abcam

ab199553

Anti-GAPDH antibody [EPR16891] - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR16891
Purification technique	Affinity purification Protein A
Concentration	1.011 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-

Notes	ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse, Rat, African green monkey, Chicken, Xenopus tropicalis, Zebrafish
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Fish, Rabbit
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat, African green monkey, Chicken, Xenopus tropicalis, Zebrafish
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Fish, Rabbit
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat, African green monkey, Chicken, Xenopus tropicalis, Zebrafish
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Fish, Rabbit
Dilution info	-
Notes	-

WB

Tested

Species	Mouse, Rat, African green monkey, Human, Chicken, Xenopus tropicalis, Zebrafish
Dilution info	-
Notes	-

Predicted

Species	Fish, Rabbit
Dilution info	-
Notes	-

IHC-P

Tested

Species	Rat
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Human
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Expected

Species	African green monkey, Chicken, Xenopus tropicalis, Zebrafish
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Fish, Rabbit
Dilution info	-
Notes	-

Target data

See full target information GAPDH

Function	Catalyzes the conversion of D-glyceraldehyde 3-phosphate (G3P) into 3-phospho-D-glyceroyl phosphate in glycolysis and the reverse reaction in gluconeogenesis (PubMed:11724794, PubMed:3170585). Also shows nitrosylase activity, thereby playing a role in nuclear functions (PubMed:11724794, PubMed:3170585). Modulates the organization and assembly of the cytoskeleton (By similarity). Facilitates the CHP1-dependent microtubule and membrane associations through its ability to stimulate the binding of CHP1 to microtubules (By similarity). Component of the GAIT (gamma interferon-activated inhibitor of translation)

complex which mediates interferon-gamma-induced transcript-selective translation inhibition in inflammation processes (PubMed:23071094). Upon interferon-gamma treatment assembles into the GAIT complex which binds to stem loop-containing GAIT elements in the 3'-UTR of diverse inflammatory mRNAs (such as ceruplasmin) and suppresses their translation (PubMed:23071094). Also plays a role in innate immunity by promoting TNF-induced NF-kappa-B activation and type I interferon production, via interaction with TRAF2 and TRAF3, respectively (PubMed:23332158, PubMed:27387501). Participates in nuclear events including transcription, RNA transport, DNA replication and apoptosis (By similarity). 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).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab199553 is the carrier-free version of ab181602.

Patented technology

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?

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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Glyceraldehyde-3-phosphate dehydrogenase commonly known as GAPDH is an enzyme involved in glycolysis. Its molecular weight (MW) is approximately 36 kDa. The protein is expressed ubiquitously in almost all tissues reflecting its essential role in energy production. GAPDH catalyzes the sixth step of glycolysis converting glyceraldehyde-3-phosphate into 13-bisphosphoglycerate. Due to its stable expression researchers often use GAPDH as a loading control in western blot experiments.
Biological function summary	GAPDH serves important metabolic functions beyond its enzymatic role in glycolysis. It functions as part of a multi-enzyme complex within the cytoplasm which facilitates efficient substrate channeling during glycolysis. Additionally GAPDH has non-glycolytic roles including involvement in nuclear processes like RNA export and DNA repair. Its ubiquitous presence across different cellular compartments indicates its multiple functions beyond metabolic pathways.
Pathways	GAPDH integrates into significant cellular functions like the glycolytic pathway and apoptotic pathways. In glycolysis GAPDH collaborates with enzymes like phosphoglycerate kinase forming a cohesive link in the energy conversion chain. Its participation in apoptotic pathways highlights GAPDH's involvement in cellular death processes interacting with proteins like Bcl-2 to influence apoptosis progression. These roles reinforce its presence in central metabolic and regulatory pathways.
Associated diseases and disorders	GAPDH has associations with neurodegenerative diseases and cancer. In neurodegenerative disorders such as Alzheimer's disease GAPDH’s altered enzymatic activity is frequently observed influencing cellular energy homeostasis. Moreover overexpression or aberrant regulation of GAPDH relates to cancer cell proliferation and metastasis implicating proteins like p53 in these pathways. The diverse functions and interactions of GAPDH emphasize its importance in both normal cellular function and disease states.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

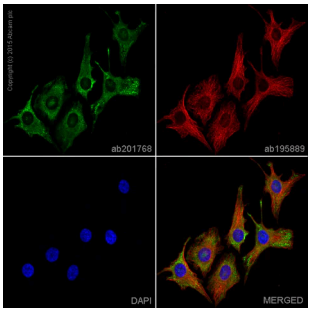
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

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11 product images

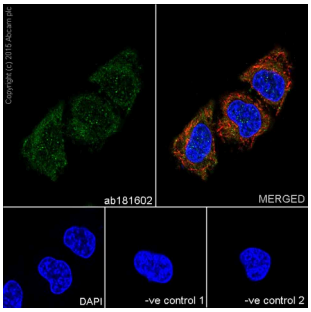


Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-GAPDH antibody

Clone EPR16891 (ab199553) has been successfully conjugated by Abcam. This image was generated using Anti-GAPDH antibody [EPR16891] (Alexa Fluor® 488). Please refer to ab201768 for protocol details.
ab201768 staining GAPDH in HeLa cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab201768 at 1/200 dilution (shown in green) and ab195889, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at 2µg/ml (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

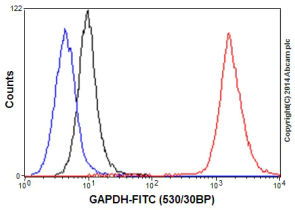
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-GAPDH antibody

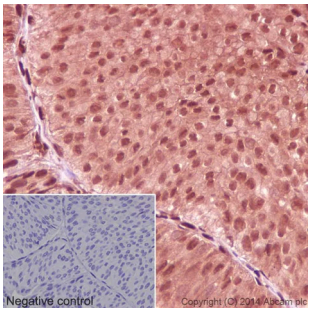
Immunocytochemistry/immunofluorescence staining of 4% paraformaldehyde fixed; 0.1% triton X 100 permeabilized HeLa (human cervix adenocarcinoma) cells labeling GAPDH with ab181602 at dilution of 1/500. The secondary antibody used was Alexa Fluor® 488; goat anti-rabbit IgG (ab150077) at a dilution of 1/400. Nucleus was counter-stained with DAPI (blue). ab7291, a mouse anti-tubulin antibody (1/500) was used to stain tubulin along with ab150120 (AlexaFluor®594 goat anti-mouse secondary, 1/500). The negative controls are shown in the bottom middle and right hand panels- for negative control 1 primary antibody (ab181602; 1/500) and secondary antibody (ab150120; 1/500) was used. For negative control 2 primary antibody (ab7291; 1/500) and secondary antibody (ab150077; 1/400) was used.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).



Flow Cytometry (Intracellular) - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Flow Cytometry (Intracellular) staining using rabbit Anti-GAPDH antibody

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling GAPDH with ab181602 at 1/180 dilution (red) compared with a rabbit monoclonal IgG isotype control (black) and an unlabeled 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.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).



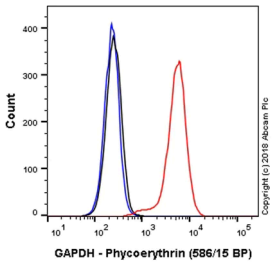
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GAPDH antibody

Immunohistochemical analysis of paraffin-embedded human transitional cell carcinoma of bladder tissue labeling GAPDH with ab181602 at 1/2000 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).

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



Flow Cytometry (Intracellular) - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

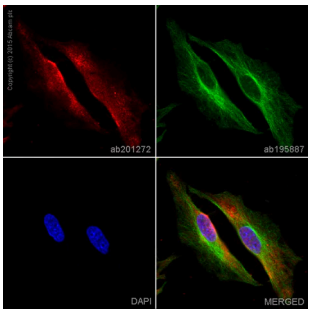
GAPDH Flow Cytometry (Intracellular) staining using rabbit Anti-GAPDH antibody

Clone EPR16891 (ab199553) has been successfully conjugated by Abcam. This image was generated using Anti-GAPDH antibody [EPR16891] (PE). Please refer to ab224004 for protocol details.
Overlay histogram showing HeLa cells stained with ab224004 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab224004, 1/1000 dilution) for 30 min at 22°C.

Isotype control antibody (black line) was Rabbit IgG (monoclonal) Phycoerythrin (ab209478) used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a 50 mW Yellow/Green laser (561nm) and 586/15 bandpass filter.

