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# **Gephyrin**

Cat.No. 147 111; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

## **Data Sheet**

Reconstitution/ Storage	100 μg purified IgG, lyophilized. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 100 μl H <sub>2</sub> O to get a 1mg/ml solution in PBS. Then aliquot and store at -20°C to -80°C until use.  Antibodies should be stored at +4°C when still lyophilized. Do not freeze!  For detailed information, see back of the data sheet.
Applications	WB: 1: 1000 up to 1: 5000 (AP staining) IP: yes ICC: 1: 500 up to 1: 1000 IHC: not recommended IHC-P: not recommended IHC-Fr: 1: 500 ExM: external data (see remarks) ELISA:
Clone	3B11
Subtype	IgG1 (κ light chain)
Immunogen	Recombinant protein corresponding to AA 307 to 735 from rat Gephyrin (UniProt Id: Q03555)
Epitop	AA 326 to 550 from rat Gephyrin (UniProt Id: Q03555)
Reactivity	Reacts with: human (Q9NQX3), rat (Q03555), mouse (Q8BUV3), zebrafish. Other species not tested yet.
Specificity	Detects all splice variants that contain a complete E-domain including the C6 domain. K.O. validated PubMed: 26829712
Remarks	This antibody is highly recommended for Western blot experiments and immunoprecipitation.  IHC-Fr: The following fixatives are possible: 4% formaldehyde/PFA, methanolacetone  ExM: This antibody has been successfully applied and published for this method by customers (see application-specific references).

TO BE USED IN VITRO / FOR RESEARCH ONLY NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

## **Background**

**Gephyrin** is a bifunctional protein which is essential for both synaptic clustering of inhibitory neurotransmitter receptors in the central nervous system and the biosynthesis of the molybdenum cofactor (MoCo) in peripheral tissues. It co-purifies with the inhibitory glycine receptor (GlyR) and is expressed abundantly in all brain areas which contain synapses.

#### Selected References for 147 111

Spinal Cord Neuronal Network Formation in a 3D Printed Reinforced Matrix-A Model System to Study Disease Mechanisms. Fischhaber N, Faber J, Bakirci E, Dalton PD, Budday S, Villmann C, Schaefer N Advanced healthcare materials (2021) 1019: e2100830.. WB, ICC, UPTAKE

TRPM2 and CaMKII Signaling Drives Excessive GABAergic Synaptic Inhibition Following Ischemia.

Burch AM, Garcia JD, O'Leary H, Haas A, Orfila JE, Tiemeier E, Chalmers N, Smith KR, Quillinan N, Herson PS

The Journal of neuroscience: the official journal of the Society for Neuroscience (2024) 4419: . . WB, ICC, IHC; tested species:

mouse

Extracellular signal-regulated kinase and glycogen synthase kinase  $3\beta$  regulate gephyrin postsynaptic aggregation and GABAergic synaptic function in a calpain-dependent mechanism.

 $Tyagarajan \, SK, \, Ghosh \, H, \, Y\acute{e}venes \, GE, \, Imanishi \, SY, \, Zeilhofer \, HU, \, Gerrits \, B, \, Fritschy \, JM \,$ 

The Journal of biological chemistry (2013) 28814: 9634-47. . WB, IP

Glycine Receptor β-Targeting Autoantibodies Contribute to the Pathology of Autoimmune Diseases. Wiessler AL, Talucci I, Piro I, Seefried S, Hörlin V, Baykan BB, Tüzün E, Schaefer N, Maric HM, Sommer C, Villmann C, et al. Neurology(R) neuroimmunology & neuroinflammation (2024) 112: e200187. . WB, ICC; tested species: human,mouse

The TMEM132B-GABAA receptor complex controls alcohol actions in the brain.

Wang G, Peng S, Reyes Mendez M, Keramidas A, Castellano D, Wu K, Han W, Tian Q, Dong L, Li Y, Lu W, et al. Cell (2024) 18723: 6649-6668.e35. . WB, ICC; tested species: mouse

LPS induces microglial activation and GABAergic synaptic deficits in the hippocampus accompanied by prolonged cognitive impairment.

Jung H, Lee D, You H, Lee M, Kim H, Cheong E, Um JW

Scientific reports (2023) 131: 6547. . WB, IHC; tested species: mouse

The complement inhibitor CD59 is required for GABAergic synaptic transmission in the dentate gyrus.

Wen L, Yang X, Wu Z, Fu S, Zhan Y, Chen Z, Bi D, Shen Y

Cell reports (2023) 424: 112349. . WB, IHC; tested species: mouse

Adamtsl3 mediates DCC signaling to selectively promote GABAergic synapse function.

Cramer TML, Pinan-Lucarre B, Cavaccini A, Damilou A, Tsai YC, Bhat MA, Panzanelli P, Rama N, Mehlen P, Benke D, Karayannis T, et al.

