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-MAGI-1
Developed in Rabbit
IgG Fraction of Antiserum

Product Number **M 5691**

Product Description

Anti-MAGI-1 is developed in rabbit using a synthetic peptide corresponding to amino acids 282-296 of MAGI-1 with a carboxy terminal added cysteine residue, conjugated to maleimide-activated KLH as immunogen. The sequence is conserved in human and mouse. Whole antiserum is fractionated and further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-MAGI-1 specifically recognizes rat MAGI-1 by immunoblotting (approx. 170, 130, and 120 kDa), and does not cross react with MAGI-2 or MAGI-3. Staining of MAGI-1 in immunoblotting is specifically inhibited by the immunizing peptide.

The MAGUK (Membrane-Associated Guanylate Kinase) family of proteins is characterized by the presence of multi-PDZ and SH3 domains, and a single region of homology to *Saccharomyces cerevisiae* guanylate kinase (GuK) domain. Its prototypic member is PSD95.[1] All MAGUKs studied to date localize to regions of cell-cell contact such as tight junctions in epithelial cells and synaptic junctions in neurons and are believed to be involved in the assembly of multi-protein complexes via their protein-protein interaction domains.[2] Three novel, closely related MAGUK proteins were isolated by means of different screening assays, and named MAGI-1/BAP1, MAGI-2/ S-SCAM /ARIP, and SLIPR/MAGI-3. Similar to other MAGUK family members, MAGI-1, -2, and -3 are characterized by the presence of multi-PDZ domains, and a guanylate kinase domain. Their unique feature consists on WW domains replacing the characteristic SH3 domains present in other MAGUK proteins.[3-8]

MAGI-1 was first identified in mouse as a protein interacting with K-RasB.[3] Three isoforms were identified, and named MAGI-1a, MAGI-1b, and MAGI-1c, of 1139, 1171, and 1374 amino acids respectively. The isolation of a growing number of MAGI-1 binding partners, together with the determination of the subcellular and tissue distribution of the different isoforms, are paving the way towards elucidation of its function.[9-11] β-catenin, mNET1 and actin-binding proteins act as binding partners of MAGI-1. In epithelia, MAGI-1 is localized at tight junctions.[9] Interestingly, MAGI-1c is localized to the nucleus, suggesting that MAGI-1 may participate in the transmission of regulatory signals from the cell surface to the nucleus.[3] Antibodies reacting specifically with MAGI-1 may be useful in studying the expression and function of the protein, as well as for distinguishing between MAGI-1, -2, and -3.

Reagent

Anti-MAGI-1 is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

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 dilution of 1:500 is recommended using rat brain extracts.

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

Procedure

For immunoblotting of rat brain extracts, we strongly recommend the addition of 5% non-fat dry milk in the blocking and antibody dilution solutions, as indicated in the following protocol.

All incubation steps should be performed at room temperature**.**

1. Separate proteins of fresh rat brain extracts from the sample extract using a standard sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) protocol.
2. Transfer proteins from the gel to nitrocellulose membrane.
3. Block the membrane for at least 60 minutes using a solution of PBS containing 5% non-fat dry milk (Product No. P 4739; or: PBS, Product No. D 8537, and non-fat dry milk, Product No. M 7409).
4. Wash the membrane three times for 10 minutes each in PBS containing 0.05% TWEEN 20^{R} (Product No. P 3563).
5. For at least 60 minutes, incubate the membrane with an optimized concentration of Anti-MAGI-1, diluted in PBS containing 0.05% TWEEN^{R} 20 and 5% non-fat dry milk for at least 60 minutes.
6. Wash the membrane three times for 10 minutes each in PBS containing 0.05% TWEEN 20^{R} at room temperature.
7. Incubate the membrane for 60 minutes with Anti-Rabbit IgG peroxidase conjugate (Product No. A 0545) as the secondary antibody, at the recommended concentration, in PBS containing 0.05% TWEEN^{R} 20 and 5% non-fat dry milk.
8. Treat the membrane with a peroxidase substrate.

References

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