Human L1CAM Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF277

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human L1CAM in direct ELISAs and Western blots. In direct ELISAs, approximately 10% cross-reactivity with recombinant human (rh) ICAM-2 and rhICAM-3 is observed and less than 1% cross-reactvity with rhCD31 and recombinant mouse VCAM-1 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human L1CAM Ile20-Glu1120 Accession # CAA42508
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.

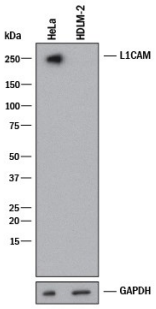
APPLICATIONS

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	0.25 - 0.5 µg/mL	HeLa human cervical epithelial carcinoma cell line, human brain (motor cortex) and rat brain
Immunohistochemistry	5-15 µg/mL	See Below
Simple Western	5 µg/mL	HeLa human cervical epithelial carcinoma cell line

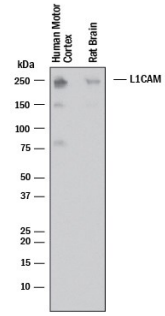
DATA

Western Blot



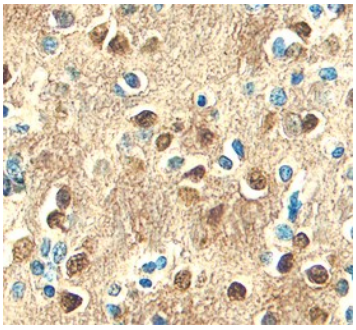
Detection of Human L1CAM by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line and HDLM-2 human Hodgkin's lymphoma cell line (negative control). PVDF membrane was probed with 0.25 µg/mL of Goat Anti-Human L1CAM Antigen Affinity-purified Polyclonal Antibody (Catalog # AF277) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for L1CAM at approximately 250 kDa (as indicated). GAPDH (Catalog # AF5718) is shown as a loading control. This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Western Blot



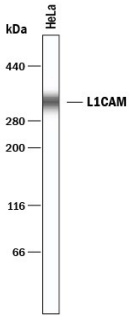
Detection of Human and rat L1CAM by Western Blot. Western blot shows lysates of human brain (motor cortex) and rat brain. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human L1CAM Antigen Affinity-purified Polyclonal Antibody (Catalog # AF277) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for L1CAM at approximately 240 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Immunohistochemistry



L1CAM in Human Brain. L1CAM was detected in immersion fixed paraffin-embedded sections of human brain (hippocampus) using Goat Anti-Human L1CAM Antigen Affinity-purified Polyclonal Antibody (Catalog # AF277) at 10 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 # Catalog # CTS013). Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS008) and counter-stained with hematoxylin (blue). Specific staining was localized to plasma membranes and cytoplasm of neurons. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western



Detection of Human L1CAM by Simple Western™. Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for L1CAM at approximately 333 kDa (as indicated) using 5 µg/mL of Goat Anti-Human L1CAM Antigen Affinity-purified Polyclonal Antibody (Catalog # AF277) . This experiment was conducted under reducing conditions and using the 66-440 kDa separation system.

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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

L1CAM (Neural cell adhesion molecule L1, also known as L1, CD171 and NCAM-L1) is a 200-230 kDa member of the L1 family, Immunoglobulin (Ig) superfamily of molecules. L1 is recognized to play a key role in cell migration, adhesion, neurite outgrowth, myelination and neuronal differentiation. It does so through a series of *cis* and *trans* interactions that involve multiple copartners and target receptors. L1 is described as forming both homotypic and heterotypic complexes, the latter with molecules as diverse as the EGFR, NCAM, CD24, neurocan and various α_V plus β_1 and β_3 integrins. Cells known to express L1 include immature oligodendrocytes, $CD4^+$ T cells, B cells and monocytes, premyelinating Schwann cells, intestinal epithelial progenitor cells, and cerebellar granule plus Purkinje cells. Mature human L1 is a 1238 amino acid (aa) type I transmembrane protein. It contains an 1101 aa extracellular region (aa 20-1120) plus a 114 aa cytoplasmic domain (aa 1144-1257). The extracellular region possesses six C2-type Ig-like domains (aa 35-607) followed by five fibronectin (FN) type III repeats (aa 612-1108). The cytoplasmic tail contains no kinase motifs, but does possess a FIGQY peptide that interacts with ankyrin, and an RSLE sequence that mediates clathrin-associated endocytosis. At least five Ser residues are known to be phosphorylated. There are two splice variants, one each in the intracellular and extracellular domain. A deletion of RSLE (aa 1177-1180) adversely affects endocytosis, while a Leu substitution for aa 26-31 interfers with numerous heterotypic interactions. In general, the full-length L1 molecule is a neuron-associated isoform. L1 is known to undergo proteolysis, either by plasmin or ADAMs. This generates soluble isoforms of varying sizes (140-200 kDa) that retain bioactivity, and which can be incorporated into the surrounding ECM. The membrane fragments (30-80 kDa) undergo further processing, most importantly by γ-secretase, to generate a soluble 28 kDa intracellular domain. This domain is SUMOylated, and believed to possess an NLS at Lys1147. Upon presumed entry into the nucleus, L1 is posited to activate L1-responsive genes. In the extracellular region, human and mouse L1 share 86% aa sequence identity.

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