# abcam

## Product datasheet

# Anti-IDH1 antibody [EPR21002] ab230949





重组 RabMAb

### 11 图像

#### 概述

产品名称 Anti-IDH1抗体[EPR21002]

描述 兔单克隆抗体[EPR21002] to IDH1

宿主 Rabbit

经测试应用 适用于: Flow Cyt (Intra), WB, IHC-P, 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: SH-SY5Y, Raji, C2C12, Neuro-2a, HepG2, HAP1, HeLa, C6 and PC-12 whole cell lysates;

Human fetal brain and fetal kidney lysates. IHC-P: Human stomach, glioblastoma and

endometrium cancer tissues; Mouse and rat kidney tissues. Flow Cyt (intra): HeLa cells. IP: SH-

SY5Y 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** EPR21002

**同种型** IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab230949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 <b>用</b>	Ab评论	说明
Flow Cyt (Intra)		1/50.
WB		1/1000. Detects a band of approximately 47 kDa (predicted molecular weight: 46 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		1/30.

应用说明 Is unsuitable for ICC/IF.

靶标

疾病相关 Glioma

Genetic variations are associated with cartilaginous tumors such as enchondroma or

chondrosarcoma. Mutations of Arg-132 to Cys, Gly or His abolish the conversion of isocitrate to

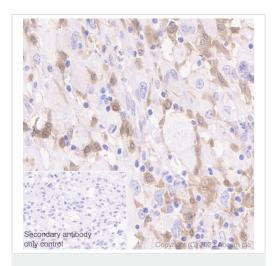
alpha-ketoglutarate. Instead, alpha-ketoglutarate is converted to R(-)-2-hydroxyglutarate.

序列相似性 Belongs to the isocitrate and isopropylmalate dehydrogenases family.

翻译后修饰 Acetylation at Lys-374 dramatically reduces catalytic activity.

细胞定位 Cytoplasm. Peroxisome.

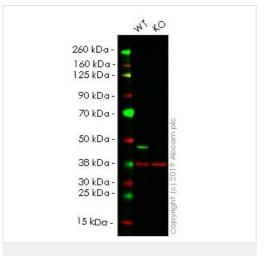
图片



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IDH1 antibody
[EPR21002] (ab230949)

Immunohistochemical analysis of paraffin-embedded human glioblastoma tissue labeling IDH1 with ab230949 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP). Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Positive staining on human glioblastoma. The section was incubated with ab230949 at 4°C overnight.



Western blot - Anti-IDH1 antibody [EPR21002] (ab230949)

**All lanes :** Anti-IDH1 antibody [EPR21002] (ab230949) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : IDH1 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

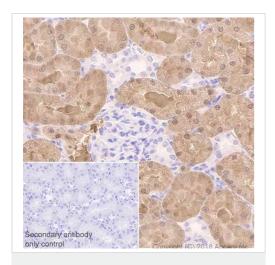
**Predicted band size:** 46 kDa **Observed band size:** 46 kDa

**Lanes 1-2:** Merged signal (red and green). Green - ab230949 observed at 46 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

ab230949 was shown to react with IDH1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab264916 (knockout cell lysate ab257221) was used. Wild-type HeLa and IDH1 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. ab230949 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) 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

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(IRDye<sup>®</sup>680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

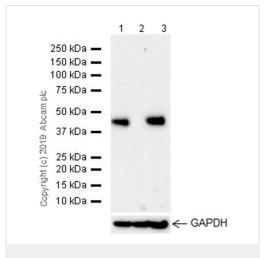


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IDH1 antibody
[EPR21002] (ab230949)

Immunohistochemical analysis of paraffin-embedded rat kidney tissue labeling IDH1 with ab230949 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic and nuclear staining in rat kidney (PMID:30153799). Counter stained with hematoxylin.

**Secondary antibody only control:** Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-IDH1 antibody [EPR21002] (ab230949)

All lanes: Anti-IDH1 antibody [EPR21002] (ab230949) at 1/1000 dilution

Lane 1: Wild type HAP1 whole cell lysate

Lane 2: IDH1 knockout HAP1 whole cell lysate

Lane 3: SH-SY5Y (human neuroblastoma epithelial cell), whole cell

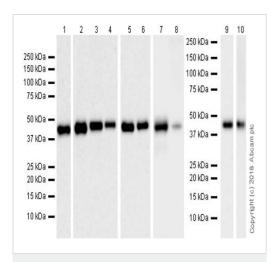
lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 46 kDa

Exposure time: 62 seconds

ab230949 was shown to specifically react with IDH1 in wild-type HAP1 cells as signal was lost in IDH1 knockout cells. Wild-type and IDH1 knockout samples were subjected to SDS-PAGE. ab230949 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.



