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Bassoon

Cat.No. 141 318; Recombinant Guinea pig antibody, 50 µg recombinant IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	50 µg purified recombinant IgG, lyophilized. 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) (see remarks) ICC: 1 : 5000 IHC: 1 : 1000 up to 1 : 2000 IHC-P: 1 : 1000 up to 1 : 2000 ExM: yes (see remarks)
Clone	Gp179H11A2
Subtype	IgG2 (κ light chain)
Immunogen	Recombinant protein corresponding to residues near the central region of rat Bassoon. (UniProt Id: O88778)
Reactivity	Reacts with: mouse (O88737), rat (O88778), zebrafish. Other species not tested yet.
Specificity	Specific for Bassoon K.O. validated
Remarks	This antibody is a chimeric antibody based on the monoclonal mouse antibody clone 179H11A2. The constant regions of the heavy and light chains have been replaced by guinea pig specific sequences. Therefore, the antibody can be used with standard anti-guinea pig secondary reagents. The antibody has been expressed in mammalian cells. WB : Due to its large size, bassoon requires special gel-electrophoresis and Western blot protocols for visualization by immunoblotting. Excellent results can be obtained with the 4-12% TRIS-glycine gradient gels from anamed or NuPAGE 3-8% TRIS-Acetate gradient gels from invitrogen. EXM : This antibody has been successfully used for the epitope-preserving magnified analysis of the proteome (eMAP) expansion microscopy method (Park et al. 2021. PMID: <u>34767453</u>). Variance from the standard protocol: Clearing after gel-embedding: overnight at 37°C followed by 10 minutes at 88°C.

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

Background

Bassoon is a large protein which consists of an N-terminal Zn²⁺ finger and several piccolo-bassoon homology domains (PBH-domains). It is generally found together with piccolo, a related huge multi-domain protein of the CAZ (cytoskeletal matrix assembled at active zones).

Bassoon was suggested to be a scaffolding element of the presynapse but deletion experiments in mice have shown that bassoon is also involved in synaptic vesicle cycling. Probably bassoon interacts with other protein factors via its Zn²⁺ domain but the potential partners have not been determined yet.

Selected References for 141 318

Rho GTPase signaling and mDia facilitate endocytosis via presynaptic actin. Oevel K, Hohensee S, Kumar A, Rosas-Brugada I, Bartolini F, Soykan T, Haucke V eLife (2024) 12:..**ICC; tested species: mouse**

Notch receptor-ligand binding facilitates extracellular vesicle-mediated neuron-to-neuron communication. Wang YZ, Castillon CCM, Gebis KK, Bartom ET, d'Azzo A, Contractor A, Savas JN Cell reports (2024) 432: 113680. . **WB; tested species: mouse**

δ-Catenin controls astrocyte morphogenesis via layer-specific astrocyte-neuron cadherin interactions. Tan CX, Bindu DS, Hardin EJ, Sakers K, Baumert R, Ramirez JJ, Savage JT, Eroglu C The Journal of cell biology (2023) 22211: . . **IHC; tested species: mouse**

Selected General References

Functional regions of the presynaptic cytomatrix protein bassoon: significance for synaptic targeting and cytomatrix anchoring. Dresbach T et al. Mol. Cell. Neurosci. (2003) PubMed:12812759

Unitary assembly of presynaptic active zones from Piccolo-Bassoon transport vesicles. Shapira M et al. Neuron (2003) PubMed:12718858

Functional inactivation of a fraction of excitatory synapses in mice deficient for the active zone protein bassoon. Altrock WD et al. Neuron (2003) PubMed:12628169

The presynaptic active zone protein bassoon is essential for photoreceptor ribbon synapse formation in the retina. Dick O et al. Neuron (2003) PubMed:12628168

Localization of the presynaptic cytomatrix protein Piccolo at ribbon and conventional synapses in the rat retina: comparison with Bassoon.

Dick O et al. J. Comp. Neurol. (2001) PubMed:11596050

Membrane association of presynaptic cytomatrix protein bassoon. Sanmartí-Vila L et al. Biochem. Biophys. Res. Commun. (2000) PubMed:10944438

Bassoon, a novel zinc-finger CAG/glutamine-repeat protein selectively localized at the active zone of presynaptic nerve terminals.

tom Dieck S et al. J. Cell Biol. (1998) PubMed:9679147

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/141318</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.