

Certificate of Analysis

Anti-GFAP⁺¹
(rabbit antiserum)
Catalog # 07-650
Lot # 27128

Immunogen: Thyroglobulin-conjugated, synthetic peptide (DRGDAGWRGH) corresponding to the C-terminus generated as a result of alternative splicing of human GFAP.

Specificity: Recognizes GFAP⁺¹, Mr 50kDa.

Species Cross-reactivity: Human.

Formulation: 100µl of rabbit antiserum containing 0.05% sodium azide and 30% glycerol.

Storage and Stability: Stable for 2 years at -20°C from date of shipment.

Handling Recommendations: Upon receipt, and prior to removing the cap, centrifuge the vial and gently mix the solution. Aliquot into microcentrifuge tubes and store at -20°C. **Avoid repeated freeze/thaw cycles, which may damage IgG and affect product performance.** Note: Variability in freezer temperatures below -20°C may cause glycerol-containing solutions to become frozen during storage.

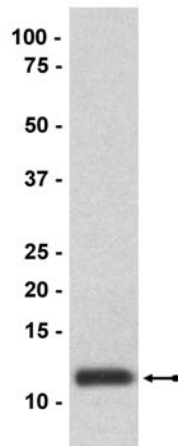
FOR IN VITRO RESEARCH USE ONLY
NOT FOR USE IN HUMANS OR ANIMALS

Quality Control Testing

Immunoblot Analysis: 1:5,000-1:20,000 dilutions of this lot detected 50ng of a recombinant protein comprising the C-terminal 119 amino acids produced by alternative splicing giving rise to the GFAP⁺¹ protein with a molecular weight of approximately 12kDa. The antibody will recognize native GFAP⁺¹ produced in the brains of Alzheimer's disease or Down syndrome patients, with an expected molecular weight of approximately 50kDa.

Additional Research Applications

Immunohistochemistry: A 1:1,000 dilution of this antibody has been reported by an independent laboratory to detect GFAP⁺¹ in human Alzheimer's brain sections.¹



Immunoblot Analysis

Recombinant C-terminal GFAP⁺¹ (~119 amino acids produced by the frameshift) was resolved by electrophoresis, transferred to nitrocellulose and probed with anti- GFAP⁺¹ (1:20,000 dilution). Proteins were visualized using a goat anti-rabbit secondary antibody conjugated to HRP and a chemiluminescence detection system. Arrow indicates GFAP⁺¹ (~12kDa).

Application References:

1. Hol, E.M., *et al.*, *Mol. Psych.* **8**: 786-796, 2003.

General References:

- Li, R., *et al.*, *Int. J. Dev. Neurosci.* **20**: 259-268, 2002.
- Lamers, K.J., *et al.*, *Brain Res. Bull.* **61**: 261-264, 2003.
- Messing, A. and Brenner, M., *Glia* **43**: 87-90, 2003.

Immunoblot Protocol

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on a fusion protein sample and transfer the proteins to nitrocellulose. Wash the blotted nitrocellulose twice with water.
2. Block the blotted nitrocellulose in freshly prepared 5% nonfat dry milk (Catalog # 20-200) in TBS with 0.05% Tween[®]-20 (TBST-MLK) for 30 minutes at room temperature with constant agitation.
3. Incubate the nitrocellulose with a **1:5,000-1:20,000 dilution of anti-GFAP⁺¹**, diluted in freshly prepared TBST-MLK overnight with agitation at 4°C.
4. Wash the nitrocellulose twice with water.
5. Incubate the nitrocellulose in the secondary reagent of choice (a goat anti-rabbit HRP conjugated IgG, Catalog # 12-348, 1:3,000 dilution was used) in TBST-MLK for 1.5 hours with agitation at room temperature.
6. Wash the nitrocellulose twice with water.
7. Wash the nitrocellulose in TBS-0.05% Tween[®]-20 for 3-5 minutes.
8. Rinse the nitrocellulose in 4-5 changes of water.
9. Use detection method of choice (enhanced chemiluminescence was used).