Western blot - Anti-IDH1 antibody [EPR21002] (ab230949)

**All lanes :** Anti-IDH1 antibody [EPR21002] (ab230949) at 1/1000 dilution

Lane 1 : SH-SY5Y (human neuroblastoma cell line from bone marrow) whole cell lysate at 20  $\mu g$ 

Lane 2 : Raji (human Burkitt's lymphoma cell line) whole cell lysate at 20 µg

Lane 3 : C2C12 (mouse myoblast cell line) whole cell lysate at 20 µg

**Lane 4 :** Neuro-2a (mouse neuroblastoma cell line) whole cell lysate at 20  $\mu g$ 

**Lane 5**: Human fetal brain lysate at 20 μg **Lane 6**: Human fetal kidney lysate at 20 μg

**Lane 7 :** HepG2 (human liver hepatocellular carcinoma cell line) whole cell lysate at 20  $\mu g$ 

Lane 8 : HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

**Lane 9 :** C6 (rat glial tumor cell line) whole cell lysate at 10  $\mu$ g **Lane 10 :** PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysate at 10  $\mu$ g

#### **Secondary**

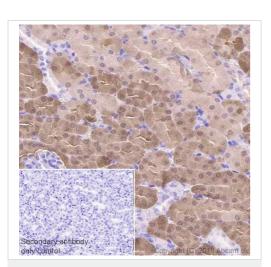
**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

**Predicted band size:** 46 kDa **Observed band size:** 47 kDa

**Exposure times:** Lanes 1-6: 3 minutes; Lanes 7-8: 15 seconds;

Lanes 9-10: 58 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

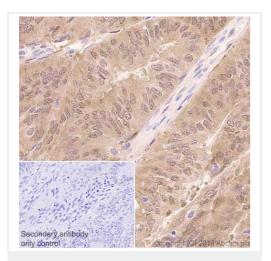


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IDH1 antibody
[EPR21002] (ab230949)

Immunohistochemical analysis of paraffin-embedded mouse kidney tissue labeling IDH1 with ab230949 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic and nuclear staining in mouse kidney (PMID:30153799). Counter stained with hematoxylin.

**Secondary antibody only control:** Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

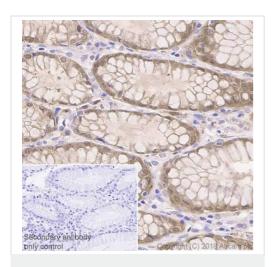


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IDH1 antibody
[EPR21002] (ab230949)

Immunohistochemical analysis of paraffin-embedded human endometrium cancer tissue labeling IDH1 with ab230949 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic and nuclear staining in human endometrium cancer (PMID:29921847). Counter stained with hematoxylin.

**Secondary antibody only control:** Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



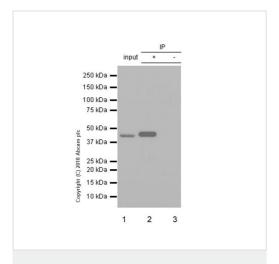
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IDH1 antibody
[EPR21002] (ab230949)

Immunohistochemical analysis of paraffin-embedded human stomach tissue labeling IDH1 with ab230949 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Cytoplasmic and nucleus staining in human stomach (PMID:27466503). Counter stained with hematoxylin.

**Secondary antibody only control:** Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-IDH1 antibody [EPR21002] (ab230949)

IDH1 was immunoprecipitated from 0.35 mg of SH-SY5Y (human neuroblastoma cell line from bone marrow) whole cell lysate with ab230949 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab230949 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

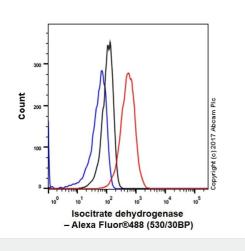
Lane 1: SH-SY5Y whole cell lysate 10 µg (Input).

Lane 2: ab230949 IP in SH-SY5Y whole cell lysate.

**Lane 3:** Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab230949 in SH-SY5Y whole cell lysate.

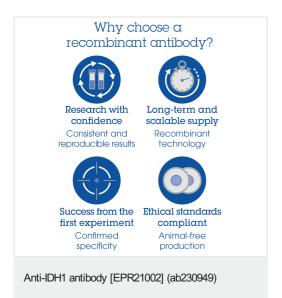
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.



Flow Cytometry (Intracellular) - Anti-IDH1 antibody [EPR21002] (ab230949)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cell line labeling IDH1 with ab230949 at 1/60 dilution (**red**) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (**ab172730**) (**black**) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (**blue**). Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>Ti</sup>¿½ 488) (**ab150077**) at 1/2000 dilution was used as the secondary antibody.



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