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REVIEW

Wnt signaling and neural stem cells: caught in the Wnt web

Theologos M. Michaelidis · D. Chichung Lie

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Abstract Wnt proteins have now been identified as major physiological regulators of multiple aspects of stem cell biology, from self-renewal and pluripotency to precursor cell competence and terminal differentiation. Neural stem cells are the cellular building blocks of the developing nervous system and provide the basis for continued neurogenesis in the adult mammalian central nervous system. Here, we outline the most recent advances in the field about the critical factors and regulatory networks involved in Wnt signaling and discuss recent findings on how this increasingly intricate pathway contributes to the shaping of the developing and adult nervous system on the level of the neural stem cell.

Keywords Signaling · Wnt · Stem cell · Neurogenesis · Development

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T. M. Michaelidis · D. C. Lie (✉)
GSF-National Research Center for Environment and Health,
Institute of Developmental Genetics,
Ingolstädter Landstrasse 1,
85764 Munich-Neuherberg, Germany
e-mail: chichung.lie@gsf.de

Present address:
T. M. Michaelidis
Department of Biological Applications & Technology,
University of Ioannina, Science and Technology School,
45110 Ioannina, Greece

T. M. Michaelidis
Foundation for Research & Technology-Hellas,
Biomedical Research Institute, University of Ioannina,
Science and Technology School,
45110 Ioannina, Greece

Introduction

In order to generate the vast number of neurons and glia of the mature nervous system, proliferation and maintenance of neural stem cells has to be balanced against their commitment towards a neural lineage. These decisions as well as the subsequent differentiation and maturation of the neural stem cell progeny are largely controlled by extrinsic signals. Wnt ligands are secreted proteins that are characterized by a high number of conserved cysteine residues. The founding member of the Wnt-protein family, i.e. Wnt-1, was identified in 1982 by Nusse and Varmus (Nusse and Varmus 1982) as a preferential integration site for mouse mammary tumor virus-induced breast tumors. Since then, numerous Wnt proteins have been described including 19 human and murine Wnt proteins (for additional information see Wnt homepage: http://www.stanford.edu/~rnusse/wntwindow.html). Work over the last 25 years has established Wnt proteins as essential regulators for the development of various organs including the nervous system as well as for adult tissue homoeostasis (for review see, e.g., (Logan and Nusse 2004; Clevers 2006). Wnt proteins participate in the regulation of almost every aspect of neural development including patterning of the neural tube, neural stem cell maintenance, proliferation, fate determination, axon guidance, dendrite development, and synapse formation. The ability of Wnt proteins to control these biologically

divergent and sometimes opposing processes in neural development may be explained by their great potential to elicit several signaling cascades and coordinate multiple cellular signals and their downstream molecular targets.

At least two distinct pathways are activated by Wnts, the so-called canonical branch, which results in the activation of β-catenin/TCF transcriptional regulatory complexes (Clevers 2006), and the non-canonical β-catenin-independent branch, which involves a wide range of downstream signaling effectors (Klein and Mlodzik 2005; Kohn and Moon 2005). The canonical Wnt/β-catenin branch is typically activated by the Wnt1 class (Wnt1, 2, 3, 3a, 8 and 8a), whereas the non-canonical pathway is triggered by the Wnt5a class of Wnt ligands (Wnt4, 5a, 5b, 6, 7a and 11). Activation of both branches is initiated by binding of the relevant Wnt ligands to the corresponding Frizzled receptors, which appear to play an active role in the final output of Wnt signaling (Mikels and Nusse 2006). Despite increasing evidence implicating the non-canonical signaling in the regulation of gastrulation and neurulation, there is only limited information with regards to the significance of this pathway in the biology of neural stem cells. Therefore, in this review we will first summarize the two Wnt branches and then focus on the critical components and interplays that adjust the output of the canonical, β-catenin-dependent pathway. We will subsequently discuss the diverse points of connection of canonical Wnt signaling with neural stem cell biology for which we have the most information and the greatest understanding.

The Wnt-signaling pathways

Canonical branch

In the absence of a Wnt signal, free cytoplasmic β-catenin is recruited in a large "scaffolding" complex consisting of Axin, APC (adenomatous polyposis coli), CK1 (casein kinase 1) and the serine/threonine kinase GSK (glucogen synthase kinase)-3β. This cytoplasmic complex allows the sequential phosphorylation of β-catenin at the N-terminal "destruction box." The phosphorylated β-catenin binds to the β-TrCP (β-transducin repeat-containing protein) component of a E3 ubiquitin ligase complex, which catalyses the ubiquitination of β-catenin and its subsequent destruction by the proteasome machinery. In the presence of Wnt, binding of the ligand to a Frizzled receptor and the LRP5/6 coreceptor triggers the recruitment of the two cytoplasmic components, Dishevelled and Axin, respectively. By an as yet ill-defined mechanism, GSK3β is then released from the scaffolding complex, resulting in accumulation of unphosphorylated β-catenin, which enters the nucleus and forms complexes with a number of transcription factors, the most common ones being the members of the Tcf (T-cell factor) and LEF (lymphocyte enhancer factor) family. In the absence of β-catenin, Tcf/LEF proteins act as transcriptional repressors by forming complexes with Groucho/TLE (transducin-like enhancer of split) proteins. The interaction with β-catenin converts these repressors into activators, which via recruitment of diverse coregulatory molecules, induce the expression of downstream target genes (for a comprehensive list, see the Wnt homepage: http://www.standford.edu/~rnusse/pathways/targets.html). The ability to switch between a repressor and an activator status is a regulatory strategy that appears to be utilized also by other transcription factors mediating signaling (Barolo and Posakony 2002), and establishes a tight control of target gene expression at the transcriptional level (van Es et al. 2003).

Non-canonical branch

The non-canonical signaling branch has at least two intracellular signaling cascades, namely the Wnt/planar cell polarity (PCP) and the Wnt/Ca^{2+} pathway. In the Wnt/PCP pathway, Wnt proteins bind to Frizzled receptors on the cell surface and activate Rho/Rac small GTPases (Habas et al. 2003) or the Jun N-terminal kinase (JNK) (Moriguchi et al. 1999), thereby regulating gene expression and subsequent cytoskeletal organization in crucial morphogenetic events during development (Veeman et al. 2003; Wang and Malbon 2003; Kuhl 2004). It is important to note that the precise orchestration of these signaling events is not yet fully characterized. In the Wnt/Ca^{2+} pathway, the binding of Wnts to Frizzled receptors stimulates heterotrimeric G proteins and phospholipase C, thereby increasing intracellular Ca^{2+} release and decreasing cyclic guanosine monophosphate (cGMP) levels. This results in activation of Ca^{2+}-calmodulin-dependent protein kinase II (CamKII) and protein kinase C (PKC), which in turn can stimulate nuclear factor (NF)-AT and other transcriptional regulators (Veeman et al. 2003; Wang and Malbon 2003; Kuhl 2004). It is not yet clear whether the PCP and Wnt/Ca^{2+} pathways are overlapping or represent distinct, context-specific noncanonical branches.

Critical players of the canonical signaling

The biogenesis of the Wnt ligands

The mechanisms that regulate synthesis, sorting and secretion of Wnts are particularly complex [for a recent review, see (Coudreuse and Korswagen 2007)]. For their secretion, Wnt proteins need to be post-translationally modified in the endoplasmic reticulum (ER) by cysteine palmitoylation (Willert et al. 2003), a modification which allows them to be loaded to lipoprotein particles (Panakova

et al. 2005). The relevant enzyme has been identified as *porcupine* in Drosophila (Hofmann 2000) and *mom-1* in C. elegans (Zhai et al. 2004). The intracellular trafficking requires a conserved seven-pass transmembrane protein, which is colocalized with Wnt in the Golgi apparatus. It is encoded by a conserved gene known as *wntless (wls)* or *evenness interrupted (evi)* (Banziger et al. 2006; Bartscherer et al. 2006). Another component of the Wnt secretory pathway is the protein VPS-35, a subunit of the "retromer" complex, which is essential for secretion of the ligand and formation of the long-range gradient (Coudreuse et al. 2006; Prasad and Clark 2006).

Receptors and co-receptors

The main receptors of Wnts, the Frizzled proteins, consist of at least ten family members that share a highly conserved seven-transmembrane domain separated by short extracellular and cytoplasmic loops, a cysteine-rich extracellular N-terminal domain and a less conserved cytoplasmic domain of variable size. An additional level of complexity is provided by the fact that a single Wnt can bind to Frizzled proteins with different affinities, suggesting that the specific action of Wnt might be determined by the individual Frizzled receptor that is expressed in the target cell (Hsieh 2004; Kikuchi et al. 2007). For the canonical Wnt signaling pathway, Frizzled receptors synergize with a single-pass transmembrane protein of the low-density-lipoprotein receptor family (LRP) known as Arrow in *Drosophila* (Wehrli et al. 2000) and LRP5/6 in vertebrates (Pinson et al. 2000; Tamai et al. 2000). The surface expression of both types of receptors is essential for the initiation of the Wnt signal, and in the case of Arrow/LRP5/6, this is dependent on a chaperone called Mesd in mice (Hsieh et al. 2003). A second, recently identified coreceptor for both canonical and non-canonical Wnt signaling is a transmembrane tyrosine kinase receptor from the Ryk family. Like the LRP5/6 coreceptor, Ryk can form a complex with Frizzled proteins and initiate the canonical pathway (Lu et al. 2004). Interestingly, Ryk can also activate the non-canonical pathway through Frizzled-independent signaling cascades (Cheyette 2004; Bejsovec 2005); however the signaling events downstream of these cascades are not known. Ryk proteins contain an extracellular domain with homology to Wnt inhibitory factor (WIF1) and a conserved PDZ-binding motif, which links Ryk to downstream molecules of the canonical Wnt pathway, such as Dishevelled (Cheyette 2004; Lu et al. 2004; Bejsovec 2005).

