

BACKGROUND

Eph family of receptor tyrosine kinases consists of at least 14 distinct receptors and has eight membrane-bound ligands, known as the ephrins. This is the largest family of receptor tyrosine kinases. Eph proteins are divided into two subfamilies: the EphA receptors (A1-A8) that bind glycosyl phosphatidylinositol (GPI)-linked ephrin-A ligands (A1-A5), and the EphB receptors (B1-B6) that bind transmembrane ephrin-B ligands (B1-B3). The only known crosstalk between the A and B subfamilies occurs with the EphA4 receptor, which can bind ephrins-B2 and -B3 as well as the entire A subclass. There is a great deal of redundancy of receptor-ligand binding specificity within each subfamily, although binding affinities vary.^{1,2} Both GPI-anchored ephrinA and transmembrane ephrinB ligands interact with the Nterminal globular domain (Glob) of Eph receptors. The Eph receptors become phosphorylated at specific tyrosine residues in the cytoplasmic domain following ligand binding. Phosphorylated motifs serve as sites of interaction with certain cytoplasmic signaling proteins to mediate downstream signaling. In addition, through their C terminus the Eph receptors associate with PDZ (postsynaptic density protein, disc large, zona occludens) domain-containing proteins. Moreover, Eph receptor contact induces tyrosine phosphorylation of the cytoplasmic domain of ephrinB proteins via an SRC-family kinase (SFK), which mediating the reverse signaling. One of the unique features of Eph/ephrin signaling is the fact that both receptors and ligands are competent to transduce a signaling cascade upon interaction. Eph-activated signaling is termed forward, and ephrin-activated signaling is termed reverse. Another level of complexity stems from the fact that interactions between Eph receptors and ephrins can happen in trans (between two opposing cells) or in cis (within the same cell). It is commonly assumed that trans interactions are activating while cis interactions are inhibiting.³ Eph-Ephrin signaling functions in a variety of biological processes including diverse assegmentation of the somites and rhombomeres, the formation of blood vessels, Axon guidance and fasciculation, migration of the neural crest and metastasis of transformed cells etc.

EphB3 and ephrin-B1 have recently been shown to be expressed in complementary compartments in the neonate intestinal epithelium where they establish a boundary between proliferative and differentiated cells. Furthermore, EphB3 knock-out mice display a highly penetrant phenotype characterized by Paneth cell mislocalization throughout the intestinal crypts instead of being restricted at the bottom of the crypt in wild type animals. These observations suggest that EphB3 signaling sets restrictions to epithelial cell migration. This is analogous to repulsive guidance of growth cones and neural crest cells in developing nervous systems. Ligation of EphB3

receptor inhibits cell adhesion in a kinase-dependent manner, whereas inhibition of migration is independent of EphB3 catalytic activity. EphB3 is widely expressed in developing and adult tissues. Kinase-dependent and -independent regulatory effects on cell motility may allow this receptor to exert diverse functions under different physiological conditions.⁴ The genetic analysis of EphB3 revealed a physiological requirement of the receptor for path-finding of specific commissural axons in the central nervous system.

References:

1. Brantley-Sieders, D.M. & Chen, J.: *Angiogenesis*. 7:17, 2004
2. Murai, K.K. & Pasquale, E.B.: *J. Cell Sci.* 116:2823, 2003
3. Arvanitis, D. & Davy, A.: *Genes & Dev.* 22:416, 2008
4. Miao, H. et al: *J. Biol. Chem.* 280:923-32, 2005

TECHNICAL INFORMATION

Source:

EphB3 antibody is a mouse monoclonal antibody raised against purified recombinant human EphB2 fragments expressed in *E. coli*.

Specificity and Sensitivity:

This antibody detects endogenous EphB3 proteins in cells without cross-reactivity with other family members.

Storage Buffer: PBS and 30% glycerol

Storage:

Store at -20°C for at least one year. Store at 4°C for frequent use. Avoid repeated freeze-thaw cycles.

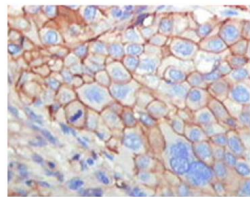
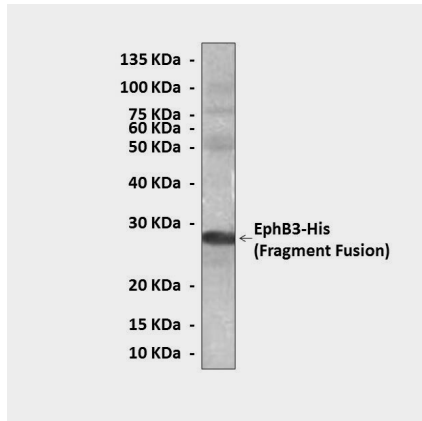
APPLICATIONS

Application:	*Dilution:
WB	1:1000
IP	n/d
ICC	n/d
IHC	1:200
FACS	n/d

*Optimal dilutions must be determined by end user.



QUALITY CONTROL DATA



Top: Western Blot detection of EphB3 proteins in bacterial lysate containing His-tagged-EphB3 fragment fusion proteins (26 kDa). **Bottom:** This antibody stains paraffin-embedded human lung squamous cell cancer tissue in IHC.

