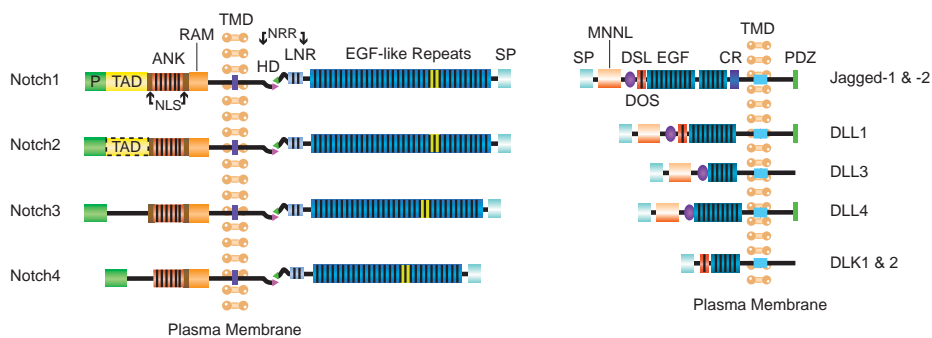


# Notch Signaling Pathway

Important for Cancer & T Cell Research

## The Importance of Notch

The highly conserved Notch signaling pathway regulates many different cell fate decisions in both vertebrate and invertebrate species. It is important for pattern formation during development such as neurogenesis, angiogenesis or myogenesis and regulates T cell development and stem cell maintenance [1]. Notch signaling is also involved in cellular processes throughout adulthood [2]. Signaling via Notch occurs between receptors and its ligands, both at the surface of neighbouring cells (see Figure 1, Notch Receptors and Their Ligands). In mammals, expression of four Notch receptors (Notch1–4) and five canonical ligands [Delta-like ligand (DLL1, 3, 4) and Jagged (Jagged-1, -2) coordinate activation of this signaling pathway [3].



**FIGURE 1: Notch Receptors and their Ligands.**

Mammals possess four Notch receptors (Notch1–4) and five ligands including Jagged-1 and -2 and Delta-like (DLL) 1, 3 and 4. Additional noncanonical Notch ligands are DLK1, DLK2.

**ANK:** Ankyrin Repeats; **CR:** Cysteine-rich Domain; **DOS:** Delta and OSM-11-like Proteins Domain; **DSL:** Delta, Serrate and LAG-2 Domain; **EGF:** Epidermal Growth Factor-like Repeats; **HD:** Heterodimerization Domain; **LNR:** Cysteine-rich Lin12-Notch Repeats; **NRR:** Negative Regulatory Region; **MNNL:** Module at N-terminal Domain of Notch Ligands; **NLS:** Nuclear Localization Signal; **P:** PEST Domain; **PDZ:** PDZ Domain; **PM:** Plasma Membrane; **RAM:** RBPJ-associated Molecule; **SP:** Signal Peptide; **TAD:** Transactivation Domain; **TMD:** Transmembrane Domain

Adapted from: *The intracellular region of Notch ligands: does the tail make the difference?* A. Pintar, et al.; Biol. Direct 2, 19 (2007), *The canonical Notch signaling pathway: unfolding the activation mechanism:* R. Kopan & M. X. Ilagan; Cell 137, 216 (2009)

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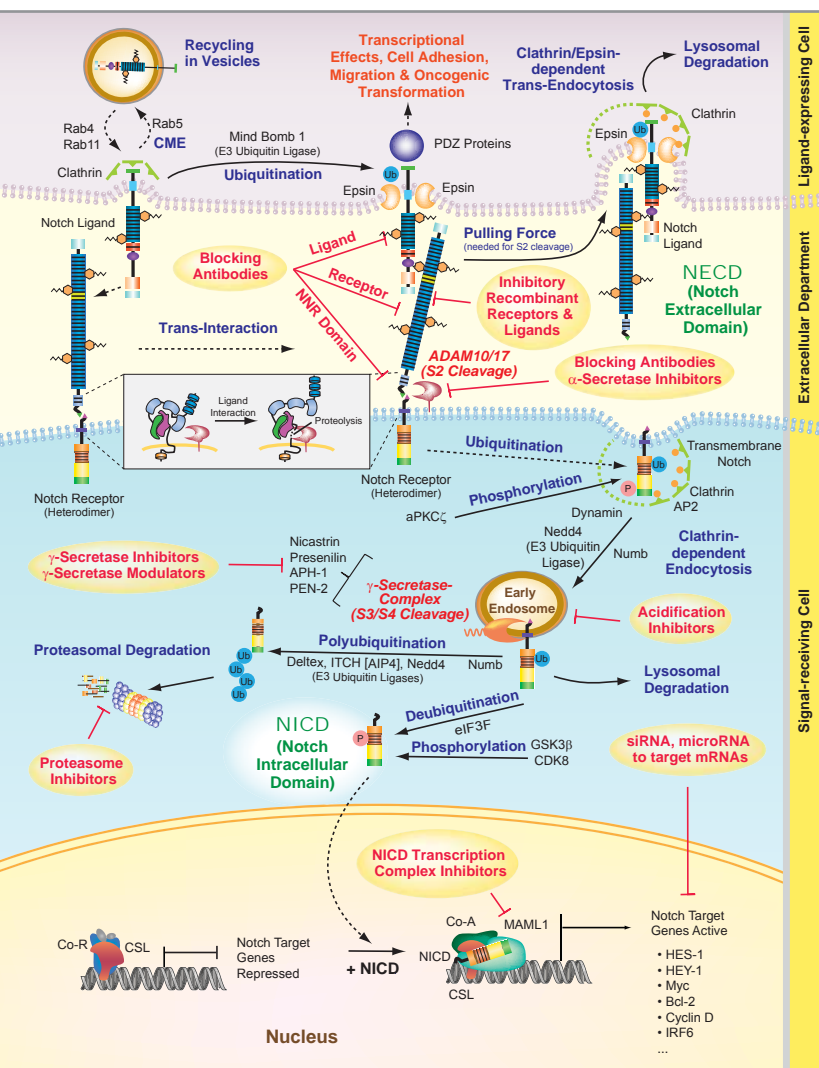
## Notch Receptors and Ligands Family

Mammalian Notch receptor homologs (Notch1 to 4) encode a Notch extracellular domain (NECD) that binds ligands, a transmembrane domain, and a Notch intracellular domain (NICD) that translocates to the nucleus to serve as a transcriptional cofactor. Mammalian NECDs consist of 29 to 36 EGF repeats followed by three Lin-Notch repeats (LNRs). EGF11 and 12 domains alone are sufficient for binding to Notch ligands (Jagged/DLL). All canonical Notch ligands are transmembrane proteins that share a largely similar structure, with an extracellular domain comprised primarily of multiple EGF repeats (6 for DLL3; 8 for DLL1 and DLL4; or 16 for Jagged-1 and Jagged-2), followed by "module at the N-terminus of Notch ligands" (MNNL) domain and by a "Delta/Serrate/Lag-2 (DSL) domain" [1].

The non-canonical Notch ligands lack the DSL domain, among these are proteins delta homolog 1 and 2 (DLK1 and DLK2) [4]. Some proteins including Contactin-3 and -6 and DNER have been postulated to act as Notch ligands, but confirmation of these observations are still needed [5].

## Activation of Notch Signaling

The Notch receptors are synthesized as single precursor proteins that are cleaved during transport to the cell surface (at cleavage site S1, not shown in the Figure 2), where they are expressed as heterodimers. Notch signal transduction is initiated upon binding of a Notch receptor heterodimer to a ligand located on a neighbour cell (see Figure 2: Notch Signaling Pathway). Upon receptor-ligand binding, ubiquitination by RING E3 ligases (such as Mind bomb (Mib) or Neuralized), marks the ligands for Epsin-dependent endocytosis. This event generates a mechanical pulling force, which drives conformational changes of the Notch receptor and facilitates its sequential proteolytic cleavages [3]. The cleavage (at S2 site) which is triggered by ligand binding and mediated by a disintegrin and metalloproteinase (ADAM family, also called TACE, tumor necrosis factor- $\alpha$ -converting enzyme) family peptidase, releases the NECD, whereas the cleavage (at S3/S4 sites) mediated by  $\gamma$ -secretase activity of a multiprotein complex (consisting of presenilin, nicastrin, APH1 and PEN2) releases the NICD. The Notch intracellular domain translocates to the nucleus where it binds with CSL/Rbpj (recombination signal binding protein for immunoglobulin  $\kappa$  j region) and recruits a transcriptional complex to activate the transcription of downstream targets, including Hairy/enhancer-of-split (Hes) and Hes-related with YRPW motif protein (Hey) family genes [6]. Activity of Notch receptors and ligands is profoundly affected by glycosylation of EGF repeats in the extracellular domain. O-fucosyltransferases, which add fucose to serine and threonine residues and O-glycosyltransferases, which add glucose to serine residues, followed by extension of the sugar by Fringe family GlcNAc-transferases are essential for modulating the binding avidity of ligand-receptor pairs. Other post-translational events, including mono- or polyubiquitination by specific E3 ubiquitin ligases and phosphorylation as well as endocytic trafficking, regulate the activities of both the Notch receptors and their ligands.



**FIGURE 2: Simplified Notch Signaling Pathway, including potential therapeutic target possibilities.**

**NUCLEUS:** Co-A: Co-Activator Proteins; Co-R: Co-Repressor Proteins; CSL: CBF1/Su(H)/LAG-1 Complex; MAML1: Mastermind-like 1. **SIGNALING:** AP2: Adaptor Protein 2; E3 Ubiquitin Ligases: Deltex, ITCH, Nedd4; eIF3F: Eukaryotic Translation Initiation Factor 3 Subunit F; NRR: Negative Regulatory Region

## Notch and Diseases

The Notch pathway plays an important role in many different processes in a wide range of tissues and deregulations in Notch signaling components have been associated with various human disorders such as cancer, immune disorders, developmental syndromes, stroke and cognitive symptoms. Other disorders affecting vertebral column such as scoliosis or the vasculature, hypertension and the developmental disorder Alagille syndrome are also caused by Notch defects [7].

## Notch and Cancer

Components of the Notch signaling pathway are altered in diseases and cancers (T and B cell lymphoproliferative disorders, liver, breast, brain, bladder, lung and prostate). Notch can act either as an oncogene or tumor suppressor depending on the cellular context. Components of the Notch signaling are not frequently mutated in most tumor types, although mutations appear to accumulate during growth of tumors. However, there are exceptions with loss-of-function mutations in Notch receptors supporting their tumor-suppressive role in multiple malignancies, including bladder cancer and squamous cell carcinoma. Constitutive activation of the Notch receptors through gene rearrangements or gain-of-function mutations leads to Notch receptors' oncogenic function in T cell acute lymphoblastic leukemia, in chronic lymphocytic leukemia and in solid tumors such as lung adenocarcinoma. In breast and prostate cancer, Notch signaling frequently appears to be upregulated, and high levels of Jagged-1 expression correlate with poor prognosis of some tumors showing that the level of Notch signaling is critical in regulation of cell proliferation, survival or death. Given that Notch signaling is dysregulated in different types of cancer, Notch inhibitors alone or in combination with chemotherapeutics are currently clinically evaluated and become an exciting new approach to fight cancer (see Figure 2).

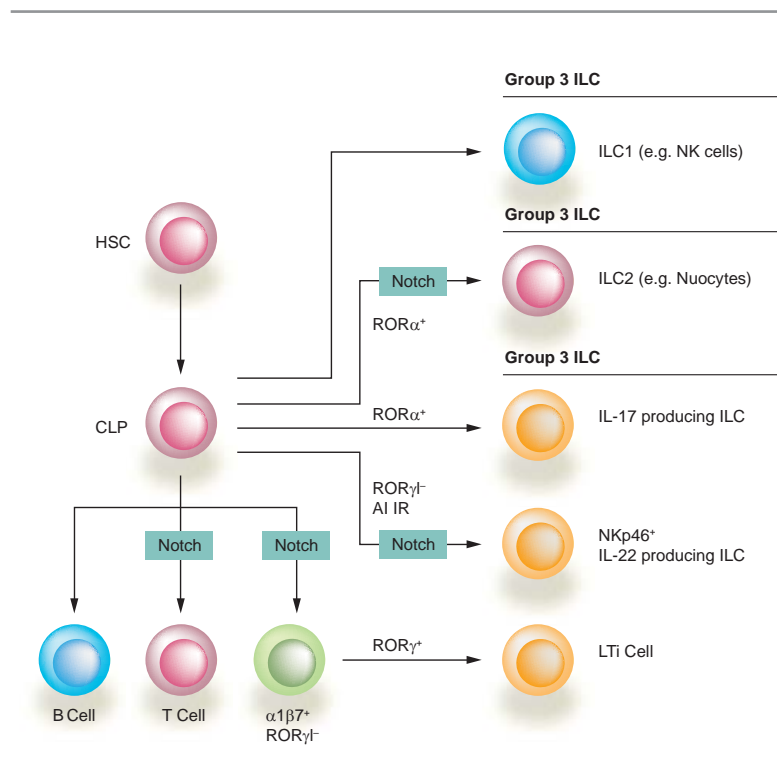
## Notch and Regulation of Innate and Adaptive Immunity

Notch signaling plays an essential role during development and differentiation of hematopoietic cells [8]. During early stages of T cell development, Notch is required continuously in the thymus while in the bone marrow, it inhibits B cell development. Notch also plays essential roles later during lymphocyte development, in particular during T cell lineage commitment and maturation in the thymus and during marginal zone B (MZB) cell development in the spleen. Notch is also a key factor in dendritic cell (DC) homeostasis. Finally, Notch functions in the development of the newly described Innate Lymphoid Cells (ILCs) playing roles in innate immune responses to infectious microorganisms, in the generation of secondary lymphoid organs and in tissue remodeling after tissue injury or infection (see Figure 3).

