



Product Information

MONOCLONAL ANTI-POLYGLUTAMYLATED TUBULIN, Clone B3

Purified Mouse Immunoglobulin

Product Number **T 9822**

Product Description

Monoclonal Anti-Polyglutamylated Tubulin (mouse IgM isotype) is derived from the B3 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a purified sea urchin (*Lytechinus pictus*) sperm axonemal proteins.¹ The isotype is determined using Sigma ImmunoType™ Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2).

The epitope recognized by the antibody is localized in the C-terminal region of α - and β -tubulins (the glutamylated motif at amino acid 445-457 of α -tubulin).¹ The product is useful in ELISA,¹ immunoblotting¹ (~ 50 kDa, possibly a doublet of α - and β -tubulins) and immunocytochemistry (4% paraformaldehyde-1% Triton X-100¹ and methanol-acetone). The product is also useful for studies involving the inhibition of flagellar motility in permeabilized sperm models.¹ Species reactivity includes: human,¹ monkey, bovine, rabbit, rat, mouse,¹ chicken, sea urchin (*Lytechinus pictus*, and three others),¹ and dinoflagellate (*Oxyrrhis marina*).¹

A typical eukaryotic centrosome consists of a pair of centrioles, constructed of microtubules and surrounded by an electron dense amorphous cloud of pericentriolar material. Many cellular functions are dependent on the proper organization of microtubules, since they are essential for mitosis, meiosis, some forms of organellar movement, intracellular transport, flagellar movement, and other cytoskeletal functions.² Thus, temporal and spatial regulation of microtubule assembly is critical for the correct assembly of the mitotic apparatus and of the cytoplasmic microtubule array.

The major building block of microtubules is tubulin, an intracellular cylindrical filamentous structure that is present in almost all eukaryotic cells. Except in the simplest eukaryotes, tubulin (100 kDa) exists in all cells as a heterodimer of two similar but non-identical polypeptides (~ 55 kDa each), designated α and β , that assemble into microtubules. Within either family of

α/β tubulin heterodimers, individual subunits diverge from each other (both within and across species) at less than 10% of the amino acid positions.³ The most extreme diversity is localized to the carboxyl-terminal 15 residues. Both α - and β -tubulins consist of various isotypes. In addition, both undergo post-translational modifications, including acetylation, phosphorylation, dephosphorylation, polyglutamylation, and polyglycylation.⁴

Polyglutamylation of tubulin consists of the addition of a lateral chain of glutamyl units on the C-terminal region of tubulin polypeptides. This modification was shown to regulate the interaction between tau, one of the major neuronal microtubule associated proteins, and tubulin.¹ Furthermore, it was shown that injection of monoclonal antibodies specific to the polyglutamylated form of tubulin into HeLa cells caused the disappearance of centrioles, an organelle known to have the most stable microtubules.⁵

The detection, localization, and characterization of proteins involved in microtubule function is fundamental to the understanding of mitosis, meiosis, organellar and flagellar movement, intracellular transport, and cytoskeletal functions. Antibodies reacting specifically with the modified forms of α - and β -tubulin isotypes serve as essential tools in the detection and study of the functional significance of these molecules.

Reagent

Monoclonal Anti-Polyglutamylated Tubulin is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 1% bovine serum albumin and 15 mM sodium azide.

Antibody concentration: Approx. 2 mg/ml

Precautions and Disclaimer

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS 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 is not recommended. Storage in frost-free freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

For immunoblotting, a minimum working antibody concentration of 0.5-1 µg/ml is recommended using a cytosolic fraction of rat brain extract.

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

References

1. Gagnon, C., et al., J. Cell Sci., **109**, 1545 (1996).
2. Oakley, B.R., Trends Cell Biol., **2**, 1 (1992).
3. Joshi, H.C., and Cleveland, D.W., Cell Motil. Cytoskel., **16**, 159 (1990).
4. Roach, M.C., et al., Cell Motil. Cytoskel., **39**, 273 (1998).
5. Bobinnec, Y., et al., J. Cell Biol. **143**, 1575, (1998).

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