SYSY Antibodies

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Collybistin

Cat.No. 261 003; 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_2O 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 up to 1 : 2000 **IP**: yes **ICC**: 1 : 500 **IHC**: yes **IHC-P**: not tested yet
Immunogen	Recombinant protein corresponding to AA 4 to 229 from mouse Collybistin (UniProt Id: Q3UTH8)
Reactivity	Reacts with: rat (Q9QX73), mouse (Q3UTH8). Other species not tested yet.
Specificity	Specific for collybistin; immunogen present in all three described splice-variants K.O. validated

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

Background

The GDP/GTP-exchange factor **collybistin** is composed of a dbl homology domain (DH) and a pleckstrin homology domain (PH) connected by a linker sequence. Three splice variants with different C-terminal regions have been described, so far.
Collybistin is supposed to be involved in the clustering of gephyrin, a scaffolding protein linking glycine and GABA receptors to microtubuli.

Selected References for 261 003

Identification of a Core Amino Acid Motif within the α Subunit of GABAARs that Promotes Inhibitory Synaptogenesis and Resilience to Seizures.
Nathanson AJ, Zhang Y, Smalley JL, Ollerhead TA, Rodriguez Santos MA, Andrews PM, Wobst HJ, Moore YE, Brandon NJ, Hines RM, Davies PA, et al.
Cell reports (2019) 283: 670-681.e8. . **WB, ICC; tested species: mouse**

A proline-rich motif in the large intracellular loop of the glycine receptor α1 subunit interacts with the Pleckstrin homology domain of collybistin.
Breitinger U, Weinländer K, Pechmann Y, Langlhofer G, Enz R, Becker CM, Sticht H, Kneussel M, Villmann C, Breitinger HG
Journal of advanced research (2021) 29: 95-106. . **WB, ICC; tested species: mouse**

Gephyrin clusters are absent from small diameter primary afferent terminals despite the presence of GABA(A) receptors.
Lorenzo LE, Godin AG, Wang F, St-Louis M, Carbonetto S, Wiseman PW, Ribeiro-da-Silva A, De Koninck Y
The Journal of neuroscience : the official journal of the Society for Neuroscience (2014) 3424: 8300-17. . **IHC**

miRNA-mediated control of gephyrin synthesis drives sustained inhibitory synaptic plasticity.
Welle TM, Rajgor D, Kareemo DJ, Garcia JD, Zych SM, Wolfe SE, Gookin SE, Martinez TP, Dell'Acqua ML, Ford CP, Kennedy MJ, et al.
EMBO reports (2024) 2511: 5141-5168. . **WB; tested species: rat**

Selective overexpression of Collybistin in mouse hippocampal pyramidal cells enhances GABAergic neurotransmission and protects against PTZ-induced seizures.
George S, James S, de Blas AL
eNeuro (2021) : . . **IHC; tested species: mouse**

Recruitment of Plasma Membrane GABA-A Receptors by Submembranous Gephyrin/Collybistin Clusters.
George S, Chiou TT, Kanamalla K, De Blas AL
Cellular and molecular neurobiology (2021) : . . **ICC; tested species: mouse**

Phosphorylation on Ser 359 of the α2 subunit in GABA type A receptors down-regulates their density at inhibitory synapses.
Nakamura Y, Morrow DH, Nathanson AJ, Henley JM, Wilkinson KA, Moss SJ
The Journal of biological chemistry (2020) : . . **WB; tested species: rat**

Alternative Splicing and the Intracellular Domain Mediate TM-agrin's Ability to Differentially Regulate the Density of Excitatory and Inhibitory Synapse-like Specializations in Developing CNS Neurons.
Handara G, Kröger S
Neuroscience (2019) : . . **ICC; tested species: mouse**

In vivo transgenic expression of collybistin in neurons of the rat cerebral cortex.
Fekete CD, Goz RU, Dinallo S, Miralles CP, Chiou TT, Bear J, Fiondella CG, LoTurco JJ, De Blas AL
The Journal of comparative neurology (2017) 5255: 1291-1311. . **IHC; tested species: rat**

Collybistin binds and inhibits mTORC1 signaling: a potential novel mechanism contributing to intellectual disability and autism.
Machado CO, Griesi-Oliveira K, Rosenberg C, Kok F, Martins S, Passos-Bueno MR, Sertie AL
European journal of human genetics : EJHG (2016) 241: 59-65. . **WB**

Proteomic Characterization of Inhibitory Synapses Using a Novel pHluorin-tagged γ-Aminobutyric Acid Receptor, Type A (GABAA), α2 Subunit Knock-in Mouse.
Nakamura Y, Morrow DH, Modgil A, Huyghe D, Deeb TZ, Lumb MJ, Davies PA, Moss SJ
The Journal of biological chemistry (2016) 29123: 12394-407. . **WB**

Access the online factsheet including applicable protocols at https://sysy.com/product/261003 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 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 freeze-thaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.