SYSY Antibodies

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Proton ATPase 116 kDa subunit

Cat.No. 109 003; Polyclonal rabbit antibody, 50 µg specific antibody (lyophilized)

Data Sheet

Reconstitution/ Storage	50 µg specific antibody, lyophilized. Affinity purified with the immunogen. Albumin was added for stabilization. For **reconstitution** add 50 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) (see remarks) **IP**: not tested yet **ICC**: 1 : 100 up to 1 : 500 (see remarks) **IHC**: 1 : 100 up to 1 : 500 (see remarks) **IHC-P (FFPE)**: not tested yet
Immunogen	Synthetic peptide corresponding to AA 826 to 838 from rat Proton ATPase (UniProt Id: P25286)
Reactivity	Reacts with: rat (P25286), mouse (Q9Z1G4), hamster. No signal: fish. Other species not tested yet.
Specificity	Specific for the α1 116kDa subunit. K.D. validated
Matching control	109-0P
Remarks	**WB**: To avoid protein aggregation, do not heat samples for SDS-PAGE. **ICC**: Methanol fixation is recommended. **IHC**: Antigen retrieval with methanol/acetic acid is required. For details see Dumoulin A, Triller A & Dieudonné S (2001).

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

Background

The **Proton ATPase**, also referred to as **vacuolar proton pump**, is involved in the acidification of many intracellular organelles. The pump is composed of more than 10 subunits, of which the 116 kDa subunit is the largest. This subunit has an N-terminal cytoplasmic domain and a C-terminal transmembrane domain with probably 6 transmembrane regions. The 116 kDa subunit is essential for proton pump activity.

Selected References for 109 003

Clathrin coat controls synaptic vesicle acidification by blocking vacuolar ATPase activity.
Farsi Z, Gowrisankaran S, Krunic M, Rammner B, Woehler A, Lafer EM, Mim C, Jahn R, Milosevic I
eLife (2018) 7: . . **WB; tested species: mouse**

The proteomic landscape of synaptic diversity across brain regions and cell types.
van Oostrum M, Blok TM, Giandomenico SL, Tom Dieck S, Tushev G, Fürst N, Langer JD, Schuman EM
Cell (2023) 18624: 5411-5427.e23. . **WB; tested species: mouse**

Suggestion of creatine as a new neurotransmitter by approaches ranging from chemical analysis and biochemistry to electrophysiology.
Bian X, Zhu J, Jia X, Liang W, Yu S, Li Z, Zhang W, Rao Y
eLife (2023) 12: . . **WB; tested species: mouse**

ATP6V0d2 controls Leishmania parasitophorous vacuole biogenesis via cholesterol homeostasis.
Pessoa CC, Reis LC, Ramos-Sanchez EM, Orikaza CM, Plaza CC, de Castro Levatti EV, Badaró ACB, Yamamoto JUDS, D'Almeida V, Goto H, Mortara RA, et al.
PLoS pathogens (2019) 156: e1007834. . **WB; tested species: mouse**

Selected General References

The synaptic vesicle cycle: a cascade of protein-protein interactions.
Südhof TC et al. Nature (1995) PubMed:7791897

Synaptic vesicles and exocytosis.
Jahn R et al. Annu. Rev. Neurosci. (1994) PubMed:8210174

Structure of the 116-kDa polypeptide of the clathrin-coated vesicle/synaptic vesicle proton pump.
Perin MS et al. J. Biol. Chem. (1991) PubMed:1704894

Access the online factsheet including applicable protocols at https://sysy.com/product/109003 or scan the QR-code.

FAQ - How should I store my antibody?

Shipping Conditions

- All SYSY antibodies and control proteins/peptides are shipped lyophilized (vacuum freeze-dried). In this form, they remain stable 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. **Do not freeze lyophilized antibodies**. Temperatures below 0°C may impair performance.
- **Fluorescence-labeled antibodies** should be reconstituted immediately upon receipt. Long-term storage of lyophilized fluorophore-conjugates may cause aggregation.
- **Control peptides** should be stored at -20°C before reconstitution.

Long Term Storage after Reconstitution (General Considerations)

- **Do not use frost-free ("no-frost") freezers.** These units periodically warm to remove ice buildup, causing freeze–thaw cycles that can damage antibodies.
- Store vials in areas with minimal temperature fluctuation - preferably toward the back of the freezer, not on the door.
- Aliquot reconstituted antibodies and store at –20°C to –80°C.
- Avoid very small aliquots (<20 µL), as evaporation and adsorption to tube surfaces can reduce antibody concentration and activity.
- Use the smallest practical storage vial to minimize surface area.
- Adding glycerol to a final concentration of 50% prevents freezing at -20°C, allowing storage in liquid form and effectively avoiding freeze–thaw cycles.

Product Specific Hints for Storage

Control proteins / peptides

- Store at -20°C to -80°C

Monoclonal Antibodies

- **Ascites and hybridoma supernatant:** Store at -20°C to -80°C
  Prolonged storage at 4°C is not recommended, as proteases present in ascites may degrade antibodies.
- **Purified IgG:** Store at -20°C to -80°C
  Adding a carrier protein (e.g., BSA) enhances long-term stability.
  Many SYSY antibodies already contain carrier proteins - refer to the respective datasheet for details.

Polyclonal Antibodies

- **Crude antisera:** Can be stored at 4°C with antimicrobials added, but -20°C to -80°C is preferred
- **Affinity-purified antibodies:** Less stable than antisera; store at -20°C to -80°C
  Adding a carrier protein such as BSA improves long-term stability.
  Most SYSY antibodies already contain carrier proteins - refer to the respective datasheet for details.

Fluorescence-labeled Antibodies

- Store as a liquid with 1:1 (v/v) glycerol at -20°C, and protect from light exposure

Avoid repeated freeze-thaw cycles for all antibodies!

FAQ - How should I reconstitute my antibody?

Reconstitution

- All purified SYSY antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add the volume of deionized water specified in the corresponding datasheet. If a larger final volume is desired, first add the recommended amount of water, then adjust with PBS and, if needed, add a stabilizing carrier protein (e.g., BSA) to a final concentration of 2%. Some SYSY antibodies already contain albumin; please take this into account before adding additional carrier protein.

For complete reconstitution, carefully remove the vial cap. After adding water, briefly vortex the solution. To collect the liquid at the bottom of the vial, place the vial inside a 50 ml centrifuge tube padded with paper and centrifuge briefly.

- If desired, small amounts of azide or thimerosal may be added to prevent microbial growth. This is particularly recommended when storing an aliquot at 4°C.
- After reconstitution of fluorescence-labeled antibodies, add glycerol 1:1 (v/v) to achieve a final concentration of 50%. This prevents freezing at –20°C and keeps the antibody in liquid form, effectively avoiding freeze–thaw cycles.
- Glycerol may also be added to unlabeled primary antibodies as a general measure to prevent freeze–thaw damage.
- For further guidance, please refer to our **storage tips** and recommendations for reconstituted antibodies, control peptides, and control proteins.