

## Product Information

### Anti- $\gamma$ -Tubulin (AK-15)

produced in rabbit, IgG fraction of antiserum

Product Number **T3320**

#### Product Description

Anti- $\gamma$ -Tubulin (AK-15) is produced in rabbit using as immunogen a synthetic peptide corresponding to the C-terminus of *Xenopus*  $\gamma$ -tubulin with N-terminal added cys-gly, conjugated to maleimide-activated KLH (keyhole limpet hemocyanin). This sequence is highly conserved in human, dog, and rat  $\gamma$ -tubulin (87% identity). It is specific for  $\gamma$ -tubulin and not found in other members of the tubulin family such as  $\alpha$ -,  $\beta$ -,  $\delta$ -, and  $\epsilon$ -tubulins. Whole antiserum is purified to provide an IgG fraction of antiserum.

Anti- $\gamma$ -Tubulin (AK-15) recognizes human, rat, and *Xenopus*  $\gamma$ -tubulin (48 kDa). Applications include immunoblotting and immunocytochemistry (immunofluorescence staining). Staining of  $\gamma$ -tubulin in immunoblotting is specifically inhibited by the  $\gamma$ -tubulin immunizing peptide.

$\gamma$ -Tubulin (48 kDa) is a widely expressed and highly conserved protein within the microtubule organizing centers (MTOCs) or centrosome in eukaryotic cells.<sup>1</sup> It is a member of the tubulin superfamily of proteins which includes  $\alpha$ - and  $\beta$ -tubulin, and the newly discovered centrosomal-associated proteins,  $\delta$ - and  $\epsilon$ -tubulins.<sup>1,2</sup> 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.<sup>1</sup> Microtubules are complex polymers of  $\alpha$ -tubulin/ $\beta$ -tubulin heterodimers. Centrosomes nucleate the assembly of microtubules and establish the polarity of microtubules.  $\gamma$ -Tubulin has an essential role in microtubule nucleation by the centrosomes.<sup>3-9</sup> It does not polymerize with  $\alpha$ -tubulin/ $\beta$ -tubulin, but instead is localized to the centrosome and to the cytoplasm.<sup>1,4-6</sup>

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

#### Reagent

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

#### Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

#### Storage/Stability

Store at  $-20\text{ }^{\circ}\text{C}$ . For continuous use, store at  $2\text{--}8\text{ }^{\circ}\text{C}$  for up to one month. For prolonged storage, freeze in working aliquots at  $-20\text{ }^{\circ}\text{C}$ . 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

Immunoblotting: a minimum antibody working dilution of 1:1,000 is determined using a whole cell extract of the human epitheloid carcinoma HeLa cell line.

Immunoblotting: a minimum working antibody dilution of 1:5,000 is determined using *Xenopus* whole egg extract.

Indirect immunofluorescence: a minimum working antibody dilution of 1:1,000 is determined using methanol-acetone-fixed rat fibroblasts (Rat-1 cell line).

Note: In order to obtain best results in various techniques and preparations, it is recommended to determine optimal working dilutions by titration.

### 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).

VS, MG,KAA,PHC,MAM 02/19-1