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# Tomosyn1

Cat.No. 183 103; Polyclonal rabbit antibody, 50 µg specific antibody (lyophilized)

# **Data Sheet**

Reconstitution/ Storage	50 $\mu g$ specific antibody, lyophilized. Affinity purified with the immunogen. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 50 $\mu l$ 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 (AP staining) IP: external data (see remarks) ICC: 1: 500 IHC: external data (see remarks) IHC-P: 1: 500
Immunogen	Recombinant protein corresponding to AA 561 to 774 from mouse Tomosyn1 (UniProt Id: Q8K400)
Reactivity	Reacts with: rat (Q9WU70), mouse (Q8K400). Other species not tested yet.
Specificity	K.O. validated PubMed: <u>24744148</u>
Remarks	IP: This antibody has been successfully applied and published for this method by customers (see application-specific references). It has not been validated using our standard protocols.  IHC: This antibody has been successfully applied for this method by our customers using mild fixation (4% PFA and 15% picric acid) according to Kirizs et al. 2014 (see gallery). It has not been validated using our standard protocol.

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

# Background

SNARE proteins play crucial roles in vesicle transport by catalyzing membrane fusion events. Several proteins like the Munc 18s and tomosyn 1 (syntaxin 1A binding protein 5) interact with the neuronal plasmalemma located SNARE protein syntaxin 1a and modulate neurotransmitter release at synaptic nerve terminals.

Tomosyn 1 contains a C-terminal synaptobrevin-like R-SNARE motif that can form a stable ternary complex with syntaxin 1A and SNAP 25.

Another isoform, tomosyn 2 (syntaxin 1A binding protein 5 like), has also been described.

# Selected References for 183 103

Tomosyn-2 is required for normal motor performance in mice and sustains neurotransmission at motor endplates. Geerts CJ, Plomp JJ, Koopmans B, Loos M, van der Pijl EM, van der Valk MA, Verhage M, Groffen AJ Brain structure & function (2015) 2204: 1971-82. . WB: KO verified

Tomosyn regulates the small RhoA GTPase to control the dendritic stability of neurons and the surface expression of AMPA receptors.

Shen W, Kilander MBC, Bridi MS, Frei JA, Niescier RF, Huang S, Lin YC

Journal of neuroscience research (2020):.. WB, IP, ICC; tested species: mouse

Tomosyn associates with secretory vesicles in neurons through its N- and C-terminal domains. Geerts CJ, Mancini R, Chen N, Koopmans FTW, Li KW, Smit AB, van Weering JRT, Verhage M, Groffen AJA PloS one (2017) 127: e0180912. . IP, ICC, EM; KO verified; tested species: mouse

The ubiquitin-proteasome system functionally links neuronal Tomosyn-1 to dendritic morphology.

Saldate JJ, Shiau J, Cazares VA, Stuenkel EL

The Journal of biological chemistry (2018) 2937: 2232-2246. . WB, IP, ICC; KD verified; tested species: rat

Genetic ablation of synaptotagmin-9 alters tomosyn-1 function to increase insulin secretion from pancreatic  $\beta$ -cells improving

Rahman MM, Pathak A, Schueler KL, Alsharif H, Michl A, Alexander J, Kim JA, Bhatnagar S

FASEB journal: official publication of the Federation of American Societies for Experimental Biology (2023) 378: e23075. . WB, ICC; tested species: mouse

Tomosyn affects dense core vesicle composition but not exocytosis in mammalian neurons. Subkhangulova A, Gonzalez-Lozano MA, Groffen AJA, van Weering JRT, Smit AB, Toonen RF, Verhage M eLife (2023) 12:.. WB, ICC; KO verified; tested species: mouse

Dynamic Partitioning of Synaptic Vesicle Pools by the SNARE-Binding Protein Tomosyn. Cazares VA, Njus MM, Manly A, Saldate JJ, Subramani A, Ben-Simon Y, Sutton MA, Ashery U, Stuenkel EL

The Journal of neuroscience: the official journal of the Society for Neuroscience (2016) 3644: 11208-11222. . WB. ICC: KD verified; tested species: rat

Mapping localization of 21 endogenous proteins in the Golgi apparatus of rodent neurons. van Bommel DM, Toonen RF, Verhage M

Scientific reports (2023) 131: 2871. . ICC; tested species: mouse

Actions of Rab27B-GTPase on mammalian central excitatory synaptic transmission. Arias-Hervert ER, Xu N, Njus M, Murphy GG, Hou Y, Williams JA, Lentz SI, Ernst SA, Stuenkel EL Physiological reports (2020) 89: e14428. . WB; tested species: mouse

#### **Selected General References**

Tomosyn negatively regulates CAPS-dependent peptide release at Caenorhabditis elegans synapses. Gracheva EO et al. J. Neurosci. (2007) PubMed:17881523

Access the online factsheet including applicable protocols at https://sysy.com/product/183103 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.