abcam

Product datasheet

Anti-GAPDH antibody - Loading Control ab9485

88 Abreviews 2835 References 9 图像

概述

产品名称	Anti-GAPDH抗体- Loading Control
描述	兔多克隆抗体to GAPDH - Loading Control
宿主	Rabbit
特异性	From Mar 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help. You may also be interested in our alternative recombinant antibody, **ab313650**.
经测试应用	**适用于:** IHC-P, WB, ICC/IF
种属反应性	**与反应:** Mouse, Human **预测可用于:** Rat, Chicken, Dog, Saccharomyces cerevisiae, Xenopus laevis, Schizosaccharomyces pombe, African green monkey
免疫原	Full length native protein (purified) corresponding to Human GAPDH.
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

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 or -80°C. Avoid freeze / thaw cycle.
存储溶液	Preservative: 0.02% Sodium azide Constituents: 98.98% PBS, 1% BSA Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

纯度	Protein A purified
克隆	**多克隆**
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab9485于以下的经测试应用**

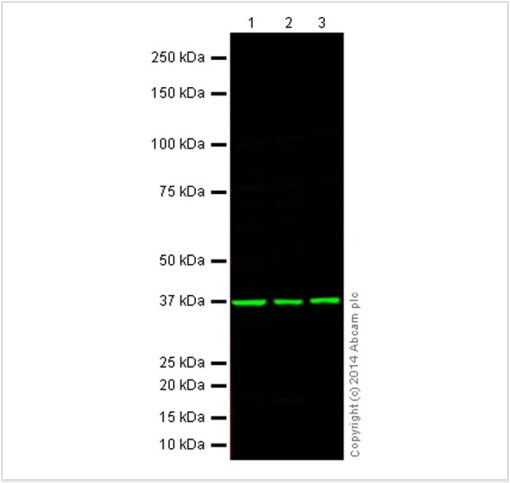
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
IHC-P		Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB	★★★★★ (74)	1/2500. Detects a band of approximately 40 kDa (predicted molecular weight: 37 kDa). Some customers have experienced that milk significantly decreases the signal in WB compared to BSA. In-house we use BSA. We recommend **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (ab175781) secondary antibody**
ICC/IF	★★★★★ (6)	Use a concentration of 5 µg/ml. We recommend **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody**.

靶标

功能	Has both glyceraldehyde-3-phosphate dehydrogenase and nitrosylase activities, thereby playing a role in glycolysis and nuclear functions, respectively. Participates in nuclear events including transcription, RNA transport, DNA replication and apoptosis. Nuclear functions are probably due to the nitrosylase activity that mediates cysteine S-nitrosylation of nuclear target proteins such as SIRT1, HDAC2 and PRKDC (By similarity). Glyceraldehyde-3-phosphate dehydrogenase is a key enzyme in glycolysis that catalyzes the first step of the pathway by converting D-glyceraldehyde 3-phosphate (G3P) into 3-phospho-D-glyceroyl phosphate.
通路	Carbohydrate degradation; glycolysis; pyruvate from D-glyceraldehyde 3-phosphate: step 1/5.
序列相似性	Belongs to the glyceraldehyde-3-phosphate dehydrogenase family.
翻译后修饰	S-nitrosylation of Cys-152 leads to interaction with SIAH1, followed by translocation to the nucleus. ISGylated.
细胞定位	Cytoplasm > cytosol. Nucleus. Cytoplasm > perinuclear region. Membrane. Translocates to the nucleus following S-nitrosylation and interaction with SIAH1, which contains a nuclear localization signal (By similarity). Postnuclear and Perinuclear regions.

图片



Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

All lanes : Anti-GAPDH antibody - Loading Control (ab9485) at 1/2500 dilution

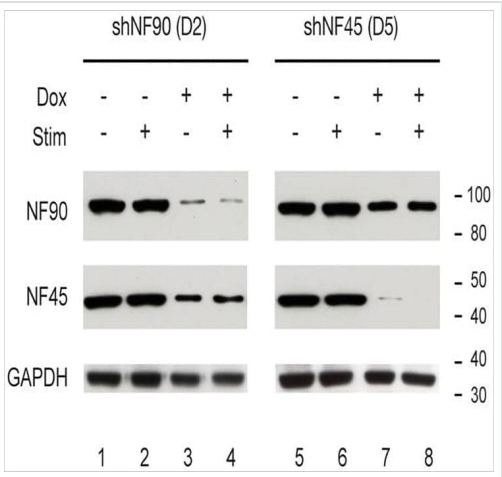
Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2 : A431 (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 3 : A549 (Human lung adenocarcinoma epithelial cell line) Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (**ab175781**) secondary antibody at 1/10000 dilution

Predicted band size: 37 kDa
Observed band size: 37 kDa

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using Licor blocking buffer before being incubated with ab9485 overnight at 4°C. Antibody binding was detected using **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (ab175781) secondary antibody** at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.

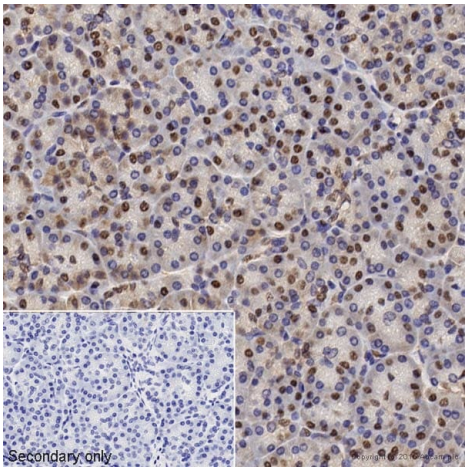


Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

HEK293 cells stably transfected with pINDUCER10-shNF90/NF110 (D2) or pINDUCER10-shNF45 (D5) were treated without or with doxycycline for 96 h, then serum starved for 12 h and treated with PMA (20 ng/mL) for 2 h. Protein lysates (20 µg/lane) were separated by SDS-PAGE and transferred to PVDF membranes.

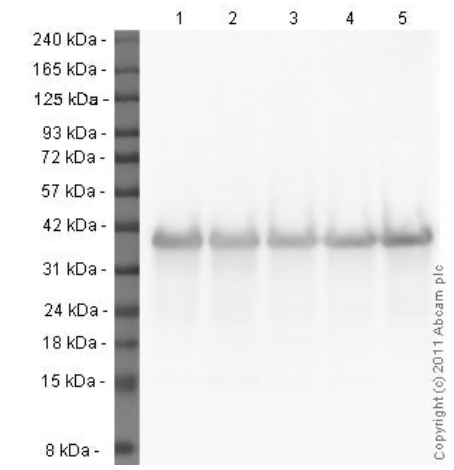
Loading control: Rabbit polyclonal to GAPDH (ab9485) at 1/1000 dilution.

Secondary antibodies (HRP) were used at 1/10,000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GAPDH antibody - Loading Control (ab9485)

IHC image of ab9485 staining GAPDH in human pancreas formalin fixed paraffin embedded tissue sections*, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab9485, 5µg/ml working concentration, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the secondary only control (shown on the inset).
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.
*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

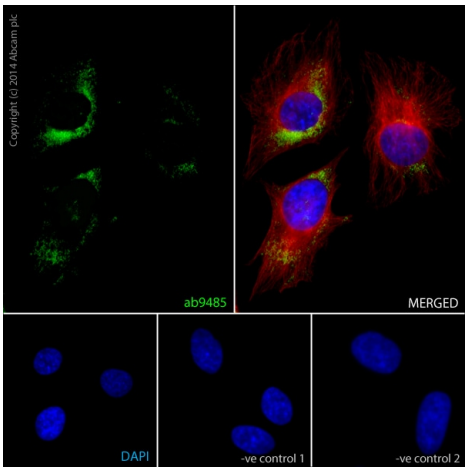
All lanes : Anti-GAPDH antibody - Loading Control (ab9485) at 1 µg/ml

Lane 1 : HeLa cell lysate
Lane 2 : Jurkat cell lysate
Lane 3 : A431 cell lysate
Lane 4 : HEK-293 cell lysate
Lane 5 : HepG2 cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 37 kDa

