

Endobrevin (Vamp8)

Cat.No. 104 303; 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 : 100 up to 1 : 500 IHC: yes IHC-P: not tested yet
Immunogen	Recombinant protein corresponding to AA 1 to 75 from rat Endobrevin (UniProt Id: Q9WUF4)
Reactivity	Reacts with: human (Q9BV40), rat (Q9WUF4), mouse (O70404). Other species not tested yet.
Specificity	K.O. validated PubMed: 25138214

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

Background

Endobrevin/VAMP8, a member of the SNARE family of proteins, is a relative of synaptobrevin that is involved in the fusion of early and late endosomes. Endobrevin is expressed in most mammalian cells but appears to be absent from neurons. The protein is predominantly localized to early and late endosomal membranes but is also found on other membranes and organelles involved in endocytic membrane traffic. In the fusion of late endosomes it forms SNARE complexes with syntaxin 7, syntaxin 8 and vti1b.

Selected References for 104 303

Vesicle associated membrane protein 8 (VAMP8)-mediated zymogen granule exocytosis is dependent on endosomal trafficking via the constitutive-like secretory pathway.

Messenger SW, Falkowski MA, Thomas DD, Jones EK, Hong W, Gaisano HY, Giasano HY, Boullis NM, Groblewski GE The Journal of biological chemistry (2014) 28940: 28040-53. . **WB, IHC; KO verified**

Plasma membrane flipping of Syntaxin-2 regulates its inhibitory action on insulin granule exocytosis.

Kang F, Xie L, Qin T, Miao Y, Kang Y, Takahashi T, Liang T, Xie H, Gaisano HY Nature communications (2022) 131: 6512. . **IP, WB; tested species: human,mouse**

Defective AP-3-dependent VAMP8 trafficking impairs Weibel-Palade body exocytosis in Hermansky-Pudlak Syndrome type 2 blood outgrowth endothelial cells.

Karampini E, Schillemans M, Hofman M, van Alphen F, de Boer M, Kuijpers TW, van den Biggelaar M, Voorberg J, Bierings R Haematologica (2019) : . . **WB, ICC; KD verified; tested species: human**

The endothelial diapedesis synapse regulates transcellular migration of human T lymphocytes in a CX3CL1- and SNAP23-dependent manner.

Schoppmeyer R, van Steen ACI, Kempers L, Timmerman AL, Nolte MA, Hombrink P, van Buul JD Cell reports (2022) 383: 110243. . **WB; tested species: human**

Fluorescence Lifetime Imaging Microscopy reveals rerouting of SNARE trafficking driving dendritic cell activation.

Verboogen DRJ, González Mancha N, Ter Beest M, van den Bogaart G eLife (2017) 6: . . **ICC; tested species: human**

Syntaxin 8 regulates platelet dense granule secretion, aggregation, and thrombus stability.

Golebiewska EM, Harper MT, Williams CM, Savage JS, Goggs R, Fischer von Mollard G, Poole AW The Journal of biological chemistry (2015) 2903: 1536-45. . **WB**

Selected General References

Membrane fusion and exocytosis.

Jahn R et al. Annu. Rev. Biochem. (1999) PubMed:10872468

Seven novel mammalian SNARE proteins localize to distinct membrane compartments.

Advani RJ et al. J. Biol. Chem. (1998) PubMed:9553086

Endobrevin, a novel synaptobrevin/VAMP-like protein preferentially associated with the early endosome.

Wong SH et al. Mol. Biol. Cell (1998) PubMed:9614193

Access the online factsheet including applicable protocols at <https://sysy.com/product/104303> 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.