

Amphiphysin

Cat.No. 120 003; Polyclonal rabbit antibody, 50 µg specific antibody (lyophilized)

Data Sheet

Reconstitution/ Storage	50 µg specific antibody, lyophilized. Affinity purified with the immunogen. Albumin was added for stabilization. For reconstitution add 50 µl H ₂ 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 (AP staining) IP: not tested yet ICC: 1 : 500 IHC: not tested yet IHC-P: 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. Other species not tested yet.
Specificity	K.D. validated PubMed: 37243578
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 120 003

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**

The phospho-regulated amphiphysin/endophilin interaction is required for synaptic vesicle endocytosis.
Kontaxi C, Kim N, Cousin MA
Journal of neurochemistry (2023) 1662: 248-264. . **ICC; KD verified; tested species: mouse**

Selected General References

Accessory factors in clathrin-dependent synaptic vesicle endocytosis.
Slepnev VI, De Camilli P
Nature reviews. Neuroscience (2000) 13: 161-72. .

The structural era of endocytosis.
Marsh M, McMahon HT
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The amphiphysin family of proteins and their role in endocytosis at the synapse.
Wigge P, McMahon HT
Trends in neurosciences (1998) 218: 339-44. .

Amphiphysin heterodimers: potential role in clathrin-mediated endocytosis.
Wigge P, Köhler K, Vallis Y, Doyle CA, Owen D, Hunt SP, McMahon HT
Molecular biology of the cell (1997) 810: 2003-15. .

A role of amphiphysin in synaptic vesicle endocytosis suggested by its binding to dynamin in nerve terminals.
David C, McPherson PS, Mundigl O, de Camilli P
Proceedings of the National Academy of Sciences of the United States of America (1996) 931: 331-5. .

The synaptic vesicle cycle: a cascade of protein-protein interactions.
Südhof TC
Nature (1995) 3756533: 645-53. .

Synaptic vesicles and exocytosis.
Jahn R, Südhof TC
Annual review of neuroscience (1994) 17: 219-46. .

The synaptic vesicle-associated protein amphiphysin is the 128-kD autoantigen of Stiff-Man syndrome with breast cancer.
De Camilli P, Thomas A, Cofield R, Folli F, Lichte B, Piccolo G, Meinck HM, Austoni M, Fassetta G, Bottazzo G, Bates D, et al.
The Journal of experimental medicine (1993) 1786: 2219-23. .

Amphiphysin, a novel protein associated with synaptic vesicles.
Lichte B, Veh RW, Meyer HE, Kilimann MW
The EMBO journal (1992) 117: 2521-30. .

Access the online factsheet including applicable protocols at <https://sysy.com/product/120003> 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.