

## Amphiphysin

Cat.No. 120 002; Polyclonal rabbit antibody, 200 µl antiserum (lyophilized)

### Data Sheet

|                         |   |
|-------------------------|---|
| Reconstitution/ Storage | 200 µl antiserum, lyophilized. For <b>reconstitution</b> add 200 µl H <sub>2</sub> O, then aliquot and store at -20°C until use.<br>Antibodies should be stored at +4°C when still lyophilized. Do not freeze!<br>For detailed information, see back of the data sheet. |
| Applications            | <b>WB:</b> 1 : 1000 (AP staining)<br><b>IP:</b> yes<br><b>ICC:</b> 1 : 500<br><b>IHC:</b> 1 : 400<br><b>IHC-P:</b> not tested yet   |
| Immunogen               | Synthetic peptide corresponding to AA 2 to 15 from rat Amphiphysin (UniProt Id: O08838)   |
| Reactivity              | Reacts with: human (P49418), rat (O08838), mouse (Q7TQF7), hamster, cow, chicken.<br>Other species not tested yet.  |
| Matching control        | 120-0P  |

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

### Background

**Amphiphysin**, a dominant autoantigen in paraneoplastic Stiff-man syndrome, is a neuronal protein highly concentrated in nerve terminals. It is an abundant presynaptic protein that interacts via its COOH-terminal src homology 3 (SH3) domain with the GTPase dynamin and the inositol-5-phosphatase synaptojanin and additionally directly to clathrin. Amphiphysin, dynamin and synaptojanin have a putative role in synaptic vesicle recycling and undergo rapid dephosphorylation in synaptosomes stimulated by a depolarizing stimulus. Two isoforms are known which seem to act in concert as a heterodimer.

### Selected References for 120 002

- Composition of isolated synaptic boutons reveals the amounts of vesicle trafficking proteins. Wilhelm BG, Mandad S, Truckenbrodt S, Kröhnert K, Schäfer C, Rammner B, Koo SJ, Claßen GA, Krauss M, Haucke V, Urlaub H, et al. *Science (New York, N.Y.)* (2014) 3446187: 1023-8. . **WB, ICC, IHC; tested species: mouse, rat**
- Evidence for a Clathrin-independent mode of endocytosis at a continuously active sensory synapse. Fuchs M, Brandstätter JH, Regus-Leidig H *Frontiers in cellular neuroscience* (2014) 8: 60. . **IHC, EM; tested species: rat**
- Trafficking proteins show limited differences in mobility across different postsynaptic spines. Mougios N, Opazo F, Rizzoli SO, Reshetniak S *iScience* (2023) 262: 105971. . **ICC; tested species: rat**
- Loss of the parkinsonism-associated protein FBXO7 in glutamatergic forebrain neurons in mice leads to abnormal motor behavior and synaptic defects. Wang J, Joseph S, Vingill S, Dere E, Tatenhorst L, Ronnenberg A, Lingor P, Preisinger C, Ehrenreich H, Schulz JB, Stegmüller J, et al. *Journal of neurochemistry* (2023) : . . **IHC; tested species: mouse**
- Mutations in Parkinsonism-linked endocytic proteins synaptojanin1 and auxilin have synergistic effects on dopaminergic axonal pathology. Ng XY, Wu Y, Lin Y, Yaqoob SM, Greene LE, De Camilli P, Cao M *NPJ Parkinson's disease* (2023) 91: 26. . **ICC; tested species: mouse**
- A novel synaptopathy-defective synaptic vesicle protein trafficking in the mutant CHMP2B mouse model of frontotemporal dementia. Clayton EL, Bonnycastle K, Isaacs AM, Cousin MA, Schorge S *Journal of neurochemistry* (2022) 1603: 412-425. . **WB; tested species: mouse**
- Modes and regulation of endocytic membrane retrieval in mouse auditory hair cells. Neef J, Jung S, Wong AB, Reuter K, Pangrsic T, Chakrabarti R, Kügler S, Lenz C, Nouvian R, Boumil RM, Frankel WN, et al. *The Journal of neuroscience : the official journal of the Society for Neuroscience* (2014) 343: 705-16. . **IHC; tested species: mouse**
- Point mutation in syntaxin-1A causes abnormal vesicle recycling, behaviors, and short term plasticity. Watanabe Y, Katayama N, Takeuchi K, Togano T, Itoh R, Sato M, Yamazaki M, Abe M, Sato T, Oda K, Yokoyama M, et al. *The Journal of biological chemistry* (2013) 28848: 34906-19. . **WB; tested species: mouse**
- Role of the clathrin terminal domain in regulating coated pit dynamics revealed by small molecule inhibition. von Kleist L, Stahlschmidt W, Bulut H, Gromova K, Puchkov D, Robertson MJ, MacGregor KA, Tomilin N, Tomlin N, Pechstein A, Chau N, et al. *Cell* (2011) 1463: 471-84. . **ICC; tested species: mouse**
- A pathologic cascade leading to synaptic dysfunction in alpha-synuclein-induced neurodegeneration. Scott DA, Tabarean I, Tang Y, Cartier A, Masliah E, Roy S *The Journal of neuroscience : the official journal of the Society for Neuroscience* (2010) 3024: 8083-95. . **ICC**

Access the online factsheet including applicable protocols at <https://sysy.com/product/120002> 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.