

#### DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human ROBO4 in direct ELISAs and Western blots. In direct ELISAs, no cross-reactivity with recombinant rat ROBO1, recombinant human (rh) ROBO2, or rhROBO3 is observed.
<b>Source</b>	Monoclonal Mouse IgG <sub>2A</sub> Clone # 265703
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	Mouse myeloma cell line NS0-derived recombinant human ROBO4 Gln28-Arg467 Accession # Q8WZ75
<b>Conjugate</b>	Alexa Fluor 700 Excitation Wavelength: 675-700 nm Emission Wavelength: 723 nm
<b>Formulation</b>	Supplied 0.2 mg/mL in a saline solution containing BSA and Sodium Azide. See Certificate of Analysis for details.  *Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to the Safety Data Sheet (SDS) for additional information and handling instructions.

#### 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.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Flow Cytometry</b>	0.25-1 µg/10 <sup>6</sup> cells	HUVEC human umbilical vein endothelial cells

#### PREPARATION AND STORAGE

<b>Shipping</b>	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<b>Protect from light. Do not freeze.</b> ● 12 months from date of receipt, 2 to 8 °C as supplied.

#### BACKGROUND

ROBO4, also called magic roundabout, is a ~150 kDa glycoprotein belonging to the ROBO family (1). ROBOs are molecular guidance receptors that typically interact with Slit ligands to regulate axon guidance and neuronal migration (2). Unlike other family members, ROBO4 is mainly restricted to the vascular endothelium (1, 2). Expression in early hematopoietic progenitors is also reported (3). The human ROBO4 cDNA encodes 1012 amino acids (aa), including a 27 aa signal sequence, a 440 aa extracellular domain (ECD) containing two C2-type Ig domains and two fibronectin type III (FNIII) domains, a transmembrane domain and an intracellular domain. ROBO4 diverges from other ROBOs in the number of Ig, FNIII and cytoplasmic CC domains (1, 4). Within the ECD, human ROBO4 shares 80%, 80%, 87% and 88% aa identity with mouse, rat, bovine and canine ROBO4, respectively. Vascular endothelial ROBO4 is expressed at highest levels in during development and vascular remodeling, including tumor angiogenesis (1, 2, 4 - 6). It is proposed to contribute to vascular stability. Consistent with this, endogenous ROBO4 is concentrated in the vascular stalk and sprouts rather than tip cells and appears to protect newly formed blood vessels against VEGF-induced vascular leak (6 - 9). ROBO4 binding of Slit proteins has been variably reported, and when detected may be mediated by ROBO4/ROBO1 heterodimers (2, 4 - 7, 9 - 13). ROBO4 is also variably reported to stimulate or inhibit cell migration or filopodia formation (2, 4 - 13). Effects on cell movement may be mediated through intracellular binding of WASP-, Ras/Rac/Rho-, Mena-, Src- or Paxillin-related proteins, all of which affect the cytoskeleton (5 - 7, 10 - 12). Recombinant soluble ROBO4 ECD can antagonize endothelial cell migration and in vivo angiogenesis (13).

#### References:

1. Huminiecki, L. *et al.* (2002) *Genomics* **79**:547.
2. Legg, J.A. *et al.* (2008) *Angiogenesis* **11**:13.
3. Shibata, F. *et al.* (2009) *Stem Cells* **27**:183.
4. Park, K.W. *et al.* (2003) *Dev. Biol.* **261**:251.
5. Seth, P. *et al.* (2005) *Biochem. Biophys. Res. Commun.* **332**:533.
6. Jones, C.A. *et al.* (2008) *Nat. Med.* **14**:448.
7. Jones, C.A. *et al.* (2009) *Nat. Cell Biol.* **11**:1325.
8. Chen, H. *et al.* (2010) *Adv. Exp. Med. Biol.* **664**:457.
9. London, N.R. *et al.* (2009) *J. Thromb. Haemost.* **7**:57.
10. Verissimo, A.R. *et al.* (2009) *Biochem. Soc. Trans.* **37**:1214.
11. Sheldon, H. *et al.* (2009) *FASEB J.* **23**:513.
12. Kaur, S. *et al.* (2008) *BMC Cell Biol.* **9**:61.
13. Suchting, S. *et al.* (2005) *FASEB J.* **19**:121.

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