**FIGURE 3: The role of Notch Signaling in the development of innate lymphoid cells.**

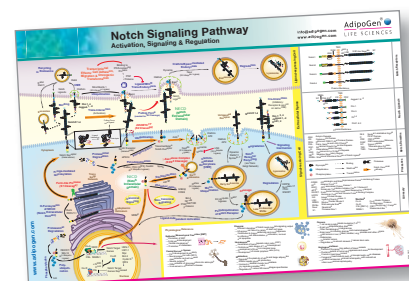
Haematopoietic stem cell (HSC)-derived common lymphoid progenitors (CLPs) give rise to adaptive immune cells, such as T cells and B cells, as well as to innate lymphoid cells (ILCs). ILCs function in innate immune responses and are grouped into three major classes: group 1, group 2 and group 3. ILCs diverge in their requirement for Notch (as indicated). AHR: aryl hydrocarbon receptor; IL: interleukin; LTi: lymphoid tissue-inducer; NK: natural killer; ROR: retinoid-related orphan receptor.

Adapted from: *Regulation of innate and adaptive immunity by Notch*: F. Radtke, et al.; *Nat. Rev. Immunol.* **13**, 427 (2013)



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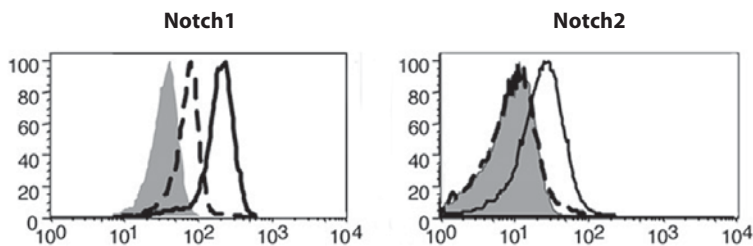
Ask for our detailed Notch Signaling Wallchart or download it from [www.adipogen.com](http://www.adipogen.com)

# Notch Receptors Notch1 & Notch2

ANTIBODIES	PID	SIZE	ISOTYPE	APPLICATION	SPECIES
<b>anti-Notch1 (mouse), mAb (22E5)</b>	AG-20B-0051	100 µg	Rat IgG2 $\alpha$	FACS	Ms
<b>anti-Notch1 (mouse), mAb (22E5) (Biotin)</b>	AG-20B-0051B	100 µg	Rat IgG2 $\alpha$	FACS	Ms
<b>anti-Notch2, mAb (16F11)</b>	AG-20B-0052	100 µg	Rat IgG1 $\kappa$	FACS	Ms
<b>anti-Notch2, mAb (16F11) (Biotin)</b>	AG-20B-0052B	100 µg	Rat IgG1 $\kappa$	FACS	Ms

PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
<b>Notch1 (mouse):Fc (human) (rec.)</b>	AG-40B-0109	50 µg   3 x 50 µg	CHO cells	<0.1EU/µg	Ms
<b>Notch2 (mouse):Fc (human) (rec.)</b>	AG-40B-0110	50 µg   3 x 50 µg	CHO cells	<0.01EU/µg	Ms



**FIGURE:** Detection of endogenous mouse Notch1 or Notch2 on resting and activated T cells with anti-Notch1 (mouse), mAb (22E5) (Prod. No. AG-20B-0051) and anti-Notch2, mAb (16F11) (Prod. No. AG-20B-0052), respectively.

**METHOD:** CD4<sup>+</sup>T cells from C57BL/6 mice were treated with anti-CD3 on plastic (solid line), IL-2 (dotted line) or medium alone as a negative control (shaded histogram) for 24h. The staining was revealed with a secondary anti-mouse IgG-PE (1/200) and then analyzed by flow cytometry.

## Canonical Notch Ligands

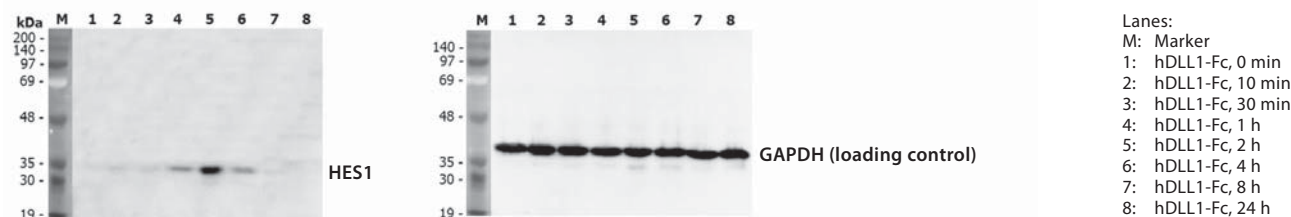
### Notch Ligand DLL1 (Delta-like Protein 1)

**BULK**

ANTIBODIES	PID	SIZE	ISOTYPE/SOURCE	APPLICATION	SPECIES
<b>anti-DLL1 (human), mAb (D1L165-6)</b>	AG-20A-0074	50 µg   100 µg	Mouse IgG1 $\kappa$	ELISA, WB	Hu
<b>anti-DLL1 (mouse), mAb (D1L357-1-4)</b>	AG-20A-0085	50 µg   100 µg	Rat IgG2 $\kappa$	ELISA, WB	Ms
<b>anti-DLL1 (mouse), mAb (30B11.1)</b>	AG-20B-0053	100 µg	Rat IgG2 $\alpha$	FACS, ICC	Ms
<b>anti-DLL1 (human), pAb</b>	AG-25A-0062	100 µg	Rabbit	ELISA, IHC, WB	Hu
<b>anti-DLL1 (human), pAb</b>	AG-25A-0079	100 µg	Rat	ELISA, WB	Hu

PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
<b>DLL1 (human) (rec.)</b>	AG-40A-0073	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Hu
<b>DLL1 (human):Fc (human) (rec.)</b>	AG-40A-0116Y	10 µg   50 µg	CHO cells	<0.01EU/µg	Hu
<b>DLL1 (mouse):Fc (human) (rec.)</b>	AG-40A-0148	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Ms



**FIGURE:** DLL1 (human):Fc (human) (rec.) (AG-40A-0116Y) induces the Notch target gene HES1 when coated on a plate.

**METHOD:** A mouse preadipocyte cell line, 3T3L1, was stimulated with 1µg/ml of human DLL1:Fc as in indicated time points and each cell lysate was prepared and subjected to Western blot by using an anti-mouse HES1 or anti-mouse GAPDH specific antibody.

## Notch Ligands DLL3 & DLL4 (Delta-like Protein 3 & 4)

**BULK**

ANTIBODIES	PID	SIZE	ISOTYPE	APPLICATION	SPECIES
<b>anti-DLL4 (human), mAb (DL86-3AG)</b>	AG-20A-0080	50 µg   100 µg	Mouse IgG1κ	ELISA, WB	Hu
<b>anti-DLL4 (mouse), mAb (9A1.5)</b>	AG-20B-0054	100 µg	Rat IgG1κ	FACS, ICC	Ms
PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
<b>DLL3 (human) (rec.)</b>	AG-40B-0151	10 µg   3 x 10 µg	HEK 293 cells	<0.02EU/µg	Hu
<b>DLL3 (extracellular domain) (mouse):Fc (human) (rec.)</b>	AG-40A-0178	10 µg	HEK 293 cells	<0.1EU/µg	Ms
<b>DLL4 (human):Fc (human) (rec.)</b>	AG-40A-0077Y	10 µg   50 µg	HEK 293 cells	<0.01EU/µg	Hu
<b>DLL4 (mouse):Fc (human) (rec.)</b>	AG-40A-0145	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Ms

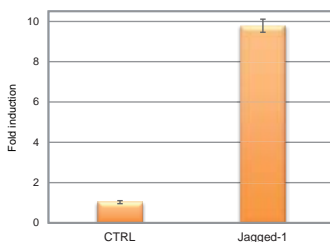
Literature Citations in High Ranking Journals using AdipoGen's DLL4 (human):Fc (human) [PID# AG-40A-0077Y]:

- Jagged2 acts as a Delta-like Notch ligand during early hematopoietic cell fate decisions: I. Van de Walle, et al.; Blood 117, 4449 (2011)
- Notch regulates BMP responsiveness and lateral branching in vessel networks via SMAD6: K.P. Mouillessaux, et al.; Nat. Commun. 7, 13247 (2016)

## Notch Ligands Jagged-1 & Jagged-2

**BULK**

ANTIBODIES	PID	SIZE	ISOTYPE	APPLICATION	SPECIES
<b>anti-Jagged-1 (human), mAb (J1G53-3)</b>	AG-20A-0049	100 µg	Mouse IgG1κ	ELISA, FACS, IHC, WB	Hu
<b>anti-Jagged-1 (human), mAb (J1G53-3) (FITC)</b>	AG-20A-0049F	50 µg	Mouse IgG1κ	FACS, WB	Hu
<b>anti-Jagged-1 (human), mAb (J1G74-7)</b>	AG-20A-0050	100 µg	Mouse IgG1κ	ELISA, FACS, WB	Hu
PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
<b>Jagged-1 (human):Fc (human) (rec.)</b>	AG-40A-0081	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Hu
<b>Jagged-1 (mouse):Fc (human) (rec.)</b>	AG-40A-0157T	10 µg   50 µg	HEK 293 cells	<0.01EU/µg	Ms
<b>Jagged-2 (human):Fc (human) (rec.)</b>	AG-40A-0155Y	10 µg	HEK 293 cells	<0.1EU/µg	Hu
<b>Jagged-2 (mouse):Fc (human) (rec.)</b>	AG-40A-0183	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Ms



**FIGURE:** Induction of IL-6 expression in human dermal fibroblasts by Jagged-1 (human):Fc (human) (rec.) (Prod. No. AG-40A-0081).

**METHOD:** Jagged-1 (human):Fc was coated on a 12-well plate at 1µg/ml overnight at 4°C. Human dermal fibroblasts were cultured in the presence or absence of Jagged-1 (human):Fc for 72 hours. Real time quantitative PCR was used to quantify the expression of IL-6.

Picture courtesy of the lab of Prof. Gian-Paolo Dotto, Department of Biochemistry, University of Lausanne

## LATEST INSIGHT

### New role of Jagged-1 & OX40L in the selective induction of Treg proliferation

P. Kumar, et al. reported that the Notch ligand Jagged-1 and the TNF superfamily ligand OX40L induce selective proliferation of functional regulatory T cells (Tregs) independent of canonical TCR signaling (in the absence of anti-CD3/CD28 activation) when used together as soluble recombinant ligands. This activation of Tregs by Jagged/OX40L works in an IL-2 dependent way without activating effector T cells. This novel "TCR-independent" strategy using Jagged-1, OX40L and IL-2 for the selective expansion of functional Tregs could have therapeutic implications in various autoimmune diseases including T1D.

**LIT:** Soluble OX40L and JAG1 Induce Selective Proliferation of Functional Regulatory T-Cells Independent of canonical TCR signaling; P. Kumar, et al.; Sci. Rep. 7, 39751 (2017)



# Non-Classical Notch Ligands

## DLK1 & DLK2 (Protein Delta Homolog 1)

ANTIBODIES	PID	SIZE	ISOTYPE/SOURCE	APPLICATION	SPECIES
anti-DLK1 (human), mAb (PF13-3)	AG-20A-0069	50 µg   100 µg	Mouse IgG1κ	ELISA, FACS, IHC, WB	Hu
anti-DLK1 (human), mAb (PF299-1)	AG-20A-0070	50 µg   100 µg	Mouse IgG1κ	ELISA, FACS, IHC, WB	Hu
anti-DLK1 (mouse), mAb (PF105B)	AG-20A-0057	50 µg   100 µg	Rat IgG2ακ	ELISA, WB	Ms
anti-DLK1 (mouse), mAb (PF183E)	AG-20A-0058Y	50 µg   100 µg	Rat IgG2ακ	ELISA, WB	Ms
anti-DLK1 (human), pAb	AG-25A-0091	100 µg	Rat	ELISA, FACS, WB	Hu
anti-DLK1 (human), pAb	AG-25A-0092	100 µg	Rabbit	ELISA, IHC, WB	Hu

PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
DLK1 (human) (rec.)	AG-40A-0133	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Hu
DLK1 (human):Fc (human) (rec.)	AG-40B-0152	10 µg   3 x 10 µg	HEK 293 cells	<0.01EU/µg	Hu
DLK1 (mouse):Fc (human) (rec.)	AG-40A-0107Y	10 µg   3 x 10 µg	HEK 293 cells	<0.1EU/µg	Ms
DLK2 (human):Fc (human) (rec.)	AG-40A-0158	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Hu

### DLK1, Soluble (human) ELISA Kit

AG-45A-0032Y

96 wells

**Species reactivity:** Human  
**Sensitivity:** 336 pg/ml  
**Range:** 0.47 to 30 ng/ml  
**Assay type:** Sandwich  
**Sample type:** Serum, Cell Culture Supernatant



## Non-Confirmed Notch Ligand

### DNER (Delta and Notch-like Epidermal Growth Factor-related Receptor)

ANTIBODIES	PID	SIZE	ISOTYPE/SOURCE	APPLICATION	SPECIES
anti-DNER (human), mAb (DR324-4)	AG-20A-0078	50 µg   100 µg	Mouse IgG2ακ	ELISA, WB	Hu
anti-DNER (human), pAb	AG-25A-0102	100 µg	Rabbit	ELISA, WB	Hu

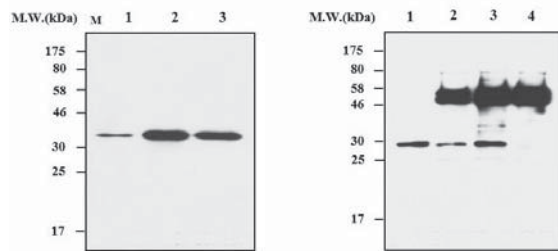
PROTEINS	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
DNER (extracellular domain) (human) (rec.)	AG-40A-0137Y	10 µg   3 x 10 µg	HEK 293 cells	<0.1EU/µg	Hu
DNER (extracellular domain) (human):Fc (human) (rec.)	AG-40A-0119	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Hu
DNER (extracellular domain) (mouse):Fc (human) (rec.)	AG-40A-0177	10 µg   50 µg	HEK 293 cells	<0.1EU/µg	Ms

## Notch Target HES1 (Hairy and Enhancer of Split 1)

Hes genes, encoding basic helix–loop–helix (HLH) transcriptional repressors, are seven members in human, expressed in many tissues and playing various roles mainly in development. Hes1, Hes5, and Hes7 are downstream effectors of canonical Notch signaling. Hes1 plays a crucial role in the control and regulation of cell cycle, proliferation, cell differentiation, survival and apoptosis in neuronal, endocrine and T-lymphocyte progenitors as well as various cancers and is a key target gene of the Notch signaling pathway.

ANTIBODY	PID	SIZE	ISOTYPE	APPLICATION	SPECIES
<b>anti-HES1, mAb (7H11)</b>	AG-20T-0400	100 µg	Mouse IgG2bκ	ELISA, FACS, IP, WB	Hu, Ms, Rt

PROTEIN	PID	SIZE	SOURCE	ENDOTOXIN	SPECIES
<b>HES1 (human) (rec.) (His)</b>	AG-40A-0180	10 µg   50 µg	E. coli	n.a.	Hu



**FIGURE (LEFT):** Western blot analysis on cell lysates HeLa cells (lane1), HT-29 cell (lane2) and BeWo cell (lane 3) using anti-HES1, mAb (7H11) at 1µg/ml.

**FIGURE (RIGHT):** Immunoprecipitation analysis on BeWo cell lysate using anti-HES1, mAb (7H11).

Lane 1: BeWo cell lysate  
 Lane 2: Precipitation from BeWo cell lysate (400µg) at 2µg  
 Lane 3: Precipitation from BeWo cell lysate (400µg) at 5µg  
 Lane 4: Precipitation from PBS at 5µg

## E3 Ubiquitin Ligases (E3UBLs) & Deubiquitinating Enzymes (DUBs)

In contrast to other signaling pathways, where a cascade of second messengers is at stake, the activated Notch receptor is itself transformed into a transcriptional activator, NICD. As a consequence, affecting NICD production and quantity directly affects Notch-dependent response. Therefore, regulating this pathway means controlling spatiotemporal production and maintenance of active receptors and ligands at the cell surface, efficiency of signal transduction and stability of NICD. These key steps all involve ubiquitination events. Dysregulations of these events might be involved in various pathological processes in which the Notch signaling is disrupted. Recently, USP12 has been shown to be important for Notch degradation. The deubiquitinating complex USP12/UAF1 is recruited by Itch to non-activated Notch and regulates Notch trafficking toward lysosomal degradation.

**LIT:** Ubiquitinations in the Notch signaling pathway: J. Moretti & C. Brou; *Int. J. Mol. Sci.* **14**, 6359 (2013)

KIT	PID	SIZE	SOURCE	SPECIES	TYPE
<b>ITCH (human) Ubiquitination Kit</b>	AG-44T-0117	Kit		Hu	E3UBLs

PROTEINS	PID	SIZE	SOURCE	SPECIES	TYPE
<b>ITCH Isoform 2 (human) (rec.)</b>	AG-40T-0284	100 µg	E. coli	Hu	E3UBLs
<b>MDM2 (human) (rec.)</b>	AG-40T-0289	50 µg	E. coli	Hu	E3UBLs
<b>MDM2 (human) (rec.) (GST)</b>	AG-40T-0290	50 µg	E. coli	Hu	E3UBLs
<b>UAF1 (human) (rec.) (His)</b>	AG-40T-0406	50 µg	Sf21 cells	Hu	DUBs
<b>USP1/UAF1 Complex (human) (rec.) (His)</b>	AG-40T-0536	50 µg	Sf21 cells	Hu	DUBs
<b>USP9x Isoform 2 (human) (rec.) (His)</b>	AG-40T-0545	25 µg	Sf21 cells	Hu	DUBs
<b>USP12 (human) (rec.) (His)</b>	AG-40T-0570	50 µg	Sf21 cells	Hu	DUBs
<b>USP12/UAF1 Complex (human) (rec.) (His)</b>	AG-40T-0571	1 vial	Sf21 cells	Hu	DUBs
<b>USP28 (human) (rec.) (His)</b>	AG-40T-0541	100 µg	Sf21 cells	Hu	DUBs

## ADAM17 – Important Sheddase in the Notch Pathway

ADAM17 (Disintegrin and metalloproteinase domain-containing protein 17), also called TACE (Tumor Necrosis Factor- $\alpha$ -Converting Enzyme) is the prototype of the ADAM family of ectodomain shedding proteases (shedase). ADAM17 is responsible for the processing of a diverse variety of membrane-anchored cytokines, cell adhesion molecules, receptors, ligands and enzymes, including processing of tumor necrosis factor  $\alpha$  at the surface of the cell and extracellular Notch Receptor 1. As the proteolytic cleavage is an indispensable activation event for many of these substrates, ADAM17 has emerged as an attractive therapeutic target for the treatment of inflammatory diseases (e.g. rheumatoid arthritis) or inflammation associated cancer.

**NEW**

### anti-ADAM17 (human), mAb (rec.) (blocking) (D1(A12)) (preservative-free)

AG-27B-6000PF 100  $\mu$ g

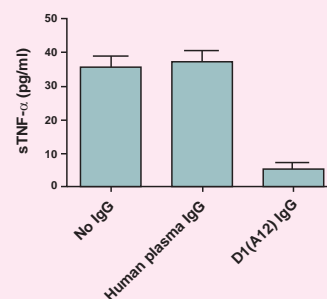
### anti-ADAM17 (human), mAb (rec.) (blocking) (D1(A12)) (Fab Fragment) (His) (preservative-free)

AG-27B-6003PF 100  $\mu$ g

Recognizes the catalytic and non-catalytic domain of human ADAM17 (TACE) through its variable light (VL) domain and variable heavy (VH) domain, respectively. Does not bind recombinant mouse ADAM17 ectodomain.

**Functional Application (Blocking):** Inhibits ADAM17 activity at 15 $\mu$ g/ml (200nM).

**LIT:** Cross-domain inhibition of TACE ectodomain: C.J. Tape, et al.; PNAS 108, 5578 (2011)



**FIGURE:** D1(A12) IgG inhibits constitutive shedding of TNF- $\alpha$  from IGROV1 (human ovarian cancer cell line) into culture medium. Medium was collected after 48 hours of incubation with or without IgGs at 200nM.

## Notch Processing / $\gamma$ -Secretase Inhibitors

### Compound E

AG-CR1-0081

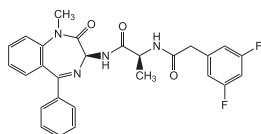
**Formula:** C<sub>27</sub>H<sub>24</sub>F<sub>2</sub>N<sub>4</sub>O<sub>3</sub>

**MW:** 490.5

**CAS:** 209986-17-4

Non-competitive  $\gamma$ -secretase inhibitor.  
Notch processing inhibitor.

250  $\mu$ g | 1 mg | 5 mg



### DAPT

AG-CR1-0016

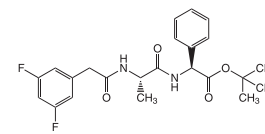
**Formula:** C<sub>23</sub>H<sub>26</sub>F<sub>2</sub>N<sub>2</sub>O<sub>4</sub>

**MW:** 432.5

**CAS:** 208255-80-5

Cell permeable  $\gamma$ -secretase inhibitor.  
Notch processing inhibitor.

5 mg | 25 mg



### Compound 34

AG-CR1-0007

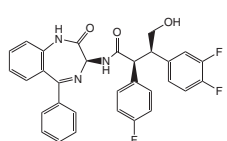
**Formula:** C<sub>31</sub>H<sub>24</sub>F<sub>3</sub>N<sub>3</sub>O<sub>3</sub>

**MW:** 543.5

**CAS:** 564462-36-8

Cell permeable, highly potent inhibitor  
of  $\gamma$ -secretase (IC<sub>50</sub> = 0.06nM).

200  $\mu$ g | 1 mg



### Withaferin A

AG-CN2-0490

**Formula:** C<sub>28</sub>H<sub>38</sub>O<sub>6</sub>

**MW:** 470.6

**CAS:** 5119-48-2

Notch receptors modulator.

**LIT:** J. Lee, et al.; Breast Cancer Res. Treat. 136, 45 (2012)

1 mg | 5 mg | 10 mg

