Store at -20°C

#48453

EEA1 (E9Q6G) Mouse mAb

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New 06/19

Entrez-Gene ID #8411
UniProt ID #Q15075

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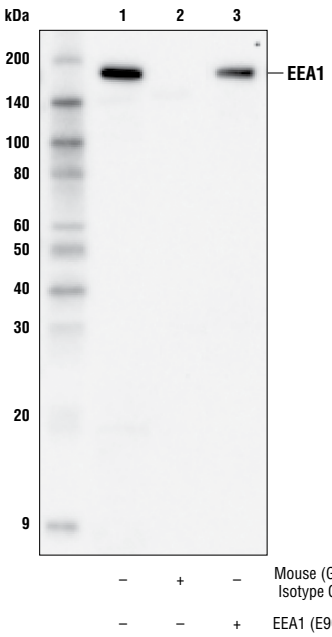
Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IF-IC	H, M, R	170 kDa	Mouse IgG1**
Endogenous			

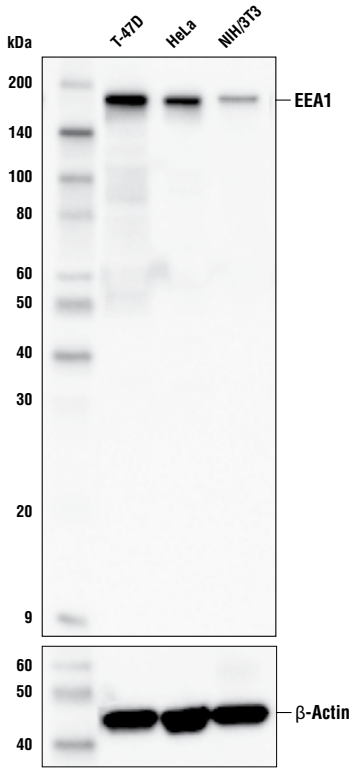
Background: EEA1 is an early endosomal marker and a Rab5 effector protein essential for early endosomal membrane fusion and trafficking (1-2). The carboxy terminus of EEA1 contains a FYVE domain which binds to phosphatidylinositol-3-phosphate (PtdIns(3)P), targeting EEA1 to early endosomes (3). The stable association of EEA1 with the endosomal membrane is regulated by PI3 kinase, Rab5 and calcium/calmodulin (4-6). Once on the membrane, EEA1 interacts with Rab5, NSF and syntaxin 13 to promote early endosomal membrane docking and fusion (7).

Specificity/Sensitivity: EEA1 (E9Q6G) Mouse mAb recognizes endogenous levels of total EEA1 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Ser70 of human EEA1 protein.



Immunoprecipitation of EEA1 protein from T-47D cell extracts. Lane 1 is 10% input, lane 2 is Mouse (G3A1) mAb IgG1 Isotype Control #5415, and lane 3 is EEA1 (E9Q6G) Mouse mAb. Western blot analysis was performed using EEA1 (C45B10) Rabbit mAb #3288.



Western blot analysis of extracts from T-47D, HeLa, and NIH/3T3 cells using EEA1 (E9Q6G) Mouse mAb (upper) and β-Actin (D6A8) Rabbit mAb #8457 (lower).

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-mouse secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunoprecipitation	1:50
Immunofluorescence (IF-IC)	1:50-1:200
Fixative:	4% Formaldehyde
Permeabilization:	0.3% Triton X-100

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

Background References:

(1) Mu, F.T. et al. (1995) *J. Biol. Chem.* 270, 13503-13511.

(2) Christoforidis, S. et al. (1999) *Nature* 397, 621-625.

(3) Gaullier, J.M. et al. (1998) *Nature* 394, 432-433.

(4) Patki, V. et al. (1997) *Proc. Natl. Acad. Sci. USA* 94, 7326-7330.

(5) Lawe, D.C. et al. (2003) *Mol. Biol. Cell* 14, 2935-2945.

(6) Simonsen, A. et al. (1998) *Nature* 394, 494-498.

(7) McBride, H.M. et al. (1999) *Cell* 98, 377-386.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween®20 at 4°C with gentle shaking, overnight.

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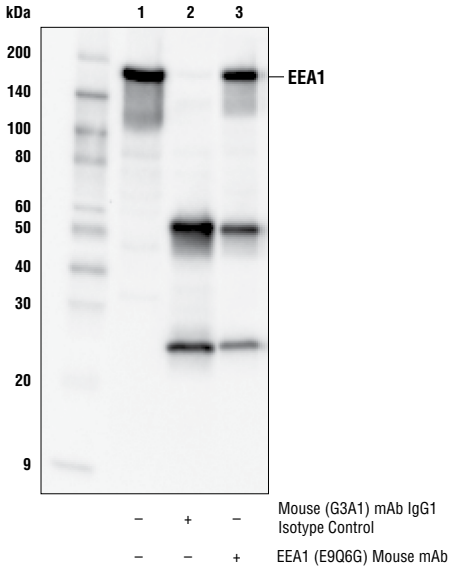
Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



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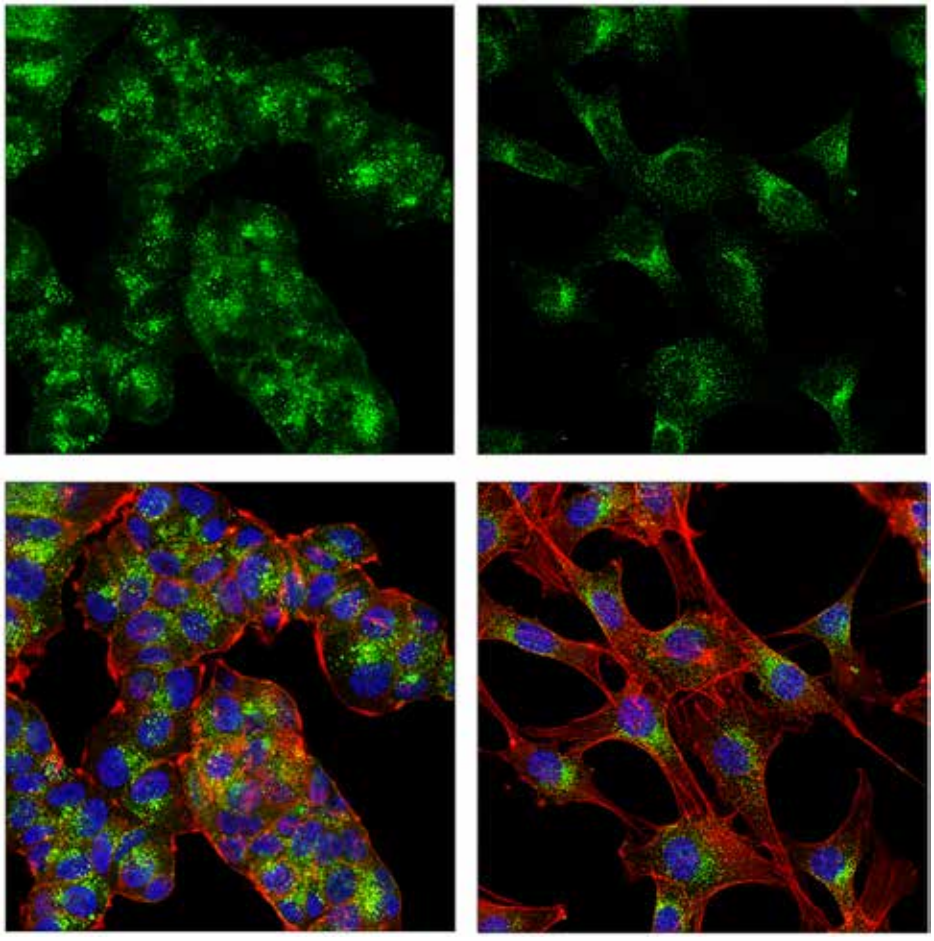

◀ *Confocal immunofluorescent analysis of T-47D cells (left) or NIH/3T3 cells (right), using EEA1 (E9Q6G) Mouse mAb (green). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red). Samples were mounted in ProLong® Gold Antifade Reagent with DAPI #8961 (blue).*