SYSY Antibodies

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EEA1

Cat.No. 237 002; Polyclonal rabbit antibody, 200 µl antiserum (lyophilized)

Data Sheet

Reconstitution/ Storage	200 µl antiserum, lyophilized. For **reconstitution** add 200 µl H_2O, then aliquot and store at -20°C until use. Antibodies should be stored at +4°C when still lyophilized. Do not freeze! For detailed information, see back of the data sheet.
Applications	**WB**: 1 : 1000 up to 1 : 5000 (AP staining) **IP**: not tested yet **ICC**: 1 : 1000 up to 1 : 2000 **IHC**: not recommended **IHC-P**: 1 : 200
Immunogen	Synthetic peptide corresponding to AA 2 to 13 from human EEA1 (UniProt Id: Q15075)
Reactivity	Reacts with: human (Q15075), rat (A0A0G2K051), mouse (Q8BL66). No signal: zebrafish. Other species not tested yet.

TO BE USED IN VITRO / FOR RESEARCH ONLY
NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

Background

Extracellular compounds are internalized by endocytosis into so called endocytic vesicles. They fuse with early endosomes, from where the endocytosed material can be shuttled to a number of alternative destinations.
Early **e**ndosomal **a**ntigen **1** (**EEA 1**) is a peripheral membrane protein that locates to early endosomes via binding to the membrane lipid phosphatidylinositol 3-phosphate (PtdIns3P) and the active form of Rab5.
Autoantibodies against EEA 1 have been shown to be associated with subacute cutaneous systemic lupus erythematosus.

Selected References for 237 002

A novel method for culturing stellate astrocytes reveals spatially distinct Ca2+ signaling and vesicle recycling in astrocytic processes.
Wolfes AC, Ahmed S, Awasthi A, Stahlberg MA, Rajput A, Magruder DS, Bonn S, Dean C
The Journal of general physiology (2017) 1491: 149-170. . **ICC**

Quantitative analysis of synaptic vesicle Rabs uncovers distinct yet overlapping roles for Rab3a and Rab27b in Ca2+-triggered exocytosis.
Pavlos NJ, Grønborg M, Riedel D, Chua JJ, Boyken J, Kloepper TH, Urlaub H, Rizzoli SO, Jahn R
The Journal of neuroscience : the official journal of the Society for Neuroscience (2010) 3040: 13441-53. . **WB**

C9orf72 hexanucleotide repeat expansions impair microglial response in ALS.
Masrori P, Bijnens B, Fumagalli L, Davie K, Poovathingal SK, Meese T, Storm A, Hersmus N, Fazal R, van den Biggelaar D, Asselbergh B, et al.
Nature neuroscience (2025) : . . **ICC; tested species: human**

Coupling of microtubule motors with AP-3 generated organelles in axons by NEEP21 Family Member Calcyon.
Shi L, Hines T, Bergson C, Smith D
Molecular biology of the cell (2018) : mbcE18010007. . **ICC; tested species: monkey**

Molecular anatomy of a trafficking organelle.
Takamori S, Holt M, Stenius K, Lemke EA, Grønborg M, Riedel D, Urlaub H, Schenck S, Brügger B, Ringler P, Müller SA, et al.
Cell (2006) 1274: 831-46. . **WB**

Sorting in early endosomes reveals connections to docking- and fusion-associated factors.
Barysch SV, Aggarwal S, Jahn R, Rizzoli SO
Proceedings of the National Academy of Sciences of the United States of America (2009) 10624: 9697-702. .

SNARE function is not involved in early endosome docking.
Geumann U, Barysch SV, Hoopmann P, Jahn R, Rizzoli SO
Molecular biology of the cell (2008) 1912: 5327-37. .

