abcam

Product datasheet

Anti-GAPDH antibody - Loading Control ab9483

6 Abreviews 86 References 3 Images

Overview

Product name	Anti-GAPDH antibody - Loading Control
Description	Goat polyclonal to GAPDH - Loading Control
Host species	Goat
Tested applications	**Suitable for:** WB, ICC/IF
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Full length native protein from human erythrocytes.
Positive control	This antibody gave a positive signal in the following whole cell lysates: HeLa; NIH3T3. This antibody also gave a positive signal in Human brain tissue lysate.

Properties

Form	Liquid
Storage instructions	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.
Storage buffer	Preservative: 0.02% Sodium azide This product may contain up to 3% BSA depending on the batch. For specific batch formulations please contact us.
Purity	Immunogen affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab9483** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★	1/1000. Detects a band of approximately 37 kDa (predicted molecular weight: 35.8 kDa).

Application	Abreviews	Notes
ICC/IF	★★★★☆	Use a concentration of 1 µg/ml.

Target

Function
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.

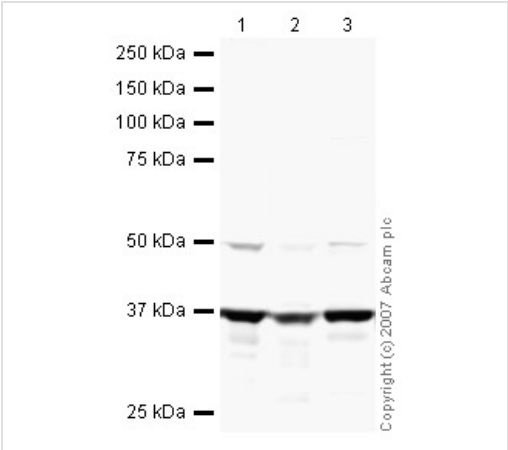
Pathway
Carbohydrate degradation; glycolysis; pyruvate from D-glyceraldehyde 3-phosphate: step 1/5.

Sequence similarities
Belongs to the glyceraldehyde-3-phosphate dehydrogenase family.

Post-translational modifications
S-nitrosylation of Cys-152 leads to interaction with SIAH1, followed by translocation to the nucleus.
ISGylated.

Cellular localization
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.

Images



Western blot - Anti-GAPDH antibody - Loading Control (ab9483)

All lanes : Anti-GAPDH antibody - Loading Control (ab9483) at 1 µg/ml

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2 : NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate
Lane 3 : Human brain tissue lysate - total protein (ab29466)

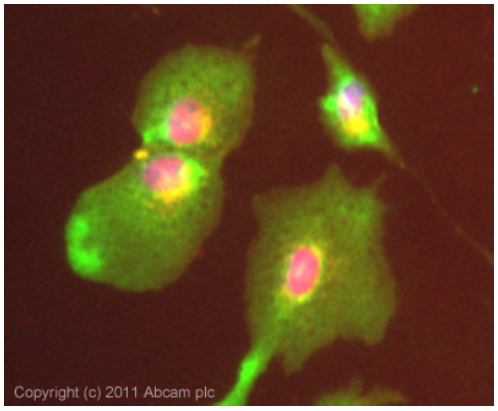
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Rabbit polyclonal to Goat IgG (Alexa Fluor® 680) at 1/10000 dilution

Performed under reducing conditions.

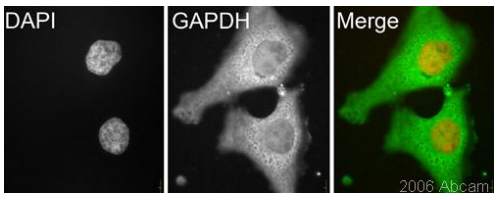
Predicted band size: 35.8 kDa
Observed band size: 37 kDa

why is the actual band size different from the predicted?



Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody - Loading Control (ab9483)

ICC/IF image of ab9483 stained HepG2 cells. The cells were 4% PFA fixed (10 min) and then incubated in 1%BSA / 10% normal donkey serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab9483, 1µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 donkey anti-goat IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM. This antibody also gave a positive result in 4% PFA fixed (10 min) HeLa cells at 1µg/ml.



Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody - Loading Control (ab9483)

This image is courtesy of an Abreview submitted by Kirk McManus

ab9483 staining GAPDH in HeLa by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehydeand. Samples were incubated with primary antibody (1/200) for 30 minutes. An undiluted Alexa Fluor® 568-conjugated Donkey anti-goat polyclonal was used as the secondary antibody.

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