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## SNAP25

Cat.No. 111 111; Monoclonal mouse antibody, 50 µg purified IgG (lyophilized)

## Data Sheet

Reconstitution/ Storage	50 $\mu$ g purified IgG, lyophilized. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 50 $\mu$ l H <sub>2</sub> 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 : 10000 (AP staining) IP: yes (see remarks) ICC: 1 : 100 up to 1 : 500 IHC: 1 : 200 IHC-P: 1 : 1000 ELISA: yes
Clone	71.2
Subtype	IgG1 (κ light chain)
Immunogen	Full-length recombinant rat SNAP25B protein (UniProt Id: P60881-1)
Epitop	AA 1 to 20 from rat SNAP25B (UniProt Id: P60881-1)
Reactivity	Reacts with: human (P60880), rat (P60881), mouse (P60879), mammals, bovine. No signal: zebrafish. Other species not tested yet.
Specificity	Detects both splice variants SNAP 25A and B. Recognizes the Botulinum neurotoxin A and E cleavage products.
Remarks	<ul> <li>IP: Immunoprecipitation not quantitative, appears to depend on the binding status of the protein.</li> <li>ELISA: The ELISA-protocol for membrane proteins is required.</li> <li>Suitable as capture antibody for sandwich-ELISA. Please refer to the protocol for suitable detector antibodies.</li> </ul>

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

### Background

**SNAP25** (synaptosome-associated protein of 25 kD) is a highly conserved protein anchored to the cytosolic face of membranes via palmitoyl side chains in the middle of the molecule. SNAP25 is the target of Botulinum neurotoxins A and E which cleave off 9 and 26 amino acids, respectively, from the C-terminus.

SNAP25 is part of the exocytotic fusion complex (v-SNARE) of neurons where it assembles with syntaxin1 and synaptobrevin. It is abundantly localized on the neuronal plasmalemma and on recycling vesicles including synaptic vesicles. It is also expressed in neuroendocrine cells. There are two splice-variants, SNAP25A and 25B.

## Selected References for 111 111

Aberrant function and structure of retinal ribbon synapses in the absence of complexin 3 and complexin 4. Reim K, Regus-Leidig H, Ammermüller J, El-Kordi A, Radyushkin K, Ehrenreich H, Brandstätter JH, Brose N Journal of cell science (2009) 122Pt 9: 1352-61. **WB, IHC; tested species: mouse** 

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

Positively charged amino acids at the SNAP-25 C terminus determine fusion rates, fusion pore properties, and energetics of tight SNARE complex zippering. Fang Q, Zhao Y, Herbst AD, Kim BN, Lindau M The Journal of neuroscience : the official journal of the Society for Neuroscience (2015) 357: 3230-9. . **ICC; tested species: cow** 

Synapsin-dependent reserve pool of synaptic vesicles supports replenishment of the readily releasable pool under intense synaptic transmission. Vasileva M, Horstmann H, Geumann C, Gitler D, Kuner T The European journal of neuroscience (2012) 368: 3005-20. . **ELISA** 

The N-ethylmaleimide-sensitive fusion protein and alpha-SNAP induce a conformational change in syntaxin. Hanson PI, Otto H, Barton N, Jahn R The Journal of biological chemistry (1995) 27028: 16955-61. . **IP** 

Catecholaminergic dysfunction drives postural and locomotor deficits in a mouse model of spinal muscular atrophy. Pagiazitis JG, Delestrée N, Sowoidnich L, Sivakumar N, Simon CM, Chatzisotiriou A, Albani M, Mentis GZ Cell reports (2025) 441: 115147. . **IHC; tested species: mouse** 

Monitoring of activity-driven trafficking of endogenous synaptic proteins through proximity labeling. Pascual-Caro C, de Juan-Sanz J PLoS biology (2024) 2210: e3002860. . **WB; tested species: rat** 

Proximity labelling reveals effects of disease-causing mutation on the DNAJC5/cysteine string protein a interactome. Barker E, Milburn A, Helassa N, Hammond D, Sanchez-Soriano N, Morgan A, Barclay J The Biochemical journal (2024) : . . **WB; tested species: rat** 

LRRTMs Organize Synapses through Differential Engagement of Neurexin and PTP $\sigma$ . Roppongi RT, Dhume SH, Padmanabhan N, Silwal P, Zahra N, Karimi B, Bomkamp C, Patil CS, Champagne-Jorgensen K, Twilley RE, Zhang P, et al. Neuron (2020) : .. **WB; tested species: mouse** 

VAMP4 maintains a Ca2+-sensitive pool of spontaneously recycling synaptic vesicles. Lin PY, Chanaday NL, Horvath PM, Ramirez DMO, Monteggia LM, Kavalali ET The Journal of neuroscience : the official journal of the Society for Neuroscience (2020) : . . **WB; tested species: rat** 



Access the online factsheet including applicable protocols at <u>https://sysy.com/product/11111</u> 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.