-PTP1B antibody [EPR22474] (ab244207) at 1/1000 dilution

Lane 1 : HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate
Lane 2 : A549 (human lung carcinoma epithelial cell) whole cell lysate
Lane 3 : MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate
Lane 4 : SW480 (human colorectal adenocarcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

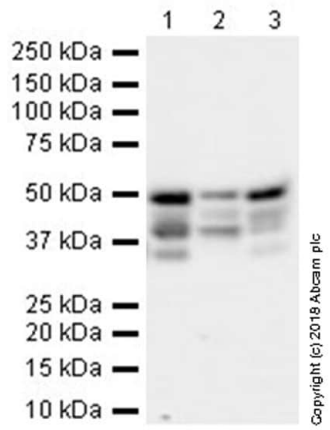
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/50000 dilution

Predicted band size: 50 kDa

The expression profile observed is consistent with what has been described in the literature (PMID: 18253097; PMID: 11895943; PMID: 19797268). The bands below 50 kDa may represent truncated forms and cleaved fragments.

Blocking/Dilution buffer: NFDM/TBST.

Exposure time: 37 seconds.



Western blot - Anti-PTP1B antibody [EPR22474] (ab244207)

All lanes : Anti-PTP1B antibody [EPR22474] (ab244207) at 1/1000 dilution

Lane 1 : RAW 264.7 (mouse abelson murine leukemia virus-induced tumor macrophage) whole cell lysate
Lane 2 : PC-12 (rat adrenal gland pheochromocytoma) whole cell lysate
Lane 3 : NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

Lysates/proteins at 10 µg per lane.

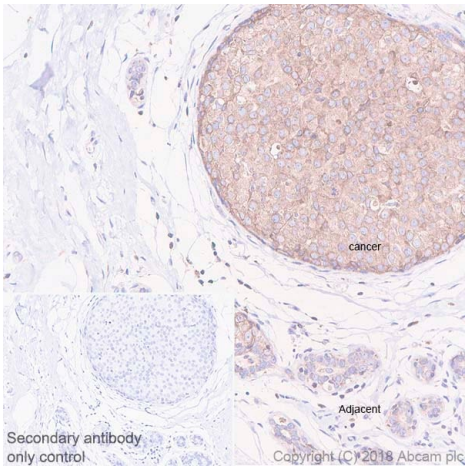
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 50 kDa

The expression profile observed is consistent with what has been described in the literature (PMID: 18253097; PMID: 11895943; PMID: 19797268). The bands below 50 kDa may represent truncated forms and cleaved fragments.

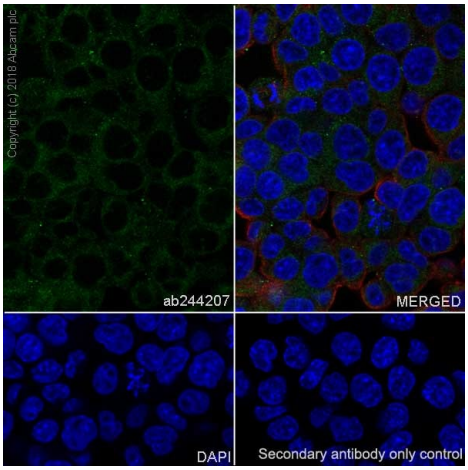
Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 48 seconds.



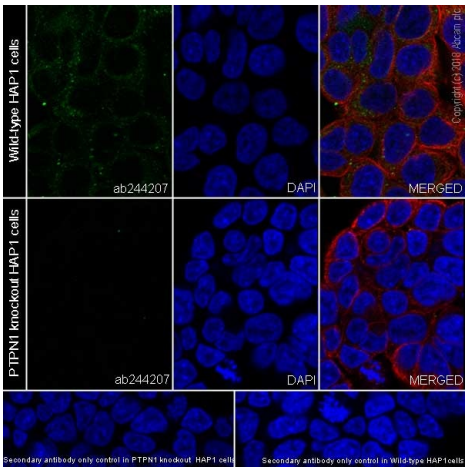
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PTP1B antibody [EPR22474] (ab244207)

Immunohistochemical analysis of paraffin-embedded human breast cancer tissue labeling PTP1B with ab244207 at 1/1000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Higher cytoplasmic expression in human breast cancer than that of adjacent normal tissues (PMID: 27465552) is observed. Counterstained with hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).
Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).



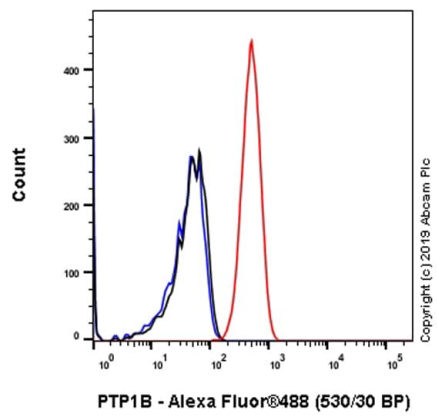
Immunocytochemistry/ Immunofluorescence - Anti-PTP1B antibody [EPR22474] (ab244207)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HCT 116 (human colorectal carcinoma epithelial cell) cells labeling PTP1B with ab244207 at 1/50 dilution, followed by a Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in HCT 116 cells. The nuclear counterstain is DAPI (blue).
Counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red).
The negative control is the secondary antibody only.



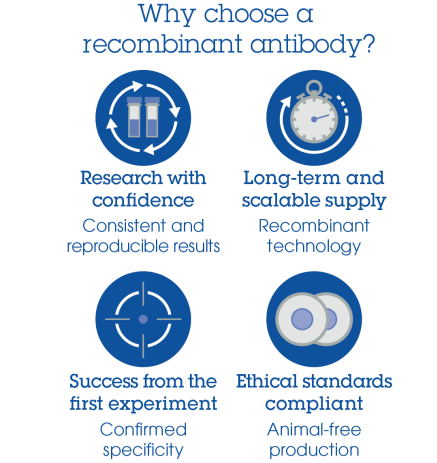
Immunocytochemistry/ Immunofluorescence - Anti-PTP1B antibody [EPR22474] (ab244207)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Wild type and PTP1B-knockout HAP1 (Human chronic myelogenous leukemia near-haploid cell line) cells labeling PTP1B with ab244207 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing no staining in PTP1B-knockout HAP1 cells. The nuclear counterstain is DAPI (blue).
Counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red).
The negative control is the secondary antibody only.



Flow Cytometry (Intracellular) - Anti-PTP1B antibody [EPR22474] (ab244207)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HCT 116 (human colorectal carcinoma epithelial cell) cell line labeling PTP1B with ab244207 at 1/50 (red) compared with a Rabbit monoclonal IgG (**ab172730**) isotype control (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**), at 1/2000 dilution was used as the secondary antibody.



Anti-PTP1B antibody [EPR22474] (ab244207)

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