Cell reports (2023) 428: 112947...ICC, IHC; tested species: mouse

The Alzheimer susceptibility gene BIN1 induces isoform-dependent neurotoxicity through early endosome defects.

Lambert E, Saha O, Soares Landeira B, Melo de Farias AR, Hermant X, Carrier A, Pelletier A, Gadaut J, Davoine L, Dupont C, Amouvel P. et al.

Acta neuropathologica communications (2022) 101: 4. . ICC, IHC; tested species: rat

A DARPin-based molecular toolset to probe gephyrin and inhibitory synapse biology.

Campbell BFN, Dittmann A, Dreier B, Plückthun A, Tyagarajan SK

eLife (2022) 11:..ICC, IHC; tested species: rat

Cross-talk between GABAergic postsynapse and microglia regulate synapse loss after brain ischemia.

Cramer T, Gill R, Thirouin ZS, Vaas M, Sampath S, Martineau F, Noya SB, Panzanelli P, Sudharshan TJJ, Colameo D, Chang PK, et al.

Science advances (2022) 89: eabj0112. . WB, IHC; tested species: mouse

Access the online factsheet including applicable protocols at https://sysy.com/product/147111 or scan the QR-code.



# FAQ - How should I store my antibody?

# **Shipping Conditions**

 All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freezedried) and are stable in this form without loss of quality at ambient temperatures for several weeks.

# Storage of Sealed Vials after Delivery

- Unlabeled and biotin-labeled antibodies and control proteins should be stored at 4°C before reconstitution. They must not be stored in the freezer when still lyophilized!
   Temperatures below zero may cause loss of performance.
- Fluorescence-labeled antibodies should be reconstituted immediately upon receipt. Long term storage (several months) may lead to aggregation.
- **Control peptides** should be kept at -20°C before reconstitution.

# Long Term Storage after Reconstitution (General Considerations)

- The storage freezer must not be of the frost-free variety ("no-frost freezer"). This cycle
  between freezing and thawing (to reduce frost-build-up), which is exactly what should be
  avoided. For the same reason, antibody vials should be placed in an area of the freezer that
  has minimal temperature fluctuations, for instance towards the back rather than on a door
  shelf.
- Aliquot the antibody and store frozen (-20°C to -80°C). Avoid very small aliquots (below 20 µl)
  and use the smallest storage vial or tube possible. The smaller the aliquot, the more the stock
  concentration is affected by evaporation and adsorption of the antibody to the surface of the
  storage vial or tube. Adsorption of the antibody to the surface leads to a substantial loss of
  activity.
- The addition of glycerol to a final concentration of 50% lowers the freezing point of your stock and keeps your antibody at -20°C in liquid state. This efficiently avoids freeze and thaw cycles.

# **Product Specific Hints for Storage**

# Control proteins / peptides

• Store at -20°C to -80°C.

#### **Monoclonal Antibodies**

- Ascites and hybridoma supernatant should be stored at -20°C up to -80°C. Prolonged storage at 4°C is not recommended! Unlike serum, ascites may contain proteases that will degrade the antibodies.
- **Purified IgG** should be stored at -20°C up to -80°C. Adding a carrier protein like BSA will increase long term stability. Many of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

#### **Polyclonal Antibodies**

- Crude antisera: With anti-microbials added, they may be stored at 4°C. However, frozen storage (-20°C up to -80°C) is preferable.
- Affinity purified antibodies: Less robust than antisera. Storage at -20°C up to -80°C is
  recommended. Adding a carrier protein like BSA will increase long term stability. Most of our
  antibodies already contain carrier proteins. Please refer to the data-sheet for detailed
  information.

#### Fluorescence-labeled Antibodies

• Store as a liquid with 1:1 (v/v) glycerol at -20°C. Protect these antibodies from light exposure.

# Avoid repeated freeze-thaw cycles for all antibodies!

# FAQ - How should I reconstitute my antibody?

#### Reconstitution

- All our purified antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add
  the amount of deionized water given in the respective datasheet. If higher volumes are
  preferred, add water as mentioned above and then the desired amount of PBS and a
  stabilizing carrier protein (e.g. BSA) to a final concentration of 2%. Some of our antibodies
  already contain albumin. Take this into account when adding more carrier protein.
   For complete reconstitution, carefully remove the lid. After adding water, briefly vortex the
  solution. You can spin down the liquid by placing the vial into a 50 ml centrifugation tube filled
  with paper.
- If desired, add small amounts of azide or thimerosal to prevent microbial growth. This is especially recommended if you want to keep an aliquot a 4°C.
- After reconstitution of fluorescence-labeled antibodies, add 1:1 (v/v) glycerol to a final
  concentration of 50%. This lowers the freezing point of your stock and keeps your antibody in
  liquid state at -20°C.
- Glycerol may also be added to unlabeled primary antibodies. It is a suitable way to avoid freezethaw cycles.
- Please refer to our tips and hints for subsequent storage of reconstituted antibodies and control peptides and proteins.