**Product datasheet**

**Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] ab76316**

**Overview**

**Product name**  
Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y]

**Description**  
Rabbit monoclonal [EPR2425Y] to Metabotropic Glutamate Receptor 5

**Host species**  
Rabbit

**Specificity**  
The Human species recommendation is based on the WB results. This antibody has been tested for IHC-P and IHC-Fr in Human samples and we obtain positive signal only in IHC-Fr. We do not recommend this antibody for IHC-P in Human samples.

**Tested applications**  
Suitable for: Electron Microscopy, Flow Cyt (Intra), WB, IHC-P, IHC-Fr  
Unsuitable for: IP

**Species reactivity**  
Reacts with: Mouse, Rat, Human

**Immunogen**  
Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

**Positive control**  
WB: Mouse brain lysate and Human hippocampus tissue. IHC-P: Rat caudate putamen and cerebrum tissue; mouse caudate putamen tissue and cerebrum IHC-Fr: Mouse caudate ptamen tissue. IHC (resin): Mouse neocortex tissue. EM: Mouse neocortex tissue. Flow Cyt (intra): SH-SYSY cells.

**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. Avoid freeze / thaw cycle.
Storage buffer
- pH: 7.20
- Preservative: 0.05% Sodium azide
- Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant

Purity
- Protein A purified

Clonality
- Monoclonal

Clone number
- EPR2425Y

Isotype
- IgG

Applications

**The Abpromise guarantee**
Our Abpromise guarantee covers the use of ab76316 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>Electron Microscopy</td>
<td></td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>Flow Cyt (Intra)</td>
<td></td>
<td>1/10. <a href="#">ab172730</a> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.</td>
</tr>
<tr>
<td>WB</td>
<td></td>
<td>1/5000 - 1/10000. Predicted molecular weight: 132 kDa. Please do not boil the sample before loading to the gel.</td>
</tr>
<tr>
<td>IHC-P</td>
<td>★★★★★ (2)</td>
<td>1/250 - 1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. Use of HRP-conjugated or polymerized HRP secondary antibodies, stronger signals have been found using the polymerized HRP secondary. We only recommend mouse and rat species for IHC-P. We do not guarantee IHC for Human species.</td>
</tr>
<tr>
<td>IHC-Fr</td>
<td>★★★★★ (2)</td>
<td>Use a concentration of 1 µg/ml. This antibody has been tested for IHC-P and IHC-Fr in Human samples and we obtain positive signal only in IHC-Fr. We do not recommend this antibody for IHC-P in Human samples.</td>
</tr>
</tbody>
</table>

**Application notes**
Is unsuitable for IP.

Target

Function
- G-protein coupled receptor for glutamate. Ligand binding causes a conformation change that triggers signaling via guanine nucleotide-binding proteins (G proteins) and modulates the activity of down-stream effectors. Signaling activates a phosphatidylinositol-calcium second messenger system and generates a calcium-activated chloride current. Plays an important role in the regulation of synaptic plasticity and the modulation of the neural network activity.

Sequence similarities
- Belongs to the G-protein coupled receptor 3 family.

Cellular localization
- Cell membrane.
Immunohistochemistry (Frozen sections) - Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316)

IHC image of Metabotropic Glutamate Receptor 5 staining in a section of frozen normal human hippocampus* performed on a Leica BOND™ system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab76316, 1 µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

Western blot - Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316)

All lanes: Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316) at 1/10000 dilution

Lane 1: Human spinal cord tissue lysate was not boiled before loading to the gel
Lane 2: Human hippocampus tissue lysate was not boiled before loading to the gel
Lane 3: Human liver tissue lysate was not boiled before loading to the gel
Lane 4: Human spinal cord tissue lysate was boiled before loading to the gel
Lane 5: Human hippocampus tissue lysate was boiled before loading to the gel
Lane 6: Human liver tissue lysate was boiled before loading to the gel

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:** 132 kDa  
**Observed band size:** 140 kDa  
**Additional bands at:** 280 kDa (possible dimer)

Buffer: 5% NFDM / TBST  
Please do not boil the sample before loading to the gel.

GRM5 is a brain enriched protein, can be observed in most of the brain areas but low in spinal cord (PMID: 12783878).

---

**Western blot - Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316)**

Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316) at 1/10000 dilution + mouse brain lysate at 10 µg

**Secondary**

Goat anti-rabbit-HRP

Developed using the ECL technique.