Selected General References

Cell-cycle-dependent binding kinetics for the early endosomal tethering factor EEA1.
Bergeland T et al. EMBO Rep. (2008) PubMed:18188183

EEA1, a tethering protein of the early sorting endosome, shows a polarized distribution in hippocampal neurons, epithelial cells, and fibroblasts.
Wilson JM et al. Mol. Biol. Cell (2000) PubMed:10930461

The Rab5 effector EEA1 interacts directly with syntaxin-6.
Simonsen A et al. J. Biol. Chem. (1999) PubMed:10506127

The endosome fusion regulator early-endosomal autoantigen 1 (EEA1) is a dimer.
Callaghan J et al. Biochem. J. (1999) PubMed:10024533

Access the online factsheet including applicable protocols at https://sysy.com/product/237002 or scan the QR-code.

FAQ - How should I store my antibody?

Shipping Conditions

- All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freeze-dried) and are stable in this form without loss of quality at ambient temperatures for several weeks.

Storage of Sealed Vials after Delivery

- **Unlabeled** and **biotin-labeled antibodies** and **control proteins** should be stored at 4°C before reconstitution. **They must not be stored in the freezer when still lyophilized!** Temperatures below zero may cause loss of performance.
- **Fluorescence-labeled antibodies** should be reconstituted immediately upon receipt. Long term storage (several months) may lead to aggregation.
- **Control peptides** should be kept at -20°C before reconstitution.

Long Term Storage after Reconstitution (General Considerations)

- The storage freezer must not be of the frost-free variety ("no-frost freezer"). This cycle between freezing and thawing (to reduce frost-build-up), which is exactly what should be avoided. For the same reason, antibody vials should be placed in an area of the freezer that has minimal temperature fluctuations, for instance towards the back rather than on a door shelf.
- Aliquot the antibody and store frozen (-20°C to -80°C). Avoid very small aliquots (below 20 µl) and use the smallest storage vial or tube possible. The smaller the aliquot, the more the stock concentration is affected by evaporation and adsorption of the antibody to the surface of the storage vial or tube. Adsorption of the antibody to the surface leads to a substantial loss of activity.
- The addition of glycerol to a final concentration of 50% lowers the freezing point of your stock and keeps your antibody at -20°C in liquid state. This efficiently avoids freeze and thaw cycles.

Product Specific Hints for Storage

Control proteins / peptides

- Store at -20°C to -80°C.

Monoclonal Antibodies

- **Ascites** and **hybridoma supernatant** should be stored at -20°C up to -80°C. **Prolonged storage at 4°C is not recommended!** Unlike serum, ascites may contain proteases that will degrade the antibodies.
- **Purified IgG** should be stored at -20°C up to -80°C. Adding a carrier protein like BSA will increase long term stability. Many of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

Polyclonal Antibodies

- **Crude antisera**: With anti-microbials added, they may be stored at 4°C. However, frozen storage (-20°C up to -80°C) is preferable.
- **Affinity purified antibodies**: Less robust than antisera. Storage at -20°C up to -80°C is recommended. Adding a carrier protein like BSA will increase long term stability. Most of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

Fluorescence-labeled Antibodies

- Store as a liquid with 1 : 1 (v/v) glycerol at -20°C. Protect these antibodies from light exposure.

Avoid repeated freeze-thaw cycles for all antibodies!

FAQ - How should I reconstitute my antibody?

Reconstitution

- All our purified antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add the amount of deionized water given in the respective datasheet. If higher volumes are preferred, add water as mentioned above and then the desired amount of PBS and a stabilizing carrier protein (e.g. BSA) to a final concentration of 2%. Some of our antibodies already contain albumin. Take this into account when adding more carrier protein.
  For complete reconstitution, carefully remove the lid. After adding water, briefly vortex the solution. You can spin down the liquid by placing the vial into a 50 ml centrifugation tube filled with paper.
- If desired, add small amounts of azide or thimerosal to prevent microbial growth. This is especially recommended if you want to keep an aliquot a 4°C.
- After reconstitution of fluorescence-labeled antibodies, add 1 : 1 (v/v) glycerol to a final concentration of 50%. This lowers the freezing point of your stock and keeps your antibody in liquid state at -20°C.
- Glycerol may also be added to unlabeled primary antibodies. It is a suitable way to avoid freeze-thaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.