PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B

Phil. Trans. R. Soc. B (2008) **363**, 123–137
doi:10.1098/rstb.2006.2016
Published online 23 February 2007

Brain micro-ecologies: neural stem cell niches in the adult mammalian brain

Patricio A. Riquelme[1], Elodie Drapeau[2] and Fiona Doetsch[1,2,3,*]

[1]*Center for Neurobiology and Behavior,* [2]*Department of Pathology, and* [3]*Department of Neurology, Columbia University, 630 West 168th Street, New York City, NY 10032, USA*

Neurogenesis persists in two germinal regions in the adult mammalian brain, the subventricular zone of the lateral ventricles and the subgranular zone in the hippocampal formation. Within these two neurogenic niches, specialized astrocytes are neural stem cells, capable of self-renewing and generating neurons and glia. Cues within the niche, from cell–cell interactions to diffusible factors, are spatially and temporally coordinated to regulate proliferation and neurogenesis, ultimately affecting stem cell fate choices. Here, we review the components of adult neural stem cell niches and how they act to regulate neurogenesis in these regions.

Keywords: stem cell; niche; astrocyte; subventricular zone; subgranular zone

1. STEM CELLS AND THEIR NICHES

Stem cells have two essential properties: self-renewal and multipotency. Self-renewal is the ability to generate an identical daughter cell and multipotency is the capacity to generate all cell types of a tissue. Although self-renewal often occurs via asymmetric divisions, to yield another stem cell and a more differentiated cell, stem cells may also undergo symmetric divisions to generate two stem cells or two differentiated daughter cells. Thus, stem cell self-renewal can occur at the single cell level or the population level, such that the stem cell population is maintained (Spradling *et al.* 2001). In many different organs, stem cells divide relatively infrequently to generate transit-amplifying cells, which in turn divide to rapidly expand their number before generating more mature progeny. This hierarchy of division and differentiation allows amplification of the number of mature cells that can be derived from a single stem cell, while minimizing the possibility of mutations due to DNA replication in the genome of long-lived stem cells (Reya *et al.* 2001). Interestingly, studies from several niches have revealed that transit-amplifying cells also retain the ability to act as stem cells when challenged by appropriate stimulation (Doetsch *et al.* 2002; Marshman *et al.* 2002; Raff 2003; Brawley & Matunis 2004; Kai & Spradling 2004).

Specialized microenvironments or niches support the lifelong self-renewal of stem cells and their production of differentiated cells (Spradling *et al.* 2001; Fuchs *et al.* 2004). Importantly, stem cells themselves extensively interact with and participate in the niche. Furthermore, niches may in fact be dynamic structures that alter their location and characteristics over time concomitant with tissue remodelling. Comparison of stem cell niches in *Drosophila* germ-line lineages, and in the mammalian haematopoietic system, intestinal epithelium, skin/hair follicle and nervous system has revealed common and unique features (figure 1; reviewed in Spradling *et al.* 2001; Doetsch 2003*b*; Fuchs *et al.* 2004; Li & Xie 2005). Cell–cell interactions (figure 1*a*) and diffusible signals (figure 1*b*) are key elements allowing feedback control of stem cell activation and differentiation from progeny and/or niche support cells. An emerging feature of several stem cell niches is the intimate association with endothelial cells, which regulate stem cell self-renewal and differentiation (figure 1*c*). Within a niche, stem cells are frequently anchored to a basal lamina or stromal cells (figure 1*c*,*d*) that can provide a substrate for oriented cell division. The basal lamina is also an important regulator of the accessibility of growth factors and other signals, as associated extracellular matrix (ECM) molecules and glycoproteins can both concentrate and sequester factors in inactive or active forms (figure 1*c*). Cell anchoring may orient cell division resulting in the segregation of key determinants into one or both daughter cells depending on the plane of division (figure 1*d*). Here, we review the role of the above components in the *in vivo* niches for adult neurogenesis.

2. ADULT NEURAL STEM CELL NICHES

Neural stem cells persist in the adult mammalian brain and exhibit the two fundamental properties of stem cells; they undergo self-renewal and are multipotent, generating neurons and macroglia (astrocytes and oligodendrocytes). While neural stem cells can be cultured *in vitro* with growth factors as either adherent cultures or non-adherent cultures called neurospheres (reviewed in Gage 2000; Temple 2001), their biology is in large part defined by their *in vivo* niche.

Neurogenesis occurs in two principal brain regions in adult mammals: the subventricular zone (SVZ), adjacent to the lateral ventricles (figure 2), and the subgranular zone (SGZ) of the hippocampal formation (figure 3) (reviewed in Doetsch & Hen 2005). The SVZ consists of a thin layer of dividing cells that extends along the length of the lateral walls of the lateral

* Author for correspondence (fkd2101@columbia.edu).

One contribution of 14 to a Theme Issue 'Stem cells and brain repair'.

© 2007 The Royal Society

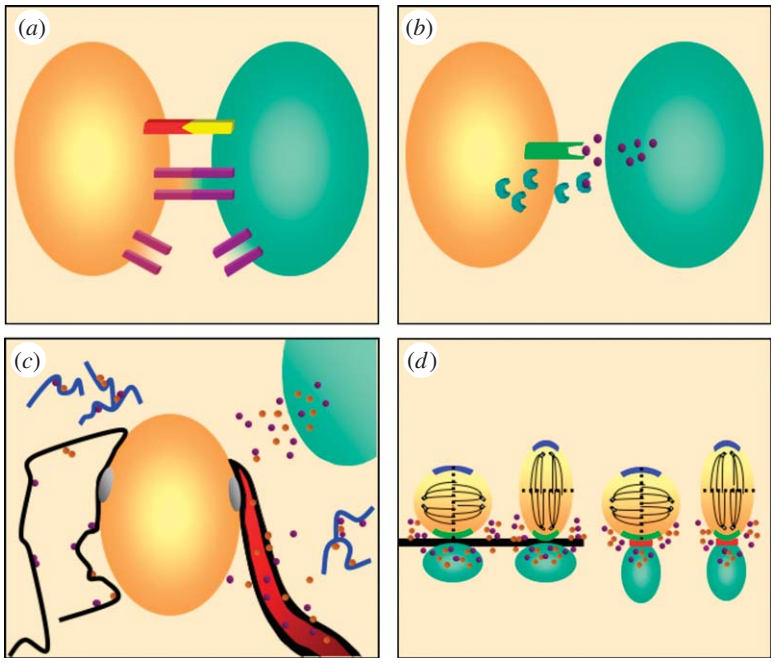

Figure 1. Components of the stem cell niche. (*a*) *Cell–cell and cell–extracellular environment interactions*. Membrane-associated receptors and ligands (red and yellow) mediate cell–cell contacts and cell fate decisions, including self-renewal and differentiation. In addition, gap junctions (purple), intercellular channels that allow the passage of ions and metabolites, both coordinate behaviour between coupled cells and can tether cells together. Hemi-channels allow communication between the cell and the environment. (*b*) *Diffusible factors*. Diffusible factors (purple spheres) can direct stem cells to either self-renew or generate differentiated progeny. The availability of diffusible factors that bind to receptors (green) in turn can be regulated by ligand inhibitors (blue half-moons), which can sequester these factors and prevent signalling. (*c*) *Basal lamina and blood vessels*. An ECM-rich basal lamina (black), which can be associated with blood vessels (red) or cells, has several functions in stem cell niches, including anchoring cells to the niche (grey spheres), sequestering and presenting diffusible signals (purple and brown spheres), and linking cells and the ECM. In addition, proteolytic fragments of ECM components (blue squiggles) may regulate stem cells. Endothelial cells and the vasculature are emerging as integral components of stem cell niches, where they can regulate stem cell fate decisions through either diffusible signals and/or direct cell–cell contact. (*d*) *Oriented cell division*. Both cell–cell contact (via adherens junctions, red) and tethering of cells to the basal lamina (black) can influence cell fate by orienting the plane of cell division, such that key intracellular determinants are symmetrically or asymmetrically distributed. Oriented segregation of these factors may influence cell fate. Short-range diffusible factors (purple and brown spheres) secreted by stromal cells (green) that promote self-renewal only influence immediately adjacent cells, allowing differentiating daughter cells to escape the niche.

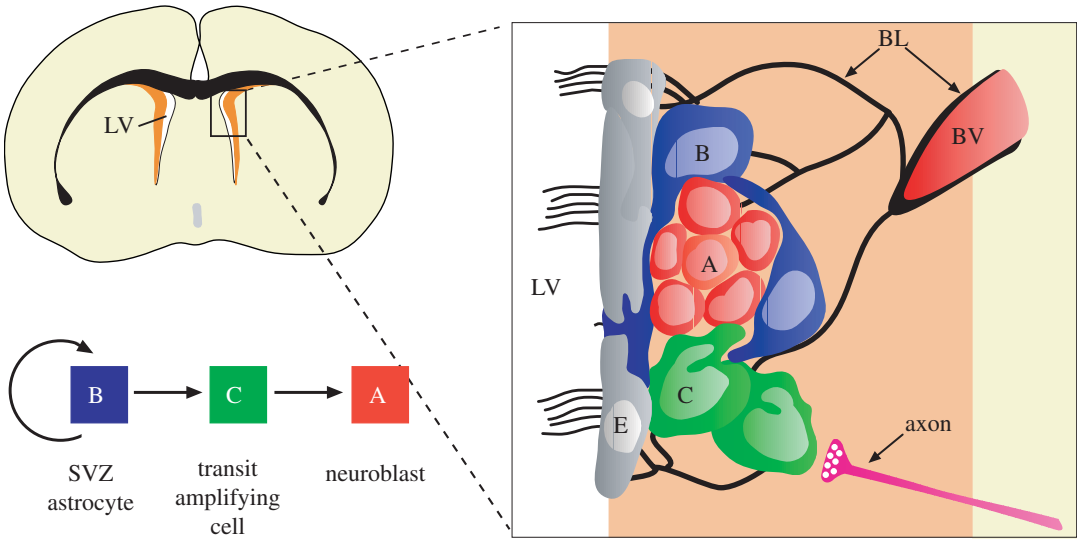

Figure 2. Cell types and anatomy of the adult SVZ niche. Schema of frontal section of the adult mouse brain showing the SVZ (orange) adjacent to the lateral ventricle (LV). SVZ astrocytes in this region (B, blue) are stem cells which generate migrating neuroblasts (A, red) destined for the olfactory bulb via a rapidly dividing transit-amplifying cell (C, green). Region in box is expanded at right to show the relationship of cells in this region and some elements of the SVZ niche. Multi-ciliated ependymal cells (E, grey) line the walls of the lateral ventricle. Chains of neuroblasts travel through tunnels formed by processes of SVZ astrocytes. Transit-amplifying cells are found in small clusters adjacent to the chains. Signals released from axons (pink) regulate proliferation and survival in this region. A specialized basal lamina (BL, black) extends from perivascular cells and contacts all cell types. Endothelial cells, blood vessels (BV) and the basal lamina are all likely key components of the niche.

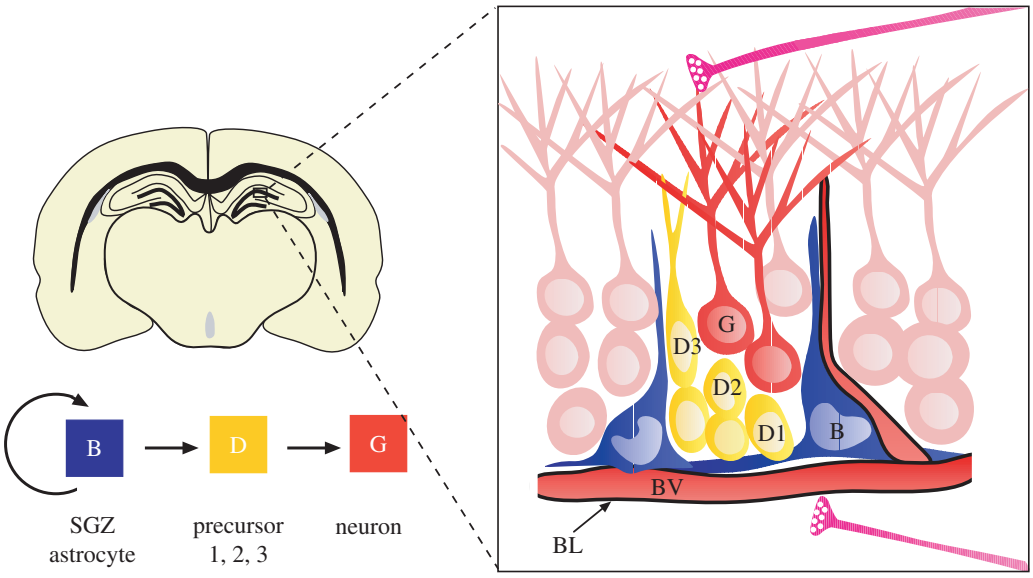

Figure 3. Cell types and anatomy of the adult SGZ niche. Schema of frontal section of the adult mouse brain showing the SGZ at the interface between the hilus (area below blood vessel) and the granule cell layer (light pink cells) of the dentate gyrus. SGZ astrocytes (B, blue) divide to generate intermediate precursors (type D cells; nomenclature according to Seri *et al.* 2004, yellow), which progressively generate more differentiated progeny (type D1 → type D2 → type D3), which mature into granule neurons (G, red). Neurogenesis occurs in pockets adjacent to blood vessels and although a specialized basal lamina has not yet been described in this region, the vascular basal lamina likely plays an important role in the niche. Afferent axons (pink) from the entorhinal cortex and axons from subcortical regions as well as from local inhibitory interneurons project to the SGZ.

ventricles and is largely separated from the cerebrospinal fluid (CSF) by a layer of multi-ciliated ependymal cells. Newly generated neuroblasts traverse a network of chains which extends throughout the SVZ to join the rostral migratory stream (RMS) that leads to the olfactory bulb. There they differentiate into two kinds of inhibitory interneurons, granule and periglomerular cells, and functionally integrate into the existing circuitry (Belluzzi *et al.* 2003; Carleton *et al.* 2003). The SGZ is located between the hilus and the granule cell layer of the dentate gyrus. Newly generated granule neurons born in the SGZ migrate only a short distance to the granule cell layer, where they extend dendrites to the molecular layer and an axon along the mossy fibre path and integrate functionally into the circuitry of the dentate gyrus (van Praag *et al.* 2002; Jessberger & Kempermann 2003; Ge *et al.* 2006).

Strikingly, in both regions, a subset of astrocytes, glial cells classically associated with support functions in the brain, are the *in vivo* primary precursors for adult neurogenesis (reviewed in Doetsch 2003*a*). These cells have been defined as astrocytes based on their ultrastructural features, markers they express and electrophysiological properties. An emerging hypothesis is that stem cells are contained within the astrocyte lineage (Alvarez-Buylla *et al.* 2001; Doetsch 2003*a*). During development, radial glia are the *in vivo* primary precursors of neurons and glia (Miyata *et al.* 2001; Noctor *et al.* 2001, 2002, 2004; Malatesta *et al.* 2003; Anthony *et al.* 2004). Post-natally, radial glia transition into astrocytes (Schmechel & Rakic 1979; Voigt 1989; Alves *et al.* 2002; Merkle *et al.* 2004), some of which are retained as stem cells in adult neurogenic niches (Merkle *et al.* 2004). Interestingly, until post-natal day 11, astrocytes from throughout the brain can generate neurospheres, but thereafter this capacity becomes limited to adult SVZ astrocytes (Laywell *et al.* 2000). An important unanswered question is whether all astrocytes (or a subset) in the adult brain retain latent neurogenic potential or whether specialized stem cell astrocytes are only found in neurogenic niches in the adult brain.

In the SVZ, stem cell astrocytes (type B cells, glial fibrillary acidic protein expressing; GFAP+) divide relatively infrequently to generate neuroblasts (type A cells, GFAP−/Dlx2+/doublecortin (Dcx)+) via a rapidly dividing transit-amplifying cell (type C cells, GFAP−/ Dlx2+) (Doetsch *et al.* 1999*a*). Oligodendrocytes can also be generated in the adult SVZ both under normal conditions (Ahn & Joyner 2005) and after demyelination (Nait-Oumesmar *et al.* 1999; Picard-Riera *et al.* 2002), but the cell type that generates them remains undefined. Whereas SVZ astrocytes have been shown to be stem cells both *in vitro* and *in vivo* (Doetsch *et al.* 1999*a*; Laywell *et al.* 2000; Imura *et al.* 2003; Morshead *et al.* 2003; Garcia *et al.* 2004; Sanai *et al.* 2004; Ahn & Joyner 2005), there is some debate as to whether the adult SGZ contains neural stem cells or only committed neurogenic precursors (Seaberg & van der Kooy 2002; Bull & Bartlett 2005). Within the SGZ, GFAP+/nestin+ astrocytes are the primary precursors that divide to generate intermediate amplifying GFAP−/nestin+ cells which in turn generate GFAP−/nestin−/Dcx+ cells that mature into differentiated granule neurons (Seri *et al.* 2001, 2004; Kronenberg *et al.* 2003; Kempermann *et al.* 2004; Steiner *et al.* 2004; Encinas *et al.* 2006). As cells progress along these lineages from SVZ and SGZ astrocytes to differentiated neurons, they sequentially acquire distinct electrophysiological properties (Carleton *et al.* 2003; Filippov *et al.* 2003; Fukuda *et al.* 2003) and express a series of transcription factors that determine their phenotype (Pleasure *et al.* 2000; Doetsch *et al.* 2002; Kronenberg *et al.* 2003;

Stenman *et al.* 2003; Parras *et al.* 2004; Seri *et al.* 2004; Hack *et al.* 2005; Kohwi *et al.* 2005; Waclaw *et al.* 2006). These molecular programmes become stabilized through a combination of epigenetic, transcriptional and post-transcriptional mechanisms (reviewed in Hsieh & Gage 2004; Cheng *et al.* 2005). Intriguingly, while common molecular mechanisms may regulate stem cell proliferation and differentiation in the embryo and the adult, some regulatory mechanisms may be unique to post-natal and/or adult neurogenesis. For example, mice deficient in the polycomb repressor Bmi1 (Molofsky *et al.* 2003), the orphan nuclear receptor Tlx (Shi *et al.* 2004) and the conditional mutants of sonic hedgehog signalling components (Machold *et al.* 2003) first reveal deficits at post-natal stages. Thus, a unique complement of regulatory mechanisms may be active in adult neurogenic regions in addition to the conserved embryonic pathways.

Stem cells harvested from non-neurogenic regions can generate neurons and astrocytes when cultured *in vitro*, but only make glia *in vivo* (reviewed in Gage 2000). In addition, primary cells from neurogenic areas transplanted into non-neurogenic regions exhibit very limited neurogenesis (reviewed in Herrera *et al.* 1999; Lim *et al.* 2000; Temple 2001). An inhibitory environment that is refractory to neurogenesis is therefore present throughout most of the brain. In contrast, upon transplantation into the SVZ, RMS or SGZ, cultured neural stem cells derived from non-neurogenic regions can generate neurons appropriate to the region (Suhonen *et al.* 1996; Shihabuddin *et al.* 2000; reviewed in Temple 2001). Thus, adult neurogenic niches have an instructive role in directing neuronal production and stem cell maintenance and shield ongoing neurogenesis from possible external inhibitory influences. Although the components of adult neurogenic niches that mediate these processes are still being elucidated, it is clear that both neural and non-neural cell types are key players (reviewed in Doetsch 2003*b*). Neurogenesis occurs in close proximity to blood vessels and may be associated with angiogenesis. Both regions are enriched in ECM proteins and a prominent basal lamina has been described in the SVZ as well as perivascular connective tissue comprising macrophages and fibroblasts. Neurotransmitter signalling probably plays a key role, as both regions are innervated by axons from distant brain regions. Finally, the proximity of unique structures, such as meningeal projections and the CSF, are likely important sources of signals.

3. STEM CELL ASTROCYTES AND NICHE ASTROCYTES

Astrocytes have classically been considered support cells in the brain, with multiple roles including forming a capsule around the surface of the brain via their endfeet, buffering extracellular potassium ion concentrations, interacting with endothelial cells to form the blood–brain barrier and taking up neurotransmitters at the synaptic cleft. However, recent work has revealed that astrocytes are dynamic regulators of many brain processes, including synaptogenesis and synaptic efficacy (Ullian *et al.* 2001; Christopherson *et al.* 2005), support adult neurogenesis (Pixley 1992; Lim & Alvarez-Buylla 1999; Song *et al.* 2002; Kornyei *et al.* 2005) and act as neural stem cells in the adult brain (Doetsch *et al.* 1999*a*; Laywell *et al.* 2000; Imura *et al.* 2003; Morshead *et al.* 2003; Garcia *et al.* 2004; Sanai *et al.* 2004; Ahn & Joyner 2005). Thus, astrocytes are emerging as key mediators of brain development, function and plasticity, highlighting the critical need to better characterize the heterogeneity and developmental specification of different subpopulations of astrocytes both within adult neurogenic regions and throughout the brain (Bachoo *et al.* 2004; Bonaguidi *et al.* 2005; Muroyama *et al.* 2005; Imura *et al.* 2006; Lim *et al.* 2006; Sakaguchi *et al.* 2006).

Within adult neurogenic niches, in addition to their role as stem cells, astrocytes are uniquely poised to be sensors and regulators of the environment. They have long processes that envelop and contact all cell types and structures in the niche (Doetsch *et al.* 1997; Seri *et al.* 2004), including blood vessels and the basal lamina (Mercier *et al.* 2002), allowing them to integrate diverse signals from many sources. Furthermore, astrocytes are often coupled via gap junctions and can form a syncytium (reviewed in Giaume & McCarthy 1996; Giaume & Venance 1998), which may allow them to propagate signals locally or throughout the entire niche, thereby regulating activation and differentiation of stem cells. Astrocytes also contribute to the neurogenic niche through contact-mediated cues and by secreting diffusible signals (Pixley 1992; Lim & Alvarez-Buylla 1999; Taupin *et al.* 2000; Song *et al.* 2002; Kornyei *et al.* 2005; Lie *et al.* 2005). It is still unknown whether the dual roles of astrocytes as stem cells and niche support cells are segregated into distinct astrocyte populations or whether individual astrocytes can have both roles.

Within the SVZ and SGZ, astrocytes are heterogeneous at the morphological, ultrastructural and molecular levels, including growth factor receptor expression, proliferation capacity and electrophysiological properties (Doetsch *et al.* 1997, 2002; Seri *et al.* 2001, 2004; Filippov *et al.* 2003; Fukuda *et al.* 2003; Kronenberg *et al.* 2003; Garcia *et al.* 2004). At least two populations of astrocytes have been defined in the SGZ: (i) radial astrocytes, which extend a process into the granule cell layer, are nestin-positive and divide and (ii) horizontal astrocytes, which extend basal processes under the granule cell layer, are nestin-negative and S100+ (Kronenberg *et al.* 2003; Seri *et al.* 2004). In the SVZ, morphologically distinct astrocytes have also been described. At the ultrastructural level, two types of astrocytes (type B1 and B2) are present that differ in their location as well as in their cytoplasmic and nuclear structure and proliferation (Doetsch *et al.* 1997). At the light microscope, both highly stellate astrocytes and unipolar and bipolar astrocytes are present, which differ in their proliferation profiles (Garcia *et al.* 2004; E. Drapeau & F. Doetsch 2004, unpublished data). Intriguingly, occasionally, the process of an SVZ astrocyte intercalates between ependymal cells and comes into contact with the lateral ventricle, and thus is exposed directly to the CSF (Doetsch *et al.* 1999*b*). The number of astrocytes in

contact with the lateral ventricle is increased during regeneration and infusion of growth factors (Doetsch *et al.* 1999*b*, 2002; Conover *et al.* 2000). SVZ astrocytes in contact with the ventricle express a single 9+0 primary cilium, which is also found on neuroepithelial stem cells during development in mammals (Sotelo & Trujillo-Cenóz 1958; Stensaas & Stensaas 1968), on radial glia (Tramontin *et al.* 2003; Spassky *et al.* 2005) and on primary precursors in adult songbirds (Alvarez-Buylla *et al.* 1998). This primary cilium may be important for transduction of signals in the CSF and for stem cell function. Despite these molecular, biophysical and morphological differences, it is still unclear whether the different subpopulations of astrocytes in both adult neurogenic regions represent functionally distinct astrocytes or are at different stages in the lineage.

4. EPENDYMAL CELLS: SUPPORT CELLS IN THE NICHE

Ependymal cells line the walls of the ventricles in the adult brain and are thus at the interface between the CSF and the brain tissue. They act both as a structural barrier and as a sensor of CSF components and osmotic pressure. Ependymal cells actively regulate the absorption of ions and transport of factors from the CSF into the parenchyma. Numerous growth factors that affect adult neurogenesis, including transforming growth factor-α (TGF-α; Seroogy *et al.* 1993), basic fibroblast growth factor (bFGF) (Hayamizu *et al.* 2001) and amphiregulin (Falk & Frisen 2002), are made by the choroid plexus, epithelial cells within the ventricles that produce the CSF. Ependymal cells may distribute and create gradients of factors produced by the choroid plexus through the directional beating of their cilia (Del Bigio 1995); thereby, they have been proposed to guide the migration of SVZ neuroblasts (Sawamoto *et al.* 2006). Importantly, within the SVZ, ependymal cells are closely apposed to astrocytes (Doetsch *et al.* 1997) as well as the specialized basal lamina (Mercier *et al.* 2002), creating the potential for transducing signals in this area. Although both ependymal cells and SVZ astrocytes are derived from radial glia (Merkle *et al.* 2004; Spassky *et al.* 2005), ependymal cells do not appear to be endogenous neural stem cells (Chiasson *et al.* 1999; Doetsch *et al.* 1999*a*; Laywell *et al.* 2000, Capela & Temple 2002; Spassky *et al.* 2005) as has been suggested (Johansson *et al.* 1999). However, they contribute to the niche in many ways, including as a source of secreted pro-neurogenic factors and of CSF components.

Ependymal cells and astrocytes are coupled by gap junctions homotypically (ependyma–ependyma and astrocyte–astrocyte) and heterotypically (ependyma—astrocyte; Zahs 1998), raising the possibility that local signals can be coordinated over long ranges within the SVZ. These intercellular channels allow the passage of ions, metabolites and second messengers between neighbouring cells (Kumar & Gilula 1996; Sohl *et al.* 2005). Connexin 43 (Cx43), a component of gap junctions that is predominantly found in glial cells in the brain (reviewed in Theis *et al.* 2005) is expressed in both adult neurogenic regions (Miragall *et al.* 1997; Menezes *et al.* 2000; Theis *et al.* 2003; Peretto *et al.* 2005; P. A. Riquelme & F. Doetsch 2005, unpublished data) and may be involved in gap junction coupling, hemi-channel signalling and cell–cell adhesion. Unlike gap junctional coupling, hemi-channels allow direct communication between the cells and the extracellular environment (Bennett *et al.* 2003). Hemi-channels have been implicated in the initiation and propagation of calcium waves between clusters of cells during cortical neurogenesis (Weissman *et al.* 2004). As such, Cx43 in adult neurogenic regions may coordinate the communication between neighbouring and distant cells within the niche either through gap junctions or hemi-channels. Cx43 may also act as a cell–cell adhesion unit to tether cells as has been suggested for glioma cells (Lin *et al.* 2002). Connexin 26, which is also expressed in the SVZ (Mercier & Hatton 2001), may have similar roles.

Ependymal cells also create a favourable environment for neurogenesis, together with astrocytes, through the secretion of noggin, an antagonist of bone morphogenic protein (BMP) signalling (Lim *et al.* 2000; Peretto *et al.* 2004). BMPs are members of the TGF-β family that are important for development. Within the adult SVZ, BMP signalling favours the production of astrocytes, but this pro-glial fate is reversed to a pro-neurogenic fate through the sequestration of BMP ligands by noggin (Lim *et al.* 2000). In addition to noggin, ependymal cells also express CXCR4 (Stumm *et al.* 2002), a chemokine receptor for SDF1 that is important for migration and survival (Dziembowska *et al.* 2005), EphA7 (Holmberg *et al.* 2005), a member of the Eph family of tyrosine kinase receptors that guide many developmental processes, and pigment epithelium-derived factor (PEDF; Ramirez-Castillejo *et al.* 2006; see §5). How these molecules and others yet to be identified interact merits further study.

5. VASCULATURE AND ADULT NEURAL STEM CELL NICHES

An important component of adult neurogenic niches is the vasculature. Within the SGZ, neurogenesis occurs in close proximity to blood vessels, with proliferative clusters containing neural progenitors, glial cells, newborn neurons and endothelial cells (Palmer *et al.* 2000), suggesting that neurogenesis and angiogenesis are coordinated processes. Indeed, common signals, which are active in adult neurogenic niches, regulate the development of the vasculature and the nervous system (reviewed in Carmeliet 2003). Thus, angiogenesis and neurogenesis are extensively interconnected and likely reciprocally influence each other in adult neurogenic regions.

Endothelial cells are emerging as critical niche cells that regulate stem cell self-renewal and neurogenesis. Co-culture of embryonic neural stem cells or adult SVZ cells with endothelial cells leads to enhanced stem cell self-renewal as well as increased neurogenesis from these expanded stem cells upon differentiation (Shen *et al.* 2004). The diffusible signals that effect these changes are as yet largely unidentified. One factor secreted by both endothelial cells and

ependymal cells is PEDF, which has been proposed to regulate SVZ astrocyte self-renewal (Ramirez-Castillejo *et al.* 2006). Endothelial cells also secrete leukaemia inhibitory factor (LIF; Mi *et al.* 2001) and brain derived neurotrophic factor (BDNF; Leventhal *et al.* 1999; Louissaint *et al.* 2002), factors known to influence proliferation and/or differentiation in adult neurogenic regions. Given LIF's role, together with BMP signalling, in promoting self-renewal in embryonic stem cells (Ying *et al.* 2003) as well as its role in astrocyte differentiation (Ross *et al.* 2003), it will be intriguing to see how LIF, BMP and noggin signalling converges within adult neurogenic niches, where astrocytes have neural stem cell properties. *In vitro*, endothelial cells in part support SVZ-derived neuron outgrowth, survival and migration through the release of BDNF (Leventhal *et al.* 1999). *In vivo* evidence for a connection between endothelial cell-derived BDNF and neuronal maturation comes from adult songbirds, in which new neurons are seasonally added to the higher vocal centre (HVC; Nottebohm 2005). Testosterone implantation induces angiogenic bursts eliciting BDNF production by endothelial cells prior to neuronal recruitment into the HVC (Louissaint *et al.* 2002). Thus, angiogenesis and neuronal maturation are coordinated in HVC.

A common factor regulating both neurogenesis and angiogenesis is vascular endothelial growth factor (VEGF), which is implicated in neurogenesis in the SVZ and SGZ (Jin *et al.* 2002; Fabel *et al.* 2003; Cao *et al.* 2004). VEGF infusion into the lateral ventricle leads to an increase in SVZ proliferation and neurogenesis, likely through VEGFR2/Flk-1 (Jin *et al.* 2002). Whether this is a direct or indirect effect on neural progenitors or cell survival is unknown. In the SGZ, exercise-induced neurogenesis in the hippocampus acts in part through VEGFR2/Flk-1 signalling (Cao *et al.* 2004). A web of factors active in adult neural stem cell niches also regulates angiogenesis, including sonic hedgehog, BMPs, Ephs/ephrins, Notch and FGF, nitric oxide and erythropoietin (reviewed in Carmeliet 2003; Alvarez-Buylla & Lim 2004, Brines & Cerami 2005; Matarredona *et al.* 2005). In addition, blood vessels are conduits for the delivery of paracrine factors, such as hormones (sex hormones, glucocorticoids and prolactin) and cytokines, from distant sources (reviewed in Gould *et al.* 2000; Lennington *et al.* 2003). These 'long-distance' cues may act directly on neural stem cells and progenitors, endothelial cells or both to regulate angiogenesis and neurogenesis.

6. BASAL LAMINA AND EXTRACELLULAR MATRIX

The ECM and associated molecules are integral components of stem cell niches creating a favourable microenvironment and architecture. They regulate signalling in the niche by providing, storing and compartmentalizing growth factors and cytokines indispensable for cell proliferation and differentiation, as well as acting as a substrate for anchoring cells.

One common component of many stem cell niches is a basal lamina. Within the SVZ, a unique basal lamina, rich in laminin and collagen-1, extends from perivascular macrophages as 'fractones' (Mercier *et al.* 2002). Each fractone consists of a base attached to perivascular macrophages, a stem crossing the SVZ and bulbs located underneath ependymal cells. The branched configuration of fractones allows for extensive interaction with all SVZ cells, especially with SVZ astrocytes and ependymal cells. This basal lamina, as well as other ECM components abundantly expressed in the SVZ, is probably a key mediator of stem cells and their progeny. Fractones may represent sites at which growth factors and other signalling molecules interact with stem cells and progenitors to regulate their proliferation, activation and differentiation by modulating the availability of signalling molecules within the stem cell niche. The source of these factors is diverse: ependymal cells, CSF, endothelial cells and SVZ cells. It will be fascinating to see whether fractones are dynamic, perhaps indicating that adult neural stem cell niches undergo constant remodelling. It is not known whether similar structures are present in the SGZ.

Other ECM components known at present to be in adult neurogenic niches are chondroitin sulphate proteoglycans (CSPG; Gates *et al.* 1995; Thomas *et al.* 1996; Bruckner *et al.* 2003), heparan sulphate proteoglycans (HSPG; Fuxe *et al.* 1994), tenascin-C (Gates *et al.* 1995; Ferhat *et al.* 1996; Jankovski & Sotelo 1996; Thomas *et al.* 1996; Deller *et al.* 1997; Heck *et al.* 2004; Peretto *et al.* 2005), laminins (Mercier *et al.* 2002; Heck *et al.* 2004) and collagen 1 (Mercier *et al.* 2002), which together probably modulate accessibility of growth factors, cytokines and other signalling molecules. Integrins are receptors that provide structural links between the ECM and the cytoskeleton, allowing for oriented cell division. In addition, they cooperate with growth factor receptors to enhance signal transduction (Comoglio *et al.* 2003). As such, the integrins coordinate spatial positioning within the niche with downstream cellular signalling and probably play a key role in maintaining adult neural stem cell niches, as they do in other stem cell niches (reviewed in Fuchs *et al.* 2004).

The cell surface carbohydrate Lewis X (LeX)/CD15/SSEA1 (fucose *N*-acetyl lactosamine), an epitope found on several stem cell populations and other cell types, is expressed by all neurosphere-forming cells in the SVZ (Capela & Temple 2002), the majority of which correspond to transit-amplifying cells and a subset of astrocytes (Doetsch *et al.* 2002). *In vivo*, LeX is also expressed by astrocytes in contact with blood vessels (Capela & Temple 2002), suggesting that LeX may capture factors from the vasculature and/or other niche cells. Indeed, LeX binds both FGFs and Wnts (Dvorak *et al.* 1998; Jirmanova *et al.* 1999; Capela & Temple 2006). LeX ectodomains are shed *in vitro* (Capela & Temple 2002); such ectodomains may modulate signalling in neurogenic niches as has been shown for proteolytic fragments of ECM which are endogenous inhibitors of angiogenesis (Sottile 2004). It will be interesting to see whether proteolytic fragments and carbohydrate ectodomains also play a role in adult neurogenic regions.

7. LONG-RANGE AND LOCAL INPUTS IN THE NICHE: NEUROTRANSMITTER REGULATION OF NEUROGENESIS

Both adult neurogenic regions are richly innervated by axonal inputs of local and distant origins. Release of neurotransmitters and other factors, such as nitric oxide and sonic hedgehog (Shh), by afferent inputs may regulate precursors at different stages of the stem cell lineage.

The SVZ receives significant axonal input from the substantia nigra, the main source of dopamine in the brain. Dopamine release from these inputs affects proliferation of the transit-amplifying C cells (Hoglinger *et al.* 2004). Loss of dopamine signalling leads to a decrease in the number of proliferating cells and subsequent neurogenesis (Baker *et al.* 2004; Hoglinger *et al.* 2004; Freundlieb *et al.* 2006), which can be rescued by giving dopamine-depleted animals dopamine analogues or D2-like receptor agonists (Hoglinger *et al.* 2004). Unlike dopamine, gamma aminobutyric acid (GABA) is a locally produced, non-synaptically released neurotransmitter that influences the SVZ niche. In contrast to its role as an inhibitory neurotransmitter in the adult brain, GABA signalling is excitatory in the adult SVZ (Wang *et al.* 2003; Bolteus & Bordey 2004; Liu *et al.* 2005), as in the developing brain (reviewed in Ben-Ari 2002; Owens & Kriegstein 2002). In addition to influencing the rate of neuronal migration in the SVZ (Bolteus & Bordey 2004), release of GABA by migrating neuroblasts negatively regulates the proliferation of GFAP+ stem cell SVZ astrocytes (Liu *et al.* 2005). Whether GABA also regulates the proliferation of transit-amplifying C cells remains to be determined. In addition, NADPH+ neurons in the striatum extensively innervate the SVZ and may regulate proliferation and neurogenesis of SVZ cells through the secretion of nitric oxide (Packer *et al.* 2003; Matarredona *et al.* 2004; Moreno-Lopez *et al.* 2004).

The SGZ receives inputs originating from distant brain regions, including the entorhinal cortex and basal forebrain/septum, and locally from interneurons within the hippocampus, which influence neurogenesis either directly or indirectly (Cameron *et al.* 1995; Lai *et al.* 2003; Fontana *et al.* 2005; Tozuka *et al.* 2005; Ge *et al.* 2006). As in the SVZ, GABA is excitatory for progenitors and newly generated neurons; however, it is released synaptically, probably by local inhibitory interneurons (Overstreet Wadiche *et al.* 2005; Tozuka *et al.* 2005; Wang *et al.* 2005; Ge *et al.* 2006). GABA decreases progenitor proliferation, promotes neuronal differentiation and is critical for the maturation of newly generated neurons (Tozuka *et al.* 2005; Ge *et al.* 2006; Overstreet-Wadiche *et al.* 2006). Like GABA, glutamatergic signalling regulates proliferation and neurogenesis in the SGZ, although it is unclear whether this is a direct or indirect effect. While *in vivo* studies have produced conflicting reports (Cameron *et al.* 1995, 1998; Gould & Cameron 1997; Deisseroth *et al.* 2004), *in vitro* studies suggest that glutamate signalling activates pro-neurogenic programmes in progenitors to stimulate neuronal production (Deisseroth *et al.* 2004). How glutamatergic and GABAergic signalling converges *in vivo* remains to be elucidated. In addition, it will be interesting to determine whether newborn or mature granule neurons feedback on to progenitors to regulate neuronal production.

Monoaminergic inputs, including serotonergic, dopaminergic and noradrenergic afferents, affect proliferation in both adult neurogenic regions (Kulkarni *et al.* 2002; Santarelli *et al.* 2003; Banasr *et al.* 2004; Hoglinger *et al.* 2004; Encinas *et al.* 2006). In contrast, cholinergic innervation is implicated in neuronal survival (Cooper-Kuhn *et al.* 2004). As such, a combination of multiple afferent inputs from distant and local brain regions regulates adult neurogenesis. Defining at which stages of the stem cell lineage different neurotransmitters and receptor subtypes regulate proliferation and differentiation will be important.

8. CELL–CELL CONTACT AND DIFFUSIBLE SIGNALS IN THE NICHE

Intrinsic genetic programmes as well as extracellular signals underlie stem cell fate choices. Whether a cell undergoes self-renewal or differentiation is the result of the spatial and temporal convergence of niche cues and the intrinsic state of the cell. The architectural elements of the niche have been discussed in detail above. Feedback signals from newly generated progeny can also regulate neural stem cells via either cell–cell contact or diffusible signals. When neuroblasts and transit-amplifying cells are depleted by an anti-mitotic treatment, SVZ astrocytes divide to rapidly regenerate the SVZ network of chains (Doetsch *et al.* 1999*a*,*b*), perhaps reflecting loss of feedback inhibition from the neuroblasts onto their ancestors, such as loss of GABA signalling (Liu *et al.* 2005). The first molecular cues that regulate stem cell lineages in the adult brain have begun to be identified (figure 4).

Notch is a transmembrane protein whose signalling regulates stem cell self-renewal in different niches (reviewed in Molofsky *et al.* 2004). Notch is activated upon binding Delta or Jagged, both membrane ligands, which causes cleavage of the intracellular tail of Notch and its translocation to the nucleus (reviewed in Gaiano & Fishell 2002). Over-expression of activated Notch1 in the embryonic brain or of activated Notch1 or 3 in adult cultured hippocampal progenitors leads to the generation of astrocytes (Gaiano *et al.* 2000; Chambers *et al.* 2001; Tanigaki *et al.* 2001). This raises the possibility that Notch is also involved in stem cell/progenitor maintenance in adult neurogenic niches, where Notch1 and Jagged1 are expressed (Stump *et al.* 2002; Irvin *et al.* 2004; Nyfeler *et al.* 2005). However, whether Notch activation promotes the acquisition of a stem cell astrocyte fate or differentiation into non-stem cell niche astrocytes awaits further evaluation.

Eph/ephrins are another large class of membrane-associated receptors and ligands with diverse roles during development, including axon guidance, cell migration and synaptogenesis (reviewed in Pasquale 2005). In the adult SVZ, different ephrins and Ephs are expressed by each SVZ cell type (Conover *et al.* 2000; Holmberg *et al.* 2005; Ricard *et al.* 2006). Eph/ephrin signalling regulates proliferation of different cell types in the adult SVZ (Conover *et al.* 2000; Holmberg *et al.* 2005; Katakowski *et al.* 2005; Ricard *et al.* 2006) and

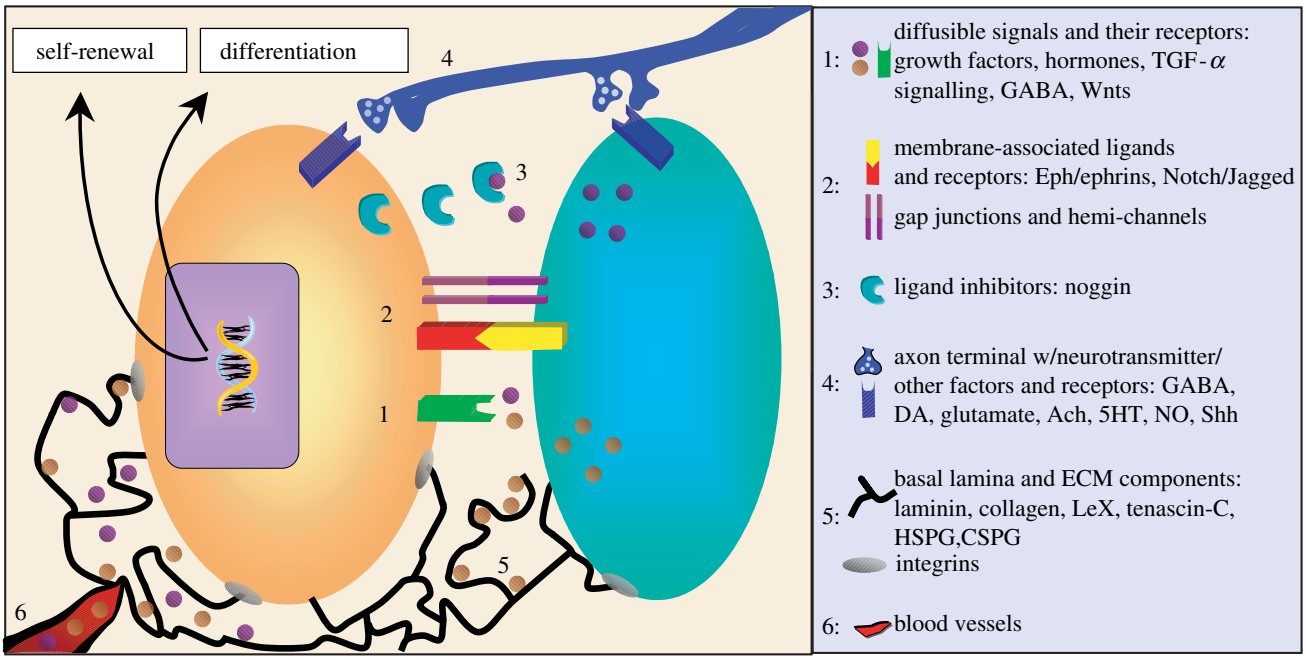

Figure 4. Neural stem cell regulation. Neural stem cells in the two adult neurogenic niches, the SVZ and SGZ, can be regulated by (1) diffusible factors (EGF, FGF, TGF-α, VEGF, PEDF, hormones, BMPs, ATP, Wnts and GABA) and their receptors, (2) cell–cell and cell–extracellular environment interactions via membrane-associated ligands and their receptors (Eph/ephrin and Notch/Jagged) or gap junctions and hemi-channels, (3) ligand inhibition (noggin), (4) release of neurotransmitters and other factors from axons (GABA, dopamine (DA), glutamate, acetylcholine (ACh), serotonin (5HT), NO and Shh), (5) basal lamina (sequestration and presentation of diffusible factors, such as growth factors) and ECM proteins (laminin, collagen-1, tenascin-C, LeX, heparan sulphate and chondroitin sulphate proteoglycans), and (6) endothelial cell/blood vessel-mediated cues. The cell types that are able to direct neural stem cell fate choices include cells within the niche (both support cells and stem cells and their progeny) (green), neurons (axonal projection; blue) and endothelial cells/blood vessels (red). Structural elements that likely regulate neural stem cell fate decisions include fractones (in the SVZ) and basal laminae (black lines). In addition, CSF and meningeal projections (not pictured in this figure) are likely important components of the niche. These elements may work in concert or independently to promote neural stem cell self-renewal or differentiation. The key to the right lists some of the factors known to be present in adult neurogenic niches.

may be an important component of feedback regulation in this region.

Another molecule involved in proliferation in adult neurogenic regions is Shh. During development, Shh acts as a morphogen, playing a crucial role in ventral patterning along the entire extent of the neuraxis, and as a mitogen, stimulating granule cell precursor proliferation in the cerebellum (reviewed in Ruiz i Altaba *et al.* 2002). In the adult SVZ, Shh regulates the proliferation of SVZ astrocytes (type B cells) and transit-amplifying type C cells (Machold *et al.* 2003; Ahn & Joyner 2005; Palma *et al.* 2005). Shh also affects proliferation in the SGZ (Lai *et al.* 2003; Machold *et al.* 2003), although it is unknown which cell type Shh acts on. As mentioned above, conditional mice deficient in *shh* signalling components first exhibit deficits in neural stem cell niches post-natally. The cellular source of Shh has not yet been identified, although for the hippocampal formation, it has been proposed that Shh is anterogradely transported to the SGZ via the fimbria–fornix (Lai *et al.* 2003; Machold *et al.* 2003). Importantly, the signals that trigger Shh release in both niches are unknown.

Members of the Wnt family of soluble ligands play critical roles in various physiological processes during development and in the adult. Wnt signalling regulates stem cell self-renewal in several stem cell niches (reviewed in Kleber & Sommer 2004). In adult neurogenic niches, Wnt signalling has thus far been shown to regulate neurogenesis in the SGZ (Lie *et al.* 2005). Increasing or decreasing Wnt activity *in vivo* leads to an increase or decrease of SGZ neurogenesis, respectively. Interestingly, astrocytes are the source of Wnts, highlighting their multiple roles in the niche.

Two mitogens widely used to culture neural stem cells *in vitro* either as neurospheres or as adherent monolayers are epidermal growth factor (EGF) and basic fibroblast growth factor (bFGF) (Reynolds & Weiss 1992; Palmer *et al.* 1995). These assays test the potential of a cell to act as a stem cell, but do not necessarily reflect its *in vivo* behaviour. Although neurospheres were believed to arise from the relatively quiescent *in vivo* SVZ stem cells, the majority of EGF-responsive neurospheres arise from the rapidly dividing transit-amplifying cells (Doetsch *et al.* 2002), which retain the capacity to act as stem cells when exposed to exogenous growth factors. These findings emphasize the need to study stem cells and their progeny *in vivo* to elucidate their roles in adult neurogenesis. Dissection of the EGF- and bFGF-responsive SVZ cell types has revealed that signalling through the EGF- and FGF-receptors occurs at distinct stages in the stem cell lineage. bFGF likely acts on more quiescent SVZ astrocytes (Zheng *et al.* 2004) and has been proposed, based on analysis of bFGF null mice, to maintain the pool of neural stem

cells (Zheng *et al.* 2004). In contrast, EGF-responsive cells are transit-amplifying C cells and a subset of SVZ astrocytes (Doetsch *et al.* 2002). This differential regulation may allow expansion of activated stem cells and transit-amplifying cells without depleting the more quiescent stem cells. The endogenous ligand for the EGF-R is likely TGF-α, which is expressed in the choroid plexus and striatum (Seroogy *et al.* 1993). Consistent with this, TGF-α null mice exhibit decreased proliferation and neurogenesis in the SVZ (Tropepe *et al.* 1997). Precise dissection of the roles of bFGF and TGF-α will require inducible genetic systems that allow one to circumvent possible developmental defects. Within adult neurogenic niches, other secreted factors, such as glycosylated cystatin which is expressed by some SGZ astrocytes, act synergistically with bFGF to influence neurogenesis (Taupin *et al.* 2000). Another signal that may synergize with bFGF is ATP (Mishra *et al.* 2006). The ecto-ATPase NTPDase2, which regulates ATP signalling, is present in both SVZ and SGZ astrocytes (Braun *et al.* 2003; Shukla *et al.* 2005). The *in vivo* role of nucleotide signalling in adult neurogenesis is yet to be explored.

Interestingly, transit-amplifying cells are emerging as key nodes in the stem cell lineage. Many signals, including dopamine (Hoglinger *et al.* 2004), soluble amyloid precursor protein (Caille *et al.* 2004), nitric oxide (Estrada *et al.* 1997; Matarredona *et al.* 2005) and Shh (Ahn & Joyner 2005; Palma *et al.* 2005), converge on the transit-amplifying cells and act cooperatively with signalling through the EGF receptor. As transit-amplifying cells are rapidly dividing, it is critical to regulate their proliferation to prevent runaway growth. Furthermore, this mode of regulation allows the relatively quiescent stem cells to divide infrequently, stopping them from accumulating mutations with division.

The structural elements and molecules important for stem cell self-renewal and differentiation, as well as for embryonic stem cells, are rapidly being elucidated. It will be fascinating to see if these pathways are conserved in other niches and, importantly, in different species. Within adult neurogenic niches, astrocytes are stem cells in the adult SVZ in both rodents (reviewed in Doetsch 2003*a*) and humans (Sanai *et al.* 2004), yet there may be fundamental differences in how neural stem cells from the two species interpret these molecular signals and ultimately affect their output. Uncovering these pathways, as well as the complex interactions within the niche, will provide invaluable insight into stem cell biology and into the potential use of neural stem cells for restorative neurogenesis in disease or trauma.

We thank Artem Kaplan and Masoud Tavazoie for critical reading of the manuscript. P.A.R. was supported by P50 AG08702 and E.D. by the Fondation pour la Recherche Medicale. F.D. is a Packard Foundation Fellow and Irma T. Hirschl Fellow and work in the laboratory is partially supported by the Jerry and Emily Spiegel Laboratory for Cell Replacement Therapies and the Anne and Bernard Spitzer Fund for Cell Replacement Therapy.

REFERENCES

Ahn, S. & Joyner, A. L. 2005 *In vivo* analysis of quiescent adult neural stem cells responding to Sonic hedgehog. *Nature* **437**, 894–897. (doi:10.1038/nature03994)

Alvarez-Buylla, A., Garcia-Verdugo, J. M. & Tramontin, A. D. 2001 A unified hypothesis on the lineage of neural stem cells. *Nat. Rev. Neurosci.* **2**, 287–293. (doi:10.1038/35067582)

Alvarez-Buylla, A., García-Verdugo, J. M., Mateo, A. & Merchant-Larios, H. 1998 Primary neural precursors and intermitotic nuclear migration in the ventricular zone of adult canaries. *J. Neurosci.* **18**, 1020–1037.

Alvarez-Buylla, A. & Lim, D. A. 2004 For the long run: maintaining germinal niches in the adult brain. *Neuron* **41**, 683–686. (doi:10.1016/S0896-6273(04)00111-4)

Alves, J. A., Barone, P., Engelender, S., Froes, M. M. & Menezes, J. R. 2002 Initial stages of radial glia astrocytic transformation in the early postnatal anterior subventricular zone. *J. Neurobiol.* **52**, 251–265. (doi:10.1002/neu.10087)

Anthony, T. E., Klein, C., Fishell, G. & Heintz, N. 2004 Radial glia serve as neuronal progenitors in all regions of the central nervous system. *Neuron* **41**, 881–890. (doi:10.1016/S0896-6273(04)00140-0)

Bachoo, R. M. *et al.* 2004 Molecular diversity of astrocytes with implications for neurological disorders. *Proc. Natl Acad. Sci. USA* **101**, 8384–8389. (doi:10.1073/pnas.0402140101)

Baker, S. A., Baker, K. A. & Hagg, T. 2004 Dopaminergic nigrostriatal projections regulate neural precursor proliferation in the adult mouse subventricular zone. *Eur. J. Neurosci.* **20**, 575–579. (doi:10.1111/j.1460-9568.2004.03486.x)

Banasr, M., Hery, M., Printemps, R. & Daszuta, A. 2004 Serotonin-induced increases in adult cell proliferation and neurogenesis are mediated through different and common 5-HT receptor subtypes in the dentate gyrus and the subventricular zone. *Neuropsychopharmacology* **29**, 450–460. (doi:10.1038/sj.npp.1300320)

Belluzzi, O., Benedusi, M., Ackman, J. & Loturco, J. J. 2003 Electrophysiological differentiation of new neurons in the olfactory bulb. *J. Neurosci.* **23**, 10 411–10 418.

Ben-Ari, Y. 2002 Excitatory actions of gaba during development: the nature of the nurture. *Nat. Rev. Neurosci.* **3**, 728–739. (doi:10.1038/nrn920)

Bennett, M. V., Contreras, J. E., Bukauskas, F. F. & Saez, J. C. 2003 New roles for astrocytes: gap junction hemichannels have something to communicate. *Trends Neurosci.* **26**, 610–617. (doi:10.1016/j.tins.2003.09.008)

Bolteus, A. J. & Bordey, A. 2004 GABA release and uptake regulate neuronal precursor migration in the postnatal subventricular zone. *J. Neurosci.* **24**, 7623–7631. (doi:10.1523/JNEUROSCI.1999-04.2004)

Bonaguidi, M. A., McGuire, T., Hu, M., Kan, L., Samanta, J. & Kessler, J. A. 2005 LIF and BMP signaling generate separate and discrete types of GFAP-expressing cells. *Development* **132**, 5503–5514. (doi:10.1242/dev.02166)

Braun, N., Sevigny, J., Mishra, S. K., Robson, S. C., Barth, S. W., Gerstberger, R., Hammer, K. & Zimmermann, H. 2003 Expression of the ecto-ATPase NTPDase2 in the germinal zones of the developing and adult rat brain. *Eur. J. Neurosci.* **17**, 1355–1364. (doi:10.1046/j.1460-9568.2003.02567.x)

Brawley, C. & Matunis, E. 2004 Regeneration of male germline stem cells by spermatogonial dedifferentiation *in vivo*. *Science* **304**, 1331–1334. (doi:10.1126/science.1097676)

Brines, M. & Cerami, A. 2005 Emerging biological roles for erythropoietin in the nervous system. *Nat. Rev. Neurosci.* **6**, 484–494. (doi:10.1038/nrn1687)

Bruckner, G., Grosche, J., Hartlage-Rubsamen, M., Schmidt, S. & Schachner, M. 2003 Region and lamina-specific

distribution of extracellular matrix proteoglycans, hyaluronan and tenascin-R in the mouse hippocampal formation. *J. Chem. Neuroanat.* **26**, 37–50. (doi:10.1016/S0891-0618(03)00036-X)

Bull, N. D. & Bartlett, P. F. 2005 The adult mouse hippocampal progenitor is neurogenic but not a stem cell. *J. Neurosci.* **25**, 10 815–10 821. (doi:10.1523/JNEUROSCI.3249-05.2005)

Caille, I., Allinquant, B., Dupont, E., Bouillot, C., Langer, A., Muller, U. & Prochiantz, A. 2004 Soluble form of amyloid precursor protein regulates proliferation of progenitors in the adult subventricular zone. *Development* **131**, 2173–2181. (doi:10.1242/dev.01103)

Cameron, H. A., McEwen, B. S. & Gould, E. 1995 Regulation of adult neurogenesis by excitatory input and NMDA receptor activation in the dentate gyrus. *J. Neurosci.* **15**, 4687–4692.

Cameron, H. A., Hazel, T. G. & McKay, R. D. G. 1998 Regulation of neurogenesis by growth factors and neurotransmitters. *J. Neurobiol.* **36**, 287–306. (doi:10.1002/(SICI)1097-4695(199808)36:2<287::AID-NEU13>3.0.CO;2-B)

Cao, L., Jiao, X., Zuzga, D. S., Liu, Y., Fong, D. M., Young, D. & During, M. J. 2004 VEGF links hippocampal activity with neurogenesis, learning and memory. *Nat. Genet.* **36**, 827–835. (doi:10.1038/ng1395)

Capela, A. & Temple, S. 2002 LeX/ssea-1 is expressed by adult mouse CNS stem cells, identifying them as non-ependymal. *Neuron* **35**, 865–875. (doi:10.1016/S0896-6273(02)00835-8)

Capela, A. & Temple, S. 2006 LeX is expressed by principle progenitor cells in the embryonic nervous system, is secreted into their environment and binds Wnt-1. *Dev. Biol.* **291**, 300–313. (doi:10.1016/j.ydbio.2005.12.030)

Carleton, A., Petreanu, L. T., Lansford, R., Alvarez-Buylla, A. & Lledo, P. M. 2003 Becoming a new neuron in the adult olfactory bulb. *Nat. Neurosci.* **6**, 507–518.

Carmeliet, P. 2003 Blood vessels and nerves: common signals, pathways and diseases. *Nat. Rev. Genet.* **4**, 710–720. (doi:10.1038/nrg1158)

Chambers, C. B., Peng, Y., Nguyen, H., Gaiano, N., Fishell, G. & Nye, J. S. 2001 Spatiotemporal selectivity of response to Notch1 signals in mammalian forebrain precursors. *Development* **128**, 689–702.

Cheng, L. C., Tavazoie, M. & Doetsch, F. 2005 Stem cells: from epigenetics to microRNAs. *Neuron* **46**, 363–367. (doi:10.1016/j.neuron.2005.04.027)

Chiasson, B. J., Tropepe, V., Morshead, C. M. & van der Kooy, D. 1999 Adult mammalian forebrain ependymal and subependymal cells demonstrate proliferative potential, but only subependymal cells have neural stem cell characteristics. *J. Neurosci.* **19**, 4462–4471.

Christopherson, K. S. *et al.* 2005 Thrombospondins are astrocyte-secreted proteins that promote CNS synaptogenesis. *Cell* **120**, 421–433. (doi:10.1016/j.cell.2004.12.020)

Comoglio, P. M., Boccaccio, C. & Trusolino, L. 2003 Interactions between growth factor receptors and adhesion molecules: breaking the rules. *Curr. Opin. Cell Biol.* **15**, 565–571. (doi:10.1016/S0955-0674(03)00096-6)

Conover, J. C., Doetsch, F., Garcia-Verdugo, J. M., Gale, N. W., Yancopoulos, G. D. & Alvarez-Buylla, A. 2000 Disruption of Eph/ephrin signaling affects migration and proliferation in the adult subventricular zone. *Nat. Neurosci.* **3**, 1091–1097. (doi:10.1038/80606)

Cooper-Kuhn, C. M., Winkler, J. & Kuhn, H. G. 2004 Decreased neurogenesis after cholinergic forebrain lesion in the adult rat. *J. Neurosci. Res.* **77**, 155–165. (doi:10.1002/jnr.20116)

Deisseroth, K., Singla, S., Toda, H., Monje, M., Palmer, T. D. & Malenka, R. C. 2004 Excitation-neurogenesis coupling in adult neural stem/progenitor cells. *Neuron* **42**, 535–552. (doi:10.1016/S0896-6273(04)00266-1)

Del Bigio, M. R. 1995 The ependyma: a protective barrier between brain and cerebrospinal fluid. *Glia* **14**, 1–13. (doi:10.1002/glia.440140102)

Deller, T., Haas, C. A., Naumann, T., Joester, A., Faissner, A. & Frotscher, M. 1997 Up-regulation of astrocyte-derived tenascin-C correlates with neurite outgrowth in the rat dentate gyrus after unilateral entorhinal cortex lesion. *Neuroscience* **81**, 829–846. (doi:10.1016/S0306-4522(97)00194-2)

Doetsch, F. 2003*a* The glial identity of neural stem cells. *Nat. Neurosci.* **6**, 1127–1134. (doi:10.1038/nn1144)

Doetsch, F. 2003*b* A niche for adult neural stem cells. *Curr. Opin. Genet Dev.* **13**, 543–550. (doi:10.1016/j.gde.2003.08.012)

Doetsch, F. & Hen, R. 2005 Young and excitable: the function of new neurons in the adult mammalian brain. *Curr. Opin. Neurobiol.* **15**, 121–128. (doi:10.1016/j.conb.2005.01.018)

Doetsch, F., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 1997 Cellular composition and three-dimensional organization of the subventricular germinal zone in the adult mammalian brain. *J. Neurosci.* **17**, 5046–5061.

Doetsch, F., Caille, I., Lim, D. A., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 1999*a* Subventricular zone astrocytes are neural stem cells in the adult mammalian brain. *Cell* **97**, 703–716. (doi:10.1016/S0092-8674(00)80783-7)

Doetsch, F., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 1999*b* Regeneration of a germinal layer in the adult mammalian brain. *Proc. Natl Acad. Sci. USA* **96**, 11 619–11 624. (doi:10.1073/pnas.96.20.11619)

Doetsch, F., Petreanu, L., Caille, I., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 2002 EGF converts transit-amplifying neurogenic precursors in the adult brain into multipotent stem cells. *Neuron* **36**, 1021–1034. (doi:10.1016/S0896-6273(02)01133-9)

Dvorak, P., Hampl, A., Jirmanova, L., Pacholikova, J. & Kusakabe, M. 1998 Embryoglycan ectodomains regulate biological activity of FGF-2 to embryonic stem cells. *J. Cell Sci.* **111**, 2945–2952. Pt 19.

Dziembowska, M., Tham, T. N., Lau, P., Vitry, S., Lazarini, F. & Dubois-Dalcq, M. 2005 A role for CXCR4 signaling in survival and migration of neural and oligodendrocyte precursors. *Glia* **50**, 258–269. (doi:10.1002/glia.20170)

Encinas, J. M., Vaahtokari, A. & Enikolopov, G. 2006 Fluoxetine targets early progenitor cells in the adult brain. *Proc. Natl Acad. Sci. USA* **103**, 8233–8238. (doi:10.1073/pnas.0601992103)

Estrada, C., Gomez, C., Martin-Nieto, J., De Frutos, T., Jimenez, A. & Villalobo, A. 1997 Nitric oxide reversibly inhibits the epidermal growth factor receptor tyrosine kinase. *Biochem. J.* **326**, 369–376. Pt 2.

Fabel, K., Fabel, K., Tam, B., Kaufer, D., Baiker, A., Simmons, N., Kuo, C. J. & Palmer, T. D. 2003 VEGF is necessary for exercise-induced adult hippocampal neurogenesis. *Eur. J. Neurosci.* **18**, 2803–2812. (doi:10.1111/j.1460-9568.2003.03041.x)

Falk, A. & Frisen, J. 2002 Amphiregulin is a mitogen for adult neural stem cells. *J. Neurosci. Res.* **69**, 757–762. (doi:10.1002/jnr.10410)

Ferhat, L., Chevassus Au Louis, N., Jorquera, I., Niquet, J., Khrestchatisky, M., Ben-Ari, Y. & Represa, A. 1996 Transient increase of tenascin-C in immature hippocampus: astroglial and neuronal expression. *J. Neurocytol.* **25**, 53–66. (doi:10.1007/BF02284785)

Filippov, V., Kronenberg, G., Pivneva, T., Reuter, K., Steiner, B., Wang, L. P., Yamaguchi, M., Kettenmann, H.

& Kempermann, G. 2003 Subpopulation of nestin-expressing progenitor cells in the adult murine hippocampus shows electrophysiological and morphological characteristics of astrocytes. *Mol. Cell Neurosci.* **23**, 373–382. (doi:10.1016/S1044-7431(03)00060-5)

Fontana, X., Nacher, J., Soriano, E. & Del Rio, J. A. 2005 Cell proliferation in the adult hippocampal formation of rodents and its modulation by entorhinal and fimbria–fornix afferents. *Cereb. Cortex* **16**, 301–312. (doi:10.1093/cercor/bhi120)

Freundlieb, N., Francois, C., Tande, D., Oertel, W. H., Hirsch, E. C. & Hoglinger, G. U. 2006 Dopaminergic substantia nigra neurons project topographically organized to the subventricular zone and stimulate precursor cell proliferation in aged primates. *J. Neurosci.* **26**, 2321–2325. (doi:10.1523/JNEUROSCI.4859-05.2006)

Fuchs, E., Tumbar, T. & Guasch, G. 2004 Socializing with the neighbors: stem cells and their niche. *Cell* **116**, 769–778. (doi:10.1016/S0092-8674(04)00255-7)

Fukuda, S., Kato, F., Tozuka, Y., Yamaguchi, M., Miyamoto, Y. & Hisatsune, T. 2003 Two distinct subpopulations of nestin-positive cells in adult mouse dentate gyrus. *J. Neurosci.* **23**, 9357–9366.

Fuxe, K., Chadi, G., Tinner, B., Agnati, L. F., Pettersson, R. & David, G. 1994 On the regional distribution of heparan sulfate proteoglycan immunoreactivity in the rat brain. *Brain Res.* **636**, 131–138. (doi:10.1016/0006-8993(94)90187-2)

Gage, F. H. 2000 Mammalian neural stem cells. *Science* **287**, 1433–1438. (doi:10.1126/science.287.5457.1433)

Gaiano, N. & Fishell, G. 2002 The role of notch in promoting glial and neural stem cell fates. *Annu. Rev. Neurosci.* **25**, 471–490. (doi:10.1146/annurev.neuro.25.030702.130823)

Gaiano, N., Nye, J. S. & Fishell, G. 2000 Radial glial identity is promoted by Notch1 signaling in the murine forebrain. *Neuron* **26**, 395–404. (doi:10.1016/S0896-6273(00)81172-1)

Garcia, A. D., Doan, N. B., Imura, T., Bush, T. G. & Sofroniew, M. V. 2004 GFAP-expressing progenitors are the principal source of constitutive neurogenesis in adult mouse forebrain. *Nat. Neurosci.* **7**, 1233–1241. (doi:10.1038/nn1340)

Gates, M. A., Thomas, L. B., Howard, E. M., Laywell, E. D., Sajin, B., Faissner, A., Gotz, B., Silver, J. & Steindler, D. A. 1995 Cell and molecular analysis of the developing and adult mouse subventricular zone of the cerebral hemispheres. *J. Comp. Neurol.* **361**, 249–266. (doi:10.1002/cne.903610205)

Ge, S., Goh, E. L., Sailor, K. A., Kitabatake, Y., Ming, G. L. & Song, H. 2006 GABA regulates synaptic integration of newly generated neurons in the adult brain. *Nature* **439**, 589–593. (doi:10.1038/nature04404)

Giaume, C. & McCarthy, K. D. 1996 Control of gap-junctional communication in astrocytic networks. *Trends Neurosci.* **19**, 319–325. (doi:10.1016/0166-2236(96) 10046-1)

Giaume, C. & Venance, L. 1998 Intercellular calcium signaling and gap junctional communication in astrocytes. *Glia* **24**, 50–64. (doi:10.1002/(SICI)1098-1136(199809)24:1<50::AID-GLIA6>3.0.CO;2-4)

Gould, E. & Cameron, H. A. 1997 Early NMDA receptor blockade impairs defensive behavior and increases cell proliferation in the dentate gyrus of developing rats. *Behav. Neurosci.* **111**, 49–56. (doi:10.1037/0735-7044.111.1.49)

Gould, E., Tanapat, P., Rydel, T. & Hastings, N. 2000 Regulation of hippocampal neurogenesis in adulthood. *Biol. Psychiatry* **48**, 715–720. (doi:10.1016/S0006-3223(00)01021-0)

Hack, M. A., Saghatelyan, A., De Chevigny, A., Pfeifer, A., Ashery-Padan, R., Lledo, P. M. & Gotz, M. 2005 Neuronal fate determinants of adult olfactory bulb neurogenesis. *Nat. Neurosci.* **8**, 865–872.

Hayamizu, T. F., Chan, P. T. & Johanson, C. E. 2001 FGF-2 immunoreactivity in adult rat ependyma and choroid plexus: responses to global forebrain ischemia and intraventricular FGF-2. *Neurol. Res.* **23**, 353–358. (doi:10.1179/016164101101198550)

Heck, N., Garwood, J., Loeffler, J. P., Larmet, Y. & Faissner, A. 2004 Differential upregulation of extracellular matrix molecules associated with the appearance of granule cell dispersion and mossy fiber sprouting during epileptogenesis in a murine model of temporal lobe epilepsy. *Neuroscience* **129**, 309–324. (doi:10.1016/j.neuroscience.2004.06.078)

Herrera, D. G., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 1999 Adult-derived neural precursors transplanted into multiple regions in the adult brain. *Ann. Neurol.* **46**, 867–877. (doi:10.1002/1531-8249(199912)46:6<867::AID-ANA9>3.0.CO;2-Z)

Hoglinger, G. U., Rizk, P., Muriel, M. P., Duyckaerts, C., Oertel, W. H., Caille, I. & Hirsch, E. C. 2004 Dopamine depletion impairs precursor cell proliferation in Parkinson disease. *Nat. Neurosci.* **7**, 726–735. (doi:10.1038/nn1265)

Holmberg, J., Armulik, A., Senti, K. A., Edoff, K., Spalding, K., Momma, S., Cassidy, R., Flanagan, J. G. & Frisen, J. 2005 Ephrin-A2 reverse signaling negatively regulates neural progenitor proliferation and neurogenesis. *Genes Dev.* **19**, 462–471. (doi:10.1101/gad.326905)

Hsieh, J. & Gage, F. H. 2004 Epigenetic control of neural stem cell fate. *Curr. Opin. Genet Dev.* **14**, 461–469. (doi:10.1016/j.gde.2004.07.006)

Imura, T., Kornblum, H. I. & Sofroniew, M. V. 2003 The predominant neural stem cell isolated from postnatal and adult forebrain but not early embryonic forebrain expresses GFAP. *J. Neurosci.* **23**, 2824–2832.

Imura, T., Nakano, I., Kornblum, H. I. & Sofroniew, M. V. 2006 Phenotypic and functional heterogeneity of GFAP-expressing cells *in vitro*: differential expression of LeX/CD15 by GFAP-expressing multipotent neural stem cells and non-neurogenic astrocytes. *Glia* **53**, 277–293. (doi:10.1002/glia.20281)

Irvin, D. K., Nakano, I., Paucar, A. & Kornblum, H. I. 2004 Patterns of Jagged1, Jagged2, Delta-like 1 and Delta-like 3 expression during late embryonic and postnatal brain development suggest multiple functional roles in progenitors and differentiated cells. *J. Neurosci. Res.* **75**, 330–343. (doi:10.1002/jnr.10843)

Jankovski, A. & Sotelo, C. 1996 Subventricular zone-olfactory bulb migratory pathway in the adult mouse: cellular composition and specificity as determined by heterochronic and heterotopic transplantation. *J. Comp. Neurol.* **371**, 376–396. (doi:10.1002/(SICI)1096-9861(19960729)371:3<376::AID-CNE3>3.0.CO;2-)

Jessberger, S. & Kempermann, G. 2003 Adult-born hippocampal neurons mature into activity-dependent responsiveness. *Eur. J. Neurosci.* **18**, 2707–2712. (doi:10.1111/j.1460-9568.2003.02986.x)

Jin, K., Zhu, Y., Sun, Y., Mao, X. O., Xie, L. & Greenberg, D. A. 2002 Vascular endothelial growth factor (VEGF) stimulates neurogenesis *in vitro* and *in vivo*. *Proc. Natl Acad. Sci. USA* **99**, 11 946–11 950. (doi:10.1073/pnas.182296499)

Jirmanova, L., Pacholikova, J., Krejci, P., Hampl, A. & Dvorak, P. 1999 O-linked carbohydrates are required for FGF-2-mediated proliferation of mouse embryonic cells. *Int. J. Dev. Biol.* **43**, 555–562.

Johansson, C. B., Momma, S., Clarke, D. L., Risling, M., Lendahl, U. & Frisen, J. 1999 Identification of a neural stem cell in the adult mammalian central nervous system. *Cell* **96**, 25–34. (doi:10.1016/S0092-8674(00)80956-3)

Kai, T. & Spradling, A. 2004 Differentiating germ cells can revert into functional stem cells in *Drosophila melanogaster* ovaries. *Nature* **428**, 564–569. (doi:10.1038/nature02436)

Katakowski, M., Zhang, Z., Decarvalho, A. C. & Chopp, M. 2005 EphB2 induces proliferation and promotes a neuronal fate in adult subventricular neural precursor cells. *Neurosci. Lett.* **385**, 204–209. (doi:10.1016/j.neulet.2005.05.060)

Kempermann, G., Jessberger, S., Steiner, B. & Kronenberg, G. 2004 Milestones of neuronal development in the adult hippocampus. *Trends Neurosci.* **27**, 447–452. (doi:10.1016/j.tins.2004.05.013)

Kleber, M. & Sommer, L. 2004 Wnt signaling and the regulation of stem cell function. *Curr. Opin. Cell Biol.* **16**, 681–687. (doi:10.1016/j.ceb.2004.08.006)

Kohwi, M., Osumi, N., Rubenstein, J. L. & Alvarez-Buylla, A. 2005 Pax6 is required for making specific subpopulations of granule and periglomerular neurons in the olfactory bulb. *J. Neurosci.* **25**, 6997–7003. (doi:10.1523/JNEUROSCI.1435-05.2005)

Kornyei, Z., Szlavik, V., Szabo, B., Gocza, E., Czirok, A. & Madarasz, E. 2005 Humoral and contact interactions in astroglia/stem cell co-cultures in the course of glia-induced neurogenesis. *Glia* **49**, 430–444. (doi:10.1002/glia.20123)

Kronenberg, G., Reuter, K., Steiner, B., Brandt, M. D., Jessberger, S., Yamaguchi, M. & Kempermann, G. 2003 Subpopulations of proliferating cells of the adult hippocampus respond differently to physiologic neurogenic stimuli. *J. Comp. Neurol.* **467**, 455–463. (doi:10.1002/cne.10945)

Kulkarni, V. A., Jha, S. & Vaidya, V. A. 2002 Depletion of norepinephrine decreases the proliferation, but does not influence the survival and differentiation, of granule cell progenitors in the adult rat hippocampus. *Eur. J. Neurosci.* **16**, 2008–2012. (doi:10.1046/j.1460-9568.2002.02268.x)

Kumar, N. M. & Gilula, N. B. 1996 The gap junction communication channel. *Cell* **84**, 381–388. (doi:10.1016/S0092-8674(00)81282-9)

Lai, K., Kaspar, B. K., Gage, F. H. & Schaffer, D. V. 2003 Sonic hedgehog regulates adult neural progenitor proliferation *in vitro* and *in vivo*. *Nat. Neurosci.* **6**, 21–27. (doi:10.1038/nn983)

Laywell, E. D., Rakic, P., Kukekov, V. G., Holland, E. C. & Steindler, D. A. 2000 Identification of a multipotent astrocytic stem cell in the immature and adult mouse brain. *Proc. Natl Acad. Sci. USA* **97**, 13 883–13 888. (doi:10.1073/pnas.250471697)

Lennington, J. B., Yang, Z. & Conover, J. C. 2003 Neural stem cells and the regulation of adult neurogenesis. *Reprod. Biol. Endocrinol.* **1**, 99. (doi:10.1186/1477-7827-1-99)

Leventhal, C., Rafii, S., Rafii, D., Shahar, A. & Goldman, S. A. 1999 Endothelial trophic support of neuronal production and recruitment from the adult mammalian subependyma. *Mol. Cell Neurosci.* **13**, 450–464. (doi:10.1006/mcne.1999.0762)

Li, L. & Xie, T. 2005 Stem cell niche: structure and function. *Annu. Rev. Cell Dev. Biol.* **17**, 605–631. (doi:10.1146/annurev.cellbio.21.012704.131525)

Lie, D. C. *et al.* 2005 Wnt signalling regulates adult hippocampal neurogenesis. *Nature* **437**, 1370–1375. (doi:10.1038/nature04108)

Lim, D. A. & Alvarez-Buylla, A. 1999 Interaction between astrocytes and adult subventricular zone precursors stimulates neurogenesis. *Proc. Natl Acad. Sci. USA* **96**, 7526–7531. (doi:10.1073/pnas.96.13.7526)

Lim, D. A., Tramontin, A. D., Trevejo, J. M., Herrera, D. G., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 2000 Noggin antagonizes BMP signaling to create a niche for adult neurogenesis. *Neuron* **28**, 713–726. (doi:10.1016/S0896-6273(00)00148-3)

Lim, D. A., Suarez-Farinas, M., Naef, F., Hacker, C. R., Menn, B., Takebayashi, H., Magnasco, M., Patil, N. & Alvarez-Buylla, A. 2006 *In vivo* transcriptional profile analysis reveals RNA splicing and chromatin remodeling as prominent processes for adult neurogenesis. *Mol. Cell Neurosci.* **31**, 131–148. (doi:10.1016/j.mcn.2005.10.005)

Lin, J. H. *et al.* 2002 Connexin 43 enhances the adhesivity and mediates the invasion of malignant glioma cells. *J. Neurosci.* **22**, 4302–4311.

Liu, X., Wang, Q., Haydar, T. F. & Bordey, A. 2005 Nonsynaptic GABA signaling in postnatal subventricular zone controls proliferation of GFAP-expressing progenitors. *Nat. Neurosci.* **8**, 1179–1187. (doi:10.1038/nn1522)

Louissaint Jr, A., Rao, S., Leventhal, C. & Goldman, S. A. 2002 Coordinated interaction of neurogenesis and angiogenesis in the adult songbird brain. *Neuron* **34**, 945–960. (doi:10.1016/S0896-6273(02)00722-5)

Machold, R. *et al.* 2003 Sonic hedgehog is required for progenitor cell maintenance in telencephalic stem cell niches. *Neuron* **39**, 937–950. (doi:10.1016/S0896-6273(03)00561-0)

Malatesta, P., Hack, M. A., 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. (doi:10.1016/S0896-6273(03) 00116-8)

Marshman, E., Booth, C. & Potten, C. S. 2002 The intestinal epithelial stem cell. *Bioessays* **24**, 91–98. (doi:10.1002/bies.10028)

Matarredona, E. R., Murillo-Carretero, M., Moreno-Lopez, B. & Estrada, C. 2004 Nitric oxide synthesis inhibition increases proliferation of neural precursors isolated from the postnatal mouse subventricular zone. *Brain Res.* **995**, 274–284. (doi:10.1016/j.brainres.2003.10.010)

Matarredona, E. R., Murillo-Carretero, M., Moreno-Lopez, B. & Estrada, C. 2005 Role of nitric oxide in subventricular zone neurogenesis. *Brain Res. Brain Res. Rev.* **49**, 355–366. (doi:10.1016/j.brainresrev.2005.01.001)

Menezes, J. R., Froes, M. M., Moura Neto, V. & Lent, R. 2000 Gap junction-mediated coupling in the postnatal anterior subventricular zone. *Dev. Neurosci.* **22**, 34–43. (doi:10.1159/000017425)

Mercier, F. & Hatton, G. I. 2001 Connexin 26 and basic fibroblast growth factor are expressed primarily in the subpial and subependymal layers in adult brain parenchyma: roles in stem cell proliferation and morphological plasticity? *J. Comp. Neurol.* **431**, 88–104. (doi:10.1002/1096-9861(20010226)431:1<88::AID-CNE1057>3.0.CO;2-D)

Mercier, F., Kitasako, J. T. & Hatton, G. I. 2002 Anatomy of the brain neurogenic zones revisited: fractones and the fibroblast/macrophage network. *J. Comp. Neurol.* **451**, 170–188. (doi:10.1002/cne.10342)

Merkle, F. T., Tramontin, A. D., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 2004 Radial glia give rise to adult neural stem cells in the subventricular zone. *Proc. Natl Acad. Sci. USA* **101**, 17 528–17 532. (doi:10.1073/pnas.0407893101)

Mi, H., Haeberle, H. & Barres, B. A. 2001 Induction of astrocyte differentiation by endothelial cells. *J. Neurosci.* **21**, 1538–1547.

Miragall, F., Albiez, P., Bartels, H., de Vries, U. & Dermietzel, R. 1997 Expression of the gap junction protein connexin43 in the subependymal layer and the rostral migratory stream of the mouse: evidence for an inverse correlation between intensity of connexin43 expression and cell proliferation activity. *Cell Tiss. Res.* **287**, 243–253. (doi:10.1007/s004410050749)

Mishra, S. K. *et al.* 2006 Extracellular nucleotide signaling in adult neural stem cells: synergism with growth factor-mediated cellular proliferation. *Development* **133**, 675–684. (doi:10.1242/dev.02233)

Miyata, T., Kawaguchi, A., Okano, H. & Ogawa, M. 2001 Asymmetric inheritance of radial glial fibers by cortical neurons. *Neuron* **31**, 727–741. (doi:10.1016/S0896-6273(01)00420-2)

Molofsky, A. V., Pardal, R., Iwashita, T., Park, I. K., Clarke, M. F. & Morrison, S. J. 2003 Bmi-1 dependence distinguishes neural stem cell self-renewal from progenitor proliferation. *Nature* **425**, 962–967. (doi:10.1038/nature02060)

Molofsky, A. V., Pardal, R. & Morrison, S. J. 2004 Diverse mechanisms regulate stem cell self-renewal. *Curr. Opin. Cell Biol.* **16**, 700–707. (doi:10.1016/j.ceb.2004.09.004)

Moreno-Lopez, B., Romero-Grimaldi, C., Noval, J. A., Murillo-Carretero, M., Matarredona, E. R. & Estrada, C. 2004 Nitric oxide is a physiological inhibitor of neurogenesis in the adult mouse subventricular zone and olfactory bulb. *J. Neurosci.* **24**, 85–95. (doi:10.1523/JNEUROSCI.1574-03.2004)

Morshead, C. M., Garcia, A. D., Sofroniew, M. V. & van der Kooy, D. 2003 The ablation of glial fibrillary acidic protein-positive cells from the adult central nervous system results in the loss of forebrain neural stem cells but not retinal stem cells. *Eur. J. Neurosci.* **18**, 76–84. (doi:10.1046/j.1460-9568.2003.02727.x)

Muroyama, Y., Fujiwara, Y., Orkin, S. H. & Rowitch, D. H. 2005 Specification of astrocytes by bHLH protein SCL in a restricted region of the neural tube. *Nature* **438**, 360–363. (doi:10.1038/nature04139)

Nait-Oumesmar, B., Decker, L., Lachapelle, F., Avellana-Adalid, V., Bachelin, C. & van Evercooren, A. B. 1999 Progenitor cells of the adult mouse subventricular zone proliferate, migrate and differentiate into oligodendrocytes after demyelination. *Eur. J. Neurosci.* **11**, 4357–4366. (doi:10.1046/j.1460-9568.1999.00873.x)

Noctor, S. C., Flint, A. C., Weissman, T. A., Dammerman, R. S. & Kriegstein, A. R. 2001 Neurons derived from radial glial cells establish radial units in neocortex. *Nature* **409**, 714–720. (doi:10.1038/35055553)

Noctor, S. C., Flint, A. C., Weissman, T. A., Wong, W. S., Clinton, B. K. & Kriegstein, A. R. 2002 Dividing precursor cells of the embryonic cortical ventricular zone have morphological and molecular characteristics of radial glia. *J. Neurosci.* **22**, 3161–3173.

Noctor, S. C., Martinez-Cerdeno, V., Ivic, L. & Kriegstein, A. R. 2004 Cortical neurons arise in symmetric and asymmetric division zones and migrate through specific phases. *Nat. Neurosci.* **7**, 136–144. (doi:10.1038/nn1172)

Nottebohm, F. 2005 The neural basis of birdsong. *PLoS Biol.* **3**, e164. (doi:10.1371/journal.pbio.0030164)

Nyfeler, Y., Kirch, R. D., Mantei, N., Leone, D. P., Radtke, F., Suter, U. & Taylor, V. 2005 Jagged1 signals in the postnatal subventricular zone are required for neural stem cell self-renewal. *EMBO J.* **24**, 3504–3515. (doi:10.1038/sj.emboj.7600816)

Overstreet Wadiche, L., Bromberg, D. A., Bensen, A. L. & Westbrook, G. L. 2005 GABAergic signaling to newborn neurons in dentate gyrus. *J. Neurophysiol.* **94**, 4528–4532. (doi:10.1152/jn.00633.2005)

Overstreet-Wadiche, L. S., Bensen, A. L. & Westbrook, G. L. 2006 Delayed development of adult-generated granule cells in dentate gyrus. *J. Neurosci.* **26**, 2326–2334. (doi:10.1523/JNEUROSCI.4111-05.2006)

Owens, D. F. & Kriegstein, A. R. 2002 Is there more to GABA than synaptic inhibition? *Nat. Rev. Neurosci.* **3**, 715–727. (doi:10.1038/nrn919)

Packer, M. A., Stasiv, Y., Benraiss, A., Chmielnicki, E., Grinberg, A., Westphal, H., Goldman, S. A. & Enikolopov, G. 2003 Nitric oxide negatively regulates mammalian adult neurogenesis. *Proc. Natl Acad. Sci. USA* **100**, 9566–9571. (doi:10.1073/pnas.1633579100)

Palma, V. *et al.* 2005 Sonic hedgehog controls stem cell behavior in the postnatal and adult brain. *Development* **132**, 335–344. (doi:10.1242/dev.01567)

Palmer, T. D., Ray, J. & Gage, F. H. 1995 FGF-2 responsive neuronal progenitors reside in proliferative and quiescent regions of the adult rodent brain. *Mol. Cell. Neurosci.* **6**, 474–486. (doi:10.1006/mcne.1995.1035)

Palmer, T. D., Willhoite, A. R. & Gage, F. H. 2000 Vascular niche for adult hippocampal neurogenesis. *J. Comp. Neurol.* **425**, 479–494. (doi:10.1002/1096-9861(20001002)425:4<479::AID-CNE2>3.0.CO;2-3)

Parras, C. M. *et al.* 2004 Mash1 specifies neurons and oligodendrocytes in the postnatal brain. *EMBO J.* **23**, 4495–4505. (doi:10.1038/sj.emboj.7600447)

Pasquale, E. B. 2005 Eph receptor signalling casts a wide net on cell behaviour. *Nat. Rev. Mol. Cell Biol.* **6**, 462–475. (doi:10.1038/nrm1662)

Peretto, P., Dati, C., De Marchis, S., Kim, H. H., Ukhanova, M., Fasolo, A. & Margolis, F. L. 2004 Expression of the secreted factors noggin and bone morphogenetic proteins in the subependymal layer and olfactory bulb of the adult mouse brain. *Neuroscience* **128**, 685–696. (doi:10.1016/j.neuroscience.2004.06.053)

Peretto, P., Giachino, C., Aimar, P., Fasolo, A. & Bonfanti, L. 2005 Chain formation and glial tube assembly in the shift from neonatal to adult subventricular zone of the rodent forebrain. *J. Comp. Neurol.* **487**, 407–427. (doi:10.1002/cne.20576)

Picard-Riera, N., Decker, L., Delarasse, C., Goude, K., Nait-Oumesmar, B., Liblau, R., Pham-Dinh, D. & Evercooren, A. B. 2002 Experimental autoimmune encephalomyelitis mobilizes neural progenitors from the subventricular zone to undergo oligodendrogenesis in adult mice. *Proc. Natl Acad. Sci. USA* **99**, 13 211–13 216. (doi:10.1073/pnas.192314199)

Pixley, S. 1992 CNS glial cells support *in vitro* survival division, and differentiation of dissociated olfactory neuronal progenitor cells. *Neuron* **8**, 1191–1204. (doi:10.1016/0896-6273(92)90139-5)

Pleasure, S. J., Collins, A. E. & Lowenstein, D. H. 2000 Unique expression patterns of cell fate molecules delineate sequential stages of dentate gyrus development. *J. Neurosci.* **20**, 6095–6105.

Raff, M. 2003 Adult stem cell plasticity: fact or artifact? *Annu. Rev. Cell Dev. Biol.* **19**, 1–22. (doi:10.1146/annurev.cellbio.19.111301.143037)

Ramirez-Castillejo, C., Sanchez-Sanchez, F., Andreu-Agullo, C., Ferron, S. R., Aroca-Aguilar, J. D., Sanchez, P., Mira, H., Escribano, J. & Farinas, I. 2006 Pigment epithelium-derived factor is a niche signal for neural stem cell renewal. *Nat. Neurosci.* **9**, 331–339. (doi:10.1038/nn1657)

Reya, T., Morrison, S. J., Clarke, M. F. & Weissman, I. L. 2001 Stem cells, cancer, and cancer stem cells. *Nature* **414**, 105–111. (doi:10.1038/35102167)

Reynolds, B. & Weiss, S. 1992 Generation of neurons and astrocytes from isolated cells of the adult mammalian central nervous system. *Science* **255**, 1707–1710. (doi:10.1126/science.1553558)

Ricard, J., Salinas, J., Garcia, L. & Liebl, D. J. 2006 EphrinB3 regulates cell proliferation and survival in adult neurogenesis. *Mol. Cell Neurosci.* **31**, 713–722. (doi:10.1016/j.mcn.2006.01.002)

Ross, S. E., Greenberg, M. E. & Stiles, C. D. 2003 Basic helix-loop-helix factors in cortical development. *Neuron* **39**, 13–25. (doi:10.1016/S0896-6273(03)00365-9)

Ruiz i Altaba, A., Sanchez, P. & Dahmane, N. 2002 Gli and hedgehog in cancer: tumours, embryos and stem cells. *Nat. Rev. Cancer* **2**, 361–372. (doi:10.1038/nrc796)

Sakaguchi, M. *et al.* 2006 A carbohydrate-binding protein, Galectin-1, promotes proliferation of adult neural stem cells. *Proc. Natl Acad. Sci. USA* **103**, 7112–7117. (doi:10.1073/pnas.0508793103)

Sanai, N. *et al.* 2004 Unique astrocyte ribbon in adult human brain contains neural stem cells but lacks chain migration. *Nature* **427**, 740–744. (doi:10.1038/nature02301)

Santarelli, L. *et al.* 2003 Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants. *Science* **301**, 805–809. (doi:10.1126/science.1083328)

Sawamoto, K. *et al.* 2006 New neurons follow the flow of cerebrospinal fluid in the adult brain. *Science* **311**, 629–632. (doi:10.1126/science.1119133)

Schmechel, D. E. & Rakic, P. 1979 A Golgi study of radial glia cells in developing monkey telencephalon: morphogenesis and transformation into astrocytes. *Anat. Embryol.* **156**, 115–152. (doi:10.1007/BF00300010)

Seaberg, R. M. & van der Kooy, D. 2002 Adult rodent neurogenic regions: the ventricular subependyma contains neural stem cells, but the dentate gyrus contains restricted progenitors. *J. Neurosci.* **22**, 1784–1793.

Seri, B., Garcia-Verdugo, J. M., Mcewen, B. S. & Alvarez-Buylla, A. 2001 Astrocytes give rise to new neurons in the adult mammalian hippocampus. *J. Neurosci.* **21**, 7153–7160.

Seri, B., Garcia-Verdugo, J. M., Collado-Morente, L., McEwen, B. S. & Alvarez-Buylla, A. 2004 Cell types, lineage, and architecture of the germinal zone in the adult dentate gyrus. *J. Comp. Neurol.* **478**, 359–378. (doi:10.1002/cne.20288)

Seroogy, K. B., Lundgren, K. H., Lee, D. C., Guthrie, K. M. & Gall, C. M. 1993 Cellular localization of transforming growth factor-a mRNA in rat forebrain. *J. Neurochem.* **60**, 1777–1782. (doi:10.1111/j.1471-4159.1993.tb13403.x)

Shen, Q., Goderie, S. K., Jin, L., Karanth, N., Sun, Y., Abramova, N., Vincent, P., Pumiglia, K. & Temple, S. 2004 Endothelial cells stimulate self-renewal and expand neurogenesis of neural stem cells. *Science* **304**, 1338–1340. (doi:10.1126/science.1095505)

Shi, Y., Chichung Lie, D., Taupin, P., Nakashima, K., Ray, J., Yu, R. T., Gage, F. H. & Evans, R. M. 2004 Expression and function of orphan nuclear receptor TLX in adult neural stem cells. *Nature* **427**, 78–83. (doi:10.1038/nature02211)

Shihabuddin, L. S., Horner, P. J., Ray, J. & Gage, F. H. 2000 Adult spinal cord stem cells generate neurons after transplantation in the adult dentate gyrus. *J. Neurosci.* **20**, 8727–8735.

Shukla, V., Zimmermann, H., Wang, L., Kettenmann, H., Raab, S., Hammer, K., Sevigny, J., Robson, S. C. & Braun, N. 2005 Functional expression of the ecto-ATPase NTPDase2 and of nucleotide receptors by neuronal progenitor cells in the adult murine hippocampus. *J. Neurosci. Res.* **80**, 600–610. (doi:10.1002/jnr.20508)

Sohl, G., Maxeiner, S. & Willecke, K. 2005 Expression and functions of neuronal gap junctions. *Nat. Rev. Neurosci.* **6**, 191–200.

Song, H., Stevens, C. F. & Gage, F. H. 2002 Astroglia induce neurogenesis from adult neural stem cells. *Nature* **417**, 39–44. (doi:10.1038/417039a)

Sotelo, J. R. & Trujillo-Cenóz, O. 1958 Electron microscope study on the development of ciliary components of the neural epithelium of the chick embryo. *Z. Zellforsch.* **49**, 1–12. (doi:10.1007/BF00335059)

Sottile, J. 2004 Regulation of angiogenesis by extracellular matrix. *Biochim. Biophys. Acta* **1654**, 13–22.

Spassky, N., Merkle, F. T., Flames, N., Tramontin, A. D., Garcia-Verdugo, J. M. & Alvarez-Buylla, A. 2005 Adult ependymal cells are postmitotic and are derived from radial glial cells during embryogenesis. *J. Neurosci.* **25**, 10–18. (doi:10.1523/JNEUROSCI.1108-04.2005)

Spradling, A., Drummond-Barbosa, D. & Kai, T. 2001 Stem cells find their niche. *Nature* **414**, 98–104. (doi:10.1038/35102160)

Steiner, B., Kronenberg, G., Jessberger, S., Brandt, M. D., Reuter, K. & Kempermann, G. 2004 Differential regulation of gliogenesis in the context of adult hippocampal neurogenesis in mice. *Glia* **46**, 41–52. (doi:10.1002/glia.10337)

Stenman, J., Toresson, H. & Campbell, K. 2003 Identification of two distinct progenitor populations in the lateral ganglionic eminence: implications for striatal and olfactory bulb neurogenesis. *J. Neurosci.* **23**, 167–174.

Stensaas, L. J. & Stensaas, S. S. 1968 An electron microscope study of cells in the matrix and intermediate laminae of the cerebral hemisphere of the 45 mm rabbit embryo. *Z. Zellforsch.* **91**, 341–365. (doi:10.1007/BF00440763)

Stumm, R. K., Rummel, J., Junker, V., Culmsee, C., Pfeiffer, M., Krieglstein, J., Hollt, V. & Schulz, S. 2002 A dual role for the SDF-1/CXCR4 chemokine receptor system in adult brain: isoform-selective regulation of SDF-1 expression modulates CXCR4-dependent neuronal plasticity and cerebral leukocyte recruitment after focal ischemia. *J. Neurosci.* **22**, 5865–5878.

Stump, G., Durrer, A., Klein, A. L., Lutolf, S., Suter, U. & Taylor, V. 2002 Notch1 and its ligands Delta-like and Jagged are expressed and active in distinct cell populations in the postnatal mouse brain. *Mech. Dev.* **114**, 153–159. (doi:10.1016/S0925-4773(02)00043-6)

Suhonen, J. O., Peterson, D. A., Ray, J. & Gage, F. H. 1996 Differentiation of adult hippocampus-derived progenitors into olfactory neurons *in vivo*. *Nature* **383**, 624–627. (doi:10.1038/383624a0)

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. (doi:10.1016/S0896-6273(01)00179-9)

Taupin, P., Ray, J., Fischer, W. H., Suhr, S. T., Hakansson, K., Grubb, A. & Gage, F. H. 2000 FGF-2-responsive neural stem cell proliferation requires CCg, a novel autocrine/paracrine cofactor. *Neuron* **28**, 385–397. (doi:10.1016/S0896-6273(00)00119-7)

Temple, S. 2001 The development of neural stem cells. *Nature* **414**, 112–117. (doi:10.1038/35102174)

Theis, M., Sohl, G., Speidel, D., Kuhn, R. & Willecke, K. 2003 Connexin43 is not expressed in principal cells of mouse cortex and hippocampus. *Eur. J. Neurosci.* **18**, 267–274. (doi:10.1046/j.1460-9568.2003.02740.x)

Theis, M., Sohl, G., Eiberger, J. & Willecke, K. 2005 Emerging complexities in identity and function of glial connexins. *Trends Neurosci.* **28**, 188–195. (doi:10.1016/j.tins.2005.02.006)

Thomas, L. B., Gates, M. A. & Steindler, D. A. 1996 Young neurons from the adult subependymal zone proliferate and migrate along an astrocyte, extracellular matrix-rich pathway. *Glia* **17**, 1–14. (doi:10.1002/(SICI)1098-1136(199605)17:1<1::AID-GLIA1>3.0.CO;2-7)

Tozuka, Y., Fukuda, S., Namba, T., Seki, T. & Hisatsune, T. 2005 GABAergic excitation promotes neuronal differentiation in adult hippocampal progenitor cells. *Neuron* **47**, 803–815. (doi:10.1016/j.neuron.2005.08.023)

Tramontin, A. D., Garcia-Verdugo, J. M., Lim, D. A. & Alvarez-Buylla, A. 2003 Postnatal development of radial glia and the ventricular zone (VZ): a continuum of the neural stem cell compartment. *Cereb. Cortex* **13**, 580–587. (doi:10.1093/cercor/13.6.580)

Tropepe, V., Craig, C. G., Morshead, C. M. & van der Kooy, D. 1997 Transforming growth factor-alpha null and senescent mice show decreased neural progenitor cell proliferation in the forebrain subependyma. *J. Neurosci.* **17**, 7850–7859.

Ullian, E. M., Sapperstein, S. K., Christopherson, K. S. & Barres, B. A. 2001 Control of synapse number by glia. *Science* **291**, 657–661. (doi:10.1126/science.291.5504.657)

van Praag, H., Schinder, A. F., Christie, B. R., Toni, N., Palmer, T. D. & Gage, F. H. 2002 Functional neurogenesis in the adult hippocampus. *Nature* **415**, 1030–1034. (doi:10.1038/4151030a)

Voigt, T. 1989 Development of glial cells in the cerebral wall of ferrets: direct tracing of their transformation from radial glia into astrocytes. *J. Comp. Neurol.* **289**, 74–88. (doi:10.1002/cne.902890106)

Waclaw, R. R., Allen II, Z. J., Bell, S. M., Erdelyi, F., Szabo, G., Potter, S. S. & Campbell, K. 2006 The zinc finger transcription factor Sp8 regulates the generation and diversity of olfactory bulb interneurons. *Neuron* **49**, 503–516. (doi:10.1016/j.neuron.2006.01.018)

Wang, D. D., Krueger, D. D. & Bordey, A. 2003 GABA depolarizes neuronal progenitors of the postnatal subventricular zone via GABAA receptor activation. *J. Physiol.* **550**, 785–800. (doi:10.1113/jphysiol.2003.042572)

Wang, L. P., Kempermann, G. & Kettenmann, H. 2005 A subpopulation of precursor cells in the mouse dentate gyrus receives synaptic GABAergic input. *Mol. Cell. Neurosci.* **29**, 181–189. (doi:10.1016/j.mcn.2005.02.002)

Weissman, T. A., Riquelme, P. A., Ivic, L., Flint, A. C. & Kriegstein, A. R. 2004 Calcium waves propagate through radial glial cells and modulate proliferation in the developing neocortex. *Neuron* **43**, 647–661. (doi:10.1016/j.neuron.2004.08.015)

Ying, Q. L., Nichols, J., Chambers, I. & Smith, A. 2003 BMP induction of Id proteins suppresses differentiation and sustains embryonic stem cell self-renewal in collaboration with STAT3. *Cell* **115**, 281–292. (doi:10.1016/S0092-8674(03)00847-X)

Zahs, K. R. 1998 Heterotypic coupling between glial cells of the mammalian central nervous system. *Glia* **24**, 85–96. (doi:10.1002/(SICI)1098-1136(199809)24:1<85::AID-GLIA9>3.0.CO;2-)

Zheng, W., Nowakowski, R. S. & Vaccarino, F. M. 2004 Fibroblast growth factor 2 is required for maintaining the neural stem cell pool in the mouse brain subventricular zone. *Dev. Neurosci.* **26**, 181–196. (doi:10.1159/000082136)