Anti-GFAP Antibody [SA03-04]

ET1601-23



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Cynomolgus monkey, Pig
Applications:	WB, IHC-P, IF-Tissue, IHC-Fr, mIHC, IP
Molecular Wt:	Predicted band size: 50 kDa
Clone number:	SA03-04
Description:	Glial fibrillary acidic protein (GFAP) is a protein that is encoded by the GFAP gene in humans. It is a type III intermediate filament (IF) protein that is expressed by numerous cell types of the central nervous system (CNS), including astrocytes and ependymal cells during development. GFAP has also been found to be expressed in glomeruli and peritubular fibroblasts taken from rat kidneys, Leydig cells of the testis in both hamsters and humans, human keratinocytes, human osteocytes and chondrocytes and stellate cells of the pancreas and liver in rats. GFAP is closely related to the other three non-epithelial type III IF family members, vimentin, desmin and peripherin, which are all involved in the structure and function of the cell's cytoskeleton. GFAP is thought to help to maintain astrocyte mechanical strength as well as the shape of cells, but its exact function remains poorly understood, despite the number of studies using it as a cell marker.
lmmunogen:	Synthetic peptide within Human GFAP aa 1-50 / 432.
Positive control:	Human brain tissue lysate, rat brain tissue lysate, human cerebellum tissue lysate, mouse brain tissue, rat brain tissue, human brain tissue, human glioblastoma tissue, mouse spinal cord tissue, rat spinal cord tissue, mouse cerebral cortex tissue, rat cerebral cortex tissue, mouse cerebellum tissue, rat cerebellum tissue, mouse hippocampus tissue, rat hippocampus tissue.
Subcellular location:	Cytoplasm
Database links:	SwissProt: P14136 Human P03995 Mouse P47819 Rat
Recommended Dilutions:	4.5.000
WB IHC-P	1:5,000
IF-Tissue	1:200-1:500
IHC-Fr	1:200-1:500
mIHC	1:1,000-1:10,000
IP	Use at an assay dependent concentration.
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4° C. Store at $+4^{\circ}$ C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20° C long term.
Purity:	Protein A affinity purified.

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Images









Fig1: Application: IHC-Fr Species: Mouse Site: Cerebral cortex Sample: Frozen section (Fresh) Antibody concentration: 1/200 Antigen retrieval: Not required Fig2: Application: IHC-Fr Species: Rat

Site: Cerebral cortex

Sample: Frozen section

Antibody concentration: 1/500

Antigen retrieval: Not required

Fig3: Application: IF-tissue

Species: Mouse

Site: Cerebellum

Sample: Paraffin-embedded section

Antibody concentration: 1/500

Fig4: Application: IF-tissue

Species: Rat

Site: Cerebral cortex

Sample: Paraffin-embedded section

Antibody concentration: 1/500

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Fig5: Fluorescence multiplex immunohistochemical analysis of mouse hippocampus (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-GFAP (ET1601-23, Green), anti-NeuN (ET1602-12, Red) and anti-c-Fos (HA722666, White) on hippocampus. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in three rounds of staining: in the order of ET1601-23 (1/1,000 dilution), ET1602-12 (1/1,000 dilution) and HA722666 (1/200 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Zeiss Observer 7 Inverted Fluorescence Microscope.



Fig6: Fluorescence multiplex immunohistochemical analysis of mouse brain (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-GFAP (ET1601-23, Green), anti-NeuN (ET1602-12, Red) and anti-c-Fos (HA722666, White) on brain. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immunostaining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in three rounds of staining: in the order of ET1601-23 (1/1,000 dilution), ET1602-12 (1/1,000 dilution) and HA722666 (1/200 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Zeiss Observer 7 Inverted Fluorescence Microscope.

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Fig7: Fluorescence multiplex immunohistochemical analysis of mouse brain (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-MAP2 (HA500177, Red), anti-Olig2 (ET1604-29, Cyan), anti-GFAP (ET1601-23, Magenta) and anti-Neun (ET1602-12, Yellow) on mouse brain. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with Sequential Immuno-staining Kit (IRISKit™MH010101, the www.luminiris.cn). The section was incubated in four rounds of staining: in the order of HA500177 (1/1,000 dilution), ET1604-29 (1/5,000 dilution), ET1601-23 (1/10,000 dilution) and ET1602-12 (1/10,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.



Fig8: Fluorescence multiplex immunohistochemical analysis of mouse hippocampus (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-MAP2 (HA500177, Green), anti-GFAP (ET1601-23, Red) and anti-NeuN (ET1602-12. Magenta) on Mouse hippocampus. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with Sequential Immuno-staining Kit (IRISKit™MH010101, the www.luminiris.cn). The section was incubated in three rounds of staining: in the order of HA500177 (1/1,000 dilution), ET1601-23 (1/1,000 dilution) and ET1602-12 (1/10,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95℃. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

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Fig9: Fluorescence multiplex immunohistochemical analysis of mouse brain (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-NeuN (ET1602-12, red), anti-Iba1 (ET1705-78, green), anti-GFAP (ET1601-23, gray), anti-Olig2 (ET1604-29, cyan), anti-MAP2 (HA500177, magenta) and anti-CD34 (ET1606-11, yellow) on mouse brain. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with Immuno-staining Kit (IRISKit™MH010101, the Sequential www.luminiris.cn). The section was incubated in six rounds of staining: in the order of ET1602-12(1/5,000 dilution), ET1705-78 (1/2,000 dilution), ET1601-23 (1/5,000 dilution), ET1604-29 (1/1,000 dilution), HA500177 (1/5,000 dilution) and ET1606-11 (1/2,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

Fig10: Western blot analysis of GFAP on different lysates with Rabbit anti-GFAP antibody (ET1601-23) at 1/5,000 dilution.

- Lane 1: Human brain tissue lysate
- Lane 2: Rat brain tissue lysate
- Lane 3: Human cerebellum tissue lysate
- Lane 4: Mouse brain tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 50 kDa Observed band size: 50 kDa

Exposure time: 11 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1601-23) at 1/5,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.









Fig11: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig12: Immunohistochemical analysis of paraffin-embedded mouse cerebral cortex tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig13: Immunohistochemical analysis of paraffin-embedded rat cerebral cortex tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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Fig14: Immunohistochemical analysis of paraffin-embedded rat cerebellum tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig15: Immunohistochemical analysis of paraffin-embedded mouse hippocampus tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig16: Immunohistochemical analysis of paraffin-embedded rat hippocampus tissue with Rabbit anti-GFAP antibody (ET1601-23) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1601-23) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Zhang N et al. A self-assembly peptide nanofibrous scaffold reduces inflammatory response and promotes functional recovery in a mouse model of intracerebral hemorrhage. Nanomedicine N/A:N/A (2016).
- 2. Green AL et al. Preclinical antitumor efficacy of selective exportin 1 inhibitors in glioblastoma. Neuro Oncol 17:697-707 (2015).

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