abcam

Product datasheet

Anti-GFAP antibody [EPR19996] ab207165

Recombinant RabMAb

★★★★★ 2 Abreviews 11 References 14 Images

Overview

Product name	Anti-GFAP antibody [EPR19996]
Description	Rabbit monoclonal [EPR19996] to GFAP
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, IHC-P, WB, IHC-Fr, IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	Flow Cyt: Mouse primary brain cell. ICC/IF: Rat hippocampal mixed glia.
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® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® 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	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR19996
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab207165 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)		1/500.
ICC/IF	★★★★★ (1)	Use a concentration of 0.1 - 1 µg/ml.
IHC-P		1/400. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/5000. Detects a band of approximately 40-54 kDa (predicted molecular weight: 49 kDa).
IHC-Fr	★★★★★ (1)	1/100. Antigen retrieval: Heated citrate solution (10mM citrate pH 6.0 + 0.05% Tween-20)
IP		1/30.

Target

Function GFAP, a class-III intermediate filament, is a cell-specific marker that, during the development of the central nervous system, distinguishes astrocytes from other glial cells.

Tissue specificity Expressed in cells lacking fibronectin.

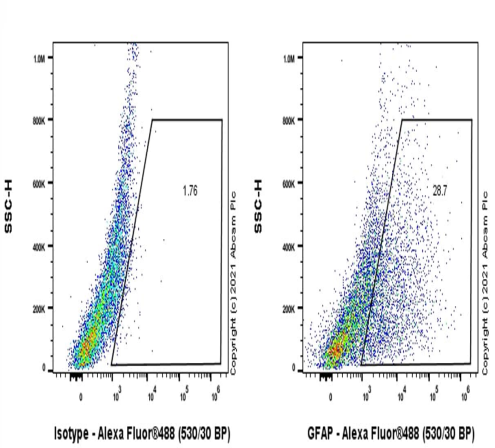
Involvement in disease Defects in GFAP are a cause of Alexander disease (ALEXD) [MIM:203450]. Alexander disease is a rare disorder of the central nervous system. It is a progressive leukoencephalopathy whose hallmark is the widespread accumulation of Rosenthal fibers which are cytoplasmic inclusions in astrocytes. The most common form affects infants and young children, and is characterized by progressive failure of central myelination, usually leading to death usually within the first decade. Infants with Alexander disease develop a leukoencephalopathy with macrocephaly, seizures, and psychomotor retardation. Patients with juvenile or adult forms typically experience ataxia, bulbar signs and spasticity, and a more slowly progressive course.

Sequence similarities Belongs to the intermediate filament family.

Post-translational modifications Phosphorylated by PKN1.

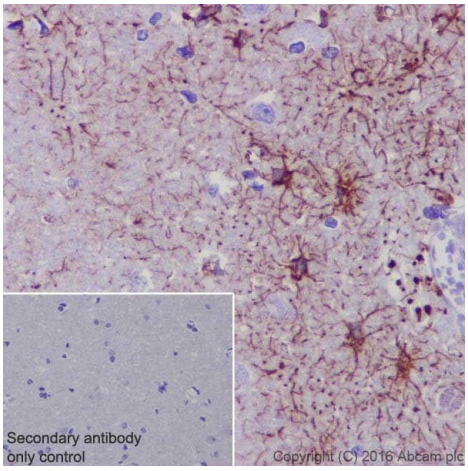
Cellular localization Cytoplasm. Associated with intermediate filaments.

