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Product Information

Anti-y-Tubulin (DQ-19) produced in rabbit, IgG fraction of antiserum

Catalog Number T3195

Product Description

Anti- γ -Tubulin (DQ-19) is produced in rabbit using a synthetic peptide corresponding to the C-terminus of human γ -tubulin (amino acids 433-451, with N-terminally added lysine), conjugated to KLH (keyhole limpet hemocyanin) as immunogen. This sequence is identical in rat γ -tubulin and highly conserved in human γ -tubulin-2, and dog and Xenopus γ -tubulin (\geq 90% identity). The product is specific for γ -tubulin and not found in other members of the tubulin family such as α -, β -, and ϵ -tubulins. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti- γ -Tubulin (DQ-19) recognizes human, rat, and *Xenopus* γ -tubulin (48 kDa). Applications include immunoblotting and immunocytochemistry (immunofluorescence staining). Staining of γ -tubulin in immunoblotting is specifically inhibited with γ -tubulin immunizing peptide (human, amino acids 433-451, with N-terminally added lysine).

γ-Tubulin (48 kDa) is a widely expressed and highly conserved protein within the microtubule organizing centers (MTOCs) or centrosome in eukaryotic cells. 1 It is a member of the tubulin superfamily of proteins which includes α - and β -tubulin and the newly discovered centrosomal-associated proteins, $\delta\text{-}$ and $\epsilon\text{-tubulin.}^{1,\,2}$ The microtubule cytoskeleton consists of a dynamic. highly polarized network of microtubules filaments, microtubule-associated proteins, microtubule motors, and microtubule-organizing proteins. The proper organization of microtubules is essential for cell division and chromosome segregation, directed cell movement, interphase cytoplasmic organization, and other cytoskeletal functions. Microtubules are complex polymers of α -tubulin/ β -tubulin heterodimers. Centrosomes nucleate the assembly of microtubules and establish the polarity of microtubules. γ-Tubulin has an essential role in microtubule nucleation by the centrosomes.³⁻⁹ It does not polymerize with α-tubulin/β-tubulin, but instead is localized to the centrosome and to the cytoplasm. 1,4-6

γ-Tubulin is found as part of a large protein complex containing at least five other proteins, and has a shape of a ring (γ -tubulin ring complex, γ -TuRC) that is roughly the same diameter as a microtubule. 9-13 It binds the microtubule minus ends and is responsible for mediating the link between microtubules and the centrosome. 1,6 γ -Tubulin binds to the β -tubulin half of the tubulin molecule, thus establishing the polarity of a microtubule, leaving the α -tubulin half exposed at the plus end. Its abundance is less than 1% of the level of either α - or β -tubulin. ⁵ γ -Tubulin shares 28-32% identity with α -tubulin from various organisms, 32-36% identity with $\beta\text{-tubulins}$ and 29-30% identity with $\delta\text{-}$ and $\epsilon\text{-}$ tubulins. Some regions including those thought to be involved in GTP binding are highly conserved among α -, β-, γ -, δ-, and ε-tubulins.²

Reagent

Supplied in a solution of 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

Product Profile

<u>Immunoblotting</u>: a minimum working dilution of 1:1,000 is determined using a whole cell extract of the human epidermoid carcinoma A431 cell line.

<u>Indirect immunofuorescent staining</u>: a minimum working dilution of 1:500 is determined using methanol-acetone-fixed chicken fibroblasts.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

- 1. Oakley, B.R., Trends Cell Biol., 2, 1 (1992).
- 2. Chang, P., and Stearns, T., *Nature Cell Biol.*, **2**, 30 (2000).
- 3. Oakley, C.E., and Oakley, B.R., *Nature*, **338**, 662 (1989).
- 4. Oakley, B.R., Cell, 61, 1289 (1990).

- 5. Stearns, T., et al., Cell, 65, 825 (1991).
- 6. Zheng, Y., et al., Cell, 65, 817 (1991).
- 7. Joshi, H.C., et al., Nature, 356, 80 (1992).
- 8. Felix, M.A., et al., J. Cell Biol., 124, 19 (1994).
- 9. Stearns, T., and Kirschner, M., Cell, 76, 623 (1994).
- 10. Oakley, B.R., Nature, 378, 555 (1995).
- 11. Zheng, Y., et al., *Nature*, **378**, 578 (1995).
- 12. Moritz., M., et al., Nature, 378, 638 (1995).
- 13. Oegema, K., et al., J. Cell Biol., 144, 721 (1999).

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