

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Notch-1 Intracellular Domain in Western blots. In Western blots, this antibody does not cross-react with the corresponding intracellular segments of recombinant human (rh) Notch-2, rhNotch-3, or rhNotch-4.
Source	Monoclonal Mouse IgG _{2A} Clone # 433802
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human Notch-1 Gly2428-Lys2556 Accession # AAG33848
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.

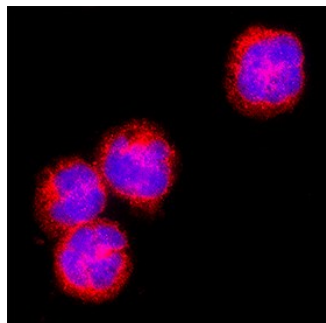
APPLICATIONS

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	1 µg/mL	Recombinant Human Notch-1 Intracellular Domain
Immunocytochemistry	8-25 µg/mL	Immersion fixed MOLT-4 human acute lymphoblastic leukemia cell line

DATA

Immunocytochemistry



Notch-1 in MOLT-4 Human Cell Line.

Notch-1 was detected in immersion fixed MOLT-4 human acute lymphoblastic leukemia cell line using Mouse Anti-Human Notch-1 Intracellular Domain Monoclonal Antibody (Catalog # MAB3647) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cell surface and cell nuclei. Staining was performed using our protocol for Fluorescent ICC Staining of Non-adherent Cells.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 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. <ul style="list-style-type: none"> 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

Human Notch-1 is a 300 kDa type I transmembrane glycoprotein that is one of four human Notch homologues involved in developmental processes (1-3). Notch signaling is important for maintaining stem cells and inducing differentiation, especially in the nervous system and lymphoid tissues (2-4). Notch can specify binary cell fates; for example, promoting T- over B-cell development from a common precursor (2). More than 50% of human T-lineage acute lymphoblastic leukemia (T-ALL) have activating mutations of Notch1 (1, 5). Human Notch-1 is synthesized as a 2556 amino acid (aa) precursor that contains an 18 aa signal sequence, a 1718 aa extracellular domain (ECD) with 36 EGF-like repeats and three Lin-12/Notch repeats (LNR), a 23 aa transmembrane (TM) segment and a 785 aa cytoplasmic domain containing six ankyrin repeats, a glutamine-rich domain and a PEST sequence. The 11th and 12th EGF-like repeats bind ligands including Jagged and Delta-like families in humans (6). O-fucosylation by Fringe family members at a site within this region can inhibit the interaction of Notch with Jagged ligands, thereby promoting Delta-like ligand interactions (7). Notch-1 receptor undergoes post-translational furin-type proteolytic cleavage, forming a heterodimer through interaction of a hydrophobic area C-terminal to the LNR on the 1647 aa ligand-binding extracellular region with the 891 aa transmembrane/cytoplasmic portion (8, 9). Upon ligand binding, additional sequential proteolysis by TNF-converting enzyme (ADAM17) and the presenilin-dependent γ -secretase results in the release of the Notch intracellular domain (NICD) which translocates into the nucleus, activating transcription of Notch-responsive genes (10). Human Notch-1 ECD aa 19-526, including the first 13 EGF repeats, shows 91% aa identity with corresponding regions of mouse and rat, 89% with canine, and 79% with chicken Notch-1. This region also exhibits 60% aa identity with human Notch-2 and Notch-3.

References:

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