**Predicted band size:** 132 kDa  
**Observed band size:** 140-150 kDa  
**Additional bands at:** >250 kDa (possible dimer)

---

**Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316)**

Immunohistochemical analysis of Paraffin-embedded mouse cerebrum tissue sections labeling Metabotropic Glutamate Receptor 5 with ab76316 at 1/400 dilution followed by Goat Anti-Rabbit IgG H&L (HRP Polymer) secondary antibody. Sections were counterstained with Hematoxylin. Antigen retrieval was heat mediated using ab93684 (Tris/EDTA buffer, pH 9.0).

Positive staining on mouse cerebrum.
Immunohistochemical analysis of Paraffin-embedded rat cerebrum tissue sections labeling Metabotropic Glutamate Receptor 5 with ab76316 at 1/400 dilution followed by Goat Anti-Rabbit IgG H&L (HRP Polymer) secondary antibody. Sections were counterstained with Hematoxylin. Antigen retrieval was heat mediated using ab93684 (Tris/EDTA buffer, pH 9.0).

Positive staining on rat cerebrum.

ab76316 staining Metabotropic Glutamate Receptor 5 in rat caudate putamen by immunohistochemistry. Tissue was fixed with formaldehyde and citrate-mediated antigen retrieval was performed. Samples were blocked with 1% BSA for 10 minutes at 21°C, before incubation with the primary antibody (1/2000) for 2 hours at 21°C. A biotin conjugated goat anti-rabbit IgG secondary was used at 1/250.
ab76316 staining Metabotropic Glutamate Receptor 5 in mouse caudate putamen/Corpus callosum by immunohistochemistry (frozen sections). Tissue was fixed with formaldehyde and samples were blocked with 1% BSA for 10 minutes at 21°C, before incubation with the primary antibody (1/2000) for 16 hours at 21°C. An alexa fluor® 594 conjugated goat anti-rabbit IgG secondary was used at 1/500.

Postembedding immunogold labeling of mouse neocortex using Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316). The tissue was embedded in Lowicryl HM20 resin. 60 nm sections were then cut and mounted on nickel mesh grids before undergoing antigen retrieval for 15 minutes in 0.01 M citrate buffer, pH 6 at 60°C.

The sections were then blocked in 1% BSA/TBSN pH 7.6 and incubated overnight at room temperature with Anti-Metabotropic Glutamate Receptor 5 antibody [EPR2425Y] (ab76316) at 1:250. Sections were washed twice in TBSN pH 7.6, treated with 1% normal donkey serum/TBSN pH 8.2 for 30 minutes, before being incubated with donkey anti-rabbit IgG-Au 10-20 nm at 1:20 for two hours at room temperature.

Sections were then washed in TBSN pH 8.2, followed by water, before undergoing post-staining with 1% uranyl acetate and Sato’s lead. They were then air dried before being transferred to an oven for 30 minutes at 60°C.

In these images you can see gold immunoparticles on the postsynaptic density of synapses.

(TBSN = 0.02M TRIS buffered saline (0.3 N, pH 7.6 or 8.2) with 0.005% Tergitol NP-10)
Intracellular flow cytometric analysis of permeabilized SH-SY5Y cells using ab76316 (red) at 1/20 or a rabbit IgG (ab172730) as a negative control (green). The cells were permeabilized with 2% PFA and a goat anti-rabbit IgG FITC was used as the secondary at 1/150.

ab76316 staining Metabotropic Glutamate Receptor 5 in mouse caudate putamen/ Corpus callosum by immunohistochemistry. Tissue was fixed with formaldehyde and citrate-mediated antigen retrieval was performed. Samples were blocked with 1% BSA for 10 minutes at 21°C, before incubation with the primary antibody (1/1000) for 2 hours at 21°C. A biotin conjugated goat anti-rabbit IgG secondary was used at 1/250.
Why choose a recombinant antibody?

- Research with confidence
- Consistent and reproducible results
- Long-term and scalable supply
- Recombinant technology
- Success from the first experiment
- Confirmed specificity
- Ethical standards compliant
- Animal-free production

Anti-Metabotropic Glutamate Receptor 5 antibody

[EPR2425Y] (ab76316)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit https://www.abcam.com/abpromise or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors