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Glu-Tubulin

Cat.No. 302 011; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 μ g purified IgG, lyophilized. Albumin and azide were added for stabilization. For reconstitution add 100 μ l H ₂ 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) (see remarks) IP: not tested yet ICC: 1 : 200 up to 1 : 500 IHC: 1 : 500 IHC-P: 1 : 1000
Clone	1D5
Subtype	IgG1 (κ light chain)
Immunogen	Synthetic peptide corresponding to residues near the c-terminus of human Glu-a- tubulin. (UniProt Id: Q71U36)
Epitop	AA 448 to 450 from human Glu-α-tubulin (UniProt Id: Q71U36)
Reactivity	Reacts with: human (Q71U36), rat (P68370), mouse (P68369), zebrafish, eukaryotes, other vertebrates. Other species not tested yet. Detects also cilia of Paramecium.
Specificity	Specific for detyrosinated α-tubulin (glu-tubulin) and polyglutamylated tubulin (also β-tubulin). No cross reaction to tyrosinated tubulin.
Remarks	WB : Tween 20 (other detergents not yet tested) should not be added to the blocking and washing solution. It greatly diminishes the intensity of the signal.

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

Background

Microtubules are involved in a wide variety of cellular activities ranging from mitosis and transport events to cell movement and the maintainance of cell shape.

Tubulin itself is a globular protein which consists of two polypeptides, **α-tubulin** and β-tubulin. α- and β-tubulin dimers are assembled to 13 protofilaments that form a microtubule of 22 nm diameter. Tyrosine ligase ads a C-terminal tyrosin to monomeric α-tubulin.

Assembled microtubules can again be detyrosinated by a cytoskeleton associated carboxypeptidase. Detyrosinated α-tubulin is referred to as **Glu-α-tubulin**. Another post-translational modification of detyrosinated α-tubulin is C-terminal polyglutamylation which is characteristic for microtubules in neuronal cells and the mitotic spindle. A third variant of detyrosinated α-tubulin is **Δ2-tubulin** which lacks the C-terminal glutamic acid. It cannot be tyrosinated by tyrosine ligase and is one of the dominant α-tubulin isoforms in neurons.

Selected References for 302 011

Tubulin detyrosination promotes monolayer formation and apical trafficking in epithelial cells. Zink S, Grosse L, Freikamp A, Bänfer S, Müksch F, Jacob R Journal of cell science (2012) 125Pt 24: 5998-6008. . **WB, ICC**

Turnover of the carboxy-terminal tyrosine of alpha-tubulin and means of reaching elevated levels of detyrosination in living cells.

Wehland J, Weber K

Journal of cell science (1987) 88 (Pt 2): 185-203. . WB, ICC

Monocilia on chicken embryonic endocardium in low shear stress areas. Van der Heiden K, Groenendijk BC, Hierck BP, Hogers B, Koerten HK, Mommaas AM, Gittenberger-de Groot AC, Poelmann RE Developmental dynamics : an official publication of the American Association of Anatomists (2006) 2351: 19-28. . **IHC**

SF-Assemblin genes in Paramecium: phylogeny and phenotypes of RNAi silencing on the ciliary-striated rootlets and surface organization.

Nabi A, Yano J, Valentine MS, Picariello T, Van Houten JL Cilia (2019) 8: 2. . **ICC**

The Ciliary Protein IFT57 in the Macronucleus of Paramecium. Shi L, Koll F, Arnaiz O, Cohen J The Journal of eukaryotic microbiology (2017) : . . **ICC**

BDNF/trkB Induction of Calcium Transients through Cav2.2 Calcium Channels in Motoneurons Corresponds to F-actin Assembly and Growth Cone Formation on β 2-Chain Laminin (221). Dombert B, Balk S, Lüningschrör P, Moradi M, Sivadasan R, Saal-Bauernschubert L, Jablonka S Frontiers in molecular neuroscience (2017) 10: 346. **ICC; tested species: mouse**

Kif26b controls endothelial cell polarity through the Dishevelled/Daam1-dependent planar cell polarity-signaling pathway. Guillabert-Gourgues A, Jaspard-Vinassa B, Bats ML, Sewduth RN, Franzl N, Peghaire C, Jeanningros S, Moreau C, Roux E, Larrieu-Lahargue F, Dufourcq P, et al. Molecular biology of the cell (2016) 276: 941-53. . **ICC**

Reduction of meckelin leads to general loss of cilia, ciliary microtubule misalignment and distorted cell surface organization. Picariello T, Valentine MS, Yano J, Van Houten J Cilia (2014) 31: 2. . ICC

Mechanisms for axon maintenance and plasticity in motoneurons: alterations in motoneuron disease. Jablonka S, Dombert B, Asan E, Sendtner M Journal of anatomy (2014) 2241: 3-14. . **ICC**

Access the online factsheet including applicable protocols at <u>https://sysy.com/product/302011</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.