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Product Information

Anti-EEA1 antibody, Mouse monoclonal
clone EEA1-N19, purified from hybridoma cell culture

Product Number **E7659**

Product Description

Monoclonal Anti-EEA1 (mouse IgG2a isotype) is derived from the hybridoma EEA1-N19 produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a synthetic peptide corresponding to a fragment of human EEA1 (GeneID: 8411), conjugated to KLH. The corresponding sequence is identical in mouse and rat. The isotype is determined by ELISA using Mouse Monoclonal Antibody Isotyping Reagents, Catalog Number ISO2. The antibody is purified from the culture supernatant of hybridoma cells grown in a bioreactor.

Monoclonal Anti-EEA1 recognizes human, rat, and mouse EEA1. The antibody may be used in several immunochemical techniques including immunoblotting (~160 kDa), immunoprecipitation and immuno-fluorescence.

Early Endosomal Antigen 1 (EEA1), a 162 kDa autoantigen associated with subacute systemic lupus erythematosus that specifically localizes to early endosomes, is a regulator of endocytic membrane docking and fusion.[1,2] EEA1 is a dimer.[3] It comprises extensive coiled-coil regions, and at its C-terminus contains a cysteine-rich zinc-finger-like domain named FYVE domain that is implicated in the specific localization of EEA1 to endosomes. This FYVE domain was found to be conserved from yeast to man among several proteins involved in intracellular trafficking.[1]

The FYVE zinc-finger domain binds specifically to the membrane lipid phosphatidylinositol 3-phosphate (PtdIns(3)P) in a Zn^{2+}-dependent manner. Anchoring of the FYVE domain to PtdIns(3)P-enriched membranes is pH-dependent due to a pair of conserved histidine residues, being enhanced by the acidic cytosolic enviroment.[4] Endosomal targeting of EEA1 also requires its binding to the active form of the small GTPase Rab5. The binding of EEA1 to PtdIns(3)P and Rab5-GTP is essential for the localization and function of EEA1 in endocytic membrane fusion.[2,5-6]

Monoclonal Anti-EEA1 may be used as an early endosome marker.

Reagent

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

Antibody concentration: ~1.0 mg/mL

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 °C. For continuous use, the product may be stored at 2–8 °C for up to one month. For extended storage, freeze at –20 °C in working aliquots. 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 working antibody concentration of 1-2 μg/mL is recommended using a whole extract of rat NRK cells.

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

References

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3. Callaghan, J., et al., *Biochem. J*., **338**, 539-543 (1999).
4. Lee, S.A., et al., *Proc. Natl. Acad. Sci. USA*, **102**, 13052-13057 (2005).
5. Simonsen, A., et al., *Nature*, **394**, 494-498 (1998).
6. Gillooly, D.J., et al., *Biochem. J.,* **355**, 249-258 (2001).

VS,DS,ST,TD,KAA,PHC,MAM 08/19-1