

## Endophilin1

Cat.No. 159 004; Polyclonal Guinea pig antibody, 100 µl antiserum (lyophilized)

### Data Sheet

Reconstitution/ Storage	100 µl antiserum, lyophilized. For <b>reconstitution</b> add 100 µl H <sub>2</sub> O, 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	<b>WB:</b> 1 : 1000 (AP staining) <b>IP:</b> yes <b>ICC:</b> 1 : 500 <b>IHC:</b> 1 : 500 <b>IHC-P:</b> 1 : 500
Immunogen	Synthetic peptide corresponding to AA 256 to 276 from mouse Endophilin1 (UniProt Id: Q62420)
Reactivity	Reacts with: rat, mouse. Other species not tested yet.
Matching control	159-0P

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

### Background

Endophilins are SH3 domain proteins involved in endocytosis. Both, **Endophilin 1** and **2** have been shown to play important roles in clathrin mediated synaptic vesicle recycling. They recruit and stabilize the polyphosphoinositide phosphatase synaptojanin at nerve terminals. The divergent C-terminal tail of VgluT1 has been reported to be a binding partner of Endophilin A1. In contrast to Endophilin 1 that shows a brain specific expression, Endophilin 2 is abundantly expressed in different tissues.

### Selected References for 159 004

Compartmentalization of soluble endocytic proteins in synaptic vesicle clusters by phase separation. Yoshida T, Takenaka KI, Sakamoto H, Kojima Y, Sakano T, Shibayama K, Nakamura K, Hanawa-Suetsugu K, Mori Y, Hirabayashi Y, Hirose K, et al. iScience (2023) 266: 106826. . **WB, ICC; tested species: mouse**

### Selected General References

Interaction between the vesicular glutamate transporter type 1 and endophilin A1, a protein essential for endocytosis. Vinatier J et al. J. Neurochem. (2006) PubMed:16606361

Endophilin is required for synaptic vesicle endocytosis by localizing synaptojanin. Schuske KR et al. Neuron (2003) PubMed:14622579

Synaptojanin is recruited by endophilin to promote synaptic vesicle uncoating. Verstreken P et al. Neuron (2003) PubMed:14622578

Endophilin and synaptojanin hook up to promote synaptic vesicle endocytosis. Song W et al. Neuron (2003) PubMed:14622570

Formation of an endophilin-Ca<sup>2+</sup> channel complex is critical for clathrin-mediated synaptic vesicle endocytosis. Chen Y et al. Cell (2003) PubMed:14532001

Endophilin-1: a multifunctional protein. Reutens AT et al. Int. J. Biochem. Cell Biol. (2002) PubMed:12127567

Endophilin mutations block clathrin-mediated endocytosis but not neurotransmitter release. Verstreken P et al. Cell (2002) PubMed:11955450

Differential expression of endophilin 1 and 2 dimers at central nervous system synapses. Ringstad N et al. J. Biol. Chem. (2001) PubMed:11518713

Fission and uncoating of synaptic clathrin-coated vesicles are perturbed by disruption of interactions with the SH3 domain of endophilin. Gad H et al. Neuron (2000) PubMed:10985350

Endophilin/SH3p4 is required for the transition from early to late stages in clathrin-mediated synaptic vesicle endocytosis. Ringstad N et al. Neuron (1999) PubMed:10677033

Endophilin I mediates synaptic vesicle formation by transfer of arachidonate to lysophosphatidic acid. Schmidt A et al. Nature (1999) PubMed:10490020

Synaptojanin forms two separate complexes in the nerve terminal. Interactions with endophilin and amphiphysin. Micheva KD et al. J. Biol. Chem. (1997) PubMed:9341169

SH3 domain-dependent interactions of endophilin with amphiphysin. Micheva KD et al. FEBS Lett. (1997) PubMed:9315708

The SH3p4/Sh3p8/SH3p13 protein family: binding partners for synaptojanin and dynamin via a Grb2-like Src homology 3 domain. Ringstad N et al. Proc. Natl. Acad. Sci. U.S.A. (1997) PubMed:9238017

Access the online factsheet including applicable protocols at <https://sysy.com/product/159004> 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 at 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.