# abcam

# Product datasheet

# Anti-GAPDH antibody [EPR16884] - Loading Control ab181603



\*\*\* \* \* 6 Abreviews 193 References 10 Images

#### Overview

Product name Anti-GAPDH antibody [EPR16884] - Loading Control

**Description** Rabbit monoclonal [EPR16884] to GAPDH - Loading Control

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF

Species reactivity Reacts with: Mouse, Rat, Chicken, Cow, Dog, Human, African green monkey

Predicted to work with: Monkey

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HeLa, MDBK, COS-1, MDCK, UMNSAH/DF-1, Jurkat, C6 and NIH/3T3 whole cell lysates;

Mouse brain and heart lysates; Rat brain, heart, kidney and spleen lysates; Human fetal brain, heart and kidney lysates. IHC-P: Human transitional cell carcinoma of bladder, Mouse spleen and

Rat spleen tissues. ICC/IF: HeLa cells. Flow: Jurkat cells.

**General notes**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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

#### **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 long

term. Avoid freeze / thaw cycle.

**Storage buffer** Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

1

ClonalityMonoclonalClone numberEPR16884

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab181603 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
Flow Cyt (Intra)		1/200. <b>ab172730</b> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	****(3)	1/10000. Detects a band of approximately 36 kDa (predicted molecular weight: 36 kDa).
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★☆ (3)	1/250.

#### **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 S-nitrosylation of Cys-152 leads to interaction with SIAH1, followed by translocation to the

modifications nucleus.

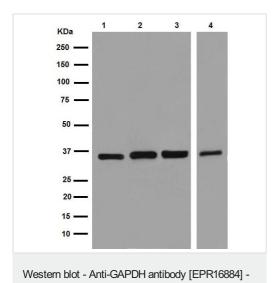
ISGylated.

**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**



Loading Control (ab181603)

**All lanes :** Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603) at 1/10000 dilution

**Lane 1**: HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysates

Lane 2: MDBK (Bovine kidney cell line) whole cell lysates

Lane 3 : COS-1 (African green monkey kidney fibroblast-like cell line) whole cell lysates

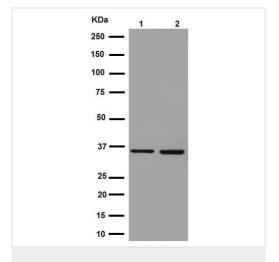
Lane 4: MDCK (Canine kidney cell line) whole cell lysates

Lysates/proteins at 20 µg per lane.

## Secondary

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

**Predicted band size:** 36 kDa **Observed band size:** 36 kDa



Western blot - Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603)

Blocking/Dilution buffer: 5% NFDM/TBST.

**All lanes :** Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603) at 1/50000 dilution

**Lane 1 :** UMNSAH/DF-1 (Transformed chicken embyronic fibroblast cells) whole cell lysates

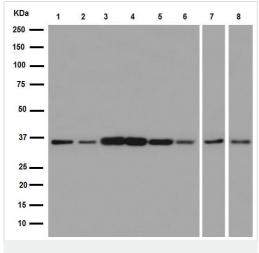
**Lane 2**: Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysates

Lysates/proteins at 20 µg per lane.

### Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L),Peroxidase conjugated at 1/1000 dilution

Predicted band size: 36 kDa Observed band size: 36 kDa Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603)

**All lanes :** Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603) at 1/10000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate

Lane 3: Rat brain lysate

Lane 4: Rat heart lysate

Lane 5 : Rat kidney lysate

Lane 6: Rat spleen lysate

Lane 7: C6 (Rat glial tumor cells) whole cell lysates

Lane 8: NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysates

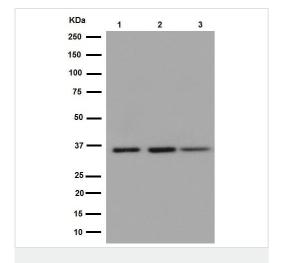
Lysates/proteins at 10 µg per lane.

# Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L),Peroxidase conjugated at 1/1000 dilution

**Predicted band size:** 36 kDa **Observed band size:** 36 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603)

**All lanes :** Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603) at 1/10000 dilution

Lane 1 : Human fetal brain lysate

Lane 2 : Human fetal heart lysate

Lane 3 : Human fetal kidney lysate

Lysates/proteins at 10 µg per lane.

#### Secondary

**All lanes :** Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

**Predicted band size:** 36 kDa **Observed band size:** 36 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

ab181603 MERGED

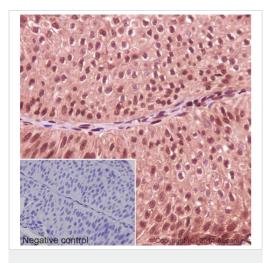
DAPI -ve control 1 -ve control 2

Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody [EPR16884] - Loading Control (ab181603)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling GAPDH with ab181603 at 1/250 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/400 dilution (green). Cytoplasm and nuclear staining on HeLa cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows;

ab181603 at 1/250 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
 <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/500 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/400 dilution.

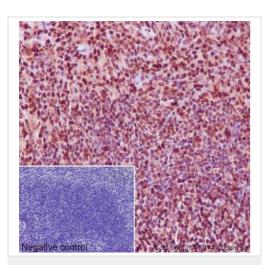


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GAPDH antibody
[EPR16884] - Loading Control (ab181603)

Immunohistochemical analysis of paraffin-embedded Human transitional cell carcinoma of bladder tissue labeling GAPDH with ab181603 at 1/500 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG. Cytoplasmic and nucleus staining on the tumor cells of transitional cell carcinoma of Human bladder is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

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



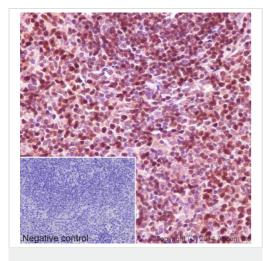
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GAPDH antibody

[EPR16884] - Loading Control (ab181603)

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling GAPDH with ab181603 at 1/500 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG. Nucleus and cytoplasmic staining on lymphocytes of mouse spleen is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

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

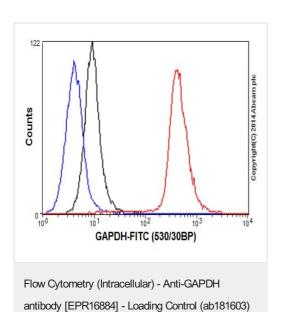


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GAPDH antibody
[EPR16884] - Loading Control (ab181603)

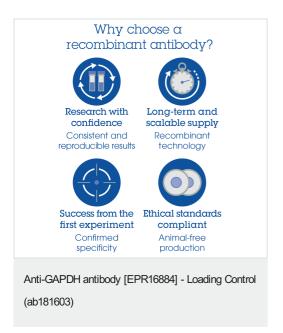
Immunohistochemical analysis of paraffin-embedded Rat spleen tissue labeling GAPDH with ab181603 at 1/500 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG. Nucleus and cytoplasmic staining on lymphocyte of rat spleen is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

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



Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling GAPDH with ab181603 at 1/200 dilution (red) compared with a rabbit monoclonal IgG isotype control (black) and a unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (FITC) at 1/150 dilution was used as the secondary antibody.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

### 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 <a href="https://www.abcam.com/abpromise">https://www.abcam.com/abpromise</a> or contact our technical team.

### Terms and conditions

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