SYSY Antibodies

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GAD2 (GAD65)

Cat.No. 198 111; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 µg purified IgG, lyophilized. Albumin and azide were added for stabilization. For **reconstitution** add 100 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 (AP staining) **IP**: yes **ICC**: 1 : 500 **IHC**: 1 : 200 up to 1 : 500 **IHC-P**: 1 : 100 up to 1 : 1000 **ELISA**: yes
Clone	26H1D1
Subtype	IgG3 (κ light chain)
Immunogen	Recombinant protein corresponding to the amino terminus of mouse GAD2 (UniProt Id: P48320)
Epitop	AA 3 to 17 from mouse GAD2 (UniProt Id: P48320)
Reactivity	Reacts with: rat (Q05683), mouse (P48320), human (Q05329). Other species not tested yet.
Specificity	Specific for GAD2 / GAD65
Matching control	198-1P
Remarks	**ELISA**: The ELISA-protocol for membrane proteins is required. Suitable as capture antibody for sandwich-ELISA. Please refer to the protocol for suitable detector antibodies.

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

Background

The **g**lutamic **a**cid **d**ecarboxylases GAD1 and GAD2, also referred to as GAD67 and GAD65 respectively, synthesize γ-aminobutyric acid (GABA), the major inhibitory neurotransmitter in the central nervous system. Therefore, GADs are widely used markers for the GABAergic system (1). The hydrophilic GAD1 can heterodimerize with the membrane achored GAD2 and a part of GAD1 is targeted to inhibitory nerve terminals by this mechanism (2). Although both proteins exhibit significant differences in their N-terminus they share high homology in the rest of the molecule (3).
GAD1 and 2 also occur in rodent pancreatic islets of Langerhans, whereas human islets mainly express GAD2 which has been identified as a major autoantigen in type 1 diabetes (3).

Selected References for 198 111

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**

Immunocytochemical identification of electroneutral Na-coupled HCO_3 transporters in freshly dissociated mouse medullary raphé neurons.
Coley AA, Ruffin VA, Moss FJ, Hopfer U, Boron WF
Neuroscience (2013) 246: 451-67. . **ICC; tested species: mouse**

ASCL1- and DLX2-induced GABAergic neurons from hiPSC-derived NPCs.
Barretto N, Zhang H, Powell SK, Fernando MB, Zhang S, Flaherty EK, Ho SM, Slesinger PA, Duan J, Brennand KJ
Journal of neuroscience methods (2020) 334: 108548. . **ICC; tested species: human**

Cardiolipin exposure on the outer mitochondrial membrane modulates α-synuclein.
Ryan T, Bamm VV, Stykel MG, Coackley CL, Humphries KM, Jamieson-Williams R, Ambasudhan R, Mosser DD, Lipton SA, Harauz G, Ryan SD, et al.
Nature communications (2018) 91: 817. . **ICC; tested species: human**

ELKS controls the pool of readily releasable vesicles at excitatory synapses through its N-terminal coiled-coil domains.
Held RG, Liu C, Kaeser PS
eLife (2016) 5: . . **ICC**

Electrical Responses and Spontaneous Activity of Human iPS-Derived Neuronal Networks Characterized for 3-month Culture with 4096-Electrode Arrays.
Amin H, Maccione A, Marinaro F, Zordan S, Nieus T, Berdondini L
Frontiers in neuroscience (2016) 10: 121. . **ICC; tested species: human**

An E3-ligase-based method for ablating inhibitory synapses.
Gross GG, Straub C, Perez-Sanchez J, Dempsey WP, Junge JA, Roberts RW, Trinh le A, Fraser SE, De Koninck Y, De Koninck P, Sabatini BL, et al.
Nature methods (2016) 138: 673-8. . **ICC; tested species: mouse**

Vesicular glutamate transporter 1 orchestrates recruitment of other synaptic vesicle cargo proteins during synaptic vesicle recycling.
Pan PY, Marrs J, Ryan TA
The Journal of biological chemistry (2015) 29037: 22593-601. . **ICC**

Evaluation of established human iPSC-derived neurons to model neurodegenerative diseases.
Meneghello G, Verheyen A, Van Ingen M, Kuijlaars J, Tuefferd M, Van Den Wyngaert I, Nuydens R
Neuroscience (2015) 301: 204-12. . **ICC; tested species: human**

The active zone protein family ELKS supports Ca2+ influx at nerve terminals of inhibitory hippocampal neurons.
Liu C, Bickford LS, Held RG, Nyitrai H, Südhof TC, Kaeser PS
The Journal of neuroscience : the official journal of the Society for Neuroscience (2014) 3437: 12289-303. . **ICC**

Access the online factsheet including applicable protocols at https://sysy.com/product/198111 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.