

Store at  
-20°C

# PhosphoPlus® Notch1 (Cleaved, Val1744) Antibody Duet

Cell Signaling  
TECHNOLOGY®

#8216

Support: +1-978-867-2388 (U.S.)  
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orders@cellsignal.comEntrez-Gene ID #  
UniProt ID #

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**For Research Use Only. Not For Use In Diagnostic Procedures.**

Products Included	Product #	Quantity	Mol. Wt.	Isotype/Source
Notch1 (D1E11) XP® Rabbit mAb	3608	100 µl	120, 300 kDa	Rabbit IgG
Cleaved Notch1 (Val1744) (D3B8) Rabbit mAb	4147	100 µl	110 kDa	Rabbit IgG

See [www.cellsignal.com](http://www.cellsignal.com) for individual component applications, species cross-reactivity, dilutions, and additional application protocols.

**Description:** PhosphoPlus® Duets from Cell Signaling Technology (CST) provide a means to assess protein activation status. Each Duet contains an activation-state and total protein antibody to your target of interest. These antibodies have been selected from CST's product offering based upon superior performance in specified applications.

**Background:** Notch proteins (Notch1-4) are a family of transmembrane receptors that play important roles in development and the determination of cell fate (1). Mature Notch receptors are processed and assembled as heterodimeric proteins, with each dimer comprised of a large extracellular ligand-binding domain, a single-pass transmembrane domain, and a smaller cytoplasmic subunit (Notch intracellular domain, NICD) (2). Binding of Notch receptors to ligands of the Delta-Serrate-Lag2 (DSL) family triggers heterodimer dissociation, exposing the receptors to proteolytic cleavages; these result in release of the NICD, which translocates to the nucleus and activates transcription of downstream target genes (3,4).

**Specificity/Sensitivity:** Notch1 (D1E11) XP® Rabbit mAb detects intracellular epitopes between 2400 and 2500 amino acids of human Notch1. It recognizes both the full-length (~300 kDa) and the NTM region (~120 kDa), which consists of a short extracellular juxtamembrane peptide, a transmembrane sequence and the intracellular domain (NICD). The antibody cannot detect the extracellular (ligand-binding) domain of Notch1 following cleavage at the S2 site by ADAM-type metalloproteases. Cleaved Notch1 (V1744) (D3B8) Rabbit mAb detects endogenous levels of the Notch1 intracellular domain (NICD) only when released by cleavage between Gly1753 and Val1754 (equivalent to Gly1743/Val1744 of murine notch1). The antibody does not recognize full-length Notch1 or Notch1 cleaved at other positions. The size of the NICD varies among cell lines due to mutations in the Notch1 C-terminus (6).

**Source/Purification:** Monoclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Pro2438 of human Notch1 or with a synthetic peptide corresponding to the sequence at the Val1754 cleavage site in human Notch1 (equivalent to Val1744 in mouse Notch1).

**Storage:** Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. *Do not aliquot the antibody.*

**Background References:**

- (1) Artavanis-Tsakonas, S. et al. (1999) *Science* 284, 770-6.
- (2) Chan, Y.M. and Jan, Y.N. (1998) *Cell* 94, 423-6.
- (3) Schroeter, E.H. et al. (1998) *Nature* 393, 382-6.
- (4) Rand, M.D. et al. (2000) *Mol Cell Biol* 20, 1825-35.

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**Applications:** W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide **Species Cross-Reactivity:** H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.