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# **VGAT** cytoplasmic domain

Cat.No. 131 005; Polyclonal Guinea pig antibody, 50 µg specific antibody (lyophilized)

# **Data Sheet**

Reconstitution/ Storage	50 $\mu g$ specific antibody, lyophilized. Affinity purified with the immunogen. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 50 $\mu$ l H <sub>2</sub> 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	<b>WB</b> : 1: 1000 up to 1: 5000 (see remarks) <b>IP</b> : not tested yet <b>ICC</b> : 1: 500 up to 1: 1000 <b>IHC</b> : 1: 500 up to 1: 1000 (see remarks) <b>IHC</b> -P: 1: 500
Immunogen	Recombinant protein corresponding to residues near the amino terminus of rat VGAT (UniProt Id: O35458)
Reactivity	Reacts with: rat (O35458), mouse (O35633). Other species not tested yet.
Matching control	131-0GP
Remarks	<b>WB</b> : VGAT aggregates after boiling, making it necessary to run SDS-PAGE only with non-boiled samples. <b>IHC</b> : Signal intensity can be enhanced by antigen retrieval at pH6 prior to blocking step.
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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 131 005

Identification and characterization of GABA(A) receptor autoantibodies in autoimmune encephalitis.

Ohkawa T, Satake S, Yokoi N, Miyazaki Y, Ohshita T, Sobue G, Takashima H, Watanabe O, Fukata Y, Fukata M

The Journal of neuroscience: the official journal of the Society for Neuroscience (2014) 3424: 8151-63. ICC; tested species:

Corelease of dopamine and GABA by a retinal dopaminergic neuron.

Hirasawa H, Betensky RA, Raviola E

The Journal of neuroscience: the official journal of the Society for Neuroscience (2012) 3238: 13281-91.. IHC; tested species: mouse

Autism risk gene Cul3 alters neuronal morphology via caspase-3 activity in mouse hippocampal neurons.

Xia QQ, Singh A, Wang J, Xuan ZX, Singer JD, Powell CM

Frontiers in cellular neuroscience (2024) 18: 1320784. . ICC; tested species: mouse

GABAergic-like dopamine synapses in the brain.

Kim HJ, Hwang B, Reva M, Lee J, Lee BE, Lee Y, Cho EJ, Jeong M, Lee SE, Myung K, Baik JH, et al.

Cell reports (2023) 4210: 113239. . IHC; tested species: mouse

Engram cell connectivity as a mechanism for information encoding and memory function.

Ortega-de San Luis C, Pezzoli M, Urrieta E, Ryan TJ

Current biology: CB (2023) 3324: 5368-5380.e5. . IHC; tested species: mouse

Deleting Mecp2 from the cerebellum rather than its neuronal subtypes causes a delay in motor learning in mice.

Achilly NP, He LJ, Kim OA, Ohmae S, Wojaczynski GJ, Lin T, Sillitoe RV, Medina JF, Zoghbi HY

eLife (2021) 10:..IHC; tested species: mouse

Direct reprogramming of oligodendrocyte precursor cells into GABAergic inhibitory neurons by a single homeodomain transcription factor Dlx2.

Boshans LL, Soh H, Wood WM, Nolan TM, Mandoiu II, Yanagawa Y, Tzingounis AV, Nishiyama A

Scientific reports (2021) 111: 3552. . ICC; tested species: mouse

Molecular self-avoidance in synaptic neurexin complexes.

Wang CY, Trotter JH, Liakath-Ali K, Lee SJ, Liu X, Südhof TC

Science advances (2021) 751: eabk1924. . IHC; tested species: mouse

Neuroligin-4 Regulates Excitatory Synaptic Transmission in Human Neurons.

Marro SG, Chanda S, Yang N, Janas JA, Valperga G, Trotter J, Zhou B, Merrill S, Yousif I, Shelby H, Vogel H, et al.

Neuron (2019) 1034: 617-626.e6. . ICC; tested species: human

## **Selected General References**

The vesicular GABA transporter, VGAT, localizes to synaptic vesicles in sets of glycinergic as well as GABAergic neurons. Chaudhry FA et al. J. Neurosci. (1998) PubMed:9822734

Identification and characterization of the vesicular GABA transporter.

McIntire SL et al. Nature (1997) PubMed:9349821

Cloning of a functional vesicular GABA and glycine transporter by screening of genome databases. Sagné C et al. FEBS Lett. (1997) PubMed:9395291

Access the online factsheet including applicable protocols at https://sysy.com/product/131005 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.