

Brn3a

Cat.No. 411 008; Recombinant rabbit antibody, 50 µg recombinant IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	50 µg purified recombinant IgG, lyophilized. Albumin and azide were 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: not tested yet IP: not tested yet ICC: not tested yet IHC: 1 : 500 (see remarks) IHC-P: 1 : 400 up to 1 : 500
Clone	Rb61A2
Subtype	IgG1 (κ light chain)
Immunogen	Synthetic peptide corresponding to residues near the amino terminus of mouse Brn3a (UniProt Id: P17208)
Reactivity	Reacts with: mouse (P17208), rat. Other species not tested yet.
Remarks	This antibody is a chimeric antibody based on the monoclonal rat antibody SY-61A2. The constant regions of the heavy and light chains have been replaced by rabbit specific sequences. The antibody can therefore be used with standard anti-rabbit secondary reagents. The antibody has been expressed in mammalian cells. IHC: For optimal results in retina tissue, follow the retina protocol according to Gierke et al. 2023 . Alternatively, Antigen retrieval with citrate buffer pH 6 can be applied.

Background

Brn3a, also referred to as POU4F1, RGC-1 or Oct-T1, is a transcription factor highly expressed in the developing peripheral sensory nervous system, in cells of the B- and T-lymphocytic lineages and in certain regions of the CNS e.g. retina, spinal cord, midbrain superior colliculus, red nucleus, nucleus ambiguus, inferior olivary nucleus and habenula. In the retina Brn3a is a well-established marker for retinal ganglion cells.

Selected General References

Brn3a and Brn3b knockout mice display unvaried retinal fine structure despite major morphological and numerical alterations of ganglion cells.

Ghinia MG et al. J. Comp. Neurol. (2016) PubMed:27391320

Brn3a and Islet1 act epistatically to regulate the gene expression program of sensory differentiation.

Dykes IM et al. J. Neurosci. (2011) PubMed:21734270

Regulation of NGF-A (Egr-1) gene expression by the POU domain transcription factor Brn-3a.

Smith MD et al. Brain Res. Mol. Brain Res. (1999) PubMed:10640682

Targeted deletion of the mouse POU domain gene Brn-3a causes selective loss of neurons in the brainstem and trigeminal ganglion, uncoordinated limb movement, and impaired suckling.

Xiang M et al. Proc. Natl. Acad. Sci. U.S.A. (1996) PubMed:8876243

The different activities of the two activation domains of the Brn-3a transcription factor are dependent on the context of the binding site.

Budhram-Mahadeo V et al. J. Biol. Chem. (1996) PubMed:8621561

The Brn-3 family of POU-domain factors: primary structure, binding specificity, and expression in subsets of retinal ganglion cells and somatosensory neurons.

Xiang M et al. J. Neurosci. (1995) PubMed:7623109

A novel POU family transcription factor is closely related to Brn-3 but has a distinct expression pattern in neuronal cells.

Lillycrop KA et al. Nucleic Acids Res. (1992) PubMed:1383937

Access the online factsheet including applicable protocols at <https://sysy.com/product/411008> or scan the QR-code.



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

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 at 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.