Images



Flow Cytometry (Intracellular) - Anti-GFAP antibody [EPR19996] (ab207165)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Mouse primary brain cells cells labelling GFAP with ab207165 at 1/500 dilution (0.1ug)/ Right compared with a Rabbit monoclonal IgG (**ab172730**) / Left isotype control. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

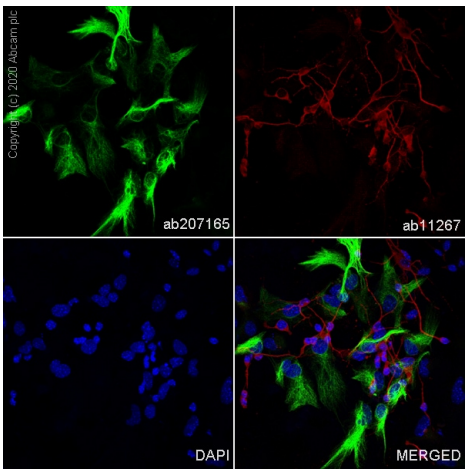


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded human cerebrum tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasmic and membrane staining on glial cells of human cerebrum [PMID: 15378652] is observed. Counter stained with Hematoxylin.

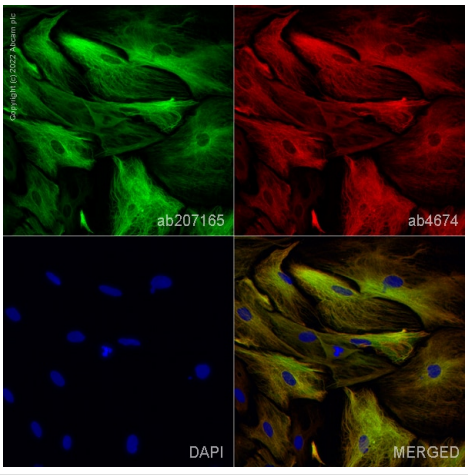
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Immunocytochemistry/ Immunofluorescence - Anti-GFAP antibody [EPR19996] (ab207165)

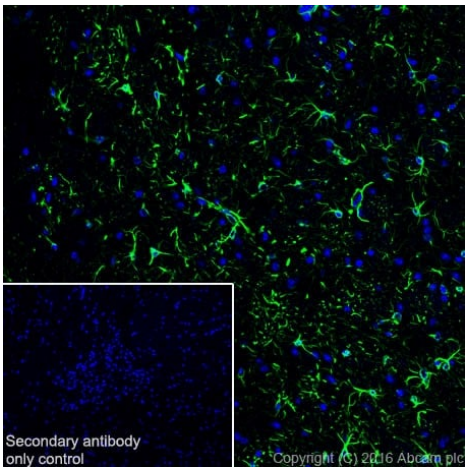
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized mouse primary neural/glia cells labelling GFAP with ab207165 at 1/1000 dilution, followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (2 µg/mL) (Green). Confocal image showing cytoplasmic staining in mouse primary astrocyte. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection. **ab11267** Anti-MAP2 mouse monoclonal antibody was used to counterstain tubulin at 1/500 dilution (4 µg/mL) followed by **ab150120** Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) at 1/1000 dilution (2 µg/mL) (Red). The Nuclear counterstain was DAPI (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-GFAP antibody [EPR19996] (ab207165)

Immunofluorescence staining of GFAP using ab207165 in primary rat hippocampal mixed glia, (prepared from P2 rat hippocampal brain area, obtained from Transnetyx Tissue by BrainBits, LLC, cat.no. SDPHP4m), DIV4. The cells were fixed with 100% MeOH (5 min), permeabilized with 0.1% Triton-X-100 (in PBS) for 5 mins 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 ab207165 at 0.1 µg/ml and **ab4674**, Anti-GFAP antibody, at 1/1000 dilution. Cells were then incubated with **ab150081**, Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and **ab150176**, Goat Anti-Chicken IgY H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Images were acquired with the Perkin Elmer Operetta HCA and a maximum intensity projection of confocal sections is shown. The antibody ab207165 gave comparable results using 4% formaldehyde fixation (10 min).



