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# VGAT (SLC32A1) cytoplasmic

## domain

Cat.No. 131 004; Polyclonal Guinea pig antibody, 100 µl antiserum (lyophilized)

## **Data Sheet**

Reconstitution/ Storage	100 µl antiserum, lyophilized. For <b>reconstitution</b> add 100 µl H <sub>2</sub> O, then aliquot and store at -20°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 (AP staining) IP: not tested yet ICC: 1 : 500 up to 1 : 1000 IHC: 1 : 200 up to 1 : 500 IHC-P: 1 : 500 IHC-Fr: 1 : 500 (see remarks) EXM: external data (see remarks) EM: external data
Immunogen	Recombinant protein corresponding to residues near the amino terminus of rat VGAT (UniProt Id: O35458)
Reactivity	Reacts with: rat (O35458), mouse (O35633), zebrafish, ape. Other species not tested yet.
Specificity	K.O. validated
Matching control	131-0GP
Remarks	<ul> <li>WB: To avoid protein aggregation, do not heat samples for SDS-PAGE.</li> <li>IHC-Fr: 5 min MeOH and PFA fixation are possible.</li> <li>EXM: This antibody has been successfully used for the magnified analysis of the proteome (MAP) expansion microscopy method (MAP; Ku et al. 2016. Nature Biotechnology 34:973-981)</li> </ul>

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

### Background

The vesicular GABA transporter VGAT is responsible for uptake and storage of GABA and glycine by synaptic vesicles in the central nervous system. For this reason it is frequently referred to as the v esicular inhibitory aminoacid transporter VIAAT. It is different from the plasma membrane transporters in that it is driven by a proton electrochemical gradient across the vesicle membrane. So far, only one isoform is known. VGAT is currently the best marker for inhibitory nerve terminals.

### Selected References for 131 004

Quantitative comparison of glutamatergic and GABAergic synaptic vesicles unveils selectivity for few proteins including MAL2, a novel synaptic vesicle protein.

Grønborg M, Pavlos NJ, Brunk I, Chua JJ, Münster-Wandowski A, Riedel D, Ahnert-Hilger G, Urlaub H, Jahn R The Journal of neuroscience : the official journal of the Society for Neuroscience (2010) 301: 2-12. . **ICC, IHC, EM** 

Colocalization of different neurotransmitter transporters on synaptic vesicles is sparse except for VGLUT1 and ZnT3. Upmanyu N, Jin J, Emde HV, Ganzella M, Bösche L, Malviya VN, Zhuleku E, Politi AZ, Ninov M, Silbern I, Leutenegger M, et al. Neuron (2022) : . . **WB, UPTAKE; tested species: rat** 

Development of dissociated cryopreserved rat cortical neurons in vitro. Schock SC, Jolin-Dahel KS, Schock PC, Theiss S, Arbuthnott GW, Garcia-Munoz M, Staines WA Journal of neuroscience methods (2012) 2052: 324-33. . **WB, IHC** 

Astrocyte-secreted neurocan controls inhibitory synapse formation and function. Irala D, Wang S, Sakers K, Nagendren L, Ulloa Severino FP, Bindu DS, Savage JT, Eroglu C Neuron (2024) 11210: 1657-1675.e10. . **ICC, IHC; tested species: mouse,rat** 

The TMEM132B-GABAA receptor complex controls alcohol actions in the brain. Wang G, Peng S, Reyes Mendez M, Keramidas A, Castellano D, Wu K, Han W, Tian Q, Dong L, Li Y, Lu W, et al. Cell (2024) 18723: 6649-6668.e35. . **WB, ICC; tested species: mouse** 

Microglia enable cross-modal plasticity by removing inhibitory synapses. Hashimoto A, Kawamura N, Tarusawa E, Takeda I, Aoyama Y, Ohno N, Inoue M, Kagamiuchi M, Kato D, Matsumoto M, Hasegawa Y, et al. Cell reports (2023) 425: 112383. WB. IHC: tested species: mouse

Spectrin-beta 2 facilitates the selective accumulation of GABAA receptors at somatodendritic synapses. Smalley JL, Cho N, Ng SFJ, Choi C, Lemons AHS, Chaudry S, Bope CE, Dengler JS, Zhang C, Rasband MN, Davies PA, et al. Communications biology (2023) 61: 11. . **WB, ICC; tested species: mouse** 

Brevican, Neurocan, Tenascin-C, and Tenascin-R Act as Important Regulators of the Interplay Between Perineuronal Nets, Synaptic Integrity, Inhibitory Interneurons, and Otx2. Mueller-Buehl C, Reinhard J, Roll L, Bader V, Winklhofer KF, Faissner A Frontiers in cell and developmental biology (2022) 10: 886527. . **WB, IHC; tested species: mouse** 

Targeted proteoform mapping uncovers specific Neurexin-3 variants required for dendritic inhibition. Hauser D, Behr K, Konno K, Schreiner D, Schmidt A, Watanabe M, Bischofberger J, Scheiffele P Neuron (2022) 11013: 2094-2109.e10. . **ICC, IHC; tested species: mouse** 

Systemic inflammation induced the delayed reduction of excitatory synapses in the CA3 during ageing. Manabe T, Rácz I, Schwartz S, Oberle L, Santarelli F, Emmrich JV, Neher JJ, Heneka MT Journal of neurochemistry (2021) 1593: 525-542. . **WB, IHC; tested species: mouse** 

Chemico-genetic discovery of astrocytic control of inhibition in vivo. Takano T, Wallace JT, Baldwin KT, Purkey AM, Uezu A, Courtland JL, Soderblom EJ, Shimogori T, Maness PF, Eroglu C, Soderling SH, et al. Nature (2020) : . . **ICC, IHC; tested species: mouse** 

Expression of Neurofilament Subunits at Neocortical Glutamatergic and GABAergic Synapses. Bragina L, Conti F Frontiers in neuroanatomy (2018) 12: 74. . **WB, IHC; tested species: rat** 

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/131004</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.