This antibody gave a positive signal in HeLa cells fixed with 4% formaldehyde (10 min)/permeabilized with 0.1% PBS-Triton X-100 for 15 min used under the same conditions.

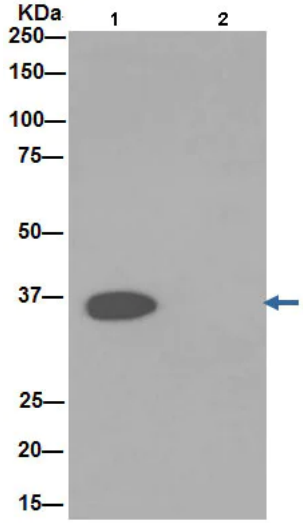


Immunocytochemistry/ Immunofluorescence - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-GAPDH antibody

Clone EPR16891 (ab199553) has been successfully conjugated by Abcam. This image was generated using Anti-GAPDH antibody [EPR16891] (Alexa Fluor® 647). Please refer to ab201272 for protocol details.
ab201272 staining GAPDH in HeLa cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab201272 at 1/100 dilution (shown in red) and ab195887, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at 2µg/ml (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunoprecipitation - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH was immunoprecipitated from 1mg of HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell extract with ab181602 at 1/60 dilution. Western blot was performed from the immunoprecipitate using ab181602 at 1/5000 dilution. Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution. **Lane 1:** HeLa whole cell extract.
Lane 2: PBS instead of HeLa whole cell extract.

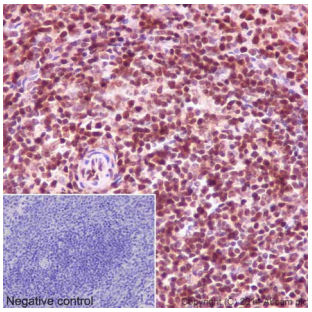
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).

All lanes:
Immunoprecipitation - Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602)

Predicted band size: 36 kDa

Observed band size: 36 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

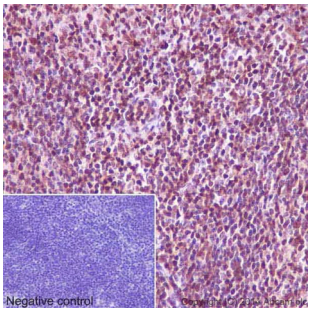
GAPDH Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GAPDH antibody

Immunohistochemical analysis of paraffin-embedded rat spleen tissue labeling GAPDH with ab181602 at 1/2000 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).

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



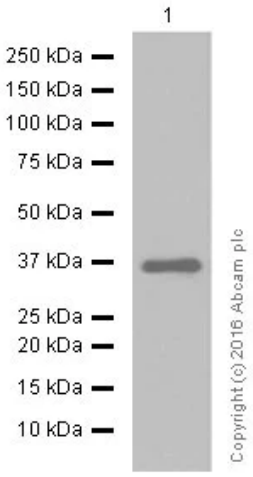
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GAPDH antibody

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling GAPDH with ab181602 at 1/2000 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181602).

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



Western blot - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Western blot staining of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysates using rabbit Anti-GAPDH antibody

Blocking and Diluting buffer and concentration:5% NFDM/TBST
Exposure time:3 seconds

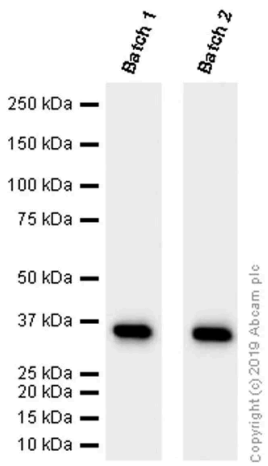
All lanes:
Western blot - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553) at 1/100000 dilution

All lanes:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysates at 15 µg

Secondary

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

Predicted band size: 36 kDa



Western blot - Anti-GAPDH antibody [EPR16891] - BSA and Azide free (ab199553)

GAPDH Western blot staining using rabbit Anti-GAPDH antibody

This data was developed using ab181602, the same antibody clone in a different buffer formulation. Different batches of ab181602 were tested on HeLa (Human cervix adenocarcinoma epithelial cell) lysate at 1.0 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 36 kDa.
All lanes:
Western blot - Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602)

Predicted band size: 36 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.