# For Research Use Only. Not for use in diagnostic procedures.



## Anti-EEA1-Alexa Fluor® 647

**CODE No.** M176-A64

CLONALITY Monoclonal SCLONE 3C10

 $\begin{array}{ll} \textbf{ISOTYPE} & \text{Mouse IgG2a } \kappa \\ \textbf{QUANTITY} & 100 \ \mu\text{L}, \ 1 \ \text{mg/mL} \end{array}$ 

SOURCE Purified IgG from hybridoma supernatant
IMMUNOGEN Human EEA1, N-terminal (synthetic peptide)
FORMURATION PBS containing 1% BSA and 0.09% NaN<sub>3</sub>.

\*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

**STORAGE** This antibody solution is stable for one year from the date of purchase when stored at 4°C.

#### APPLICATIONS-CONFIRMED

Immunocytochemistry 10 μg/mL

#### SPECIES CROSS REACTIVITY on WB

Species	Human	Mouse	Rat	Hamster
Cells	HeLa, A549	NIH/3T3, MEF	NRK	Not tested
Reactivity	+	+	+	

**Entrez Gene ID** 8411 (Human), 216238 (Mouse), 314764 (Rat)

**REFERENCES** 1) Gaullier, J. M., et al., J. Biol. Chem. **275**, 24595-24600 (2000)

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

For more information, please visit our web site https://ruo.mbl.co.jp/

#### LABEL LICENSES:

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Alexa Fluor® is a registered trademark of Molecular Probes, Inc.



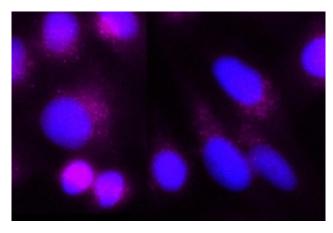
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RELATED PRODUCTS
 M175-3
            anti-α-Tubulin (2F9)
 M175-A48 anti-α-Tubulin Alexa Fluor® 488 (2F9)
 M175-A59 anti-α-Tubulin Alexa Fluor<sup>®</sup> 594 (2F9)
 M175-A64 anti-α-Tubulin Alexa Fluor® 647 (2F9)
 PM054
            anti-α-Tubulin (polyclonal)
 M176-3
            anti-EEA1 (3C10)
 M176-A48 anti-EEA1 Alexa Fluor® 488 (3C10)
 M176-A59 anti-EEA1 Alexa Fluor® 594 (3C10)
            anti-EEA1 (polyclonal)
 PM062
 M178-3
            anti-Calnexin (4F10)
 M178-A48 anti-Calnexin Alexa Fluor® 488 (4F10)
 M178-A59 anti-Calnexin Alexa Fluor® 594 (4F10)
 M178-A64 anti-Calnexin Alexa Fluor® 647 (4F10)
            anti-Calnexin (polyclonal)
 PM060
 M181-3
            anti-KDEL (1D5)
 PM059
            anti-KDEL (polyclonal)
            anti-GM130 (5G8)
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M179-A64 anti-GM130 Alexa Fluor<sup>®</sup> 647 (5G8)
 PM061
            anti-GM130 (polyclonal)
            anti-COX4 (polyclonal)
 PM063
 PM064
            anti-Lamin B1 (polyclonal)
 D115-3
            anti-CENP-A (3-19)
 PD030
            anti-CENP-C (polyclonal)
 K0171-3
            anti-CENP-E (1H12)
 PD031
            anti-CENP-H (polyclonal)
 PD032
            anti-CENP-I/hMis6 (polyclonal)
 D282-3
            anti-CENP-K/ICEN37 (46F1)
 PD018
            anti-CENP-K (polyclonal)
            anti-CENP-L/ICEN33 (27E10)
 D283-3
 D284-3
            anti-CENP-M/ICEN39 (23F6)
 D285-3
            anti-CENP-N/ICEN32 (22F4)
 PD020
            anti-CENP-O (polyclonal)
 D286-3
            anti-CENP-T/ICEN22 (42F10)
 PD019
            anti-CENP-50 (polyclonal)
 PD014
            anti-LC3 (polyclonal) [WB]
 PD015
            anti-LC3 (polyclonal) [IC]
 PM036
            anti-LC3 (polyclonal) [WB, IP, IC, IHC, FCM]
 PM046
            anti-LC3 (polyclonal) [WB, IC]
 M115-3
            anti-LC3 (51-11)
                                   [WB]
                                   [WB, IP, IC, FCM]
 M152-3
            anti-LC3 (4E12)
 M186-3
            anti-LC3 (8E10)
                                   [WB]
 M162-3
            anti-p62 (5F2)
 PM045
            anti-p62 (polyclonal)
            anti-p62 C-terminal (polyclonal)
 PM066
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WB: Western blotting IP: Immunoprecipitation IC: Immunocytochemistry IHC: Immunohistochemistry FCM: Flow cytometry

### **Immunocytochemistry**

- 1) Spread the cells in the nutrient condition on a glass slide, then incubate in a CO<sub>2</sub> incubator for one night.
- 2) Remove the culture supernatant by careful aspiration.
- 3) Fix the cells by immersing the slide in 4% paraformaldehyde (PFA)/PBS for 10 minutes at room temperature (20~25°C).
- 4) Prepare a wash container such as a 500 mL beaker with a magnetic stirrer. Then wash the fixed cells on the glass slide by soaking the slide with a plenty of PBS in the wash container for 5 minutes. Take care not to touch the cells. Repeat another wash twice more.
- 5) Immerse the slide in 0.2% Triton X-100/PBS for 10 minutes at room temperature.
- 6) Wash the slide 2 times with PBS.
- 7) Add 200 μL of the primary antibody diluted with 2% fetal calf serum (FCS)/PBS as suggested in the **APPLICATIONS** onto the cells and incubate for 30 minutes at room temperature. (Optimization of antibody concentration or incubation condition is recommended if necessary.)
- 8) Wash the slide 2 times with PBS.
- 9) Counter stain with DAPI for 5 minutes at room temperature.
- 10) Wash the slide 2 times with PBS.
- 11) Wipe excess liquid from slide but take care not to touch the cells. Never leave the cells to dry.
- 12) Promptly add mounting medium onto the slide, then put a cover slip on it.

(Positive control for Immunocytechemistry; HeLa)



Immunocytochemical detection of EEA1 in HeLa

Magenta: M176-A64

Blue: DAPI