SYSY Antibodies

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Gephyrin

Cat.No. 147 021; Monoclonal mouse antibody, 300 µl hybridoma supernatant (lyophilized)

Data Sheet

Reconstitution/ Storage	300 µl hybridoma supernatant, lyophilized. For **reconstitution** add 300 µl H_2O, then aliquot and store at -20°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**: not recommended (see remarks) **IP**: not recommended (see remarks) **ICC**: 1 : 250 up to 1 : 500 **IHC**: 1 : 100 up to 1 : 250 (see remarks) **IHC-P**: not recommended **IHC-Fr**: 1 : 500 (see remarks) **IHC-G**: yes (see remarks) **ExM**: yes **EM**: yes
Clone	mAb7a
Subtype	IgG1 (κ light chain)
Immunogen	Nativ Protein corresponding to AA 1 to 768 from rat Gephyrin (UniProt Id: Q03555)
Epitop	AA 264 to 276 from rat Gephyrin (UniProt Id: Q03555)
Reactivity	Reacts with: human (Q9NQX3), rat (Q03555), mouse (Q8BUV3), pig, goldfish, zebrafish, frog. Other species not tested yet.
Specificity	Specific for the brain specific 93 kDa splice variant phosphorylated at Ser-270. K.O. validated
Remarks	**WB**: Clone 3B11 (cat. no. 147 111) is highly recommended. **IP**: Clone 3B11 (cat. no. 147 111) highly recommended. **IHC**: Hybridoma supernatant highly recommended. For best results use the protocol of Schneider Gasser et al., 2006. Alternatively antigen retrieval (10 mM citrate, pH 6.0, over night at 60°C) can be applied. Glyoxal fixation according to Konno et al. 2023 has improved signal quality and strength. **IHC-Fr**: Fixation with acetone or PFA/formaldehyde is recommended. Postfixation with methanol is not advised. **IHC-G**: Fixation with 9% glyoxal, 8% acetic acid in ddH2O according to Konno et al. 2023 are recommended.

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 147 021

Molecular Dissection of Neuroligin 2 and Slitrk3 Reveals an Essential Framework for GABAergic Synapse Development.
Li J, Han W, Pelkey KA, Duan J, Mao X, Wang YX, Craig MT, Dong L, Petralia RS, McBain CJ, Lu W, et al.
Neuron (2017) 964: 808-826.e8. . **WB, ICC, IHC; tested species: mouse**

A protocol for concurrent high-quality immunohistochemical and biochemical analyses in adult mouse central nervous system.
Notter T, Panzanelli P, Pfister S, Mircsof D, Fritschy JM
The European journal of neuroscience (2014) 392: 165-75. . **IHC, EM**

Interference With Complex IV as a Model of Age-Related Decline in Synaptic Connectivity.
Kriebel M, Ebel J, Battke F, Griesbach S, Volkmer H
Frontiers in molecular neuroscience (2020) 13: 43. . **WB, ICC; tested species: rat**

Expression of Neurofilament Subunits at Neocortical Glutamatergic and GABAergic Synapses.
Bragina L, Conti F
Frontiers in neuroanatomy (2018) 12: 74. . **WB, IHC; tested species: rat**

Extrasynaptic homomeric glycine receptors in neurons of the rat trigeminal mesencephalic nucleus.
Bae JY, Lee JS, Ko SJ, Cho YS, Rah JC, Cho HJ, Park MJ, Bae YC
Brain structure & function (2018) : . . **IHC, EM; tested species: rat**

Electron tomography on γ-aminobutyric acid-ergic synapses reveals a discontinuous postsynaptic network of filaments.
Linsalata AE, Chen X, Winters CA, Reese TS
The Journal of comparative neurology (2014) 5224: 921-36. . **EM, ICC; tested species: rat**

Spatial proteomics in neurons at single-protein resolution.
Unterauer EM, Shetab Boushehri S, Jevdokimenko K, Masullo LA, Ganji M, Sograte-Idrissi S, Kowalewski R, Strauss S, Reinhardt SCM, Perovic A, Marr C, et al.
Cell (2024) 1877: 1785-1800.e16. . **DNA_PAINT; tested species: rat**

Structural Heterogeneity of the GABAergic Tripartite Synapse.
Brunskine C, Passlick S, Henneberger C
Cells (2022) 1119: . . **EXM; tested species: mouse**

iPSC-derived models of PACS1 syndrome reveal transcriptional and functional deficits in neuron activity.
Rylaarsdam L, Rakotomamonjy J, Pope E, Guemez-Gamboa A
Nature communications (2024) 151: 827. . **IHC; tested species: human**

Synaptic and dendritic architecture of different types of hippocampal somatostatin interneurons.
Takács V, Bardóczi Z, Orosz Á, Major A, Tar L, Berki P, Papp P, Mayer MI, Sebők H, Zsolt L, Sos KE, et al.
PLoS biology (2024) 223: e3002539. . **IHC; tested species: mouse**

Disruption of the autism-associated Pcdh9 gene leads to transcriptional alterations, synapse overgrowth, and defective network activity in the CA1.
Miozzo F, Murru L, Maiellano G, di Iasio I, Zippo AG, Zambrano Avendano A, Metodieva VD, Riccardi S, D'Aliberti D, Spinelli S, Canu T, et al.
The Journal of neuroscience : the official journal of the Society for Neuroscience (2024) 4450: . . **ICC; tested species: rat**

Multimodal sensory control of motor performance by glycinergic interneurons of the mouse spinal cord deep dorsal horn.
Gradwell MA, Ozeri-Engelhard N, Eisdorfer JT, Laflamme OD, Gonzalez M, Upadhyay A, Medlock L, Shrier T, Patel KR, Aoki A, Gandhi M, et al.
Neuron (2024) 1128: 1302-1327.e13. . **IHC; tested species: mouse**

Access the online factsheet including applicable protocols at https://sysy.com/product/147021 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 freeze-dried) 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 freeze-thaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.