Human Plexin D1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF4160

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Plexin D1 in direct ELISAs and Western blots. In direct ELISAs and Western blots, less than 1% cross-reactivity with recombinant mouse (rm) Plexin A1, rmPlexin A3, and recombinant human Plexin B1 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human Plexin D1 Leu47-Ala1271 Accession # Q9Y4D7
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 as a 0.2 μm filtered solution in PBS.

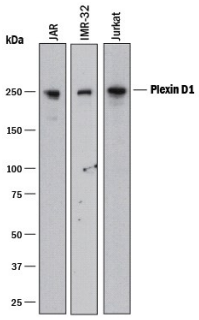
APPLICATIONS

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	0.5 µg/mL	See Below
Flow Cytometry	0.25 µg/10^6 cells	See Below
Immunohistochemistry	3-15 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.	
Neutralization	Measured by its ability to neutralize Semaphorin 3E-induced inhibition of the HUVEC human umbilical vein endothelial cells. Sakurai, A. et al. (2010) Mol Cell Bio 12:3086-98. The Neutralization Dose (ND_{50}) is typically 0.5-2 ug/mL.	

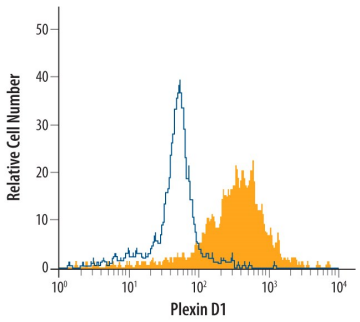
DATA

Western Blot



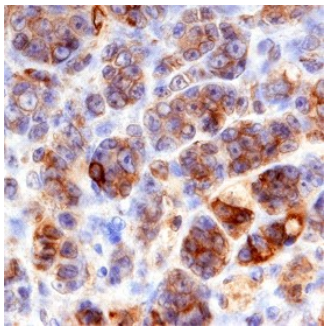
Detection of Human Plexin D1 by Western Blot. Western blot shows lysates of JAR human choriocarcinoma cell line, IMR-32 human neuroblastoma cell line, and Jurkat human acute T cell leukemia cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human Plexin D1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4160) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for Plexin D1 at approximately 250 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Flow Cytometry



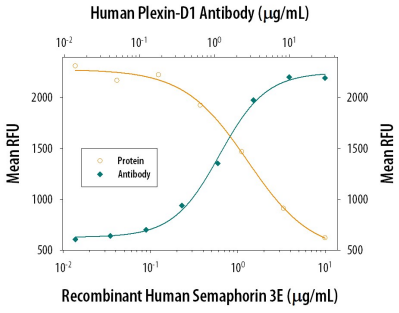
Detection of Plexin D1 in Human peripheral blood monocytes by Flow Cytometry. Human peripheral blood monocytes were stained with Goat Anti-Human Plexin D1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4160, filled histogram) or control antibody (Catalog # AB-108-C, open histogram), followed by Phycoerythrin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0107).

Immunohistochemistry



Plexin D1 in Human Melanoma. Plexin D1 was detected in immersion fixed paraffin-embedded sections of human melanoma using Goat Anti-Human Plexin D1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4160) at 3 µg/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific staining was localized to plasma membranes. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Neutralization



Proliferation Inhibited by Semaphorin 3E and Neutralization by Human Plexin D1 Antibody. Recombinant Human Semaphorin 3E (Catalog # 3239-S3) inhibits proliferation in the HUVEC human umbilical vein endothelial cells in a dose-dependent manner (orange line). Plexin D1 mediated inhibition elicited by Recombinant Human Semaphorin 3E (5 ug/mL) is neutralized (green line) by increasing concentrations of Goat Anti-Human Plexin D1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4160). The ND_{50} is typically 0.5-2 ug/mL.

bio-techne
bio-techne.com
Global info@bio-techne.com techsupport@bio-techne.com
North America TEL 800 343 7475 Europe | Middle East | Africa TEL +44 (0)1235 529449
China info.cn@bio-techne.com TEL +86 (21) 52380373
Rest of World bio-techne.com/find-us/distributors TEL +1 612 379 2956

Human Plexin D1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF4160

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.

BACKGROUND

Plexin D1 is a type I transmembrane glycoprotein that is the prototype of the plexin D subfamily of semaphorin receptors (1, 2). Human Plexin D1 contains a 46 amino acid (aa) signal sequence, a 1225 aa extracellular domain (ECD), a 21 aa transmembrane domain, and a 633 aa cytoplasmic domain that includes features common to other plexins (1). The human Plexin D1 ECD shares 89% identity with mouse Plexin D1, and ~84-92% aa identity based on incomplete sequences of rat, bovine, porcine and canine Plexin D1. It contains a sema domain, two plexin-semaphorin-integrin (PSI) or Met-related sequence (MRS) cysteine-rich motifs, and three glycine/proline-rich IPT/TIG domains which are immunoglobulin-like domains found in plexins, transcription factors, and the scatter factor receptors Met and Ron (1, 2). Isoforms of 1787 and 1747 aa have been sequenced; these contain a 178 aa N-terminal deletion with or without a longer alternate C-terminus (3). Like other Sema/plexin interactions, Plexin D1 interacts with Sema3C or Sema4A via neuropilins. Interaction with Sema3E, however, is direct (4). Plexin D1/Sema3E interaction mediates vascular guidance during development or angiogenesis; deletion of either molecule results in similar, profound cardiac abnormalities (4, 5). Plexin D1 is also expressed in lymphocytes, osteoblasts, the neural crest and the central nervous system during development (2, 6). In the brain, the presence of neuropilin can change Plexin D1/Sema3E interaction from an attractive to a repulsive signal (7, 8). Plexin D1 directs migration of thymocytes to the thymic medulla, probably through repulsion of Sema3E (9). Endothelial cell Plexin D1 binding to Sema4A can oppose VEGF and suppresses tumor angiogenesis, and expression of Sema3E correlates inversely with tumor metastasis, indicating that Plexin D1 is anti-metastatic in the presence of its ligands (10, 11).

References:

1. Negishi, M. *et al.* (2005) Cell. Mol. Life Sci. **62**:1363.
2. Van Der Zwaag, B. *et al.* (2002) Dev. Dyn. **225**:336.
3. Entrez protein Accession # Q9Y4D7, EAW79239, EAW79240.
4. Gu, C. *et al.* (2005) Science **307**:265.
5. Gitler, A.D. *et al.* (2004) Developmental Cell **7**:107.
6. Zhang, Y. *et al.* (2009) Dev. Biol. **325**:82.
7. Chauvet, S. *et al.* (2007) Neuron **56**:807.
8. Pecho-Vrieseling, E. *et al.* (2009) Nature **459**:842.
9. Choi, Y.I. *et al.* (2008) Immunity **29**:888.
10. Toyofuku, T. *et al.* (2007) EMBO J. **26**:1373.
11. Roodink, I. *et al.* (2008) Am. J. Pathol. **173**:1873.

bio-techne
bio-techne.com
Global info@bio-techne.com techsupport@bio-techne.com
North America TEL 800 343 7475 Europe | Middle East | Africa TEL +44 (0)1235 529449
China info.cn@bio-techne.com TEL +86 (21) 52380373
Rest of World bio-techne.com/find-us/distributors TEL +1 612 379 2956