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# Dynamin3

Cat.No. 115 302; 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	<b>WB</b> : 1 : 1000 up to 1 : 5000 (AP staining) <b>IP</b> : yes <b>ICC</b> : 1 : 500 up to 1 : 1000 <b>IHC</b> : 1 : 1000 up to 1 : 5000 <b>IHC_P</b> : 1 : 500
Immunogen	Synthetic peptide corresponding to AA 773 to 794 from mouse Dynamin3 (UniProt Id: Q8BZ98)
Reactivity	Reacts with: rat (Q08877), mouse (Q8BZ98). Other species not tested yet.
Specificity	Detects dynamin 3 with weak cross-reactivity for Dynamin1
Matching control	115-3P

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

# **Background**

**Dynamin** was discovered because of its binding to microtubules. It was later shown not to function in the cytoskeleton but in endocytosis. Dynamin is required for clathrin - mediated endocytosis. It contains a NH2 - terminal GTPase domain, a middle pleckstrin - homology domain, and a COOH-terminal proline - rich sequence. The COOH - terminal sequence binds to amphiphilin which contains a SH3 domain that recognizes the proline - rich sequence of dynamin.

There are at least three isoforms of dynamin: Dynamin 1 is enriched in synapses whereas dynamin 2 is ubiquitous and dynamin 3 is expressed in brain and testis. Neuronal dynamin 1 is phosphorylated by protein kinase C and dephosphorylated by calcineurin during an action potential in the nerve terminal. It is possible that the dephosphorylation provides a trigger for endocytosis.

#### Selected References for 115 302

Evidence for a Clathrin-independent mode of endocytosis at a continuously active sensory synapse.

Fuchs M, Brandstätter JH, Regus-Leidig H

Frontiers in cellular neuroscience (2014) 8: 60. . IHC, EM; tested species: rat

Reduced dynamin-1 levels in neurons lacking MUNC18-1.
Lammertse HCA, Moro A, Saarloos I, Toonen RF, Verhage M
Journal of cell science (2022) 13522: . . WB; tested species: mouse

The first synapse in vision in the aging mouse retina. Gierke K, Lux UT, Regus-Leidig H, Brandstätter JH

Frontiers in cellular neuroscience (2023) 17: 1291054. . IHC; tested species: mouse

Quantitative Fluorescent in situ Hybridization Reveals Differential Transcription Profile Sharpening of Endocytic Proteins in Cochlear Hair Cells Upon Maturation.

Huang G, Eckrich S

Frontiers in cellular neuroscience (2021) 15: 643517. . IHC; tested species: mouse

Modes and regulation of endocytic membrane retrieval in mouse auditory hair cells.

Neef J, Jung S, Wong AB, Reuter K, Pangrsic T, Chakrabarti R, Kügler S, Lenz C, Nouvian R, Boumil RM, Frankel WN, et al. The Journal of neuroscience: the official journal of the Society for Neuroscience (2014) 343: 705-16. IHC; tested species: mouse

#### **Selected General References**

Dynamin and its role in membrane fission.

Hinshaw JE

Annual review of cell and developmental biology (2000) 16: 483-519. .

Accessory factors in clathrin-dependent synaptic vesicle endocytosis.

Slepnev VI, De Camilli P

Nature reviews. Neuroscience (2000) 13: 161-72...

Sequential steps in clathrin-mediated synaptic vesicle endocytosis.

Brodin L, Löw P, Shupliakov O

Current opinion in neurobiology (2000) 103: 312-20..

Synaptic vesicle biogenesis.

Hannah MJ. Schmidt AA. Huttner WB

Annual review of cell and developmental biology (1999) 15: 733-98.

The synaptic vesicle cycle: a cascade of protein-protein interactions.

Südhof TC

Nature (1995) 3756533: 645-53. .

Access the online factsheet including applicable protocols at https://sysy.com/product/115302 or scan the OR-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.