# abcam

## Product datasheet

## Anti-Gephyrin antibody [EPR12650] - BSA and Azide free ab250503





RabMAb

## 6 Images

#### Overview

**Product name** Anti-Gephyrin antibody [EPR12650] - BSA and Azide free

**Description** Rabbit monoclonal [EPR12650] to Gephyrin - BSA and Azide free

**Host species** Rabbit

Specificity 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: WB, IHC-P

Unsuitable for: ICC/IF

Species reactivity Reacts with: Mouse, Rat, Human

Predicted to work with: Chicken, Cow, Dog, Pig, Zebrafish, Rhesus monkey, Xenopus

tropicalis -

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

Positive control WB: HEK-293, Mouse heart, Fetal brain, SH-SY5Y, Neuro-2a, HAP1, MCF7, U2OS, and C6

lysates; IHC-P: Human kidney and brain tissue.

**General notes** ab250503 is the carrier-free version of ab181382.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for

increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR12650

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromi

Our <u>Abpromise guarantee</u> covers the use of ab250503 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	
WB		1/1000 - 1/5000. Detects a band of approximately 93 kDa (predicted molecular weight: 80 kDa).	
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.  The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.	

**Application notes** Is unsuitable for ICC/IF.

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**Function** Microtubule-associated protein involved in membrane protein-cytoskeleton interactions. It is

thought to anchor the inhibitory glycine receptor (GLYR) to subsynaptic microtubules (By similarity). Catalyzes two steps in the biosynthesis of the molybdenum cofactor. In the first step, molybdopterin is adenylated. Subsequently, molybdate is inserted into adenylated molybdopterin

and AMP is released.

**Pathway** Cofactor biosynthesis; molybdopterin biosynthesis.

**Involvement in disease** Defects in GPHN are the cause of molybdenum cofactor deficiency type C (MOCOD type C)

[MIM:252150]. MOCOD type C is an autosomal recessive disease which leads to the pleiotropic

loss of all molybdoenzyme activities and is characterized by severe neurological damage, neonatal seizures and early childhood death.

Defects in GPHN are a cause of startle disease (STHE) [MIM:149400]; also known as hyperekplexia. STHE is a genetically heterogeneous neurologic disorder characterized by muscular rigidity of central nervous system origin, particularly in the neonatal period, and by an exaggerated startle response to unexpected acoustic or tactile stimuli.

Sequence similarities In the N-terminal section; belongs to the moaB/mog family.

In the C-terminal section; belongs to the moeA family.

Cellular localization Cell junction > synapse. Cell junction > synapse > postsynaptic cell membrane. Cytoplasm >

cytoskeleton. Cytoplasmic face of glycinergic postsynaptic membranes.

#### **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Gephyrin antibody
[EPR12650] - BSA and Azide free (ab250503)

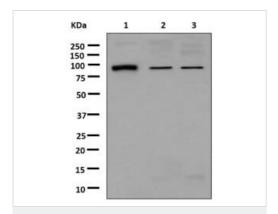
This data was developed using <u>ab181382</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of parafiin embedded human brain tissue using <u>ab181382</u> (unpurified).

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.

Immunohistochemical analysis of parafiin embedded human brain tissue using **ab181382** (unpurified).

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-Gephyrin antibody [EPR12650] - BSA and Azide free (ab250503)

**All lanes :** Anti-Gephyrin antibody [EPR12650] (<u>ab181382</u>) at 1/1000 dilution (Unpurified)

Lane 1: Fetal brain tissue lysate

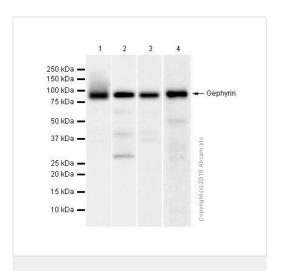
Lane 2: 293T cell lysate

Lane 3: SH-SY5Y cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 80 kDa

This data was developed using <u>ab181382</u>, the same antibody clone in a different buffer formulation.



Western blot - Anti-Gephyrin antibody [EPR12650] - BSA and Azide free (ab250503)

**All lanes :** Anti-Gephyrin antibody [EPR12650] (**ab181382**) at 1/1000 dilution (purified)

**Lane 1 :** HEK-293 (Human embryonic kidney epithelial cell) whole cell lysates

Lane 2: Mouse heart lysates

**Lane 3 :** Neuro-2a (Mouse neuroblastoma neuroblast) whole cell lysates

Lane 4: C6 (Rat glial tumor glial cell) whole cell lysates

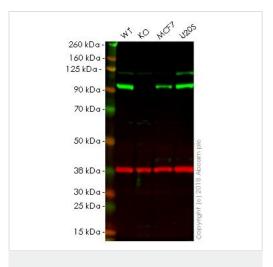
Lysates/proteins at 20 µg per lane.

### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

**Predicted band size:** 80 kDa **Observed band size:** 93 kDa

This data was developed using <u>ab181382</u>, the same antibody clone in a different buffer formulation.



Western blot - Anti-Gephyrin antibody [EPR12650] - BSA and Azide free (ab250503)

**All lanes :** Anti-Gephyrin antibody [EPR12650] ( $\underline{ab181382}$ ) at 1  $\mu g/ml$  (unpurified)

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: GPHN (Gephyrin) knockout HAP1 whole cell lysate

Lane 3: MCF7 whole cell lysate
Lane 4: U2OS whole cell lysate

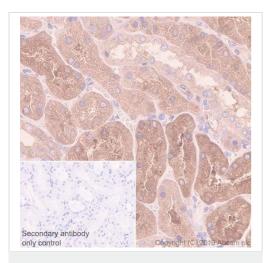
Lysates/proteins at 20 µg per lane.

Predicted band size: 80 kDa

This data was developed using <u>ab181382</u>, the same antibody clone in a different buffer formulation.

**Lanes 1 - 4:** Merged signal (red and green). Green - <u>ab181382</u> observed at 90 kDa. Red - loading control, <u>ab9484</u>, observed at 37 kDa.

ab181382 was shown to recognize Gephyrin in wild-type HAP1 cells as signal was lost at the expected MW in GPHN (Gephyrin) knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and GPHN (Gephyrin) knockout samples were subjected to SDS-PAGE. Ab181382 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1 μg/ml and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Gephyrin antibody

[EPR12650] - BSA and Azide free (ab250503)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue sections labeling Gephyrin with purified <u>ab181382</u> at 1/100 dilution (1.62 µg/ml). Heat mediated antigen retrieval was performed Perform heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab181382)



Azide free (ab250503)

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