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Ankyrin G

Cat.No. 386 006; Polyclonal chicken antibody, 50 µg specific antibody (lyophilized)

Data Sheet

Reconstitution/ Storage	50 μg specific antibody, lyophilized. Affinity purified with the immunogen. Albumin and azide 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 up to 1 : 2000 (AP staining) IP: not tested yet ICC: 1 : 1000 up to 1 : 2000 IHC: 1 : 500 (see remarks) IHC-P: not tested yet IHC-Fr: 1 : 500 (see remarks)
Immunogen	Recombinant protein corresponding to residues near the carboxy terminus of mouse Ankyrin G. (UniProt Id: G5E8K5-1)
Reactivity	Reacts with: rat (O70511-1), mouse (G5E8K5-1). Other species not tested yet.
Specificity	Specific for Ankyrin G; detects all described splice variants.
Remarks	 IHC: Antigen retrieval (10mM Tris, 1mM EDTA, pH 9.0, overnight at 60°C) is recommended. IHC-Fr: Fixation with acetone or PFA/formaldehyde is recommended. Postfixation with acetone-methanol or methanol is not advised.

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

Background

Ankyrin G is a membrane-cytoskeleton linker. It may participate in the targeting and clustering of ion channels and cell adhesion molecules at the nodes of Ranvier and axonal initial segments (AIS).

Selected References for 386 006

INSIHGT: an accessible multi-scale, multi-modal 3D spatial biology platform. Yau CN, Hung JTS, Campbell RAA, Wong TCY, Huang B, Wong BTY, Chow NKN, Zhang L, Tsoi EPL, Tan Y, Li JJX, et al. Nature communications (2024) 151: 10888. . **IHC; tested species: mouse**

Endocytosis in the axon initial segment maintains neuronal polarity. Eichel K, Uenaka T, Belapurkar V, Lu R, Cheng S, Pak JS, Taylor CA, Südhof TC, Malenka R, Wernig M, Özkan E, et al. Nature (2022):.. **ICC; tested species: mouse**

Immunoproximity biotinylation reveals the axon initial segment proteome. Zhang W, Fu Y, Peng L, Ogawa Y, Ding X, Rasband A, Zhou X, Shelly M, Rasband MN, Zou P Nature communications (2023) 141: 8201. . **ICC; tested species: rat**

Selected General References

Structural basis for the membrane association of ankyrinG via palmitoylation. Fujiwara Y et al. Sci Rep (2016) PubMed:27046665

betalV spectrin is recruited to axon initial segments and nodes of Ranvier by ankyrinG. Yang Y et al. J. Cell Biol. (2007) PubMed:17283186

AnkyrinG is required for clustering of voltage-gated Na channels at axon initial segments and for normal action potential firing. Zhou D et al. J. Cell Biol. (1998) PubMed:9832557

beta-Spectrin is colocalized with both voltage-gated sodium channels and ankyrinG at the adult rat neuromuscular junction. Wood SJ et al. J. Cell Biol. (1998) PubMed:9456326

AnkyrinG. A new ankyrin gene with neural-specific isoforms localized at the axonal initial segment and node of Ranvier. Kordeli E et al. J. Biol. Chem. (1995) PubMed:7836469

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/386006</u> 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.