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# Rab3a

Cat.No. 107 102; Polyclonal rabbit antibody, 200 µl antiserum (lyophilized)

### **Data Sheet**

Reconstitution/ Storage	200 $\mu$ l antiserum, lyophilized. For <b>reconstitution</b> add 200 $\mu$ l H <sub>2</sub> O, then aliquot and store at -20°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) IP: yes ICC: 1: 1000 IHC: 1: 500 up to 1: 1000 IHC-P: not tested yet ELISA: yes
Immunogen	Synthetic peptide corresponding to AA 2 to 14 from rat Rab3a (UniProt Id: P63012)
Reactivity	Reacts with: human (P20336), rat (P63012), mouse (P63011), hamster, cow, zebrafish. Other species not tested yet.
Matching control	107-1P
Remarks	<b>ELISA</b> : The ELISA-protocol for membrane proteins is required. Suitable as detector antibody for sandwich-ELISA. Please refer to the protocol for suitable capture antibodies.

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

#### **Background**

Rab3 proteins belong to the Rab family, a subset of the Ras-related superfamily of small monomeric GTPases. There are four known isoforms: Rab3a, Rab3b, Rab3c, and Rab3d (1, 2). Rab3a and Rab3c are primarily found in neuronal and neuroendocrine cells, whereas Rab3b and Rab3d are predominantly expressed in non-neuronal tissues such as the parotid gland, pancreas, mast cells, and adipose tissue (2, 3).

Rab3a, the most abundant and well-characterized isoform, is highly enriched in synaptic vesicles. It regulates vesicle transport, docking, fusion, and Ca<sup>2</sup>II-dependent neurotransmitter release by cycling between a GDP-bound inactive state and a GTP-bound vesicle-associated active state, interacting with other synaptic proteins in the process (1, 2).

Unlike integral membrane proteins of synaptic vesicles, Rab3a and Rab3c are absent from the Golgi complex, preventing immunostaining of the axo-dendritic region, which can occur with proteins such as synaptophysin, synaptobrevin/VAMP, or synaptogyrin (1).

Beyond the nervous system, Rab3a is also expressed in the pancreas, where it is predominantly localized to  $\beta$ -cells of the islets of Langerhans. It plays a crucial role in regulated insulin secretion, while Rab3d is primarily involved in exocrine pancreatic enzyme secretion (3).

#### Selected References for 107 102

Localization versus function of Rab3 proteins. Evidence for a common regulatory role in controlling fusion.

Schlüter OM, Khvotchev M, Jahn R, Südhof TC

The Journal of biological chemistry (2002) 27743: 40919-29. . WB, IHC

SV31 is a Zn2+-binding synaptic vesicle protein.

Barth J, Zimmermann H, Volknandt W

Journal of neurochemistry (2011) 1184: 558-70. . WB, ICC

Regulation of density of functional presynaptic terminals by local energy supply.

Zhou H, Liu G

Molecular brain (2015) 8: 42. . WB, ICC

Distribution of synaptic vesicle proteins in the mammalian retina identifies obligatory and facultative components of ribbon synapses.

Von Kriegstein K, Schmitz F, Link E, Südhof TC

The European journal of neuroscience (1999) 114: 1335-48. . WB, IHC

Synapsin-dependent reserve pool of synaptic vesicles supports replenishment of the readily releasable pool under intense synaptic transmission.

Vasileva M, Horstmann H, Geumann C, Gitler D, Kuner T

The European journal of neuroscience (2012) 368: 3005-20. . ELISA

Cochlear ablation in neonatal rats disrupts inhibitory transmission in the medial nucleus of the trapezoid body.

Hruskova B, Trojanova J, Kralikova M, Melichar A, Suchankova S, Bartosova J, Burianova JS, Popelar J, Syka J, Turecek R

Neuroscience letters (2019): .. IHC; tested species: rat

The GTPase Rab26 links synaptic vesicles to the autophagy pathway.

Binotti B, Pavlos NJ, Riedel D, Wenzel D, Vorbrüggen G, Schalk AM, Kühnel K, Boyken J, Erck C, Martens H, Chua JJ, et al. eLife (2015) 4: e05597. . **WB** 

Proteomic screening of glutamatergic mouse brain synaptosomes isolated by fluorescence activated sorting. Biesemann C, Grønborg M, Luquet E, Wichert SP, Bernard V, Bungers SR, Cooper B, Varoqueaux F, Li L, Byrne JA, Urlaub H, et al. The EMBO journal (2014) 332: 157-70. • WB; tested species: mouse

Composition of isolated synaptic boutons reveals the amounts of vesicle trafficking proteins.

Wilhelm BG, Mandad S, Truckenbrodt S, Kröhnert K, Schäfer C, Rammner B, Koo SJ, Claßen GA, Krauss M, Haucke V, Urlaub H, et al.

Science (New York, N.Y.) (2014) 3446187: 1023-8. . IHC; tested species: mouse

Access the online factsheet including applicable protocols at <a href="https://sysy.com/product/107102">https://sysy.com/product/107102</a> 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.