



3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

Product Information

Anti-Ephrin-A2

produced in goat, affinity isolated antibody

Catalog Number **E5154**

Product Description

Anti-Ephrin-A2 is produced in goat using as immunogen a purified recombinant mouse ephrin-A2 extracellular domain (Met 1-Asn 184) expressed in mouse NSO cells. Affinity isolated antibody is obtained from goat Anti-Ephrin-A2 antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to the peptide.

Anti-Ephrin-A2 recognizes recombinant mouse ephrin-A2 extracellular domain by immunoblotting, immunohistochemistry, ELISA, and blockade of receptor binding. The antibody shows less than 1% cross-reactivity with recombinant mouse ephrin-A1, recombinant human ephrin-A3, recombinant mouse ephrin-A4, recombinant human ephrin-A5, recombinant mouse ephrin-B1, recombinant mouse ephrin-B2, and recombinant human ephrin-B3.

Ephrin-A2, also known as ELF-1, Cek7-L, and LERK-6, is a member of the ephrin ligand family, which binds members of the Eph receptor family. All ligands share a conserved extracellular sequence, thought to correspond to the receptor binding domain. The conserved sequence contains ~125 amino acids including four invariant cysteines. A-class ligands have a GPI anchor following the conserved sequence. The calculated molecular mass of the reduced mouse ephrin-A2/Fc monomer is ~46 kDa. As a result of glycosylation, the recombinant protein migrates as an ~57 kDa protein under reducing conditions in SDS-PAGE. The extracellular domains of human and mouse ephrin-A2 share 93% amino acid identity.

Ephrin-A2 binds EphA2, EphA3, EphA4, EphA5, EphA6, EphA7, and EphA8.^{1,2} Only membrane-bound or Fc-clustered ligands have been shown to activate the receptor *in vitro*. Soluble monomeric ligands can bind the receptor, but do not induce receptor auto-phosphorylation and activation.¹ The ephrin ligands and Eph receptors display reciprocal expression *in vivo*.²

Developing and adult neural tissue express nearly all of the Eph receptors and ephrin ligands.² Ephs and ephrins play a significant role in angiogenesis.²

Reagent

Supplied lyophilized from a 0.2 µm filtered solution of phosphate buffered saline containing 5% trehalose.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

To one vial of lyophilized powder, add 1 mL of sterile phosphate buffered saline to produce a 0.1 mg/mL stock solution of antibody.

Storage/Stability

Prior to reconstitution, store at -20 °C. Reconstituted product may be stored at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. Avoid repeated freezing and thawing. Do not store in frost-free freezer.

Product Profile

Immunoblotting: a working concentration of 0.1-0.2 µg/mL is recommended. The detection limit for recombinant mouse ephrin-A2 is ~2 ng/lane under non-reducing and reducing conditions

Indirect ELISA: a working concentration of 0.5-1.0 µg/mL is recommended. The detection limit for recombinant mouse ephrin-A2 is ~0.6 ng/well.

For blockade of receptor ligand interaction, 1-5 µg/mL of the antibody will block 50% of the binding of 10 ng/mL of recombinant mouse Ephrin-A2/Fc to immobilized recombinant rat Eph-A5/Fc (100 µL of a 2 µg/mL solution coated in each well) in an ELISA assay.

Immunohistochemistry: the antibody can be used to detect mouse Ephrin-A2. Cells in culture may be fixed for 20 minutes at 37 °C with freshly prepared 1-2% paraformaldehyde/PBS (pH7.4). Three to five washes of cells in PBS for 15 minutes each is usually required after fixation before application of primary antibodies. Labeling for Ephrin-A2 may be obtained by incubating cells overnight at 4 °C with 1-3 µg/ml of Anti-Ephrin-A2. Detection of bound primary Anti-Ephrin-A2 antibodies may be done by using either fluorescent or chromogenic labels.

Note: In order to obtain the best results in various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

1. Flanagan, J.G. and Vanderhaegen, P., The ephrins and Eph receptors in neural development. *Annu. Rev. Neurosci.*, **21**, 309–345 (1998)
2. Pasquale, E.B., The Eph family of receptors. *Curr. Opin. Cell Biol.*, **9**, 608–615 (1997).

KAA,PHC 09/06-1

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.