

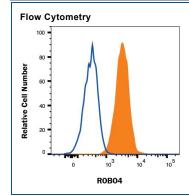
Human ROBO4 Antibody

Monoclonal Mouse IgG_{2B} Clone # 265721 Catalog Number: MAB24541

DESCRIPTION		
Species Reactivity	Human	
Specificity	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.	
Source	Monoclonal Mouse IgG _{2B} Clone # 265721	
Purification	Protein A or G purified from hybridoma culture supernatant	
Immunogen	Mouse myeloma cell line NS0-derived recombinant human ROBO4 Gln28-Arg467 Accession # Q8WZ75	
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.			
Western Blot	1 μg/mL	Human ROBO4	
Flow Cytometry	0.25 μg/10 ⁶ cells	See Below	
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.		

DATA



Detection of ROBO4 in HUVEC cells by Flow Cytometry HUVEC cells were stained with Mouse Anti-Human ROBO4 Monoclonal Antibody (Catalog # MAB24541, filled histogram) or isotype control antibody (Catalog # MAB004, open histogram) followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B). View our protocol for Staining Membrane-associated Proteins.

PREPARATION AND STORAGE

 Reconstitution
 Reconstitute at 0.5 mg/mL in sterile PBS. For liquid material, refer to CoA for concentration.

 Shipping
 Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.

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.

Rev. 6/2/2025 Page 1 of 2



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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 ROBO proteins 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:

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Rev. 6/2/2025 Page 2 of 2