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# **ProductInformation**

MONOCLONAL ANTI-b-TUBULIN IV CLONE ONS.1A6 Mouse Ascites Fluid

Product Number T7941

## **Product Description**

Monoclonal Anti- $\beta$ -Tubulin IV (mouse IgG1 isotype) is derived from the ONS.1A6 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a BALB/c mouse immunized with a synthetic peptide corresponding to the C-terminal sequence of  $\beta$ -tubulin isotype IV coupled to BSA. 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).

Monoclonal Anti- $\beta$ -Tubulin IV specifically recognizes an epitope located in the C-terminal sequence of  $\beta$ -tubulin isotype IV of human, <sup>1,2</sup> bovine, <sup>3-6</sup> hamster, rat, <sup>1</sup> mouse<sup>1,3</sup> and chicken. No reactivity with other tubulin isotypes is observed. The antibody may be used for localization of  $\beta$ -tubulin isotype IV by ELISA<sup>1</sup>, and in cultured human and animal cells by immunoblotting<sup>1,3,5,6</sup> (approx. 55 kDa, and possibly also the 100 kDa dimer), immunohistochemistry (10% neutral buffered formalinalcoholic formalin-xylene, paraffin-embedded<sup>2</sup>, or 1-2% glutaraldehyde-4% formaldehyde fixed, LR Gold embedded<sup>3</sup>) and immunoaffinity purification. <sup>1,4,6</sup>

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. 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 exists in all cells as a 100 kDa protein, a heterodimer of two similar but not identical polypeptides (approx. 55 kDa each), designated  $\alpha$  and  $\beta$ , that

assembles into microtubules. Within either family of  $\alpha/\beta$ tubulin heterodimer, individual subunits diverge from each other (both within and across species) at less than 10% of the amino acid positions. 8 The most extreme diversity is localized to the carboxy-terminal 15 residues. Both  $\alpha$ - and  $\beta$ -tubulins consist of various isotypes. In addition, both undergo post-translational modifications, including acetylation, phosphorylation detyrosination, polyglutamylation, and polyglycylation.<sup>2</sup> For β-tubulin, six evolutionarily conserved isotypes were identified (designated  $\beta_{l}$ - $\beta_{Vl}$ ). These are nearly absolutely conserved in the subunits utilized in the same cell types of different species, with the exception of the hematopoietic β-tubulin, which is highly divergent in sequence and which is not conserved between species. Research has been centered around the hypothesis that these β-tubulin isotypes contribute to unique functional properties, since the different isotypes of tubulin differ from each other in their ability to polymerize into microtubules. <sup>9</sup> The most complex pattern of isotype distribution in tissues is seen in the vertebrate  $\beta$ -tubulins. <sup>10</sup> In mammals and birds,  $\beta_I$  is constitutive and found in most tissues.  $\beta_{II}$  is found in many tissues, but largely in the brain; its synthesis increases in regeneration and development of neurons. B<sub>III</sub> is found in the brain and in dorsal root ganglia: it appears to be localized to neurons, where its expression seems to increase during axonal outgrowth. It is also found in Sertoli cells of the testis, and in certain tumors of non-neural origin, such as lymphoma, squamous cell carcinoma, and malignant melanoma, but does not appear to be expressed in those tissues before transformation.  $\beta_{IV}$  is somewhat complex: in mammals, it exists at two subtypes, differing from each other at 10 positions.  $\beta_{IVa}$  is brain specific, whereas  $\beta_{IVb}$ is ubiquitous, and both appear to be constitutive. In chickens, there is only one form of  $\beta_{IV}$  that is expressed at low levels in many tissues, but is the major  $\beta$  isotype in the testis.  $\beta_V$  in chickens is apparently ubiquitous outside of the brain, and is also expressed in a variety of cultured mammalian cells.  $\beta_{\text{VI}}$  is apparently restricted

to hematopoietic tissues, being expressed in chicken erythrocytes and in mammalian platelets, spleen, bone marrow, and other blood-forming tissues. 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  $\alpha$ - and  $\beta$ -tubulin isotypes serve as an essential tool in the detection of the presence and functional significance of these molecules in various cellular settings.

## Reagents

Monoclonal Anti-β-Tubulin IV is supplied as ascites fluid containing 15 mM sodium azide.

## **Precautions and Disclaimer**

Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution.

Consult the MSDS for information regarding hazardous and safe handling practices.

#### Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. 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**

A minimum working dilution of 1:400 is determined by immunoblotting, using cultured human breast adenocarcinoma cell line MCF-7.

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

#### References

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