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# Rab5

Cat.No. 108 111; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

## Data Sheet

Reconstitution/ Storage	100 $\mu$ g purified IgG, lyophilized. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 100 $\mu$ l 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	WB: 1 : 1000 up to 1 : 10000 (AP staining) IP: yes ICC: 1 : 100 up to 1 : 1000 (see remarks) IHC: 1 : 50 up to 1 : 500 IHC-P: not tested yet
Clone	621.4
Subtype	IgG2a (κ light chain)
Immunogen	Recombinant protein corresponding to AA 1 to 215 from rat Rab5 (UniProt Id: O88565)
Reactivity	Reacts with: human (Q9UJ41), rat (M0RC99), mouse (Q9JM13). Other species not tested yet.
Specificity	Specific for rab 5 (probably only rab 5a). No cross-reactivity to other rab proteins.
Remarks	This antibody was used very successfully for immunoisolation of early endosomes and for the differentiation of early endosomes from related trafficking organelles in neurons and nonneuronal cells. <b>ICC</b> : Not suitable for ICC of PFA-fixed cells. Methanol fixation is recommended.

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

### Background

**Rab 5** is a member of the Rab protein family that belongs to the ras-related superfamily of small monomeric GTPases. Rab 5 is ubiquitously expressed in all tissues where it functions in the fusion of early endosomes which is the first fusion step of endocytic organelles after their formation and detachment from the plasma membrane. It is presently the best marker with selectivity for this compartment.

## Selected References for 108 111

Hyperactivation of RAB5 disrupts the endosomal Rab cascade leading to endolysosomal dysregulation in Down syndrome: A necessary role for increased APP gene dose.

Chen XQ, Zuo X, Becker A, Mobley WC

Alzheimer's & dementia : the journal of the Alzheimer's Association (2025) 215: e70046. . IP; tested species: mouse

Vti1a/b regulate synaptic vesicle and dense core vesicle secretion via protein sorting at the Golgi. Emperador-Melero J, Huson V, van Weering J, Bollmann C, Fischer von Mollard G, Toonen RF, Verhage M Nature communications (2018) 91: 3421. . **ICC; tested species: mouse** 

SNARE proteins are highly enriched in lipid rafts in PC12 cells: implications for the spatial control of exocytosis. Chamberlain LH, Burgoyne RD, Gould GW Proceedings of the National Academy of Sciences of the United States of America (2001) 9810: 5619-24. . **WB; tested species:** rat

Increased expression of heme-binding protein 1 early in Alzheimer's disease is linked to neurotoxicity. Yagensky O, Kohansal-Nodehi M, Gunaseelan S, Rabe T, Zafar S, Zerr I, Härtig W, Urlaub H, Chua JJ eLife (2019) 8: . . **WB; tested species: mouse** 

Cardiac recovery via extended cell-free delivery of extracellular vesicles secreted by cardiomyocytes derived from induced pluripotent stem cells.

Liu B, Lee BW, Nakanishi K, Villasante A, Williamson R, Metz J, Kim J, Kanai M, Bi L, Brown K, Di Paolo G, et al. Nature biomedical engineering (2018) 25: 293-303. . **WB; tested species: human** 

### **Selected General References**

Rab GTPases coordinate endocytosis. Somsel Rodman J et al. J. Cell. Sci. (2000) PubMed:10633070

Alzheimer amyloid protein precursor is localized in nerve terminal preparations to Rab5-containing vesicular organelles distinct from those implicated in the synaptic vesicle pathway.

Ikin AF et al. J. Biol. Chem. (1996) PubMed:8943215

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/108111</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.