Human/Mouse/Rat Ephrin-B2 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF496

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects mouse Ephrin-B2 in direct ELISAs and Western blots. In direct ELISAs and Western blots, approximately 5% cross-reactivity with recombinant mouse Ephrin-B1 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant mouse Ephrin-B2 Arg27-Ala227 Accession # AAA82934
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

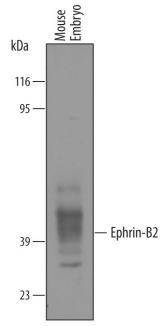
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Flow Cytometry	0.25 µg/10^6 cells	See Below
Immunocytochemistry	5-15 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

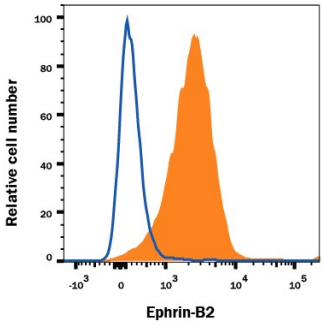
DATA

Western Blot



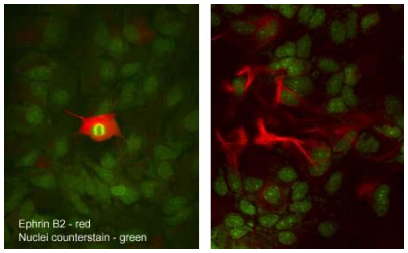
Detection of Mouse Ephrin-B2 by Western Blot. Western blot shows lysates of mouse embryo tissue. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human/Mouse/Rat Ephrin-B2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF496) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF019). Specific bands were detected for Ephrin-B2 at approximately 40-45 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 8.

Flow Cytometry



Detection of Ephrin-B2 in SH-SY5Y Human Cell Line by Flow Cytometry. SH-SY5Y human neuroblastoma cell line was stained with Goat Anti-Human/Mouse/Rat Ephrin-B2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF496, filled histogram) or isotype control antibody (Catalog # AB-108-C, open histogram), followed by Phycoerythrin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0107).

Immunocytochemistry



Ephrin-B2 in Rat Hippocampal Neurons. Ephrin-B2 was detected in immersion fixed rat hippocampal neurons using 2 µg/mL Goat Anti-Human/Mouse/Rat Ephrin-B2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF496) for 3 hours at room temperature. Cells were stained (red) and counterstained (green). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

bio-techne®
Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 Canada TEL 855 668 8722 China TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449

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BACKGROUND

Ephrin-B2, also known as Htk-L, ELF-2, LERK-5, and NLERK-1 (1), is a member of the ephrin ligand family which binds members of the Eph receptor family. All ligands share a conserved extracellular sequence, which most likely corresponds to the receptor binding domain. This conserved sequence consists of approximately 125 amino acids and includes four invariant cysteines. The B-class ligands are transmembrane proteins which can become tyrosine phosphorylated upon receptor ligation. The cytoplasmic domains are approximately 80 amino acids long and are highly conserved, especially the last 33 amino acids. Several signaling molecules have been shown to interact with the cytoplasmic region, although specific signaling roles have yet to be elucidated. Ephrin-B2 has been shown to bind EphA4, EphB1, EphB2, EphB3, and EphB4 (2, 3). The extracellular domains of murine and human Ephrin-B2 share 98% amino acid identity. Only membrane-bound or Fc-clustered ligands are capable of activating the receptor *in vitro*. While soluble monomeric ligands bind the receptor, they do not induce receptor autophosphorylation and activation (2). *In vivo*, the ligands and receptors display reciprocal expression (3). It has been found that nearly all the receptors and ligands are expressed in developing and adult neural tissue (3). The Ephrin/Eph families also appear to play a role in angiogenesis (3).

References:

1. Eph Nomenclature Committee [letter]. (1997) Cell **90**:403.
2. Flanagan, J.G. and P. Vanderhaeghen (1998) Annu. Rev. Neurosci. **21**:309.
3. Pasquale, E.B. (1997) Curr. Opin. Cell Biol. **9**:608.

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Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 **Canada** TEL 855 668 8722 **China** TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449