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# **ProductInformation**

## EphB6 EXTRACELLULAR DOMAIN/Fc CHIMERA

Mouse, Recombinant Expressed in NSO mouse myeloma cells

Product Number E 9777 Storage Temperature –20 °C

Synonyms: Mep

### **Product Description**

Recombinant mouse EphB6 extracellular domain/Fc chimera consists of amino acid residues 1-587 (extracellular domain of mouse EphB6)<sup>1</sup> that was fused by means of a polypeptide linker to the Fc portion of human  $IgG_1$  that is 6X histidine-tagged at the carboxyl termnal. The chimeric protein is expressed in a mouse myeloma cell line, NSO. Recombinant EphB6 is a disulfide-linked homodimer. The amino-terminus is Leu(33) based on N-terminal sequencing. The calculated molecular mass of the reduced protein is approximately 87.3 kDa, but as a result of glycosylation, the recombinant EphB6/Fc migrates as an approximately 100 kDa protein on reducing SDS-PAGE.

The Eph receptor family, of which EphB6 is a member, binds members of the Ephrin ligand family. Two classes of receptors exist, designated A and B, that have an extracellular domain made up of a globular domain, a cysteine-rich domain, and two fibronectin type III domains, followed by the transmembrane region and cytoplasmic region. The cytoplasmic region is a juxtamembrane region with two tyrosines, the major phosphorylation sites, and a conserved sterile alpha motif (SAM) in the carboxyl terminus, the latter including one conserved tyrosine.<sup>2,3</sup> EphB6 lacks intrinsic kinase activity<sup>1</sup>, however, cross-linking of the Eph6 receptor leads to activation of the cellular kinase activity.<sup>4</sup> EphB6 binds to Ephrin-B2 and Ephrin B-3.<sup>5</sup> Human and mouse EphB6 extracellular domains share approximately 92% homology. Only membrane-bound or Fc-clustered ligands have been shown to activate the receptor in vitro.

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

### Reagents

Recombinant mouse EphB6 extracellular domain/Fc chimera is supplied as approximately 200 µg of protein 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  $100 \ \mu g/ml$ .

#### Storage/Stability

Lyophilized samples are stable for more than six months at -20 °C. Upon reconstitution, store at 2-4 °C for up to one month. For extended storage, store in working aliquots at -20 °C. Repeated freeze-thaw cycles should be avoided. Do not store in frost-free freezer.

### **Product Profile**

EphB6/Fc activity is measured by its ability to bind immobilized recombinant mouse Ephrin-B2/Fc in a functional ELISA assay. Immobilized recombinant mouse EphB6/Fc (2 mg/ml, 100 ml/well) binds recombinant mouse Ephrin-B2/Fc with a linear range of 0.078-5 ng/ml. Optimal dilutions should be determined by each laboratory for each application.

Purity: >90% by SDS-PAGE, visualized by silver stain.

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

#### References

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- 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).
- Luo, H., et al., Cross-linking of EphB6 resulting in signal transduction and apoptosis in Jurkat cells. J. Immunol., **167**, 1362-1370 (2001).
- Tang. X.X., et al., Implications of EPHB6, EFNB2, and EFNB3 expressions in human neuroblastoma. Proc. Natl. Acad. Sci. USA, 97, 10936-10941 (2000).

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