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MINI-REVIEW

Opportunities lost and gained: Changes in progenitor competence during nervous system development

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ABSTRACT

During development of the central nervous system, a small pool of stem cells and progenitors generate the vast neural diversity required for neural circuit formation and behavior. Neural stem and progenitor cells often generate different progeny in response to the same signaling cue (e.g. Notch or Hedgehog), including no response at all. How does stem cell competence to respond to signaling cues change over time? Recently, epigenetics particularly chromatin remodeling – has emerged as a powerful mechanism to control stem cell competence. Here we review recent Drosophila and vertebrate literature describing the effect of epigenetic changes on neural stem cell competence.

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Introduction

Understanding the genetic and molecular mechanisms that allow stem cells to generate distinct cell types over time is critical to our broader understanding of animal development and how to reprogram adult stem cells to regenerate tissues damaged from injury or disease. It is well known that extrinsic niche-derived cues can alter stem cells' self-renewal and differentiation,[1,2,3,4] but stem cells often have heterogeneous responses to a single cue (Fig. 1), and how stem cells change their competence to respond to a specific cue has only recently been characterized. Here we use the term "competence" to describe the ability of a stem cell to respond to an extrinsic or intrinsic cue – for example, a progenitor at one stage of development may be competent to proliferate in response to active Notch signaling, but the same progenitor at a later state of development may be non-competent to respond to the same Notch signal. There are many ways a stem cell might change its competence to respond to a cue, but recently the role of epigenetic remodeling of the stem cell genome has emerged as an important process in controlling stem cell competence.

Epigenetic changes to the genomes of neural stem cell lineages represent a powerful mechanism for regulating competence during development. When loci that are targets of activation by molecular cues are epigenetically remodeled, the competence of neural progenitors to respond to the cue changes. For example, histone-modifying protein complexes, such as BAF, can be recruited to suites of loci by DNA-binding proteins, and induce changes in chromatin conformation that permit gene expression.[5] Additionally, the Polycomb repressive complex (PRC) can have the opposite effect, inducing the formation of heterochromatin and gene silencing.[6] Thus, the relative activity BAF and PRC at target genes can determine the competence of neural progenitors to respond to a molecular cue.

Here we focus on evidence that neural stem cells change competence over time to generate different responses to a single cue; and highlight examples where changes in stem cell competence are due to

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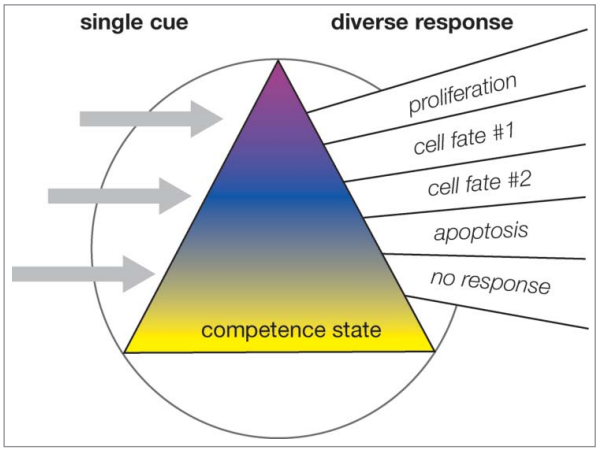

Figure 1. Illustration showing how a single developmental cue (gray arrow) can produce multiple outcomes depending on variations in stem cell competence (multicolored triangle). Note that in some cases a potent signal (gray arrow) can generate no response if the cell has lost competence to respond (bottom right output).

epigenetic modifications. We highlight how findings in multiple model organisms demonstrate that changes in stem cell competence are relevant for generating neuronal diversity during embryogenesis, as well as preventing tumorigenesis in adult stem cells.

To maintain the focus of this review, we do not cover work in non-neural stem cells, which has been reviewed elsewhere.[7] The gain and loss of stem cell competence via known or likely epigenetic modification is a conserved developmental strategy to generate neuronal diversity from a relatively small pool of neural stem cells.

Drosophila

The Drosophila CNS is generated by neural stem cells called neuroblasts, which undergo a series of asymmetric divisions to generate progeny with a more restricted fate. The most common "type I" neuroblast lineage produces ganglion mother cells (GMCs) which undergo a terminal division to generate a pair of neurons or glia; these neuroblasts can be found in the embryo, the larval optic lobe, and the larval central brain (Fig. 2a, b).[8,9,10] The rarer Type II neuroblasts are located in the dorsomedial region of the central brain (Fig. 2a).[10,11,12] Type II neuroblasts produce a series of intermediate neural progenitors (INPs) that each divide asymmetrically to generate 4–6 GMCs which make 8–12 neurons, and thus they give rise to large clones of neurons that contribute to the adult central complex (Fig. 2c).[13,14,15] In this section, we will discuss how (a) embryonic type I neuroblasts lose competence to respond to early temporal transcription factors due to changes in subnuclear gene position, (b) larval type I neuroblasts lose competence to respond to oncogenic mutations, (c) larval INPs lose competence to respond to Notch signaling, (d) larval type II neuroblasts use Trithorax to maintain competence to generate INPs, and (e) sensory neuron progenitors change competence to respond to Notch signaling.

Aging embryonic neuroblasts lose competence to respond to early temporal transcription factors via subnuclear genome reorganization and PRC1/2 complex function

Embryonic neuroblasts of the ventral nerve cord (VNC) can be uniquely identified by their position, molecular markers, and the stereotyped clone of neural progeny they produce.[16] Neuronal identity is determined by the spatial identity of the parental neuroblast in combination with "temporal transcription factors" which are sequentially expressed by most neuroblasts as they progress through their lineage. The temporal transcription factor cascade is Hunchback (Hb; Ikaros in mammals), Kruppel (Kr), Nubbin/Pou domain 2 (Pdm), and Castor (Casz1 in mammals). Loss of Hb or Kr leads to failure to specify the neurons born during these expression windows, whereas forced misexpression of Hb or Kr results in ectopic first-born or second-born neuron subtypes,[17,18,19] in part by Hb positively regulating its own expression.[20] However, pulses of Hb or Kr later in the embryonic neuroblast lineages fail to induce early neuronal fates: the neuroblast has lost competence to respond to these transcription factors.[21,22] Recent work has shown that loss of competence to respond to Hb is due to movement of the *hb* locus to the nuclear lamina in aging neuroblasts, thereby preventing ectopic Hb protein from inducing endogenous hb transcription.[20] In contrast, loss of competence to respond to Kr is due to activity of Polycomb repressive complex (PRC) activity,[23] presumably by making Kr target genes inaccessible. It will be interesting to see if both mechanisms are linked: do both Hb and Kr target genes move to the nuclear lamina? Is this movement a

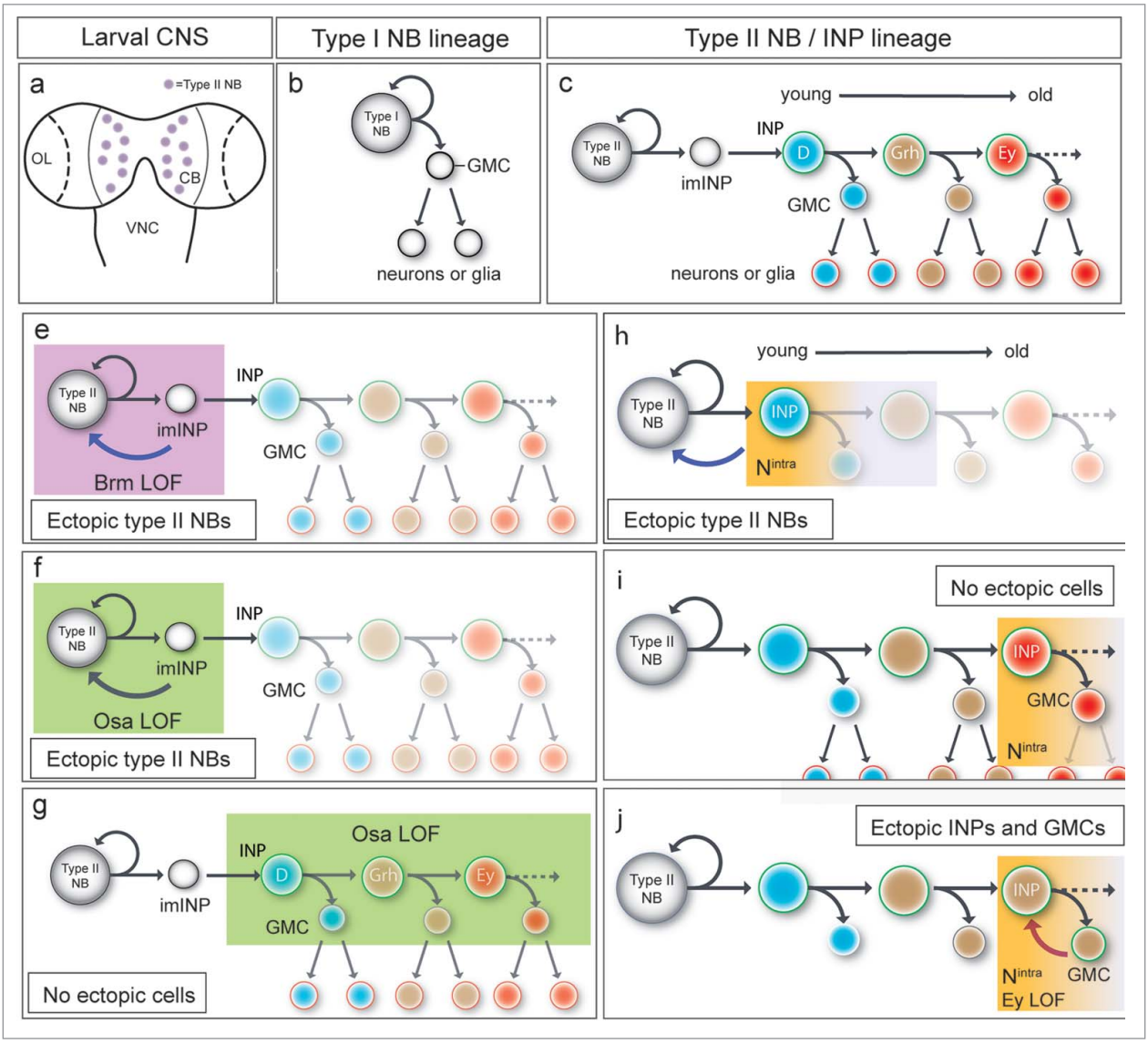

Figure 2. (a-c) Drosophila neural stem cells in the central brain (a) undergo a type I lineage (b) or a more elaborate type II lineage (c). (e-g) Epigenetic regulation of Drosophila neural stem cells alters their ability to respond to Notch signaling, which is normally present in stem cell progeny but suppressed by Brm (e) or Osa (f,g), Eyeless (i,j) or other Notch pathway repressors (h).

cause or consequence of PRC recruitment to these loci?

Larval type I neuroblasts lose competence to respond to oncogenic mutations

Drosophila embryonic and early larval type I neuroblasts coordinately express 2 RNA-binding proteins (Imp/IGF2BP and Lin-28) and a transcription factor (Chinmo); they are all downregulated in neuroblasts during the second half of larval life.[24,25,26,27,28] Recent work has shown that this suite of factors gives neuroblasts competence to form malignant tumors in response to several oncogenic mutations, including mutants in transcription factors (Prospero, Nerfin-1), and an RNA-binding protein (Brain tumor; Brat). Importantly, older neuroblasts during the second half of larval life are still proliferating but have little or no response to the same oncogenic mutations.[24] The normal function of Chinmo, Lin-28 and Imp is to specify early-born larval neurons and glia,[25,26] but they also open a competence window for "single hit" tumor formation; it is unknown if these two functions are related. This suite of proteins is unlikely to act on a single locus or a highly specific process because they provide tumor-forming competence to a diverse array of oncogenic mutations, including mutants in two different transcription factors (Prospero and Nerfin-1)

and an RNA-binding protein (Brain tumor). All 3 proteins are conserved in mammals,[29,30,31] raising the question of whether they may have a similar function, and making it important to determine their mechanism of action in both *Drosophila* and mammals.

Larval type II neuroblasts require Trithorax to maintain competence to produce INPs

Larval type II neuroblasts are defined by expression of the transcription factor PointedP1 and lack of expression of the transcription factor Asense; only 8 neuroblasts in each brain lobe are type II neuroblasts.[13,14,15,32] Type II neuroblasts divide asymmetrically to generate a series of INPs that produce an average of 10 neurons each, whereas larval type I neuroblasts make GMCs that only produce a pair of neurons.[13,14,15] How do type II neuroblasts generate INPs rather than GMCs? Recent work demonstrated that type II neuroblasts require the Buttonhead (Btd) transcription factor to maintain INP production, and that Trithorax (member of the SET1/MLL histone methyltransferase complex) is required to maintain the *btd* locus in a permissive chromatin state, allowing its expression in type II neuroblasts.[33] Loss of Trithorax led to lack of Btd, and loss of either Trithorax or Btd led to type II neuroblasts switching to GMC production[33] (Fig. 2e). They next showed that the loss of type II neuroblast identity was specifically caused by a loss of Trx histone methylation activity. Similarly, RNAi knockdown of several members of the SET1/MLL histone methyltransferase complex that co-purified with Trx also led to loss of INP production from type II neuroblasts.[33] Thus, Trx histone methylase activity is required to maintain INPs identity by opposing differentiation cues.

What prevents INPs from taking the opposite path, and dedifferentiating into neuroblasts or tumors? The Wang laboratory showed that the Brahma/histone deacetylase 3/Earmuff (Brm/HDAC3/Erm) complex is required to maintain INP identity and prevent dedifferentiation into type II neuroblasts[34] (Fig. 2e). This requires the activation of Erm in the new-born INP to prevent dedifferentiation. Interestingly, recent work from the Lee laboratory provides insight into this process. They showed that the *erm* enhancer is maintained in a poised state within type II neuroblasts by the Hdac1/Rpd3 histone deacetylase complex.[35] This prevents the activation of differentiation programs in parental neuroblasts, but enables the rapid activation of *erm* in their immature INP progeny, and their subsequently limited proliferative potential. In addition, Rpd3 deacetylation activity was required for type II neuroblast responsiveness to the self-renewal factors Deadpan, Klumpfuss and E(spl)m-gamma, indicating that these transcription factors utilize a specific epigenetic landscape in type II NBs to promote self-renewal. Collectively, these results show that type II neuroblast competence to produce INPs, and ability of INPs to initiate a program of differentiation, is regulated by the cell type-specific actions of multiple chromatin remodeling complexes.

Aging INPs lose competence to proliferate in response to Notch signaling

Type II neuroblasts divide asymmetrically to produce a series of INPs, which have a limited ability to proliferate, dividing only 4–6 times. We recently showed that aging INPs express a series of 3 temporal transcription factors: Dichaete (Sox family), Grainy head (CP2 family), and Eyeless (Pax family), which are important for generating neuronal and glial diversity within the short INP lineages.[36] An interesting question is what limits INP proliferation to 4–6 divisions, when their parental neuroblast can divide ~50 times. Recent work has shown that the chromatin remodeler Osa (SWI/SNF complex member) and Prdm family member Hamlet limit INP proliferation.[37] Osa is required for expression of Hamlet in INPs (but not in other cell types of the lineage), and reducing Osa or Hamlet levels in INPs led to extension of INP lineages (Fig. 2f, g). This is not due to derepression of Notch target genes (none were upregulated by transcriptional profiling of *osa* mutant INPs), but rather due to changes in INP temporal transcription factor expression: prolonged Grainy head and reduced Eyeless.[37] These data suggest a model in which INP chromatin remodeling is required for proper expression of the anti-proliferation factor Eyeless, which helps terminate INP proliferation.

How might Eyeless restrict INP proliferation? Many stem cells and progenitors require Notch signaling to maintain proliferation, so we asked whether Eyeless limits Notch signaling in aging INPs. It is well known that misexpression of the Notch intracellular domain (NICD), a potent inducer of Notch target gene expression,[38] in Type II NBs and young Eyeless-

negative INPs results in tumor formation[15,39,40,41,42] (Fig. 2h). In contrast, we found that NICD expression in old Eyeless+ INPs had no effect on the fate or proliferation of INPs, even when the exact same promoter was used to drive expression to ensure equal levels of Notch activity[39] (Fig. 2i). Furthermore, removal of the late temporal transcription factor Eyeless restored competence to generate ectopic cells by de-repressing Notch target genes in INP progeny (Fig. 2j). Thus, aging INPs lose competence to respond to Notch signaling, and Eyeless is required to block Notch-induced proliferation in old INP progeny.[39] How does Ey block Notch signaling? An attractive model is that Ey recruits SWI/SNF proteins to prevent activation of Notch target genes in GMCs.[43,44] Consistent with this model, murine Pax6 protein directly binds the SWI/SNF-related BAF complex to promote neuronal differentiation in murine adult neural progenitors.[45] In addition, a BAF subunit switch triggers the transition from proliferation to differentiation in mammalian neural progenitors,[46] raising the possibility that both Drosophila and mammals use similar pathways to regulate progenitor choice of differentiation or proliferation.

Aging sensory neuron progenitors change competence to respond to Notch signaling

Drosophila olfactory receptor neurons (ORNs) are specified from neuronal progenitors called sensory organ precursors (SOPs), which undergo three rounds of division to generate 8 cells, three of which are distinct ORNs. One ORN is specified by an absence of Notch signaling (Nab) while 2 distinct ORNs are specified by high level Notch signaling (Naa and Nba) – in the absence of Notch signaling these 2 neurons take an alternate fate.[47] How does one signal, Notch, generate the 2 different ORN fates: Naa and Nba? The authors found that only Naa expressed the Prdm member Hamlet, and that Hamlet was necessary and sufficient to induce Naa identity including axon projection to the appropriate olfactory glomeruli and odorant receptor expression.[47] How are Notch signaling and Hamlet expression integrated to generate distinct ORN fates? Genetic experiments indicate that Hamlet suppresses Notch activity, and biochemical data support this conclusion. Hamlet directly binds the CtBP co-repressor, and this binding is required for Hamlet suppression of Notch signaling. Furthermore, forced expression of Hamlet in a Drosophila cell line resulted in altered chromatin structure at Notch target loci, likely through enhancing H3K27 tri-methylation (associated with a repressive chromatin state) and diminishing H3K4 tri-methylation (associated with an active chromatin state).[47] For example, Hamlet expression decreased the Notch nuclear effector Su(H) occupancy at the Notch target gene *E(spl)m3*. Thus, Hamlet appears to bias Notch signaling by creating repressive chromatin structure around at least one Notch target gene, such that high Notch signaling without Hamlet gives the Nba fate, partial or differential Notch signaling with Hamlet gives the Naa fate, and no Notch signaling gives the Nab fate.[47] These findings illustrate how histone modifications can drive changes in competence by altering the local chromatin structure of target genes important for neuronal specification and function, and how neuronal diversity can be expanded in a stem cell lineage through changes in competence while reusing the same extrinsic cue.

Mouse

Competence to respond to extrinsic cues depends on Sox2-regulated chromatin state in neural progenitor lineages

The Sox family of transcription factors are important for maintaining stem cell/progenitor identity in many contexts.[48] Sox2 loss of function results in premature expression of neuronal differentiation genes, and Sox2 overexpression represses neuronal differentiation.[49] However, recent work reveals that Sox2 also has a role in promoting competence of young neurons to initiate neuronal differentiation in response to extrinsic Wnt signaling. Conditional Sox2 deletion in adult hippocampal neural progenitor cells (NPCs) has shown that Sox2 limits Polycomb Repressive Complex 2 (PRC2) activity to maintain a “poised” bivalently marked H3K4me3/H3K27me3 chromatin state at neuronal differentiation loci such as NeuroD1 and Bdnf[50] (Fig. 3). In this manner, Sox2 prevents the formation of a “closed” H3K27me3 chromatin state, which would otherwise block Wnt-induced expression of neuronal differentiation loci.[50,51] Sox2 conditional knockout mice had lower NeuroD1 levels and apoptosis of young neurons; the latter phenotype could be rescued by targeted re-expression of NeuroD1.[50] The authors propose a model where Sox2 limits PRC2 activity to maintain a poised chromatin state at

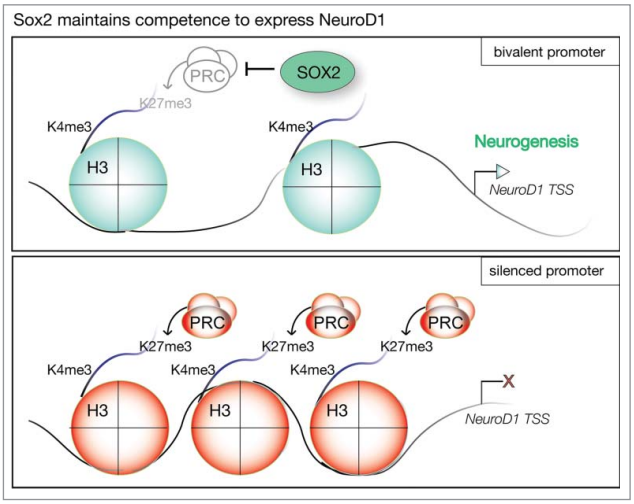

Figure 3. Sox2 prevents the Polycomb Repressive Complex (PRC) from silencing proneural genes such as NeuroD1 via histone H3 lysine 27 trimethylation (K27me3).

neuronal differentiation genes, thereby giving them competence to respond to Wnt-induced expression and subsequent neuronal differentiation (Fig. 3).

Interestingly, this is a different mode of Sox2 action than the authors described previously for maintaining progenitor identity of hippocampal NPCs. In that study, they found that Sox2 recruited the TRRAP/GCN5 histone acetyltransferase complex to maintain "open" H3K9ac chromatin at the *LIN28* locus, allowing this self-renewal promoting gene to be expressed in NPCs.[52] Furthermore, recent work has illuminated a non-histone, acetyltransferase activity for GCN5 that is crucial for retinoic acid responsiveness and proper control of Wnt, Gli3 and Shh signaling during early diencephalic development in embryonic mice.[53] Specifically, their model posits that in the presence of retinoic acid, GCN-5 mediated acetylation of TACC1 causes transcriptional de-repression of retinoic acid response elements.

Sox2 is also required to maintain the proliferative potential of retinal progenitor cells by modulating responsiveness to the Notch signaling pathway.[54] Conditional mutations and knockdown of Sox2 resulted in decreased expression of the Notch1 receptor, and chromatin immunoprecipitation experiments showed association of Sox2 and the *Notch1* locus. The authors propose a model in which Sox2 promotes Notch1 receptor expression in retinal progenitors, giving them competence to respond to Notch ligands and activating expression of Notch target genes such as Hes-5, which are important for maintaining the proliferative capacity of retinal progenitors.[54]

Epigenetic silencing of Notch target genes restricts INP competence to respond to Notch

Neural stem cells (NSCs) in the germinal zone of developing mammalian cortex have active Notch signaling via the canonical CBF1/SuH/Lag-1 (CSL) nuclear effector, and express target genes such as Hes-5 to maintain proliferation and block differentiation. In contrast, NSC progeny called intermediate neural progenitors (INPs) are exposed to Notch ligands but fail to express Notch target genes including a CSL reporter construct or Hes5, and thus initiate neuronal differentiation.[55,56] What limits INP competence to respond to Notch/CSL signaling? Recent work has shown that the Bcl6 oncogene is required to blunt Notch signaling in INPs. Bcl6 is detected at low levels in NSCs and high levels in INPs, where it reduces occupancy of the Mam-1 co-activator protein at the *Hes5* locus, increases occupancy of the Sirt-1 deacetylase, leading to silencing of the Hes5 gene.[56] The authors conclude that epigenetic silencing of the *Hes5* locus blocks productive Notch signaling in INPs leading to neuronal differentiation. It will be interesting to compare this mechanism to that of Eyeless blocking Notch signaling in Drosophila old INPs (see above); perhaps in both cases loss of competence to respond to Notch will be due to epigenetic silencing of specific Notch target genes.

The role of extrinsic cues and epigenetic modification in subdividing a single progenitor competence window

In the developing mammalian hindbrain, Nkx2.2+ progenitors produce motor neurons (MNs) during early neurogenesis, and then switch to making serotonergic neurons (5HTNs). The homeodomain transcription factor Phox2b is expressed in young progenitors during MN production, and *Phox2b* mutant progenitors fail to make MNs and instead prematurely give rise to 5HTNs, showing that young progenitors have an intrinsic competence to generate 5HTNs.[57] More recently, Dias *et al* identified TGFβ signaling as a temporally regulated cue that downregulates Phox2b expression; reduced TGFβ signaling delayed the MN-to-5HTN switch, altering the

number of neurons in each population.[58] This system illustrates how temporal regulation of cell fate determinates (e.g., Phox2b) can subdivide a single competence window to generate neuronal diversity, and how an extrinsic cue can determine the timing of the switch between neuronal cell types.

In contrast, a different mechanism times the neuronal-to-glial switch that occurs in many regions of the murine CNS. Although the switch requires an extrinsic cue, in this case the CNTF/LIF cytokine, there is also a requirement for epigenomic modification. Early cortical progenitors are exposed to CNTF, yet they still produce neurons.[59,60] Similarly, young cortical progenitors were less competent to produce glia than older progenitors when exposed to gliogenic cytokines in culture.[61] Even overexpression of CNTF in young cortical progenitors only generates a slight increase in glial production.[62] What prevents CNTF from inducing gliogenesis in young progenitors? It appears that at least one key glial differentiation gene, *Gfap*, is highly methylated and thus epigenetically silenced in young cortical progenitors; elimination of DNA methyltransferase 1 (Dnmt1) activity leads to robust precocious production of $GFAP^+$ astrocytes in response to CNTF.[63,64]

Zebrafish

Insight into how progenitors change competence to respond to extrinsic cues has come from studies in the developing zebrafish spinal cord. Zebrafish lateral floor plate progenitors (LFPs) require Hedgehog (Hh) signaling to maintain proliferation.[65] Progenitors stop dividing and initiate differentiation by diminishing their response to Hh signaling; this is achieved, at least in part, by a regulatory network that restricts apical cilia formation – a process implicated in perceiving Hh signaling,[66] and controlled by apically restricted proteins – a hallmark of asymmetrically dividing neural stem cells and progenitors.[67] Previous work revealed that expression of the apically restricted Par proteins, Pard3 and Prkci, are repressed by the expression of miR-219.[68] The authors put forth a model where spinal cord progenitors in the early embryo proliferate in response to Hh signaling, but the onset of miR-219 expression leads to Par protein repression and loss of apical cilia, thereby rendering progenitors non-competent to respond to Hh signaling and triggering differentiation. This model is supported by the observations that miR-219 knockdown caused an extension of Hh signaling, as measured by *patched2* expression and an increased number of Sox2+ progenitors in the spinal cord. Furthermore, these effects of miR-219 knockdown could be rescued by treating embryos with cyclopamine, an inhibitor of Hh signaling. Importantly, expression of Shh ligands in the developing spinal cord does not diminish from one to 3 day post fertilization (dpf), although expression of *patched2* is lost by 3 dpf, suggesting that progenitors are no longer competent to respond to Shh ligands. Thus, microRNAs regulate the competence of neural progenitors to respond to Hh signaling, leading to a transition from proliferation to neurogenesis.

Another example of altered neural progenitor competence comes from the analysis of Kolmer-Agduhr (KA") interneuron development. LFPs generate KA" neurons via combinatorial interactions between the Notch and Hh signaling pathways.[69] Notch signaling is required transiently to maintain LFP progenitors and to convey competence to respond to Hh, which is required in LFP progenitors for the subsequent specification of KA" interneurons.[69] Early activation of Hh caused the formation of ectopic KA" interneurons, while late activation the Hh effector Gli1 inhibited the differentiation of LFP progeny into KA" interneurons. Thus, Hh signaling could only promote the specification of KA" interneurons in LFP progenitors where Notch signaling was active.[69] How Notch signaling provides competence to respond to Hh remains unknown, but Notch signaling is known to alter chromatin state,[70,71] and it a likely mechanism for altering progenitor competence in this system.

Future directions

Work in many systems has now shown that epigenetic remodeling can alter neural stem cell competence, thereby resulting in a single neural stem cell generating diverse progeny in response to a common signaling pathway (e.g., Notch). This allows a limited number of highly conserved signaling pathways to generate myriad cell fate outcomes during neural development. Future work will be needed to identify factors that trigger chromatin alterations, the precise nature of the alterations at a genome-wide level, and the mechanism by which these changes lead to distinct neuronal and glial cell types. A better understanding

of how temporally regulated changes in stem cell epigenomes bias the response to signaling pathways is likely to help guide in vitro production of neural cell types for clinical neurotherapeutics.

Disclosure of potential conflicts of interest

No potential conflicts of interest were disclosed.

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References

[1] Huang G, Ye S, Zhou X, Liu D, Ying QL. Molecular basis of embryonic stem cell self-renewal: from signaling pathways to pluripotency network. Cell Mol Life Sci 2015; 72 (9):1741–57; PMID:25595304; https://doi.org/10.1007/s00018-015-1833-2

[2] Mooney CJ, Hakimjavadi R, Fitzpatrick E, Kennedy E, Walls D, Morrow D, Redmond EM, Cahill PA. Hedgehog and resident vascular stem cell fate. Stem Cells Int 2015; 2015:468428; PMID:26064136; https://doi.org/10.1155/2015/468428

[3] Pauklin S, Vallier L. Activin/Nodal signalling in stem cells. Development 2015; 142(4):607–19; PMID:25670788; https://doi.org/10.1242/dev.091769

[4] Serio RN. Wnt of the two horizons: putting stem cell self-renewal and cell fate determination into context. Stem Cells Dev 2014; 23(17):1975–90; PMID:24762106; https://doi.org/10.1089/scd.2014.0055

[5] Ronan JL, Wu W, Crabtree GR. From neural development to cognition: unexpected roles for chromatin. Nat Rev Genet 2013; 14(5):347–59; PMID:23568486; https://doi.org/10.1038/nrg3413

[6] Di Croce L, Helin K. Transcriptional regulation by Polycomb group proteins. Nat Struct Mol Biol 2013; 20 (10):1147–55; PMID:24096405; https://doi.org/10.1038/nsmb.2669

[7] Segales J, Perdiguero E, Munoz-Canoves P. Epigenetic control of adult skeletal muscle stem cell functions. Febs J 2015; 282(9):1571–88; PMID:25251895; https://doi.org/10.1111/febs.13065

[8] Kang KH, Reichert H. Control of neural stem cell self-renewal and differentiation in Drosophila. Cell Tissue Res 2015; 359(1):33–45; PMID:24902665; https://doi.org/10.1007/s00441-014-1914-9

[9] Viktorin G, Riebli N, Reichert H. A multipotent transit-amplifying neuroblast lineage in the central brain gives rise to optic lobe glial cells in Drosophila. Dev Biol 2013; 379(2):182–94; PMID:23628691; https://doi.org/10.1016/j.ydbio.2013.04.020

[10] Bayraktar OA, Boone JQ, Drummond ML, Doe CQ. Drosophila type II neuroblast lineages keep Prospero levels low to generate large clones that contribute to the adult brain central complex. Neural Dev 2010; 5:26; PMID:20920301; https://doi.org/10.1186/1749-8104-5-26

[11] Ito M, Masuda N, Shinomiya K, Endo K, Ito K. Systematic analysis of neural projections reveals clonal composition of the Drosophila brain. Curr Biol 2013; 23(8):644–55; PMID:23541729; https://doi.org/10.1016/j.cub.2013.03.015

[12] Yang JS, Awasaki T, Yu HH, He Y, Ding P, Kao JC, Lee T. Diverse neuronal lineages make stereotyped contributions to the Drosophila locomotor control center, the central complex. J Comp Neurol 2013; 521(12):2645–62; PMID:23696496; https://doi.org/10.1002/cne.23339

[13] Bello BC, Izergina N, Caussinus E, Reichert H. Amplification of neural stem cell proliferation by intermediate progenitor cells in Drosophila brain development. Neural Dev 2008; 3:5; PMID:18284664; https://doi.org/10.1186/1749-8104-3-5

[14] Boone JQ, Doe CQ. Identification of Drosophila type II neuroblast lineages containing transit amplifying ganglion mother cells. Dev Neurobiol 2008; 68(9):1185–95; PMID:18548484; https://doi.org/10.1002/dneu.20648

[15] Bowman SK, Rolland V, Betschinger J, Kinsey KA, Emery G, Knoblich JA. The tumor suppressors Brat and Numb regulate transit-amplifying neuroblast lineages in Drosophila. Dev Cell 2008; 14(4):535–46; PMID:18342578; https://doi.org/10.1016/j.devcel.2008.03.004

[16] Skeath JB, Thor S. Genetic control of Drosophila nerve cord development. Curr Opin Neurobiol 2003; 13(1):8–15; PMID:12593977; https://doi.org/10.1016/S0959-4388(03)00007-2

[17] Isshiki T, Pearson B, Holbrook S, Doe CQ. Drosophila neuroblasts sequentially express transcription factors which specify the temporal identity of their neuronal progeny. Cell 2001; 106(4):511–21; PMID:11525736; https://doi.org/10.1016/S0092-8674(01)00465-2

[18] Novotny T, Eiselt R, Urban J. Hunchback is required for the specification of the early sublineage of neuroblast 7-3 in the Drosophila central nervous system. Development 2002; 129(4):1027–36; PMID:11861485

[19] Kanai MI, Okabe M, Hiromi Y. seven-up Controls switching of transcription factors that specify temporal identities of Drosophila neuroblasts. Dev Cell 2005; 8 (2):203–13; PMID:15691762; https://doi.org/10.1016/j.devcel.2004.12.014

[20] Kohwi M, Lupton JR, Lai SL, Miller MR, Doe CQ. Developmentally regulated subnuclear genome reorganization restricts neural progenitor competence in Drosophila. Cell 2013; 152(1-2):97–108; PMID:23332748; https://doi.org/10.1016/j.cell.2012.11.049

[21] Cleary MD, Doe CQ. Regulation of neuroblast competence: multiple temporal identity factors specify distinct neuronal fates within a single early competence window.

Genes Dev 2006; 20(4):429–34; PMID:16481472; https://doi.org/10.1101/gad.1382206

[22] Pearson BJ, Doe CQ. Regulation of neuroblast competence in Drosophila. Nature 2003; 425(6958):624–8; PMID:14534589; https://doi.org/10.1038/nature01910

[23] Touma JJ, Weckerle FF, Cleary MD. Drosophila Polycomb complexes restrict neuroblast competence to generate motoneurons. Development 2012; 139(4):657–66; PMID:22219354; https://doi.org/10.1242/dev.071589

[24] Narbonne-Reveau K, Lanet E, Dillard C, Foppolo S, Chen CH, Parrinello H, Rialle S, Sokol NS, Maurange C. Neural stem cell-encoded temporal patterning delineates an early window of malignant susceptibility in Drosophila. Elife 2016; 5; https://doi.org/10.7554/eLife.13463

[25] Zhu S, Lin S, Kao CF, Awasaki T, Chiang AS, Lee T. Gradients of the Drosophila Chinmo BTB-zinc finger protein govern neuronal temporal identity. Cell 2006; 127 (2):409–22; PMID:17055440; https://doi.org/10.1016/j.cell.2006.08.045

[26] Liu Z, Yang CP, Sugino K, Fu CC, Liu LY, Yao X, Lee LP, Lee T. Opposing intrinsic temporal gradients guide neural stem cell production of varied neuronal fates. Science 2015; 350(6258):317–20; PMID:26472907; https://doi.org/10.1126/science.aad1886

[27] Ren, Q, Yang CP, Liu Z, Sugino K, Mok K, He Y, Ito M, Nern A, Otsuna H, Lee T. Stem cell intrinsic, Seven-up-triggered temporal factor gradients diversify intermediate neural progenitors. Curr Biol 2017; 27:1–11.

[28] Syed MH, Mark BJ, Doe CQ. Steroid hormone induction of temporal gene expression in Drosophila brain neuroblasts generates neuronal and glial diversity. eLife 2017; 6:e26287; https://doi.org/10.7554/eLife.26287

[29] Bell JL, Wächter K, Mühleck B, Pazaitis N, Köhn M, Lederer M, Hüttelmaier S. Insulin-like growth factor 2 mRNA-binding proteins (IGF2BPs): post-transcriptional drivers of cancer progression? Cell Mol Life Sci 2013; 70 (15):2657–75; PMID:23069990; https://doi.org/10.1007/s00018-012-1186-z

[30] Yang M, Yang SL, Herrlinger S, Liang C, Dzieciatkowska M, Hansen KC, Desai R, Nagy A, Niswander L, Moss EG, et al. Lin28 promotes the proliferative capacity of neural progenitor cells in brain development. Development 2015; 142(9):1616–27; PMID:25922525; https://doi.org/10.1242/dev.120543

[31] Dykes IM, van Bueren KL, Ashmore RJ, Floss T, Wurst W, Szumska D, Bhattacharya S, Scambler PJ. HIC2 is a novel dosage-dependent regulator of cardiac development located within the distal 22q11 deletion syndrome region. Circ Res 2014; 115(1):23–31; PMID:24748541; https://doi.org/10.1161/CIRCRESAHA.115.303300

[32] Zhu S, Barshow S, Wildonger J, Jan LY, Jan YN. Ets transcription factor Pointed promotes the generation of intermediate neural progenitors in Drosophila larval brains. Proc Natl Acad Sci U S A 2011; 108 (51):20615–20; PMID:22143802; https://doi.org/10.1073/pnas.1118595109

[33] Komori H, , Xiao Q, Janssens DH, Dou Y, Lee CY. Trithorax maintains the functional heterogeneity of neural stem cells through the transcription factor buttonhead. Elife 2014; 3; PMID:25285447; https://doi.org/10.7554/eLife.03502

[34] Koe CT, Li S, Rossi F, Wong JJ, Wang Y, Zhang Z, Chen K, Aw SS, Richardson HE, Robson P, et al. The Brm-HDAC3-Erm repressor complex suppresses dedifferentiation in Drosophila type II neuroblast lineages. Elife 2014; 3:e01906; PMID:24618901; https://doi.org/10.7554/eLife.01906

[35] Janssens DH, Hamm DC, Anhezini L, Xiao Q, Siller KH, Siegrist SE, Harrison MM, Lee CY. An Hdac1/Rpd3-poised circuit balances continual self-renewal and rapid restriction of developmental potential during asymmetric stem cell division. Dev Cell 2017; 40(4):367–80 e7; PMID:28245922; https://doi.org/10.1016/j.devcel.2017.01.014

[36] Bayraktar OA, Doe CQ. Combinatorial temporal patterning in progenitors expands neural diversity. Nature 2013; 498:445–55; https://doi.org/10.1038/nature12266

[37] Eroglu E, Burkard TR, Jiang Y, Saini N, Homem CC, Reichert H, Knoblich JA. SWI/SNF complex prevents lineage reversion and induces temporal patterning in neural stem cells. Cell 2014; 156(6):1259–73; PMID:24630726; https://doi.org/10.1016/j.cell.2014.01.053

[38] Borggrefe T, Lauth M, Zwijsen A, Huylebroeck D, Oswald F, Giaimo BD. The Notch intracellular domain integrates signals from Wnt, Hedgehog, TGFbeta/BMP and hypoxia pathways. Biochim Biophys Acta 2016; 1863 (2):303–13; PMID:26592459; https://doi.org/10.1016/j.bbamcr.2015.11.020

[39] Farnsworth DR, Bayraktar OA, Doe CQ. Aging neural progenitors lose competence to respond to mitogenic notch signaling. Curr Biol 2015; 25(23):3058–68; PMID:26585279; https://doi.org/10.1016/j.cub.2015.10.027

[40] Weng M, Golden KL, Lee CY. dFezf/Earmuff maintains the restricted developmental potential of intermediate neural progenitors in Drosophila. Dev Cell 2010; 18 (1):126–35; PMID:20152183; https://doi.org/10.1016/j.devcel.2009.12.007

[41] Xiao Q, Komori H, Lee CY. klumpfuss distinguishes stem cells from progenitor cells during asymmetric neuroblast division. Development 2012; 139(15):2670–80; PMID:22745313; https://doi.org/10.1242/dev.081687

[42] Wang H, Somers GW, Bashirullah A, Heberlein U, Yu F, Chia W. Aurora-A acts as a tumor suppressor and regulates self-renewal of Drosophila neuroblasts. Genes & Development 2006; 20(24):3453–63; https://doi.org/10.1101/gad.1487506

[43] Housden BE, Li J, Bray SJ, Visualizing Notch signaling in vivo in Drosophila tissues. Methods Mol Biol 2014; 1187:101–13; PMID:25053484

[44] San-Juan BP, Baonza A. The bHLH factor deadpan is a direct target of Notch signaling and regulates neuroblast self-renewal in Drosophila. Dev Biol 2011; 352 (1):70–82; PMID:21262215; https://doi.org/10.1016/j.ydbio.2011.01.019

[45] Ninkovic J, Steiner-Mezzadri A, Jawerka M, Akinci U, Masserdotti G, Petricca S, Fischer J, von Holst A, Beckers J, Lie CD, et al. The BAF complex interacts with Pax6 in adult neural progenitors to establish a neurogenic cross-regulatory transcriptional network. Cell Stem Cell 2013; 13(4):403–18; PMID:23933087; https://doi.org/10.1016/j.stem.2013.07.002

[46] Lessard J, Wu JI, Ranish JA, Wan M, Winslow MM, Staahl BT, Wu H, Aebersold R, Graef IA, Crabtree GR. An essential switch in subunit composition of a chromatin remodeling complex during neural development. Neuron 2007; 55(2):201–15; PMID:17640523; https://doi.org/10.1016/j.neuron.2007.06.019

[47] Endo K, Karim MR, Taniguchi H, Krejci A, Kinameri E, Siebert M, Ito K, Bray SJ, Moore AW. Chromatin modification of Notch targets in olfactory receptor neuron diversification. Nat Neurosci 2012; 15(2):224–33; https://doi.org/10.1038/nn.2998

[48] Miyagi S, Kato H, Okuda A. Role of SoxB1 transcription factors in development. Cell Mol Life Sci 2009; 66 (23):3675–84; PMID:19633813; https://doi.org/10.1007/s00018-009-0097-0

[49] Graham V, Khudyakov J, Ellis P, Pevny L. SOX2 functions to maintain neural progenitor identity. Neuron 2003; 39(5):749–65; PMID:12948443; https://doi.org/10.1016/S0896-6273(03)00497-5

[50] Amador-Arjona A, Cimadamore F, Huang CT, Wright R, Lewis S, Gage FH, Terskikh AV. SOX2 primes the epigenetic landscape in neural precursors enabling proper gene activation during hippocampal neurogenesis. Proc Natl Acad Sci U S A 2015; 112(15):E1936–45; PMID:25825708; https://doi.org/10.1073/pnas.1421480112

[51] Kuwabara T, Hsieh J, Muotri A, Yeo G, Warashina M, Lie DC, Moore L, Nakashima K, Asashima M, Gage FH. Wnt-mediated activation of NeuroD1 and retro-elements during adult neurogenesis. Nat Neurosci 2009; 12 (9):1097–105; PMID:19701198; https://doi.org/10.1038/nn.2360

[52] Cimadamore F, Amador-Arjona A, Chen C, Huang CT, Terskikh AV. SOX2-LIN28/let-7 pathway regulates proliferation and neurogenesis in neural precursors. Proc Natl Acad Sci U S A 2013; 110(32):E3017–26; PMID:23884650; https://doi.org/10.1073/pnas.1220176110

[53] Wilde JJ, Siegenthaler JA, Dent SY, Niswander LA. Diencephalic size is restricted by a novel interplay between GCN5 Acetyltransferase activity and retinoic acid signaling. J Neurosci 2017; 37(10):2565–79; PMID:28154153; https://doi.org/10.1523/JNEUROSCI.2121-16.2017

[54] Taranova OV, Magness ST, Fagan BM, Wu Y, Surzenko N, Hutton SR, Pevny LH. SOX2 is a dose-dependent regulator of retinal neural progenitor competence. Genes Dev 2006; 20(9):1187–202; PMID:16651659; https://doi.org/10.1101/gad.1407906

[55] Mizutani K, Yoon K, Dang L, Tokunaga A, Gaiano N. Differential Notch signalling distinguishes neural stem cells from intermediate progenitors. Nature 2007; 449 (7160):351–5; PMID:17721509; https://doi.org/10.1038/nature06090

[56] Tiberi L, van den Ameele J, Dimidschstein J, Piccirilli J, Gall D, Herpoel A, Bilheu A, Bonnefont J, Iacovino M, Kyba M, et al. BCL6 controls neurogenesis through Sirt1-dependent epigenetic repression of selective Notch targets. Nat Neurosci 2012; 15(12):1627–35; PMID:23160044; https://doi.org/10.1038/nn.3264

[57] Pattyn A, Vallstedt A, Dias JM, Samad OA, Krumlauf R, Rijli FM, Brunet JF, Ericson J. Coordinated temporal and spatial control of motor neuron and serotonergic neuron generation from a common pool of CNS progenitors. Genes Dev 2003; 17(6):729–37; PMID:12651891; https://doi.org/10.1101/gad.255803

[58] Dias JM, Alekseenko Z, Applequist JM, Ericson J. Tgfbeta signaling regulates temporal neurogenesis and potency of neural stem cells in the CNS. Neuron 2014; 84(5):927–39; PMID:25467979; https://doi.org/10.1016/j.neuron.2014.10.033

[59] Uemura A, Takizawa T, Ochiai W, Yanagisawa M, Nakashima K, Taga T. Cardiotrophin-like cytokine induces astrocyte differentiation of fetal neuroepithelial cells via activation of STAT3. Cytokine 2002; 18 (1):1–7; PMID:12090754; https://doi.org/10.1006/cyto.2002.1006

[60] Derouet D, Rousseau F, Alfonsi F, Froger J, Hermann J, Barbier F, Perret D, Diveu C, Guillet C, Preisser L, et al. Neuropoietin, a new IL-6-related cytokine signaling through the ciliary neurotrophic factor receptor. Proc Natl Acad Sci U S A 2004; 101(14):4827–32; PMID:15051883; https://doi.org/10.1073/pnas.0306178101

[61] He F, Ge W, Martinowich K, Becker-Catania S, Coskun V, Zhu W, Wu H, Castro D, Guillemot F, Fan G, et al. A positive autoregulatory loop of Jak-STAT signaling controls the onset of astrogliogenesis. Nat Neurosci 2005; 8 (5):616–25; PMID:15852015; https://doi.org/10.1038/nn1440

[62] Barnabe-Heider F, Wasylnka JA, Fernandes KJ, Porsche C, Sendtner M, Kaplan DR, Miller FD. Evidence that embryonic neurons regulate the onset of cortical gliogenesis via cardiotrophin-1. Neuron 2005; 48 (2):253–65; PMID:16242406; https://doi.org/10.1016/j.neuron.2005.08.037

[63] Fan G, Martinowich K, Chin MH, He F, Fouse SD, Hutnick L, Hattori D, Ge W, Shen Y, Wu H, et al. DNA methylation controls the timing of astrogliogenesis through regulation of JAK-STAT signaling. Development 2005; 132(15):3345–56; PMID:16014513; https://doi.org/10.1242/dev.01912

[64] Takizawa T, Nakashima K, Namihira M, Ochiai W, Uemura A, Yanagisawa M, Fujita N, Nakao M, Taga T. DNA methylation is a critical cell-intrinsic determinant of astrocyte differentiation in the fetal brain. Dev Cell 2001; 1(6):749–58; PMID:11740937; https://doi.org/10.1016/S1534-5807(01)00101-0

[65] Hudish LI, Galati DF, Ravanelli AM, Pearson CG, Huang P, Appel B. miR-219 regulates neural progenitors by dampening apical Par protein-dependent Hedgehog signaling. Development 2016; 143 (13):2292–304; PMID:27226318; https://doi.org/10.1242/dev.137844

[66] Huang P, Schier AF. Dampened Hedgehog signaling but normal Wnt signaling in zebrafish without cilia. Development 2009; 136(18):3089–98; PMID:19700616; https://doi.org/10.1242/dev.041343

[67] Lee CY, Robinson KJ, Doe CQ. Lgl, Pins and aPKC regulate neuroblast self-renewal versus differentiation. Nature 2006; 439(7076):594–8; PMID:16357871; https://doi.org/10.1038/nature04299

[68] Hudish LI, Blasky AJ, Appel B. miR-219 regulates neural precursor differentiation by direct inhibition of apical par polarity proteins. Dev Cell 2013; 27(4):387–98; PMID:24239515; https://doi.org/10.1016/j.devcel.2013.10.015

[69] Huang P, Xiong F, Megason SG, Schier AF. Attenuation of Notch and Hedgehog signaling is required for fate specification in the spinal cord. PLoS Genet 2012; 8(6): e1002762; PMID:22685423; https://doi.org/10.1371/journal.pgen.1002762

[70] Schwanbeck R. The role of epigenetic mechanisms in Notch signaling during development. J Cell Physiol 2015; 230(5):969–81; PMID:25336183; https://doi.org/10.1002/jcp.24851

[71] Wang H, Zang C, Liu XS, Aster JC. The role of Notch receptors in transcriptional regulation. J Cell Physiol 2015; 230(5):982–8; PMID:25418913; https://doi.org/10.1002/jcp.24872