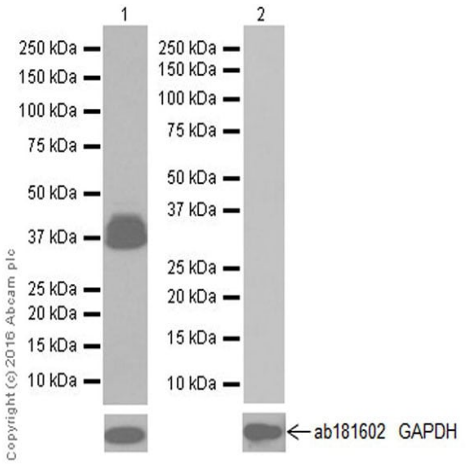
Immunohistochemistry (Frozen sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse cerebrum tissue labeling GFAP with ab207165 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

Cytoplasmic and membrane staining on glial cells of mouse cerebrum is observed.

The nuclear counterstain is DAPI (blue).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution.



Western blot - Anti-GFAP antibody [EPR19996] (ab207165)

Lane 1 : Anti-GFAP antibody [EPR19996] (ab207165) at 1/10000 dilution
Lane 2 : Anti-GFAP antibody [EPR19996] (ab207165) at 1/1000 dilution

Lane 1 : Human brain lysate
Lane 2 : NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

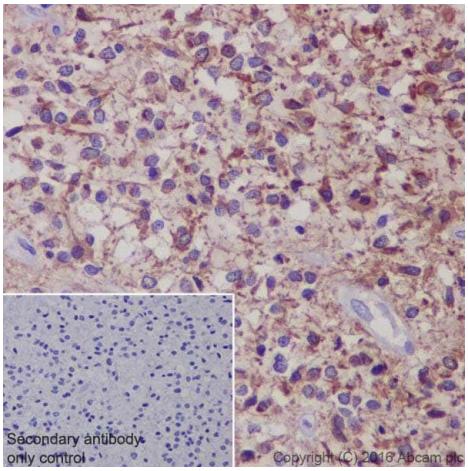
Predicted band size: 49 kDa
Observed band size: 40-54 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with the literature.

Negative control: NIH/3T3 (PMID: 824020; PMID:2294; PMID: 6340792; PMID: 9466565).

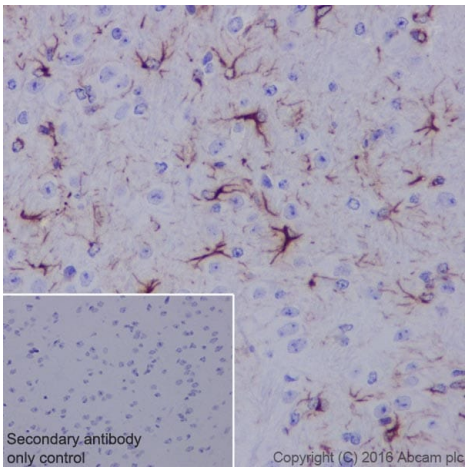


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded human glioma tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasmic staining on tumor cells of human glioma is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

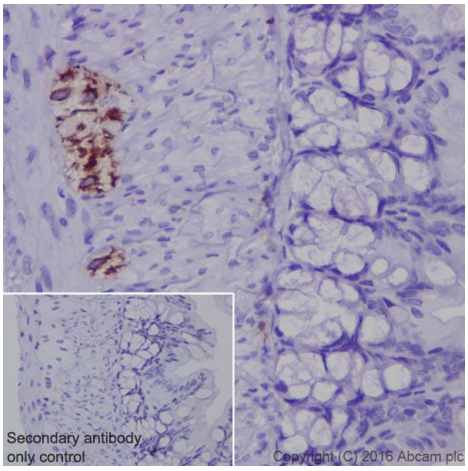


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasmic and membrane staining on glial cells of mouse cerebrum is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

