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Кіг4.1

Cat.No. 472 005; Polyclonal Guinea pig antibody, 50 µg specific antibody (lyophilized)

Data Sheet

Reconstitution/ Storage	50 μg specific antibody, lyophilized. Affinity purified with the immunogen. 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: 1 : 1000 up to 1 : 2000 (AP-staining) IP: yes ICC: 1 : 1000 up to 1 : 3000 IHC: 1 : 500 up to 1 : 1000 IHC-P: 1 : 500 up to 1 : 1000 IHC-Fr: 1 : 500 up to 1 : 1000
Immunogen	Recombinant protein corresponding to residues near the carboxy terminus of human Kir4.1. (UniProt Id: P78508)
Reactivity	Reacts with: mouse (Q9JM63), rat (P49655), human (P78508). Other species not tested yet.
Remarks	 WB: To avoid protein aggregation, do not heat samples for SDS-PAGE. IHC: Antigen retrieval with citrate buffer pH 6 can be applied to improve the signal to noise ratio. IHC-Fr: Methanol fixation is recommended.

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

Background

Astrocyte membranes are highly permeable to K⁺ ions, leading to a hyperpolarized resting membrane potential and low input membrane resistance. The main player in mediating these properties is the Kir4.1 inward rectifying K⁺ channel [1, 2]. Without Kir4.1, astrocytes lack their signature K⁺ currents, which are sensitive to K⁺ blockers such as Ba2⁺ [3, 4]. The Kir4.1 channels are expressed throughout the brain, but are found in the highest concentrations in the olfactory bulb, cerebellum, brain stem, spinal cord and midbrain. While Kir4.1 is not expressed in neurons, it is expressed in a variety of CNS glia, including oligodendrocytes, and astrocytes. Protoplasmic astrocytes within the grey matter have higher Kir4.1 expression than fibrous astrocytes within the white matter [2, 5]. The expression of Kir4.1 increases with age, particularly within the first 10 days postnatally. This increase is associated with both an increase of the inward current in developing astrocytes and a shift away from oligodedroglial expression of Kir4.1 [6, 3].

Selected General References

Turning down the volume: Astrocyte volume change in the generation and termination of epileptic seizures. Murphy TR et al. Neurobiol Dis (2017) PubMed:28438505

The role of glial-specific Kir4.1 in normal and pathological states of the CNS. Nwaobi SE et al. Acta Neuropathol (2016) PubMed:26961251

Subcellular localization of K+ channels in mammalian brain neurons: remarkable precision in the midst of extraordinary complexity.

Trimmer JS et al. Neuron (2015) PubMed:25611506

Analysis of astroglial K+ channel expression in the developing hippocampus reveals a predominant role of the Kir4.1 subunit. Seifert G et al. J Neurosci (2009) PubMed:19515915

Conditional knock-out of Kir4.1 leads to glial membrane depolarization, inhibition of potassium and glutamate uptake, and enhanced short-term synaptic potentiation. Djukic B et al. J Neurosci (2007) PubMed:17942730

Kir4.1 channels regulate swelling of astroglial processes in experimental spinal cord edema. Dibaj P et al. J Neurochem (2007) PubMed:17953658

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/472005</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.