

## 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 <b>reconstitution</b> add 50 µl H <sub>2</sub> 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	<b>WB:</b> 1 : 1000 (AP staining) (see remarks) <b>IP:</b> not tested yet <b>ICC:</b> 1 : 100 (see remarks) <b>IHC:</b> 1 : 100 up to 1 : 500 (see remarks) <b>IHC_P:</b> 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.
Matching control	109-0P
Remarks	<b>WB:</b> Proton ATPase aggregates after boiling, making it necessary to run SDS-PAGE with non-boiled samples. <b>ICC:</b> Methanol fixation is recommended. <b>IHC:</b> Tissue sections require additional antigen retrieval with methanol/acetic acid prior to antibody incubation. 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**

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  
Nature (1995) 3756533: 645-53. .

Synaptic vesicles and exocytosis.  
Jahn R, Südhof TC  
Annual review of neuroscience (1994) 17: 219-46. .

Structure of the 116-kDa polypeptide of the clathrin-coated vesicle/synaptic vesicle proton pump.  
Perin MS, Fried VA, Stone DK, Xie XS, Südhof TC  
The Journal of biological chemistry (1991) 2666: 3877-81. .

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 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.