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Munc13-1

Cat.No. 126 103; Polyclonal rabbit antibody, 50 µg specific antibody (lyophilized)

Data Sheet

Reconstitution/ Storage	50 μg specific antibody, lyophilized. Affinity purified with the immunogen. Albumin and azide were 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: yes ICC: 1: 500 up to 1: 1000 IHC: 1: 200 IHC-P: not tested yet ExM: external data (see remarks) DNA-PAINT: external data EM: ELISA:
Immunogen	Recombinant protein corresponding to AA 3 to 317 from rat Munc13-1 (UniProt Id: Q62768)
Reactivity	Reacts with: human (Q9UPW8), rat (Q62768), mouse (Q4KUS2), zebrafish. Other species not tested yet.
Specificity	K.O. validated PubMed: <u>28772123</u>
Remarks	 IP: Denaturing IP-protocol is recommended. Protein-protein interactions may be affected. IHC: Antigen retrieval with citrate buffer pH 6 is required. ExM: This antibody has been successfully used for super resolution expansion microscopy according to (Eilts et al. 20223).

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

Background

Munc13s are homologues of the C. elegans unc-13 gene product. Three brain-specific isoforms, Munc 13-1, -13-2, and -13-3, are expressed in rat, where they localize to presynaptic terminals. All three isoforms share multiple regulatory domains that may mediate phorbol ester and diacylglycerol binding.

Munc13-1 shows the broadest expression pattern and is found in cortex, cerebellum, olfactory bulb, and hippocampus. Munc13-2 is mainly expressed in cortex and hippocampus, whereas **Munc13-3** exhibits highest expression levels in cerebellum and pons. Munc13-1 interacts directly with a putative coiled-coil domain in the N-terminal part of syntaxin and is involved in synaptic vesicle priming. For Munc13-2 an additional ubiquitously expressed N-terminal splice variant (ubMunc13-2) has been described.

Munc13-3 has been shown to be involved in the regulation of cerebellar synaptic transmission and motor learning.

Selected References for 126 103

Non-additive potentiation of glutamate release by phorbol esters and metabotropic mGlu7 receptor in cerebrocortical nerve terminals.

Martín R, Bartolomé-Martín D, Torres M, Sánchez-Prieto J

Journal of neurochemistry (2011) 1164: 476-85.. ICC, WB; tested species: mouse

Molecular definition of distinct active zone protein machineries for Ca2+ channel clustering and synaptic vesicle priming. Emperador-Melero J, Andersen JW, Metzbower SR, Levy AD, Dharmasri PA, de Nola G, Blanpied TA, Kaeser PS bioRxiv: the preprint server for biology (2023):..ICC, DNA_PAINT; tested species: mouse

Variability in the Munc13-1 content of excitatory release sites. Karlocai MR, Heredi J, Benedek T, Holderith N, Lorincz A, Nusser Z

eLife (2021) 10:.. IHC, EM; tested species: mouse

Munc13-1 is a Ca2+-phospholipid-dependent vesicle priming hub that shapes synaptic short-term plasticity and enables sustained neurotransmission.

Lipstein N, Chang S, Lin KH, López-Murcia FJ, Neher E, Taschenberger H, Brose N

Neuron (2021):.. WB, IHC; tested species: mouse

Distinct active zone protein machineries mediate Ca2+ channel clustering and vesicle priming at hippocampal synapses. Emperador-Melero J, Andersen JW, Metzbower SR, Levy AD, Dharmasri PA, de Nola G, Blanpied TA, Kaeser PS Nature neuroscience (2024) 279: 1680-1694.. **DNA PAINT, ICC; tested species: mouse**

Loss of postsynaptic NMDARs drives nanoscale reorganization of Munc13-1 and PSD-95.

Dharmasri PA, DeMarco EM, Anderson MC, Levy AD, Blanpied TA

bioRxiv: the preprint server for biology (2024):..ICC, DNA_PAINT; tested species: rat

 $\label{liprin-a} \mbox{Liprin-a proteins are master regulators of human presynapse assembly.}$

Marcó de la Cruz B, Campos J, Molinaro A, Xie X, Jin G, Wei Z, Acuna C, Sterky FH

Nature neuroscience (2024) : . . WB, ICC; tested species: human

Munc13 supports fusogenicity of non-docked vesicles at synapses with disrupted active zones.

Tan C, de Nola G, Qiao C, Imig C, Born RT, Brose N, Kaeser PS

eLife (2022) 11:.. WB, ICC; KO verified; tested species: mouse

Bassoon controls synaptic vesicle release via regulation of presynaptic phosphorylation and cAMP.

Montenegro-Venegas C, Guhathakurta D, Pina-Fernandez E, Andres-Alonso M, Plattner F, Gundelfinger ED, Fejtova A EMBO reports (2022) 238: e53659. . WB, ICC; tested species: mouse

An active vesicle priming machinery suppresses axon regeneration upon adult CNS injury.

Hilton BJ, Husch A, Schaffran B, Lin TC, Burnside ER, Dupraz S, Schelski M, Kim J, Müller JA, Schoch S, Imig C, et al.

Neuron (2021):.. WB, ICC; KO verified; tested species: mouse

Access the online factsheet including applicable protocols at https://sysy.com/product/126103 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.