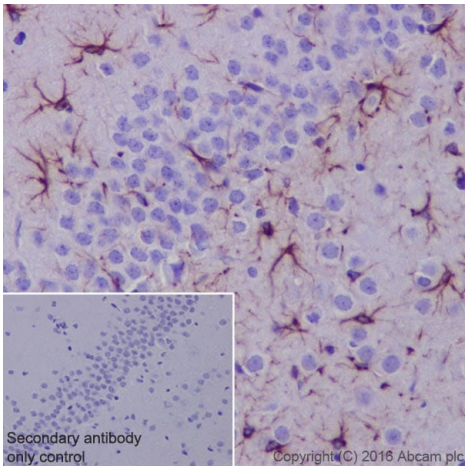


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded mouse colon tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Specific cytoplasmic staining on myenteric nerve plexus, and negative on epithelial cells and smooth muscle cells of mouse colon is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

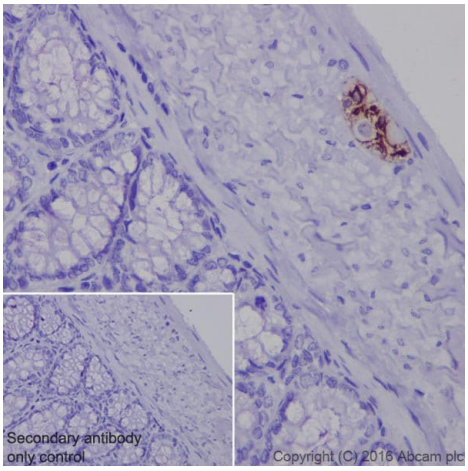


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded rat hippocampus tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasmic and membrane staining on glial cells of rat hippocampus is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

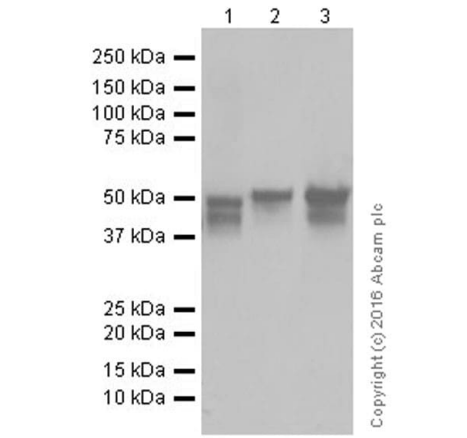


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFAP antibody [EPR19996] (ab207165)

Immunohistochemical analysis of paraffin-embedded rat colon tissue labeling GFAP with ab207165 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Specific cytoplasmic staining on myenteric nerve plexus, and negative on epithelial cells and smooth muscle cells of rat colon is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Western blot - Anti-GFAP antibody [EPR19996] (ab207165)

All lanes : Anti-GFAP antibody [EPR19996] (ab207165) at 1/5000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Rat brain lysate
Lane 3 : Rat cerebellum lysate

Lysates/proteins at 10 µg per lane.

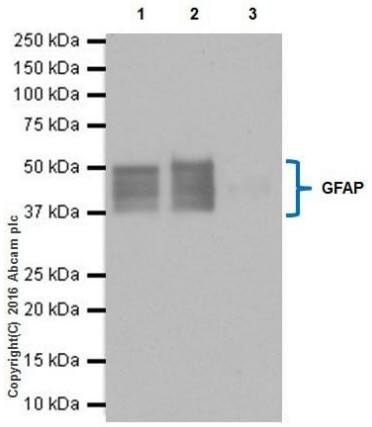
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 49 kDa
Observed band size: 40-54 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with the literature (PMID: 824020; PMID:2294; PMID: 6340792).



Immunoprecipitation - Anti-GFAP antibody [EPR19996] (ab207165)

GFAP was immunoprecipitated from 0.35 mg of human brain lysate with ab207165 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab207165 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

Lane 1: Human brain lysate 10µg (Input).

Lane 2: ab207165 IP in human brain lysate.

Lane 3: Rabbit IgG, monoclonal [EPR25A] - Isotype Control (**ab172730**) instead of ab207165 in human brain lysate.

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

Exposure time: 1 second.

Anti-GFAP antibody [EPR19996] (ab207165)

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