**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.

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

**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-109-C, open histogram), followed by Phycoerythrin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0107).

**Immunocytochemistry**  
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.

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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 96% 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: