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Product Information

Anti-Ephrin-B1
produced in goat, affinity isolated antibody

Catalog Number **E5404**

Product Description

Anti-Ephrin-B1 is produced in goat using as immunogen a purified recombinant mouse ephrin-B1 extracellular domain expressed in mouse NSO cells. The antibody was purified by mouse ephrin-B1 affinity chromatography.

Anti-Ephrin-B1 recognizes mouse ephrin-B1 by immunoblotting, immunohistochemistry, and ELISA. The antibody shows less than 1% cross-reactivity (based on ELISA) with recombinant human ephrin-A3, recombinant human ephrin-A4, recombinant mouse ephrin-A1, recombinant mouse ephrin-B2, recombinant human ephrin-A5, recombinant human ephrin-B3, and rzfEprin-B2.

Ephrin-B1, also known as Cek5-L, ELK-L, EFL-3, LERK-2, and STRA-1, 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. B-class ligands are transmembrane proteins and may be phosphorylated on tyrosine upon receptor ligation. The cytoplasmic domains consist of ~80 amino acids and are highly conserved, especially the last 33 amino acids. Several signaling molecules interact with the cytoplasmic region, but specific signaling roles are still unknown. 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 autophosphorylation and activation.[2]

The recombinant mouse ephrin-B1 used for the testing of this antibody consists of the extracellular domain of mouse ephrin-B1 (amino acid 1-229)[1] fused by means of a polypeptide linker to the Fc portion of human IgG1 that is histidine-tagged at the C-terminus. N-terminal sequencing indicates that recombinant mouse ephrin-B1/Fc has Lys30 at the amino terminus. The calculated molecular mass of the reduced mouse protein is ~49.2 kDa. As a result of glycosylation, recombinant mouse ephrin-B1 migrates as an ~60 kDa protein under reducing conditions in SDS-PAGE. Ephrin-B1 binds EphA3, EphB1, EphB2, EphB3, and EphB4.[2, 3] Human and mouse ephrin-B1 extracellular domains share ~94% homology.

The ephrin ligands and Eph receptors display reciprocal expression *in vivo*.[3] Developing and adult neural tissue express nearly all of the Eph receptors and ephrin ligands.[3] Ephs and ephrins play a significant role in angiogenesis.

Reagent

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

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of sterile phosphate buffered saline (PBS) 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 antibody concentration of 0.1-0.2 μg/ml is recommended. The detection limit for recombinant mouse ephrin-B1 is ~2 ng/lane under non-reducing and reducing conditions.

ELISA: a working antibody concentration of 0.5-1.0 μg/ml is recommended. The detection limit for recombinant mouse ephrin-B1 is ~0.3 ng/well.

Immunohistochemistry: a working antibody concentration of 2-15 μg/mL is recommended using mouse cells and tissues.

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

References

1. Bouillet, P., et al., Efficient cloning of cDNAs of retinoic acid-responsive genes in P19 embryonal carcinoma cells and characterization of a novel mouse gene, Stra1 (mouse LERK-2/Eplg2). *Dev. Biol*., **170**, 420-433 (1995).
2. Flanagan, J.G. and Vanderhaegen, P., The ephrins and Eph receptors in neural development. *Annu. Rev. Neurosci.*, **21,** 309–345 (1998)
3. Pasquale, E.B., The Eph family of receptors. *Curr. Opin. Cell Biol*., **9,** 608–615 (1997).

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