

Product No. S-0664

Lot 096H4854

Monoclonal Anti-Syntaxin

Mouse Ascites Fluid

Clone HPC-1

Monoclonal Anti-Syntaxin (mouse IgG1 isotype) is derived from the hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. A synaptosomal plasma-membrane fraction from adult rat hippocampus was used as the immunogen.¹ The isotype is determined using Sigma ImmunoType™ Kit (Sigma Stock No. ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Sigma Stock No. ISO-2). The product is provided as ascites fluid with 0.1% sodium azide (see MSDS)* as a preservative.

Specificity

Monoclonal Anti-Syntaxin recognizes an epitope located in the extracellular domain of the 35 kD membrane protein syntaxin, expressed predominantly in neuronal tissues.^{2,3} The antibody has also been used to isolate syntaxin cDNA clones by screening a λ gt 11 library from rat hippocampus.³ In hippocampus, the antibody recognizes the stratum radiatum, the layer formed by the fibers of dentate granule cells, as they synapse onto hippocampal pyramidal cells. In rat retina, it stains the membrane of the amacrine cell bodies and inner plexiform layer. It recognizes some neurons early in development and can be detected on migrating amacrine cells in embryonic retina. In immunoblotting, cross reactivity has been observed with bovine, rabbit and rat, but not with guinea pig. The product may be used to label monolayer cultures of neonatal retinal cells⁴ and frozen paraformaldehyde-fixed tissue sections.

Titer

A titer of 1:2,000 was determined by indirect immunoblotting using a crude preparation of synaptic vesicles from rat cerebral cortex.

In order to obtain best results, it is recommended that each individual user determine their working dilution by titration assay.

Description

Chemical neurotransmitters are stored within the nerve terminal in synaptic vesicles that are often found associated with cytoskeletal components or the presynaptic plasma membrane.⁵ Upon nerve stimulation, activation of voltage-gated Ca²⁺ channels in the nerve terminal plasma membrane results in an influx of Ca²⁺. The increase in cytosolic Ca²⁺ concentration triggers the fusion of a portion of the synaptic vesicle population with the presynaptic plasma membrane, resulting in neurotransmitter release. The docking and subsequent fusion of synaptic vesicles with the presynaptic plasma membrane occur at a restricted, morphologically distinct domain known as the active zone. The process of synaptic vesicle docking with the presynaptic membrane may represent the assembly of a pre-fusion complex that is likely to include components of each membrane. Three synaptic vesicle membrane proteins, synaptotagmin, synaptophysin,⁶ and synapsin I, exhibit properties suggestive of a role in synaptic vesicle docking or fusion. Syntaxin (also cited as HPC-1 antigen),^{5,7} a 35 kD molecule with carboxyl-terminal membrane anchor, is a synaptic protein identified by its ability to interact with the synaptic vesicle protein synaptotagmin. It has been implicated in docking at synaptic vesicles of presynaptic neurotransmitter release sites.^{5,7,8} The molecular machinery for secretion seems to be conserved from yeast to neurons, since 3 genes have been identified in yeast, that encode proteins with carboxyl-terminal membrane anchor and displaying significant homology to syntaxin, primarily over a 70 amino acid segment near the membrane anchor.^{8,9} In addition, epimorphin, a protein expressed in mesenchymal cells and regulates the morphogenesis of adjacent epithelial cells, is also closely related (63% identical) to syntaxins A and B.⁹ Monoclonal antibodies reacting specifically against syntaxin are useful for studies on the molecular machinery of secretion, cellular heterogeneity and the development of the central nervous system.

Uses

Monoclonal Anti-Syntaxin may be used for the localization of syntaxin using various immunochemical assays such as immunoblotting and immunohistochemistry.

Storage

For continuous use, store at 2-8°C. For extended storage, the solution may be frozen 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.

*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.

References

1. Barnstable, C., et al., *Dev. Brain Res.*, **20**, 286 (1985).
2. Barnstable, C., "Monoclonal Antibodies in Studies of Neuronal Diversity", in: *Molecular Biology of the Human Brain*, Jones, E., (ed.), Alan R. Liss, New York, pp. 27 (1988).
3. Inoue, A., et al., *J. Biol. Chem.*, **267**, 10613 (1992).
4. Akagawa, K., and Barnstable, C., *Brain Res.* **383**, 110 (1986).
5. Bennet, M., et al., *Science*, **257**, 255 (1992).
6. Devoto, S., and Barnstable, C., *Ann. N.Y. Acad. Sci.*, **493**, 493 (1987).
7. Barinaga, M., (ed.), *Science*, **260**, 487 (1993).
8. Bennet, M., and Scheller, R., *Proc. Natl. Acad. Sci. (USA)*, **90**, 2559 (1993).
9. Hirai, Y., et al., *Cell*, **73**, 425 (1993).