

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

# **ProductInformation**

# Anti-Munc-18-1

Developed in Rabbit, IgG Fraction of Antiserum

Product Number M 2694

# **Product Description**

Anti-Munc-18-1 is developed in rabbit using a synthetic peptide corresponding to amino acids 577-594 located at the C-terminus of rat Munc-18-1 conjugated to KLH as immunogen. This sequence is identical in mouse, chicken, bovine and human Munc-18-1. It does not share homology with the Munc-18-2 and Munc-18-3 isoforms. 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-Munc-18-1 recognizes specifically Munc-18-1 (67 kDa). Applications include the detection of Munc-18-1 by immunoblotting and immunofluorescence. Staining of the Munc-18-1 band in immunoblotting is specifically inhibited with the Munc-18-1 immunizing peptide (rat, amino acids 577-594).

Synaptic transmission involves the regulated exocytosis of vesicles containing neurotransmitters. Munc-18-1 (syntaxin-binding protein 1, STXBP1, rb-Sec-1, n-Sec-1, 67 kDa) is an abundant neuronal protein that tightly binds to the synaptic fusion protein syntaxin and functions in synaptic vesicle exocytosis. 1-5 Munc-18 is highly homologous to the c. elegans unc-18 gene product, and weakly related to the yeast sec-1, sly-1, and slp1 genes. 1,2 There are three known Munc-18 isoforms in mammalian cells; Munc-18-1 (Munc-18a), Munc-18-2 (Munc-18b) and Munc-18-3 (Munc-18c). 6,7 Munc-18-1 is primarily expressed in neuronal tissues, whereas Munc-18-2 and Munc-18-3 are ubiquitously expressed. Munc-18-1 shows only 62% amino acid sequence homology with Munc-18-2 and 51% sequence identity with Munc-18-3. Both Munc-18-1 and -2 isoforms bind tightly to syntaxins 1A, 2, and 3 but not to syntaxin 4. Mint1 localizes Munc-18-1 to the plasma membrane, where the cytoplasmic tail of neurexins binds Munc-18-1 through a multiprotein complex with Mint1.8 Syntaxin 1A has been implicated as a key component in SNARE assembly and synaptic vesicle docking. Munc-18-1 has been suggested to modulate neurotransmission by

associating with syntaxin 1. Munc-18 binds to both syntaxin 1A and 1B isoforms, inhibiting the binding of t-SNARE synaptobrevin/VAMP and SNAP25 to syntaxin 1 and preventing SNARE assembly. Munc-18 is required for neurotransmitter secretion from synaptic vesicles throughout development and has been implicated in axonal branching in hippocampal neurons. 9,10

### Reagent

The product is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

#### **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 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 working dilution of 1:2,000-1:4,000 is determined by immunoblotting, using a rat brain extract (S1 fraction).

A working dilution of 1:2,000-1:4,000 is determined by immunoblotting using a whole cell extract of the rat pheochromocytoma PC12 cell line.

A working dilution of 1:250-1:500 is determined by immunofluorescence staining of mouse fibroblast NIH3T3 cell line.

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

#### References

- Hata, Y., et al., Synaptic vesicle fusion complex contains unc-18 homologue bound to syntaxin., Nature, 366, 347-351 (1993).
- Garcia, E.P., et al., A rat brain Sec1 homologue related to Rop and UNC18 interacts with syntaxin., Proc. Natl. Acad. Sci. USA, 91, 2003-2007 (1994).
- Pevsner, J., et al., n-Sec1: a neural-specific syntaxin-binding protein., Proc. Natl. Acad. Sci. USA, 91, 1445-1449 (1994).
- Swanson, D.A., et al., Identification and characterization of the human ortholog of rat STXBP1, a protein implicated in vesicle trafficking and neurotransmitter release., Genomics, 48, 373-376 (1998).
- 5. Jahn, R., Sec1/Munc18 proteins: mediators of membrane fusion moving to center stage., Neuron, **27**, 201-204 (2000).

- 6. Tellam, J.T., et al., Molecular identification of two novel Munc-18 isoforms expressed in non-neuronal tissues .,J. Biol. Chem., **270**, 5857-5863 (1995).
- 7. Hata, Y., and Südhof, T.C., A novel ubiquitous form of Munc-18 interacts with multiple syntaxins. Use of the yeast two-hybrid system to study interactions between proteins involved in membrane traffic., J. Biol. Chem., **270**, 13022-13028 (1995).
- 8. Biederer, T., and Südhof, T.C., Mints as adaptors. Direct binding to neurexins and recruitment of munc18., J. Biol. Chem., **275**, 39803-39806 (2000).
- 9. Verhage, M., et al., Synaptic assembly of the brain in the absence of neurotransmitter secretion., Science, **287**, 864-869 (2000).
- 10. Steiner, P., et al., Overexpression of neuronal Sec1 enhances axonal branching in hippocampal neurons., Neuroscience, **113**, 893-905 (2002).

ER,MCT,PHC 03/05-1