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VGAT luminal domain

Cat.No. 131 103; 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) (see remarks) IP: yes ICC: 1 : 100 up to 1 : 200 (see remarks) IHC: 1 : 100 up to 1 : 500 IHC-P: not tested yet
Immunogen	Synthetic peptide corresponding to residues near the carboxy terminus of rat VGAT (UniProt Id: O35458)
Reactivity	Reacts with: human (Q9H598), rat (O35458), mouse (O35633). Other species not tested yet.
Specificity	K.O. validated PubMed: <u>19052203</u>
Remarks	 WB: VGAT aggregates after boiling, making it necessary to run SDS-PAGE only with non-boiled samples. ICC: This antibody is intended for <u>labeling of recycling synaptic vesicles</u> at inhibitory nerve terminals by adding to living neurons. Further details see <u>Martens</u> et al. 2008.

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 131 103

Unique luminal localization of VGAT-C terminus allows for selective labeling of active cortical GABAergic synapses. Martens H, Weston MC, Boulland JL, Grønborg M, Grosche J, Kacza J, Hoffmann A, Matteoli M, Takamori S, Harkany T, Chaudhry FA, et al.

The Journal of neuroscience : the official journal of the Society for Neuroscience (2008) 2849: 13125-31. WB, ICC, IHC, UPTAKE; KO verified; tested species: mouse,rat

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**

A new method for isolation and purification of fusion-competent inhibitory synaptic vesicles. Gopal N, Leitz J, Wang C, Esquivies L, Pfuetzner RA, Brunger AT Current research in physiology (2024) 7: 100121. . **WB; tested species: mouse**

NaV1.1 haploinsufficiency impairs glutamatergic and GABAergic neuron function in the thalamus. Studtmann C, Ladislav M, Topolski MA, Safari M, Swanger SA Neurobiology of disease (2022) 167: 105672. . **IHC; tested species: mouse**

Excitatory neuronal CHD8 in the regulation of neocortical development and sensory-motor behaviors. Kweon H, Jung WB, Im GH, Ryoo J, Lee JH, Do H, Choi Y, Song YH, Jung H, Park H, Qiu LR, et al. Cell reports (2021) 348: 108780. . **IHC; tested species: mouse**

Inhibitory control in neuronal networks relies on the extracellular matrix integrity. Dzyubenko E, Fleischer M, Manrique-Castano D, Borbor M, Kleinschnitz C, Faissner A, Hermann DM Cellular and molecular life sciences : CMLS (2021) 7814: 5647-5663. . **IHC; tested species: mouse**

Extracellular matrix remodeling through endocytosis and resurfacing of Tenascin-R. Dankovich TM, Kaushik R, Olsthoorn LHM, Petersen GC, Giro PE, Kluever V, Agüi-Gonzalez P, Grewe K, Bao G, Beuermann S, Hadi HA, et al.

Nature communications (2021) 121: 7129. . ICC; tested species: rat

The potassium channel subunit Kvβ1 serves as a major control point for synaptic facilitation. Cho IH, Panzera LC, Chin M, Alpizar SA, Olveda GE, Hill RA, Hoppa MB Proceedings of the National Academy of Sciences of the United States of America (2020) 11747: 29937-29947. . **UPTAKE;** tested species: rat

Biallelic DMXL2 mutations impair autophagy and cause Ohtahara syndrome with progressive course. Esposito A, Falace A, Wagner M, Gal M, Mei D, Conti V, Pisano T, Aprile D, Cerullo MS, De Fusco A, Giovedì S, et al. Brain : a journal of neurology (2019) : . . **ICC; tested species: mouse**

Nuclei-specific differences in nerve terminal distribution, morphology, and development in mouse visual thalamus. Hammer S, Carrillo GL, Govindaiah G, Monavarfeshani A, Bircher JS, Su J, Guido W, Fox MA Neural development (2014) 9: 16. I**HC**

Inhibitory synapse dynamics: coordinated presynaptic and postsynaptic mobility and the major contribution of recycled vesicles to new synapse formation.

Dobie FA, Craig AM

The Journal of neuroscience : the official journal of the Society for Neuroscience (2011) 3129: 10481-93. . ICC



Access the online factsheet including applicable protocols at https://sysy.com/product/131103 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.