0031-3998/05/5705-0104R
PEDIATRIC RESEARCH

Vol. 57, No. 5, Pt 2, 2005
Printed in U.S.A.

Notch Signaling, Brain Development, and Human Disease

JOSEPH L. LASKY AND HONG WU

University of California, Los Angeles School of Medicine, Department of Molecular and Medical Pharmacology, Los Angeles, California, 90025

ABSTRACT

The Notch signaling pathway is central to a wide array of developmental processes in a number of organ systems, including hematopoiesis, somitogenesis, vasculogenesis, and neurogenesis. These processes involve maintenance of stem cell self-renewal, proliferation, specification of cell fate or differentiation, and apoptosis. Recent studies have led to the recognition of the role of the Notch pathway in early neurodevelopment, learning, and memory, as well as late-life neurodegeneration. This review summarizes what is currently known about the role of the Notch pathway in neural stem cells, gliogenesis, learning and memory, and neurologic disease. **(*Pediatr Res* 57: 104R–109R, 2005)**

Abbreviations
FCD, focal cortical dysplasia
ICD, intracellular domain
PS1, presenilin1

The formation of the mammalian nervous system takes place *via* a number of developmental steps. All phases of brain development involve the recurrent themes of induction, cell proliferation, cell fate determination (differentiation), cell movement (migration), cell process formation, and targeting (synapse formation) (1). Signaling pathways involved in these processes are regulated not only in space, but in time and intensity as well. A vast array of signaling events are coordinated such that cells proliferate and differentiate at the correct time, space, and orientation to generate an amazingly organized structure capable of adaptability and plasticity. The Notch signaling pathway, originally discovered in *Drosophila*, impinges on a wide array of cellular processes including maintenance of stem cell self-renewal, proliferation, specification of cell fate or differentiation, and apoptosis. What follows is a review of the role of the Notch signaling pathway in neurodevelopmental processes and its role in the pathogenesis of certain human neurologic diseases.

DISCUSSION

Notch signaling. The Notch gene encodes a receptor with a single transmembrane domain. Although initially synthesized as a single protein, it is cleaved in two and exists as a heterodimeric receptor embedded in the plasma membrane. Signaling is initiated when a Notch receptor on one cell interacts with Notch ligands, such as Delta or Serrate (in *Drosophila*), on an adjacent cell (Fig. 1). This interaction triggers two proteolytic events culminating in the release of the Notch ICD. The free intracellular fragment then translocates to the nucleus where it binds to the transcriptional regulator CSL [for CBF-1, Su(H), and LAG-1], resulting in displacement of co-repressors previously bound to CSL and recruitment of co-activators. The co-activators then induce expression of the Hairy-Enhancer of Split (HES) and Hes-related proteins (HERP) gene families, although recent data suggest that CSL-independent pathways may also exist (2–4). It is also important to note that in mammals there exist multiple subtypes of each "actor" in this pathway including Notch1–4, Delta-like ligands (Dll)-1,-3, and -4, and Serrate-like ligands (Jagged-1 and -2) (3). This becomes relevant later on in this review in that various human diseases are usually associated with a particular Notch gene. Notch is involved in mediating two distinct types of cell-cell signaling interactions: lateral and inductive signaling (5,6). Studies in *Drosophila* revealed the importance of Notch in the control of cellular differentiation by lateral inhibition, which ensures that two distinct cell types are produced in correct numbers from a population of initially equipotent cells (7). For example, in *Drosophila*, Delta signaling to Notch induces the Notch responding cell to remain a progenitor cell, whereas the Delta-expressing cell differentiates into a neural cell (8). Although the equipotent cells initially express equivalent levels of both ligands and receptors, *via* a negative feedback mechanism, the Notch responding cell down-regulates its own Delta ligand expression effectively blocking Notch receptor pathway activity in the Delta signaling cell

Received January 4, 2005; accepted January 31, 2005.
Correspondence: Joseph L. Lasky, M.D., Department of Molecular and Medical Pharmacology, UCLA School of Medicine, 650 Charles Young, Dr, 23-234 CHS, P.O. Box 951735, Los Angeles, CA 90095-1735; E-mail: jlasky@mednet.ucla.edu
Supported in part by NCI Cancer Education Grant R25 CA 098010.

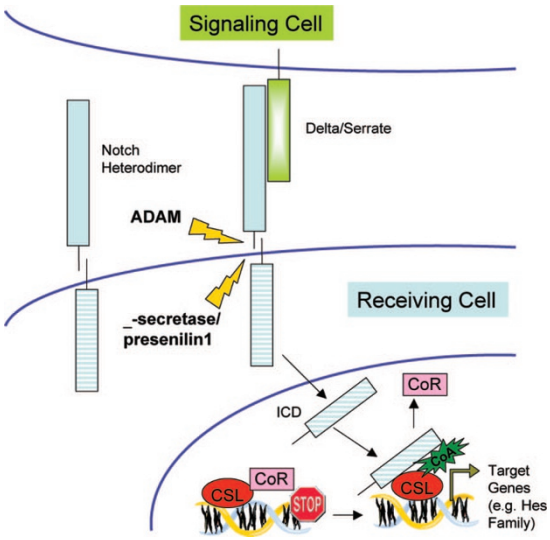

Figure 1. The heterodimeric Notch receptor, upon contact with its ligand (*e.g.* Delta) undergoes proteolytic cleavage first by an ADAM family protease and then by γ-secretase/presenilin1. This liberates the intracellular domain of Notch (ICD) allowing it to translocate into the nucleus where it displaces co-repressors (CoR) from the CSL transcription factor. Subsequent binding to CSL then occurs and recruitment of co-activators (CoA) results in the transcriptional activation of downstream target genes.

(Fig. 2*A*). Inductive signaling, on the other hand, involves Notch receptor and ligand expressed on two different cell types such that Notch is only activated in the receptor-bearing cell, resulting in a cell-fate decision (Fig. 2*B*). This role for Notch signaling has been demonstrated in multiple settings including T-cell lineage specification (9,10), mammalian keratinocyte differentiation (11), and mammalian gliogenesis (12–14). The importance of these mechanisms is illustrated by their conservation across multiple species, from *Drosophila* and *Caenorhabditis elegans* to amphibians and mice (8,15–18).

Stem cell maintenance. In *Drosophila*, Notch prevents early neurocompetence in ectodermal cells *via* interplay with the Wnt signaling pathway (19,20). Although this particular role has not yet been shown to be active in vertebrates, other Notch1-mediated signaling pathways are crucial for mammalian CNS development *via* maintenance of a neural stem cell (progenitor) state, inhibition of neuronal commitment, and promotion of glial fates (12,21). Notch1 mouse mutant embryos die before E11.5, near to the time when the first neurons express their mature, differentiated phenotype (17). However, close examination of mutant embryos revealed hypoplastic brains and neural tubes secondary to a loss of neuroblasts and premature neuronal differentiation (22). Further support for the role of the Notch pathway in the inhibition of neuronal differentiation came from studies on PS1 (presenilin), which is a component of the γ-secretase complex that allows for the efficient proteolytic release of the Notch ICD. PS1 knock-out mice (PS1−/−) also experience a reduction in the neural progenitor population (23). Consistent with these findings, overexpression of Notch also promotes maintenance of neural precursors (13,24). The downstream signaling pathways responsible for these effects are being elucidated. The basic helix-loop-helix genes Hes1 and Hes5 are the major effectors

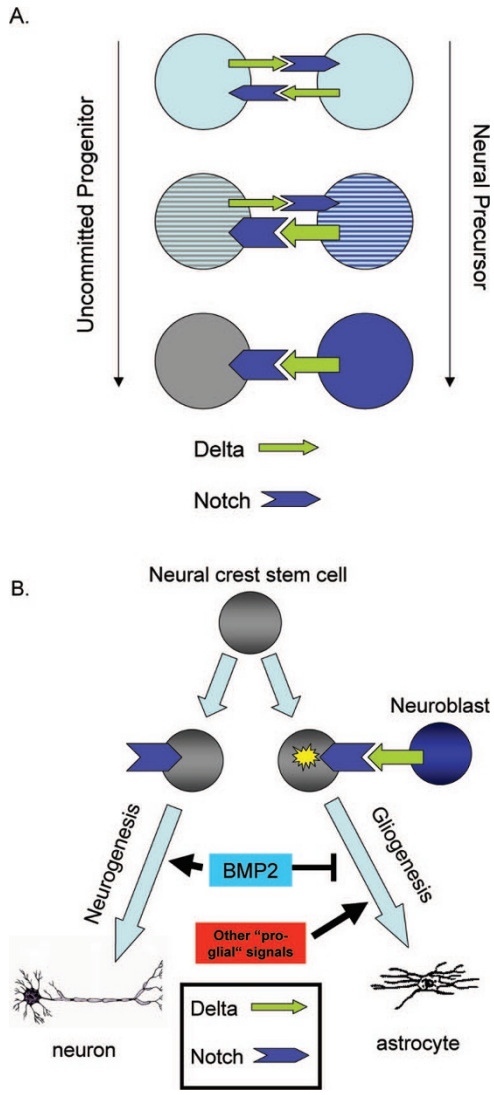

Figure 2. (*A*) Model of lateral inhibition: Two initially equipotent cells express equal levels of the Notch receptor and Delta ligand. If levels of a "proneural" fluctuate so as to increase in the cell on the right, Delta ligand expression increases and Notch decreases in that cell, and neural differentiation commences. Secondary to increased Delta ligand exposure, an opposite pattern in the left cell results such that Delta ligand expression is down-regulated and Notch signaling predominates resulting in maintenance of an uncommitted progenitor state. (*B*) Example of inductive signaling: A neural stem cell expressing the Notch receptor may interact with a variety of cells depending on its environmental context. In this figure, for example, the Notch expressing cell may come into contact with an already committed neurogenic precursor expressing the Delta ligand. This contact initiates a cascade of signaling events that, along with other cell extrinsic signals, results in glial fate commitment even in the setting of a strong pro-neurogenic signal such as BMP2 (14). In the absence of this cell contact, the neural progenitor is instructed toward neuronal development.

for Notch signaling (12). In Hes1/Hes5 double mutant mice, virtually all the neural stem cells differentiate prematurely into neurons resulting in severely disorganized neural tube morphology and absence of development of normal brain structures (25). However, initial stem cell generation seems independent of Notch signaling (26). Recently, two forms of the mammalian neural stem cell have been proposed: the "primitive" neural stem cell, isolated from E5.5 to E7.5 mouse

embryos that possess self-renewal capacity and neural multipotentiality, but also contain some non-neural properties (27); and the "definitive" neural stem cell, which, generally isolated initially at E8.5, are FGF2 responsive (28). Notch1 appears to be required for the transition from the primitive neural stem cell to the definitive neural stem cell and subsequent maintenance of the definitive neural stem cell state (27).

Gliogenesis. Although the Notch pathway appears to be critical for neural stem cell regulation and maintenance, it also appears to have a role in later neuroglial development (29–31). Cells of glial lineage comprise more than 90% of the human brain. While astrocytes and oligodendrocytes make up the bulk of these, other cell types also comprise this "non-neural" component of the brain. Microglia clear debris and mediate inflammation and may be of hematopoietic origin and the radial glia of the embryonic brain have been shown to be neural progenitors (32). In 1994, the observation was made using the P19 mouse embryonic carcinoma cell line, that Notch could inhibit retinoic acid induced P19 cell differentiation into neurons and myocytes. However, differentiation into glial (astrocyte) lineages was maintained (29). At that time, it was assumed that Notch simply "permitted" differentiation under the control of other undefined pathways. However, studies since then have revealed a more complex role for Notch and gliogenesis. Gain-of-function studies have revealed that Notch not only inhibits the differentiation of some cell types but can also promote a glial fate, such as the Müller glia cells in the retina, the radial glial cells in the neocortex, and astrocytes from the hippocampus (12–14,21,31,33). Interestingly, all of these cell types in certain contexts have the potential to act as neural stem cells, thus reiterating a potential role for Notch in neural progenitor maintenance even in these "glial" specified cell types (33–35). A recent study examining Dll1 (mammalian delta ligand homologue) knock-out neurospheres used time-dependent modulation of Notch pathway activation to construct a model of neural stem cell differentiation (36). The authors suggest the existence of two progenitors, arising from the primitive neural stem cell, P1 which is committed to a neural fate, and P2 which is committed to a glial fate. Notch is initially responsible for inhibiting P2 from developing into neurons. After other differentiation signals commence, Notch guides the now committed glial precursor into GFAP+ (glial fibrillary acidic protein) astrocyte rather than oligodendrocyte development. This is supported by another recent *in vivo* study showing that Notch signaling in the mouse embryonic brain is activated in certain GE (ganglionic eminence) cells, a known site of neural stem cells, and yet exclusive from those neuronal precursors expressing Mash1 a pro-neural bHLH (basic helix-loop-helix) gene (37). Notch expression can be observed during the maintenance and proliferation of the radial glia (glial precursors) and reappears again transiently during immature glial precursor differentiation into astrocytes.

Learning and memory. Now that the molecular and physiologic roles of notch in the nervous system have been discussed, the implications of these roles can be discussed in the setting of learning, memory, psychiatric disorders, and specific neurologic pathology. In addition to its embryonic role in determining cell fates, Notch has also been found to play a role in postnatal developmental processes. Interest in Notch with regards to plasticity and other higher brain functioning began with its link to the presenilins that were initially discovered during early research on the genetic basis of Alzheimer's disease, which will not be further discussed, given excellent reviews elsewhere (38–40). Notch has been shown to regulate cortical neurite (axon and dendrite) growth as well as dendrite branching in murine postmitotic neurons (41–43). Presenilin (also known as sel-12) mutants generated in *C. elegans* displayed defects in their ability to "learn" to find their optimal temperature in a gradient (44). It was postulated that this neurodeficit was mediated by defects in Notch signaling. Presente *et al*. (45) generated temperature sensitive Notch mutants in *Drosophila*, and after inactivation the flies suffered from a progressive neurodegenerative syndrome characterized by loss of flying ability and early death. More recently, the same group using the temperature-sensitive mutants as well as RNA-mediated inactivation of Notch demonstrated specific neurocognitive deficits in *Drosophila* related to long-term memory formation (46). Similar findings have been documented in mammalian systems as well. Mice with heterozygous null mutations in the Notch1 gene (Notch1+/−) were tested using a water maze method, which revealed long-term spatial memory deficits. Identical in nature, but more severe deficits were seen in mice lacking one copy of the CSL gene (47), where CSL is the downstream effector of all four mammalian Notch genes. Wang *et al*. (48) used knock-down Notch mutant mice (*via* an anti-sense transgene) to specifically examine the effects on hippocampal neuron plasticity. Detailed neurophysiologic testing of hippocampal neurons revealed that long-term potentiation (a phenomenon associated with long-term memory formation) was inhibited in the mutants but rescued by exogenous addition of the Notch ligand, Jag-1 (48).

The study of learning and memory in humans is difficult due to the complexity of the human brain, and elucidation of the molecular processes responsible has just begun. By studying known developmental syndromes and linking their phenotype with genotype we can begin to dissect the complex pathways that lead from the embryonic neural tube to the developed brain and then to the unique attributes that define humans, the ability to learn and retrieve memories. A recent study on Down's syndrome (DS) identified a possible linkage with a presenilin1 polymorphism (49). Individuals with DS are likely to develop neuropathologic changes characteristic of Alzheimer's disease after only 40 y of age, including build-up of amyloid protein secondary to abnormal processing of the β-amyloid precursor protein (APP). Again, PS1 is a component of the γ-secretase complex that plays a role in both APP and Notch processing (39). Additionally, from birth, individuals with DS display defects in domains of learning and memory often attributed to hippocampal and prefrontal systems (50). Examining the possible role for defective Notch pathway signaling in these patients and in the DS mouse model (51) will provide for not only a deeper understanding of DS and Alzheimer's disease neuropathology but potential understanding of other developmental disorders as well.

Neurologic disease. As described above, Notch plays key roles in both embryonic neural development and later brain

plasticity in animal models. Human diseases are often multifactorial and impossible to attribute to a single gene defect. The Notch pathway is ubiquitously present throughout mammalian development and is integral to the proper formation and maintenance of multiple tissues and organs including but not limited to the hematopoietic system, the vasculoendothelial system, as well as the nervous system (3). It is becoming apparent that dysregulation of the Notch pathway can be found in a multitude of human diseases including T-cell leukemia, spondylocostal dysostosis, schizophrenia, CADASIL syndrome (to be described shortly), and Alagille syndrome among others (52–56).

CADASIL

Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) is characterized by the adult-onset of recurrent strokes, progressive vascular dementia, migraines, psychiatric disturbances, and pseudobulbar palsy. Mutations in Notch3 have been identified as the causative mutation in this syndrome (57). Although the disease is characterized by predominately neurologic pathology, the mechanism behind the disease appears to be vascular in nature. The cerebral vessels are narrowed by expansion of the extracellular matrix and by degeneration of the vascular smooth muscle cells and subsequent deposition of granular osmiophilic material (GOM). MRI reveals a microangiopathic pattern of signal abnormalities and white matter changes suggestive of infarcts and leukoencephalopathy. Interestingly, there is also an accumulation of the ectodomain of the Notch3 receptor (58,59). In fact, this accumulation in the small vessels of the skin has been used for diagnostic purposes with a MAb to the ectodomain of Notch3 (60). However, the vascular pathology begins before the deposition of either the ectodomain or GOM. There have been suggestions of an autoimmune component to the vasculopathy, however, there are no studies documenting this as yet. The exact molecular basis behind this pathology has yet to be elucidated. Primary loss or gain of function of Notch3 signaling has not consistently been demonstrated in multiple studies using CADASIL-associated Notch3 mutants, suggesting these mechanisms are unlikely candidates for the resultant pathology (61–64). Another possibility is a direct toxic effect from the accumulation of the Notch3 ectodomain, as mentioned above, however the mechanism behind this aggregation has yet to be determined.

SCHIZOPHRENIA

Schizophrenia is a complex mental illness with multiple phenotypic presentations. The underlying heritability of this illness likely arises from a multitude of genetic and/or epigenetic factors, and the final phenotype depends on environmental modulation as well. Early linkage studies identified a susceptibility locus on chromosome 6p (65), and subsequently a more detailed analysis looked specifically at the Notch4 gene. Linkage disequilibrium (LD) mapping of 80 British parent-offspring trios revealed LD with an A to G substitution in the promoter region and mutations in a CTG repeat site in exon 1 of the Notch4 gene as candidate susceptibility sites (56). Multiple similar analysis have been performed on various populations since then, but not all find similar linkage patterns (66–69). However, a study of 210 affected individuals comparing neuropsychological testing and brain volumes showed no significant link between the presence or absence of schizophrenia, but did show an association between smaller frontal lobe volume and worse performance on the Wisconsin Card Sort Test (WCST), a measure of frontal lobe function and integrity in those patients with schizophrenia (70). The existence of subtle neurodevelopmental anomalies and anatomic findings in these patients would be consistent with either an early developmental event in which migration or neurogenesis was affected, or a later loss of brain matter, possibly due to lack of neural stem cells. Obviously, these are merely speculations, and further epidemiologic analysis and animal model development is needed to define the role of Notch signaling in schizophrenia.

CORTICAL DYSPLASIA

Focal cortical dysplasia (FCD) was originally described as focal developmental anomalies of cortical structure characterized histologically by cortical dyslamination, the presence of abnormal giant neurons throughout the resected cortex and adjacent white matter, and accompanied in many cases by balloon-shaped cells of uncertain lineage (71). However, FCD includes a spectrum of disordered white and gray matter entities that range from mild cortical disruption to complete derangement of cortical lamination (72). Clinically, patients present usually in childhood with refractory partial epilepsy, and subsequent magnetic resonance imaging (MRI) reveals the area of disordered cortex. Treatment is centered on relieving the seizures and often involves surgical removal of the affected cortex. The defect is thought to arise from a migrational or apoptotic defect occurring early in development. In humans, the earliest migrations occur around wk 6 of gestation. At this time, radial glial cell fibers begin directing the migration of neural precursors from the ventricular to pial surface (73). PS1-deficient mice develop global cortical dysplasia characterized by overmigration of cortical plate neurons. This is associated with alterations in the distribution of Notch1 in the Cajal-Retzius neurons, cortical plate neurons responsible for regulating radial neuronal migration (74). Furthermore, specific analysis of tissue specimens from human FCD revealed greatly altered levels of both Notch-1 and Dvl-1 (disheveled), an integral effector of the Wnt pathway, in the abnormal neurons and balloon cells (75). In light of the known role that Notch plays in embryonic neural fate specification, it is not surprising that the Notch pathway plays a role in disorders of cortical formation. Further studies using animal models of cortical dysplasia are needed to define the early embryonic role of Notch in the genesis of these disorders.

BRAIN TUMORS

In addition to playing a central role in neurodevelopmental processes as described, Notch dysregulation has been implicated in a number of human malignancies, including T-cell leukemia, breast and colon adenocarcinomas, and cervical

cancer among others. The potential role for the Notch pathway in brain tumorigenesis has been given recent attention. The existence of neural stem cell-like tumor stem cells was postulated many years ago (76). Several studies have shown the existence of neural stem-like cells from human glial tumors (77,78). A recent analysis of pediatric brain tumors revealed the existence of progenitor cells with multipotentiality and certain expression characteristics similar to those of neural stem cells (79). Ignatova *et al*. (78) showed abnormal Notch ligand (Delta-1 and Jagged-1 and -2) expression from human tumors of glial origin (glioblastoma multiforme and anaplastic astrocytoma). In the presence of growth factors, tumor cell lines down-regulated Delta-1 expression whereas the control cell lines continued to express it suggesting a disordered signaling pathway tending toward persistent "stemness" as opposed to differentiation. Other studies have revealed elevated expression of Musashi1, an RNA-binding protein that acts to negatively regulate m-Numb translation, in medulloblastomas and high-grade astrocytomas (77,80,81). Numb is a well-known Notch1 antagonist, thus implicating up-regulation of the Notch pathway as part of the pathogenesis of these tumors. The prognosis of malignant brain tumors remains poor even with advances in surgical approaches, radiation therapy, and chemotherapy. Therapies targeted at the molecular mechanisms behind these tumors are desperately needed to improve the outcomes of these devastating tumors, underscoring the need for further examination of these crucial signaling pathways.

CONCLUSION

Although there has been significant progress in the past decade with regard to understanding the molecular mechanisms behind neurodevelopment, much still remains to be discovered. Hundreds of interacting signaling pathways likely lie behind even the most simple of neurodevelopmental events and only further analysis of animal models, basic biochemistry, and human diseases will bring us closer to elucidating these mechanisms. The Notch signaling pathway likely plays a key role in these events and further study is needed to define the developmental context in which the different Notch receptors and ligands act to direct development and affect disease.

REFERENCES

1. Clark GD 2002 Brain development and the genetics of brain development. Neurol Clin 20:917–939
2. Baker NE 2000 Notch signaling in the nervous system. Pieces still missing from the puzzle. Bioessays 22:264–273
3. Harper JA, Yuan JS, Tan JB, Visan I, Guidos CJ 2003 Notch signaling in development and disease. Clin Genet 64:461–472
4. Radtke F, Raj K 2003 The role of Notch in tumorigenesis: oncogene or tumour suppressor? Nat Rev Cancer 3:756–767
5. Greenwald I 1998 LIN-12/Notch signaling: lessons from worms and flies. Genes Dev 12:1751–1762
6. Artavanis-Tsakonas S, Rand MD, Lake RJ 1999 Notch signaling: cell fate control and signal integration in development. Science 284:770–776
7. Beatus P, Lendahl U 1998 Notch and neurogenesis. J Neurosci Res 54:125–136
8. Chitnis A, Henrique D, Lewis J, Ish-Horowicz D, Kintner C 1995 Primary neurogenesis in *Xenopus* embryos regulated by a homologue of the *Drosophila* neurogenic gene Delta. Nature 375:761–766
9. Deftos ML, Huang E, Ojala EW, Forbush KA, Bevan MJ 2000 Notch1 signaling promotes the maturation of CD4 and CD8 SP thymocytes. Immunity 13:73–84
10. Robey E, Chang D, Itano A, Cado D, Alexander H, Lans D, Weinmaster G, Salmon P 1996 An activated form of Notch influences the choice between CD4 and CD8 T cell lineages. Cell 87:483–492
11. Lefort K, Dotto GP 2004 Notch signaling in the integrated control of keratinocyte growth/differentiation and tumor suppression. Semin Cancer Biol 14:374–386
12. Gaiano N, Fishell G 2002 The role of notch in promoting glial and neural stem cell fates. Annu Rev Neurosci 25:471–490
13. Gaiano N, Nye JS, Fishell G 2000 Radial glial identity is promoted by Notch1 signaling in the murine forebrain. Neuron 26:395–404
14. Morrison SJ, Perez SE, Qiao Z, Verdi JM, Hicks C, Weinmaster G, Anderson DJ 2000 Transient Notch activation initiates an irreversible switch from neurogenesis to gliogenesis by neural crest stem cells. Cell 101:499–510
15. Greenwald IS, Sternberg PW, Horvitz HR 1983 The lin-12 locus specifies cell fates in *Caenorhabditis elegans*. Cell 34:435–444
16. Stollewerk A 2002 Recruitment of cell groups through Delta/Notch signalling during spider neurogenesis. Development 129:5339–5348
17. Swiatek PJ, Lindsell CE, del Amo FF, Weinmaster G, Gridley T 1994 Notch1 is essential for postimplantation development in mice. Genes Dev 8:707–719
18. Weinmaster G, Roberts VJ, Lemke G 1991 A homolog of *Drosophila* Notch expressed during mammalian development. Development 113:199–205
19. Ramain P, Khechumian K, Seugnet L, Arbogast N, Ackermann C, Heitzler P 2001 Novel Notch alleles reveal a Deltex-dependent pathway repressing neural fate. Curr Biol 11:1729–1738
20. Arias AM 2002 New alleles of Notch draw a blueprint for multifunctionality. Trends Genet 18:168–170
21. Wang S, Barres BA 2000 Up a notch: instructing gliogenesis. Neuron 27:197–200
22. de la Pompa JL, Wakeham A, Correia K, Samper E, Brown S, Aguilera R, Nakano T, Honjo T, Mak T, Rossant J, Conlon R 1997 Conservation of the Notch signalling pathway in mammalian neurogenesis. Development 124:1139–1148
23. Handler M, Yang X, Shen J 2000 Presenilin-1 regulates neuronal differentiation during neurogenesis. Development 127:2593–2606
24. Chambers CB, Peng Y, Nguyen H, Gaiano N, Fishell G, Nye J 2001 Spatiotemporal selectivity of response to Notch1 signals in mammalian forebrain precursors. Development 128:689–702
25. Hatakeyama J, Bessho Y, Katoh K, Ookawara S, Fujioka M, Guillemot F, Kageyama R 2004 Hes genes regulate size, shape and histogenesis of the nervous system by control of the timing of neural stem cell differentiation. Development 131:5539–5550
26. Hitoshi S, Alexson T, Tropepe V, Donoviel D, Elia AJ, Nye JS, Conlon RA, Mak TW, Bernstein A, van der Kooy D 2002 Notch pathway molecules are essential for the maintenance, but not the generation, of mammalian neural stem cells. Genes Dev 16:846–858
27. Hitoshi S, Seaberg RM, Koscik C, Alexson T, Kusunoki S, Kanazawa I, Tsuji S, van der Kooy D 2004 Primitive neural stem cells from the mammalian epiblast differentiate to definitive neural stem cells under the control of Notch signaling. Genes Dev 18:1806–1811
28. Tropepe V, Sibilia M, Ciruna BG, Rossant J, Wagner EF, van der Kooy D 1999 Distinct neural stem cells proliferate in response to EGF and FGF in the developing mouse telencephalon. Dev Biol 208:166–188
29. Nye JS, Kopan R, Axel R 1994 An activated Notch suppresses neurogenesis and myogenesis but not gliogenesis in mammalian cells. Development 120:2421–2430
30. Lutolf S, Radtke F, Aguet M, Suter U, Taylor V 2002 Notch1 is required for neuronal and glial differentiation in the cerebellum. Development 129:373–385
31. Tanigaki K, Nogaki F, Takahashi J, Tashiro K, Kurooka H, Honjo T 2001 Notch1 and Notch3 instructively restrict bFGF-responsive multipotent neural progenitor cells to an astroglial fate. Neuron 29:45–55
32. Rowitch DH 2004 Glial specification in the vertebrate neural tube. Nat Rev Neurosci 5:409–419
33. Furukawa T, Mukherjee S, Bao ZZ, Morrow EM, Cepko CL 2000 rax, Hes1, and notch1 promote the formation of Muller glia by postnatal retinal progenitor cells. Neuron 26:383–394
34. Doetsch F, Garcia-Verdugo JM, Alvarez-Buylla A 1999 Regeneration of a germinal layer in the adult mammalian brain. Proc Natl Acad Sci U S A 96:11619–11624
35. Malatesta P, Hack MA, Hartfuss E, Kettenmann H, Klinkert W, Kirchhoff F, Gotz M 2003 Neuronal or glial progeny: regional differences in radial glia fate. Neuron 37:751–764
36. Grandbarbe L, Bouissac J, Rand M, Hrabe de Angelis M, Artavanis-Tsakonas S, Mohier E 2003 Delta-Notch signaling controls the generation of neurons/glia from neural stem cells in a stepwise process. Development 130:1391–1402
37. Tokunaga A, Kohyama J, Yoshida T, Nakao K, Sawamoto K, Okano H 2004 Mapping spatio-temporal activation of Notch signaling during neurogenesis and gliogenesis in the developing mouse brain. J Neurochem 90:142–154
38. Morishima-Kawashima M, Ihara Y 2002 Alzheimer's disease: beta-Amyloid protein and tau. J Neurosci Res 70:392–401
39. Selkoe D, Kopan R 2003 Notch and Presenilin: regulated intramembrane proteolysis links development and degeneration. Annu Rev Neurosci 26:565–597
40. Cummings JL 2004 Alzheimer's disease. N Engl J Med 351:56–67
41. Sestan N, Artavanis-Tsakonas S, Rakic P 1999 Contact-dependent inhibition of cortical neurite growth mediated by notch signaling. Science 286:741–746
42. Redmond L, Oh SR, Hicks C, Weinmaster G, Ghosh A 2000 Nuclear Notch1 signaling and the regulation of dendritic development. Nat Neurosci 3:30–40
43. Whitford KL, Dijkhuizen P, Polleux F, Ghosh A 2002 Molecular control of cortical dendrite development. Annu Rev Neurosci 25:127–149
44. Wittenburg N, Eimer S, Lakowski B, Rohrig S, Rudolph C, Baumeister R 2000 Presenilin is required for proper morphology and function of neurons in *C. elegans*. Nature 406:306–309

45. Presente A, Andres A, Nye JS 2001 Requirement of Notch in adulthood for neurological function and longevity. Neuroreport 12:3321–3325
46. Presente A, Boyles RS, Serway CN, de Belle JS, Andres AJ 2004 Notch is required for long-term memory in *Drosophila*. Proc Natl Acad Sci U S A 101:1764–1768
47. Costa RM, Honjo T, Silva AJ 2003 Learning and memory deficits in Notch mutant mice. Curr Biol 13:1348–1354
48. Wang Y, Chan SL, Miele L, Yao PJ, Mackes J, Ingram DK, Mattson MP, Furukawa K 2004 Involvement of Notch signaling in hippocampal synaptic plasticity. Proc Natl Acad Sci U S A 101:9458–9462
49. Lucarelli P, Piciullo A, Palmarino M, Verdecchia M, Saccucci P, Arpino C, Curatolo P 2004 Association between presenilin-1-48C/T polymorphism and Down's syndrome. Neurosci Lett 367:88–91
50. Nadel L 2003 Down's syndrome: a genetic disorder in biobehavioral perspective. Genes Brain Behav 2:156–166
51. Reeves RH, Irving NG, Moran TH, Wohn A, Kitt C, Sisodia SS, Schmidt C, Bronson RT, Davisson MT 1995 A mouse model for Down syndrome exhibits learning and behaviour deficits. Nat Genet 11:177–184
52. Weng AP, Ferrando AA, Lee W, Morris JP 4th, Silverman LB, Sanchez-Irizarry C, Blacklow SC, Look AT, Aster JC 2004 Activating mutations of NOTCH1 in human T hell acute lymphoblastic leukemia. Science 306:269–271
53. Ellisen LW, Bird J, West DC, Soreng AL, Reynolds TC, Smith SD, Sklar J 1991 TAN-1, the human homolog of the *Drosophila* notch gene, is broken by chromosomal translocations in T lymphoblastic neoplasms. Cell 66:649–661
54. Joutel A, Corpechot C, Ducros A, Vahedi K, Chabriat H, Mouton P, Alamowitch S, Domenga V, Cecillion M, Marechal E, Maciazek J, Vayssiere C, Cruaud C, Cabanis EA, Ruchoux MM, Weissenbach J, Bach JF, Bousser MG, Tournier-Lasserve E 1996 Notch3 mutations in CADASIL, a hereditary adult-onset condition causing stroke and dementia. Nature 383:707–710
55. Li L, Krantz ID, Deng Y, Genin A, Banta AB, Collins CC, Qi M, Trask BJ, Kuo WL, Cochran J, Costa T, Pierpont ME, Rand EB, Piccoli DA, Hood L, Spinner NB 1997 Alagille syndrome is caused by mutations in human Jagged1, which encodes a ligand for Notch1. Nat Genet 16:243–251
56. Wei J, Hemmings GP 2000 The NOTCH4 locus is associated with susceptibility to schizophrenia. Nat Genet 25:376–377
57. Abe K, Murakami T, Matsubara E, Manabe Y, Nagano I, Shoji M 2002 Clinical Features of CADASIL. Ann N Y Acad Sci 977:266–272
58. Joutel A, Andreux F, Gaulis S, Domenga V, Cecillon M, Battail N, Piga N, Chapon F, Godfrain C, Tournier-Lasserve E 2000 The ectodomain of the Notch3 receptor accumulates within the cerebrovasculature of CADASIL patients. J Clin Invest 105:597–605
59. Ruchoux MM, Domenga V, Brulin P, Maciazek J, Limol S, Tournier-Lasserve E, Joutel A 2003 Transgenic mice expressing mutant Notch3 develop vascular alterations characteristic of cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy. Am J Pathol 162:329–342
60. Kalaria RN, Viitanen M, Kalimo H, Dichgans M, Tabira T 2004 The pathogenesis of CADASIL: an update. J Neurol Sci 226:35–39
61. Karlstrom H, Beatus P, Dannaeus K, Chapman G, Lendahl U, Lundkvist J 2002 A CADASIL-mutated Notch 3 receptor exhibits impaired intracellular trafficking and maturation but normal ligand-induced signaling. Proc Natl Acad Sci U S A 99:17119–17124
62. Haritunians T, Boulter J, Hicks C, Buhrman J, DiSibio G, Shawber C, Weinmaster G, Nofziger D, Schanen C 2002 CADASIL Notch3 mutant proteins localize to the cell surface and bind ligand. Circ Res 90:506–508
63. Joutel A, Monet M, Domenga V, Riant F, Tournier-Lasserve E 2004 Pathogenic mutations associated with cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy differently affect Jagged1 binding and Notch3 activity via the RBP/JK signaling pathway. Am J Hum Genet 74:338–347
64. Peters N, Opherk C, Zacherle S, Capell A, Gempel P, Dichgans M 2004 CADASIL-associated Notch3 mutations have differential effects both on ligand binding and ligand-induced Notch3 receptor signaling through RBP-Jk. Exp Cell Res 299:454–464
65. Wang S, Sun CE, Walczak CA, Ziegle JS, Kipps BR, Goldin LR, Diehl SR 1995 Evidence for a susceptibility locus for schizophrenia on chromosome 6pter-p22. Nat Genet 10:41–46
66. Takahashi S, Cui YH, Kojima T, Han YH, Yu SY, Tanabe E, Yara K, Matsuura M, Matsushima E, Nakayama J, Arinami T, Shen YC, Faraone SV, Tsuang MT 2003 Family-based association study of the NOTCH4 gene in schizophrenia using Japanese and Chinese samples. Biol Psychiatry 54:129–135
67. Skol AD, Young KA, Tsuang DW, Faraone SV, Haverstock SL, Bingham S, Prabhudesai S, Mena F, Menon AS, Yu CE, Rundell P, Pepple J, Sauter F, Baldwin C, Weiss D, Collins J, Keith T, Boehnke M, Schellenberg GD, Tsuang MT 2003 Modest evidence for linkage and possible confirmation of association between NOTCH4 and schizophrenia in a large Veterans Affairs Cooperative Study sample. Am J Med Genet 118:8–15
68. McGinnis RE, Fox H, Yates P, Cameron LA, Barnes MR, Gray IC, Spurr NK, Hurko O, St Clair D 2001 Failure to confirm NOTCH4 association with schizophrenia in a large population-based sample from Scotland. Nat Genet 28:128–129
69. Anttila S, Kampman O, Illi A, Roivas M, Mattila KM, Lassila V, Lehtimaki T, Leinonen E 2003 NOTCH4 gene promoter polymorphism is associated with the age of onset in schizophrenia. Psychiatr Genet 13:61–64
70. Wassink TH, Nopoulos P, Pietila J, Crowe RR, Andreasen NC 2003 NOTCH4 and the frontal lobe in schizophrenia. Am J Med Genet B Neuropsychiatr Genet 118:1–7
71. Taylor DC, Falconer MA, Bruton CJ, Corsellis JA 1971 Focal dysplasia of the cerebral cortex in epilepsy. J Neurol Neurosurg Psychiatry 34:369–387
72. Colombo N, Tassi L, Galli C, Citterio A, Lo Russo G, Scialfa G, Spreafico R 2003 Focal cortical dysplasias: MR imaging, histopathologic, and clinical correlations in surgically treated patients with epilepsy. AJNR Am J Neuroradiol 24:724–733
73. Cotter DR, Honavar M, Everall I 1999 Focal cortical dysplasia: a neuropathological and developmental perspective. Epilepsy Res 36:155–164
74. Hartmann D, De Strooper B, Saftig P 1999 Presenilin-1 deficiency leads to loss of Cajal-Retzius neurons and cortical dysplasia similar to human type 2 lissencephaly. Curr Biol 9:719–727
75. Cotter D, Honavar M, Lovestone S, Raymond L, Kerwin R, Anderton B, Everall I 1999 Disturbance of Notch-1 and Wnt signalling proteins in neuroglial balloon cells and abnormal large neurons in focal cortical dysplasia in human cortex. Acta Neuropathol (Berl) 98:465–472
76. Rubinstein LJ 1985 Embryonal central neuroepithelial tumors and their differentiating potential. A cytogenetic view of a complex neuro-oncological problem. J Neurosurg 62:795–805
77. Toda M, Iizuka Y, Yu W, Imai T, Ikeda E, Yoshida K, Kawase T, Kawakami Y, Okano H, Uyemura K 2001 Expression of the neural RNA-binding protein Musashi1 in human gliomas. Glia 34:1–7
78. Ignatova TN, Kukekov VG, Laywell ED, Suslov ON, Vrionis FD, Steindler DA 2002 Human cortical glial tumors contain neural stem-like cells expressing astroglial and neuronal markers *in vitro*. Glia 39:193–206
79. Hemmati HD, Nakano I, Lazareff JA, Masterman-Smith M, Geschwind DH, Bronner-Fraser M, Kornblum HI 2003 Cancerous stem cells can arise from pediatric brain tumors. Proc Natl Acad Sci U S A 100:15178–15183
80. Okano H, Imai T, Okabe M 2002 Musashi: a translational regulator of cell fate. J Cell Sci 115:1355–1359
81. Yokota N, Mainprize TG, Taylor MD, Kohata T, Loreto M, Ueda S, Dura W, Grajkowska W, Kuo JS, Rutka JT 2004 Identification of differentially expressed and developmentally regulated genes in medulloblastoma using suppression subtraction hybridization. Oncogene 23:3444–3453