

## GAD2 / GAD65

Cat.No. 198 102; Polyclonal rabbit antibody, 200 µl antiserum (lyophilized)

### Data Sheet

Reconstitution/ Storage	200 µl antiserum, lyophilized. For <b>reconstitution</b> add 200 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	<b>WB:</b> not recommended (see remarks) <b>IP:</b> not tested yet <b>ICC:</b> 1 : 2000 <b>IHC:</b> not recommended <b>IHC-P:</b> not tested yet
Immunogen	Recombinant protein corresponding to AA 3 to 96 from mouse GAD2 (UniProt Id: P48320)
Reactivity	Reacts with: rat (P18088), mouse (P48318). Other species not tested yet.
Specificity	Specific for GAD 2 / GAD 65.
Matching control	198-1P
Remarks	<b>WB:</b> The affinity purified antibody is recommended.

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

### Background

The **glutamic acid decarboxylases GAD 2**, also referred to as **GAD 65**, and GAD 1 / GAD 67 synthesize  $\gamma$ -aminobutyric acid (GABA), the major inhibitory neurotransmitter in the central nervous system. The hydrophilic GAD 1 can heterodimerize with the membrane anchored GAD 2 and part of GAD 1 is targeted to inhibitory nerve terminals by this mechanisms. Although both proteins exhibit significant differences in their N-terminus they share high homology in the rest of the molecule.

GADs are a widely used markers for the GABAergic system. In type 1 diabetes GAD 1 has been identified as a major autoantigen.

### Selected References for 198 102

Mechanism and effects of pulsatile GABA secretion from cytosolic pools in the human beta cell.  
Menegaz D, Hagan DW, Almacá J, Cianciaruso C, Rodriguez-Diaz R, Molina J, Dolan RM, Becker MW, Schwalie PC, Nano R, Lebreton F, et al.

Nature metabolism (2019) 111: 1110-1126. . **ICC, IHC; tested species: human, rat**

Blocking L-type voltage-gated Ca<sup>2+</sup> channels with dihydropyridines reduces gamma-aminobutyric acid type A receptor expression and synaptic inhibition.

Saliba RS, Gu Z, Yan Z, Moss SJ

The Journal of biological chemistry (2009) 28447: 32544-50. . **WB**

Neuron-astrocyte interaction enhance GABAergic synaptic transmission in a manner dependent on key metabolic enzymes.

Kaczor P, Rakus D, Mozrzyms JW

Frontiers in cellular neuroscience (2015) 9: 120. . **ICC**

Function coupling of otoferlin with GAD65 acts to modulate GABAergic activity.

Wu W, Rahman MN, Guo J, Roy N, Xue L, Cahill CM, Zhang S, Jia Z

Journal of molecular cell biology (2015) 72: 168-79. . **IHC**

### Selected General References

A palmitoylation cycle dynamically regulates partitioning of the GABA-synthesizing enzyme GAD65 between ER-Golgi and post-Golgi membranes.

Kanaani J, Patterson G, Schaufele F, Lippincott-Schwartz J, Baekkeskov S

Journal of cell science (2008) 121Pt 4: 437-49. .

The hydrophilic isoform of glutamate decarboxylase, GAD67, is targeted to membranes and nerve terminals independent of dimerization with the hydrophobic membrane-anchored isoform, GAD65.

Kanaani J, Lissin D, Kash SF, Baekkeskov S

The Journal of biological chemistry (1999) 27452: 37200-9. .

Differential expression of GAD65 and GAD67 in human, rat, and mouse pancreatic islets.

Kim J, Richter W, Aanstoot HJ, Shi Y, Fu Q, Rajotte R, Warnock G, Baekkeskov S

Diabetes (1993) 4212: 1799-808. .

Glutamate decarboxylases in nonneural cells of rat testis and oviduct: differential expression of GAD65 and GAD67.

Tillakaratne NJ, Erlander MG, Collard MW, Greif KF, Tobin AJ

Journal of neurochemistry (1992) 582: 618-27. .

Access the online factsheet including applicable protocols at <https://sysy.com/product/198102> 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.