

Human Notch-3 Antibody

Antigen Affinity-purified Polyclonal Sheep IgG Catalog Number: AF1559

DESCRIPTION			
Species Reactivity	Human		
Specificity	Detects human Notch-3 in direct ELISAs and Western blots. In direct ELISAs and Western blot, approximately 30% cross-reactivity with recombinant mouse Notch-3 is observed and less than 5% cross-reactivity with recombinant rat Notch-2 is observed.		
Source	Polyclonal Sheep IgG		
Purification	Antigen Affinity-purified		
Immunogen	recombinant human Notch-3 extracellular domain Ala40-Glu467 Accession # Q9UM47		
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.		
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.		
APPLICATIONS Please Note: Optimal dilution	ons should be determined by each	laboratory for each application	n. General Protocols are available in the Technical Information section on our website.
		Recommended Concentration	Sample
Western Blot		0.1 μg/mL	Recombinant Human Notch-3 Fc Chimera, aa 40-467 (Catalog # 1559-NT)
Blockade of Receptor-ligand Interaction		In a functional ELISA, 1-3 µg/mL of this antibody will block 50% of the binding of 250 ng/mL of Recombinant Rat Jagged 1 Fc Chimera (Catalog # 599-JG) to immobilized Recombinant Human Notch-3 Fc Chimera (Catalog # 1559-NT) coated at 5 µg/mL (100 µL/well). At 10 µg/mL, this antibody will block >90% of the binding.	
PREPARATION AND S	TORAGE		
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.		
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.		

BACKGROUND

Human Notch-3 is part of the Notch family of type I transmembrane glycoproteins involved in a number of early-event developmental processes (1). The extracellular domain of Notch receptors interact with the extracellular domain of transmembrane ligands Jagged, Delta, and Serrate expressed on the surface of a neighboring cell. In both vertebrates and invertebrates, Notch signaling is important for specifying cell fates and for defining boundaries between different cell types. The Notch molecule is synthesized as a 2321amino acid (aa) precursor that contains an 39 aa signal sequence, a 1603 aa extracellular region, a 21aa transmembrane (TM) segment and a 658 aa cytoplasmic domain. The large Notch extracellular domain has 34 EGF-like repeats followed by three notch/Lin-12 repeats (LNR) (2). The 11th and 12th EGF-like repeats of Notch have been shown to be both necessary and sufficient for binding the ligands Serrate and Delta, in Drosophila (3). Notch-3 has the same biochemical mechanism of signal tranduction as Notch-1, where a series of cleavage events result in the release of the Notch intracellular domain (NICD). NICD translocates into the nucleus and initiates transcription of Notch-responsive genes (4). Thus, Notch acts as both a ligand-binding receptor and a nuclear factor that regulates transcription.

Mutations in Notch-3 in humans cause an autosomal dominant condition called CADASIL (cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy). This disorder is characterized by recurrent ischemic strokes at an early age without any underlying vascular risk and progressive dementia. Nearly all mutations leading to this disorder are clustered in the first 5 EGF repeats of the Notch-3 gene (5). Human Notch-3 shows 90% aa identity to mouse Notch-3 over the entire protein.

References:

- 1. Weinmaster, G. (2000) Curr. Opin. Genet. Dev. **10**:363.
- 2. Joutel, A. et al. (1996) Nature 383:707.
- 3. Rebay, I. et al. (1991) Cell **67**:687.
- 4. Mizutani, T. et al. (2001) Proc. Natl. Acad. Sci. USA 98:9026.
- 5. Joutel, A. and E. Tounier-Lasserve (1998) Sem Cell & Dev Biol. 9:619.

1 month from date of receipt, 2 to 8 °C, reconstituted. 6 months from date of receipt, -20 to -70 °C, reconstituted.

> RED SYSTEMS