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EAAT1 cytoplasmic domain

Cat.No. 250-11P; control peptide, 100 µg peptide (lyophilized)

Data Sheet

Reconstitution/ Storage	100 µg peptide, lyophilized. For **reconstitution** add 100 µl H_2O to get a 1mg/ml solution in PBS. Then aliquot and store at -20°C to -80°C until use. Control peptides should be stored at -20°C when still lyophilized! For detailed information, see back of the data sheet.
Immunogen	Synthetic peptide corresponding to AA 522 to 541 from rat EAAT1 (UniProt Id: P24942)
Recommended dilution	Optimal concentrations should be determined by the end-user.
Matching antibodies	250 113, 250 114, 250 116
Remarks	This control peptide consists of the synthetic peptide (aa 522 - 541 of rodent EAAT 1) that has been used for immunization. It has been tested in preadsorption experiments and blocks efficiently and specifically the corresponding signal in Western blots. The amount of peptide needed for efficient blocking depends on the titer and on the affinity of the antibody to the antigen.

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

Background

Glutamate is the major excitatory neurotransmitter in the mammalian central nervous system. After the release of glutamate from synaptic vesicles into the synaptic cleft during neurotransmission, excitatory amino acid transporters (EAATs) remove extracellular glutamate to avoid excitotoxic levels (1).
Five EAATs with differential expression patterns have been described so far: **EAAT1**, also referred to as GLAST and SLC1A3, has neuroprotective potential following ischemia and occurs in reactive astrocytes and activated microglia. EAAT2 (GLT-1, SLC1A2) is the most abundant isoform and is primarily expressed in astrocytes. Both variants show high levels in brain and retina. EAAT3 / SLC1A1, EAAT4 / SLC1A6 and EAAT5 / SLC1A7 are expressed in neurons (2). EAAT4 shows weak expression in the forebrain and high levels in the cerebellum, where it mainly locates to Purkinje cells (3). EAAT5 primarily occurs in the retina, where it locates very close to glutamate release sites. In K.O. mice flicker resolution is considerably compromised (4). Recent findings suggest that EAAT5 is an abundant isoform, expressed also in non-neuronal peripheral tissues (5).

Selected General References

Phosphatidylcholine metabolism after transfer from lipid emulsions injected intravenously in rats. Implications for high-density lipoprotein metabolism.
Martins IJ et al. Biochim Biophys Acta (1989) PubMed:2804050

Climbing Fiber-Mediated Spillover Transmission to Interneurons Is Regulated by EAAT4.
Malhotra S et al. J Neurosci (2021) PubMed:34400517

Excitatory amino acid transporter EAAT5 improves temporal resolution in the retina.
Gehlen J et al. eNeuro (2021) PubMed:34772693

Excitatory amino acid transporter 5 is widely expressed in peripheral tissues.
Lee A et al. Eur J Histochem (2013) PubMed:23549460

Association of excitatory amino acid transporters, especially EAAT2, with cholesterol-rich lipid raft microdomains: importance for excitatory amino acid transporter localization and function.
Butchbach ME et al. J Biol Chem (2004) PubMed:15187084

Access the online factsheet including applicable protocols at https://sysy.com/product/250-11P 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.