Product datasheet

Anti-BDNF antibody [EPR1292] ab108319

Overview

Product name: Anti-BDNF antibody [EPR1292]
Description: Rabbit monoclonal [EPR1292] to BDNF
Host species: Rabbit
Specificity: This product may cross react with the following family members: NGF beta, neurotrophin 3, neurotrophin 4. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.

Tested applications: Suitable for: Flow Cyt, WB, IHC-P, IHC-Fr, ICC/IF

Species reactivity: Reacts with: Mouse, Rat, Human

Immunogen: Synthetic peptide within Human BDNF aa 150 to the C-terminus. The exact sequence is proprietary.
Database link: P23560
(Peptide available as ab182199)

Positive control: WB: Human, rat and mouse brain, hippocampus and cerebellum lysates; IHC-P: Human brain tissue, human bladder cancer tissue; Coronal sections from fetal (SNC) and post-natal (VTA) WT and Rgs6-/- mice.; ICC/IF: HeLa cells; Flow Cyt: HeLa cells; IHC-Fr: Mouse and Rat cerebrum tissue.

General notes: For BDNF, multiple WB bands are possible and expected. The human protein has 5 isoforms (precursors: 28 – 37 kDa) and can be glycosylated (Uniprot: http://www.uniprot.org/uniprot/P23560). The mature form is expected at ~14 kDa (monomer) and the dimer at ~28 kDa.

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.

We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is
designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.

Properties

Form
Liquid

Storage instructions

Storage buffer
pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity
Protein A purified

Clonality
Monoclonal

Clone number
EPR1292

Isotype
IgG

Applications

Our Abpromise guarantee covers the use of ab108319 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flow Cyt</td>
<td></td>
<td>1/30.</td>
</tr>
<tr>
<td>WB</td>
<td>★★★★★</td>
<td>1/1000 - 1/10000. Predicted molecular weight: 15 kDa. Can be blocked with Human BDNF peptide (ab182199).</td>
</tr>
<tr>
<td>IHC-P</td>
<td>★★★★★</td>
<td>1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.</td>
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<tr>
<td></td>
<td></td>
<td>See IHC antigen retrieval protocols.</td>
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<tr>
<td></td>
<td></td>
<td>Heat up to 98 degrees C, below boiling, and then let cool for 10-20 min.</td>
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<tr>
<td></td>
<td></td>
<td>For unpurified use at 1/100 - 1/250 dilution.</td>
</tr>
<tr>
<td>IHC-Fr</td>
<td></td>
<td>1/100.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>★★★★★</td>
<td>1/500.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>For unpurified use at 1/750 dilution.</td>
</tr>
</tbody>
</table>

Target

Function
During development, promotes the survival and differentiation of selected neuronal populations of
the peripheral and central nervous systems. Participates in axonal growth, pathfinding and in the modulation of dendritic growth and morphology. Major regulator of synaptic transmission and plasticity at adult synapses in many regions of the CNS. The versatility of BDNF is emphasized by its contribution to a range of adaptive neuronal responses including long-term potentiation (LTP), long-term depression (LTD), certain forms of short-term synaptic plasticity, as well as homeostatic regulation of intrinsic neuronal excitability.

**Tissue specificity**
Brain. Highly expressed in hippocampus, amygdala, cerebral cortex and cerebellum. Also expressed in heart, lung, skeletal muscle, testis, prostate and placenta.

**Involvement in disease**
Bulimia nervosa
Congenital central hypoventilation syndrome

**Sequence similarities**
Belongs to the NGF-beta family.

**Post-translational modifications**
The propeptide is N-glycosylated and glycosulfated.
Converted into mature BDNF by plasmin (PLG).

**Cellular localization**
Secreted.

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**Images**

**All lanes**: Anti-BDNF antibody [EPR1292] (ab108319) at 1/1000 dilution (unpurified)

**Lane 1**: Human hippocampus lysate
**Lane 2**: Rat hippocampus lysate
**Lane 3**: Mouse hippocampus lysate

Lysates/proteins at 20 µg per lane.

**Secondary**
All lanes: Gt anti Rb IR680 at 1/10000 dilution

Performed under reducing conditions.

**Predicted band size**: 15 kDa

**Additional bands at**: 15 kDa (possible mature (processed) protein), 28 kDa (possible multimer), 35 kDa, 45 kDa (possible immature (unprocessed)). We are unsure as to the identity of these extra bands.

This blot was produced using a 4-12% Bis-tris gel under the MES buffer system. The gel was run at 200V for 35 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using Licor blocking buffer before being incubated with unpurified ab108319 (1/1000)
overnight at 4°C. Ab8245 (mouse anti-GAPDH; 0.05 µg/mL) was included as a loading control. Antibody binding was detected using goat anti-rabbit IgG IR-680 (green) and goat anti-mouse IgG IR800 (red) at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.

Immunohistochemistry (Frozen sections) analysis of rat cerebral cortex tissue sections labeling BDNF with Purified ab108319 at 1/100 (2.8 µg/ml). Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human bladder cancer tissue sections labeling BDNF with Purified ab108319 at 1:500 dilution (0.56 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0)
Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling BDNF with purified ab108319 at 1:30 dilution (10 µg/ml) (red). Cells were fixed with 80% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1:2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

Immunohistochemistry (Frozen sections) analysis of mouse cerebrum tissue sections labeling BDNF with Purified ab108319 at 1/100 (2.8 µg/ml). Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.
All lanes: Anti-BDNF antibody [EPR1292] (ab108319) at 1/1000 dilution (Purified)

Lane 1: Human brain lysates with 5% NFDM/TBST
Lane 2: Mouse brain lysates with 5% NFDM/TBST
Lane 3: Rat brain lysates with 5% NFDM/TBST
Lane 4: Human hippocampus lysates with 5% NFDM/TBST
Lane 5: Mouse hippocampus lysates with 5% NFDM/TBST
Lane 6: Rat hippocampus lysates with 5% NFDM/TBST
Lane 7: Human cerebellum lysates with 5% NFDM/TBST
Lane 8: Mouse cerebellum lysates with 5% NFDM/TBST
Lane 9: Rat cerebellum lysates with 5% NFDM/TBST

Lysates/proteins at 20 µg per lane.

Secondary
All lanes: Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 15 kDa
Observed band size: 15-45 kDa
why is the actual band size different from the predicted?
Co-immunofluorescence staining against tyrosine hydroxylase (TH; green), nuclear Dapi (blue) and BDNF (red, ab108319) in coronal sections from fetal (SNc) and post-natal (VTA) WT and Rgs6−/− mice. Scale bar 20 µm.

Immunohistochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling BDNF with Purified ab108319 at 1:500 (0.6 µg/ml). Cells were fixed in 100% Methanol and permeabilized with None. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.
Immunohistochemical analysis of paraffin-embedded human brain tissue using unpurified ab108319 at 1/100 dilution.
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Immunocytochemistry/Immunofluorescence analysis of HeLa (human cervix adenocarcinoma) cells labelling BDNF with unpurified ab108319 at a dilution of 1/750. Cells were fixed with paraformaldehyde and permeabilized with 0.5% Triton-X100 in PBS. ab150081 (1/200) was used as the secondary antibody.
The antibody produces a strong, golgi-associated labelling pattern in both PF and MeOH fixed samples.

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