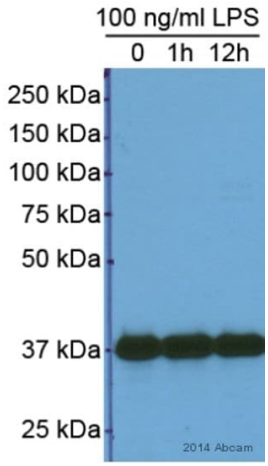
Western blot image using 4-20% Optiblot gel with the Prism Ultra Protein Ladder (**ab116028**) 5µl used. We recommend using our ECL substrate kit (**ab65623**).
20ug of Lysate per lane and detection using ab9485 diluted to 1ug/ml.
Lane 1: HeLa cell lysate
Lane 2: Jurkat cell lysate
Lane 3: A431 cell lysate
Lane 4: HEK-293 cell lysate
Lane 5: HepG2 cell lysate.



Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody - Loading Control (ab9485)

ab9485 staining GAPDH in HeLa cells. The cells were fixed with 100% methanol (5min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab9485 at 5µg/ml and **ab7291** at 1µg/ml overnight at +4°C, followed by a further incubation at room temperature for 1h with **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody** at 2 µg/ml (shown in green) and **Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) secondary antibody** at 2 µg/ml (shown in pseudo color red). Nuclear DNA was labelled in blue with DAPI.

Negative controls: 1– Rabbit primary antibody and anti-mouse secondary antibody; 2 – Mouse primary antibody and anti-rabbit secondary antibody. Controls 1 and 2 indicate that there is no unspecific reaction between primary and secondary antibodies used.



Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

This image is courtesy of an anonymous Abreview

All lanes : Anti-GAPDH antibody - Loading Control (ab9485) at 1/1000 dilution

Lane 1 : Mouse hepatocytes - untreated
Lane 2 : Mouse hepatocytes - treated with LPS (100 ng/mL) for 1 hour
Lane 3 : Mouse hepatocytes - treated with LPS (100 ng/mL) for 12 hours

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-rabbit secondary antibody (HRP) at 1/10000 dilution

Developed using the ECL technique.

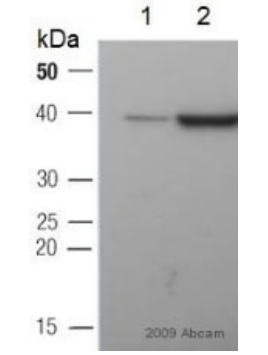
Performed under reducing conditions.

Predicted band size: 37 kDa
Observed band size: 37 kDa

Exposure time: 1 minute

Primary incubation: 16 hours at 4°C

Blocking: 5% milk for 1 hour at room temperature



Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

This image is a courtesy of Anonymous Abreview

All lanes : Anti-GAPDH antibody - Loading Control (ab9485) at 1/2500 dilution

Lane 1 : Lysate prepared from human Huh-7 cells at 2 µg
Lane 2 : Lysate prepared from human Huh-7 cells at 20 µg

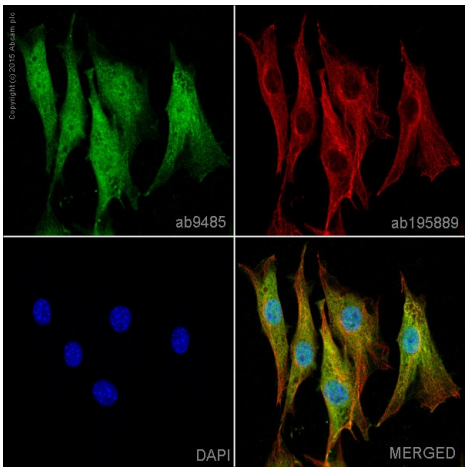
Secondary
All lanes : HRP-conjugated sheep polyclonal to rabbit IgG at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 37 kDa

Observed band size: 40 kDa

Exposure time: 5 minutes



Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody - Loading Control (ab9485)

ab9485 staining GAPDH in NIH3T3 cells. The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab9485 at 5µg/ml and **ab195889** at 1/250 overnight at +4°C, followed by a further incubation at room temperature for 1h with **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody** at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Western blot - Anti-GAPDH antibody - Loading Control (ab9485)

Anti-GAPDH antibody - Loading Control (ab9485) at 1/1000 dilution + Mouse Embryonic lung whole tissue lysate at 30 µg

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 37 kDa

Exposure time: 15 seconds

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish

- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.cn/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors