

Product Information

EPHRIN-B1 EXTRACELLULAR DOMAIN/Fc CHIMERA

Mouse, Recombinant
Expressed in NSO mouse myeloma cells

Product Number **E 0653**
Storage Temperature $-20\text{ }^{\circ}\text{C}$

Synonyms: Cek5-L; ELK-L; EFL-3; LERK-2; and STRA-1

Product Description

Recombinant mouse Ephrin-B1 extracellular domain/Fc chimera consists of amino acid residues 1-229 (extracellular domain of mouse Ephrin-B1)¹ that was fused by means of a polypeptide linker to the Fc portion of human IgG₁ that is histidine-tagged at the carboxyl terminus. The chimeric protein is expressed in a mouse myeloma cell line, NSO. Recombinant Ephrin B1 is a disulfide-linked homodimer. The amino-terminus is Lys(30) determined by N-terminal sequencing. The calculated molecular mass of the reduced protein is approximately 49.2 kDa, but as a result of glycosylation, recombinant Ephrin-B1/Fc chimera migrates as an approximately 60 kDa protein on reducing SDS-PAGE.

The Ephrin ligand family, of which Ephrin-B1 is a member, 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 approximately 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 approximately 80 highly conserved amino acids, especially the last 33. Several signaling molecules interact with the cytoplasmic region, but specific signaling roles are still unknown. Ephrin-B1 can bind EphA3, EphB1, EphB2, EphB3, and EphB4.^{2,3} Human and mouse Ephrin-B1 extracellular domains share approximately 94% homology. 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 ligands and receptors display reciprocal expression *in vivo*.³

Nearly all Ephrin-related receptors and ligands have been found to be expressed in developing and adult neural tissue.³ The Eph/Ephrin families may also play a role in angiogenesis.³

Reagent

Recombinant mouse Ephrin-B1 extracellular domain/Fc chimera is lyophilized from a sterile-filtered phosphate-buffered saline (PBS) solution.

Preparation Instructions

Reconstitute the vial contents with sterile PBS. Stock solution concentration should be no less than 100 $\mu\text{g/ml}$.

Storage/Stability

Lyophilized samples are stable for at least six months at $-20\text{ }^{\circ}\text{C}$. Upon reconstitution, store at 2-4 $^{\circ}\text{C}$ for up to one month. For extended storage, store in working aliquots at $-20\text{ }^{\circ}\text{C}$. Repeated freeze-thaw cycles should be avoided. Do not store in a frost-free freezer.

Product Profile

Ephrin-B1/Fc activity is measured by its ability to bind immobilized recombinant mouse EphB3/Fc in a functional ELISA assay. Immobilized recombinant mouse EphB1/Fc (2 $\mu\text{g/ml}$, 100 $\mu\text{l/well}$) binds recombinant mouse Ephrin-B3/Fc with a linear range of 0.01 – 0.5 ng/ml. Optimal dilutions should be determined by each laboratory for each application.

Purity: >95% by SDS-PAGE, visualized by silver stain.

Endotoxin level: < 0.1 ng/ μg of protein as determined by the LAL (Limulus amoebocyte lysate) method.

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 P. Vanderhaegen, 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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