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# **AP180**

Cat.No. 155 002; 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 up to 1: 5000 (AP staining) IP: not tested yet ICC: not recommended (see remarks) IHC: not recommended IHC-P: not tested yet
Immunogen	Synthetic peptide corresponding to AA 279 to 297 from rat AP180 (UniProt Id: Q05140)
Reactivity	Reacts with: human (O60641), rat (Q05140), mouse (Q61548), dog. Other species not tested yet.
Specificity	Specific for AP 180.
Matching control	155-0P
Remarks	ICC: The affinity-purified antibody ( <u>cat. no. 155 003</u> ) is recommended.

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

#### Background

During neurotransmitter release synaptic vesicles fuse with the presynaptic plasma membrane. A whole protein machinery consisting of e.g. amphiphysin, clathrin, endophilin and synaptojanin is involved in the subsequent endocytotic recycling of the synaptic vesicles.

**AP 180** also known as **pp155**, **NP185**, **F1-20**, and **SNAP 91** is a clathrin binding phospho-protein and faciliates the formation of clathrin coats.

#### Selected References for 155 002

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

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

Endophilin-A coordinates priming and fusion of neurosecretory vesicles via intersectin.

Gowrisankaran S, Houy S, Del Castillo JGP, Steubler V, Gelker M, Kroll J, Pinheiro PS, Schwitters D, Halbsgut N, Pechstein A, van Weering JRT, et al.

Nature communications (2020) 111: 1266. . WB; tested species: mouse

CSPa knockout causes neurodegeneration by impairing SNAP-25 function.

Sharma M, Burré J, Bronk P, Zhang Y, Xu W, Südhof TC

The EMBO journal (2012) 314: 829-41.. WB; tested species: mouse

Endosomal sorting of readily releasable synaptic vesicles.

Hoopmann P, Punge A, Barysch SV, Westphal V, Bückers J, Opazo F, Bethani I, Lauterbach MA, Hell SW, Rizzoli SO Proceedings of the National Academy of Sciences of the United States of America (2010) 10744: 19055-60.

#### **Selected General References**

AP180 maintains the distribution of synaptic and vesicle proteins in the nerve terminal and indirectly regulates the efficacy of Ca2+-triggered exocytosis.

Bao H et al. J. Neurophysiol. (2005) PubMed:15888532

Synaptic distribution of the endocytic accessory proteins AP180 and CALM.

Yao PJ et al. J. Comp. Neurol. (2005) PubMed:15558718

High-resolution localization of clathrin assembly protein AP180 in the presynaptic terminals of mammalian neurons.

Yao PJ et al. J. Comp. Neurol. (2002) PubMed:11977118

Unusual structural organization of the endocytic proteins AP180 and epsin 1.

Kalthoff C et al. J. Biol. Chem. (2002) PubMed:11756460

Changes in synaptic expression of clathrin assembly protein AP180 in Alzheimer's disease analysed by immunohistochemistry. Yao PJ et al. Neuroscience (1999) PubMed:10579202

AP180 and AP-2 interact directly in a complex that cooperatively assembles clathrin.

Hao W et al. J. Biol. Chem. (1999) PubMed:10428863

Clathrin assembly protein AP180: primary structure, domain organization and identification of a clathrin binding site.

Morris SA et al. EMBO J. (1993) PubMed:8440257

Molecular characterization of the AP180 coated vesicle assembly protein.

Prasad K et al. Biochemistry (1988) PubMed:3191110

Access the online factsheet including applicable protocols at https://sysy.com/product/155002 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.