Extracellular modulators of Wnt signaling

A common theme in many potent signaling pathways is the presence of regulatory proteins capable of dampening down the effects of the ligands. On this level, the Wnt activity is controlled by transmembrane as well as secreted proteins. Secreted antagonists include soluble forms of Frizzled-related proteins (sFRPs), members of the Dkk family of proteins, Cerberus and WIF proteins, which inhibit Wnt signaling either by binding directly to Wnts or to the receptor components (Kawano and Kypta 2003). The sFRP proteins contain the cystein-rich domain (CRD) of the Frizzled family that is sufficient and necessary for Wnt binding, but no transmembrane regions (Hoang et al. 1996). Therefore, they are able to bind and sequester Wnts preventing their interaction with Frizzled and LRP5/6 receptors and inhibiting both canonical and non-canonical signaling (Finch et al. 1997; Leyns et al. 1997; Hsieh et al. 1999). In contrast, Dkk proteins such as Dkk1 do not bind to Wnt ligands, but rather to the coreceptor LRP5/6, and therefore, they specifically inhibit the Wnt canonical pathway (Glinka et al. 1998). Through the interaction with LRP5/6, Dkk1 recruits a different class of transmembrane molecules, the Kremens (Mao et al. 2002). In the simplest model, the Dkk1-LRP5/6-Kremen complex is internalized and degraded, thus depleting the cell surface from the LRP5/6 coreceptor, which is essential for the canonical signaling. Interestingly, there are three members in the Dkk family. While Dkk1 acts as a specific Wnt antagonist, Dkk2 also binds to LRP5/6, but depending on the cellular context it can either activate or inhibit the canonical pathway (Li et al. 2002), whereas Dkk3 fails to bind to LRP5/6 and inhibit Wnt signaling (Krupnik et al. 1999). Cerberus functions as a more general, multivalent growth-factor antagonist, as in addition to Wnt it binds to Nodal and BMP ligands via independent sites (Piccolo et al. 1999). The synchronized inhibition of all three pathways by Cerberus results in simultaneous head formation and trunk inhibition. WIF1 also appears to antagonize Wnt signaling by sequestering Wnts from their Frizzled receptors; however in contrast to sFRPs, it lacks a CRD-like domain (Hsieh et al. 1999).

The regulation of the Wnt pathway at the extracellular level becomes even more complex considering that depending on the context, extracellular inhibitors can also enhance signaling by facilitating Wnt secretion, transport or stability (Logan and Nusse 2004). In addition, the canonical Wnts are sensitive to the action of other non-canonical Wnts, which are cabable of interfering with the canonical pathway (Logan and Nusse 2004; He and Axelrod 2006). For example, Wnt5a can antagonize the Wnt3a canonical pathway via the tyrosine kinase receptor Ror2 (Mikels and Nusse 2006). Finally, the Frizzled/LRP receptors can also be activated by factors unrelated to Wnt such as the cysteine-knot protein Norrin, which induces the canonical Wnt pathway (Xu et al. 2004), and the R-spondins, which can activate β-catenin (Kazanskaya et al. 2004; Nam et al. 2006) and promote proliferation on the intestinal epithelium (Kim et al. 2005).

Downstream of the activated receptors, the complexity of Wnt signaling is conveyed in the regulation and subcellular distribution of the two crucial transducers, Dishevelled and β-catenin.

Dishevelled

In both canonical and non-canonical pathways the Wnt signal is transmitted through the cytoplasmic multifunctional phosphoprotein Dishevelled (a single member in Drosophila, *Dsh* and three members in mammals *Dvl-1*, *-2* and *-3*) (Wallingford and Habas 2005). The function of Dsh/Dvl is required for multiple Wnt/Frizzled cascades, and also in this case, the final response to Wnt activation depends on the cellular context (Ilyas 2005). Recent studies from numerous laboratories have revealed that Dsh/Dvl proteins act as core scaffolds for multicomponent signaling complexes, providing docking sites for a wide range of protein kinases, phosphatases, adaptor proteins as well as for other scaffolding molecules, such as Axin. Dsh/Dvl proteins can self-associate in vitro and in vivo and generate dynamic assemblies via their DIX (Dishevelled-Axin) domain, which are important for the efficient recruitment of Axin (Schwarz-Romond et al. 2005). These proteins display a remarkably dynamic spatial localization. Accordingly, Dsh/Dvl can act at the plasma membrane, in the cytoplasm or even within the nucleus with the nuclear import of Dsh/Dvl being essential for its function in the pathway (Habas and Dawid 2005; Itoh et al. 2005; Weitzman 2005). The Dsh/Dvl proteins comprise three highly conserved primary domains (Wharton 2003), which are involved in multiple protein-protein interactions with downstream effectors and are essential for subcellular trafficking. This modular structure allows the separate activation of at least three different signaling cascades: (1) the Wnt/β-catenin canonical pathway that regulates nuclear translocation of β-catenin and activation of the Tcf/LEF transcriptional regulators, (2) the Wnt/ROCK(Rho-associated protein kinase)/JNK pathway that controls the non-canonical PCP cascade that modulates the actin cytoskeleton and (3) the Wnt/Ca^{2+}/cGMP pathways that control the activation of Ca^{2+}-sensitive transcription factors as well as the cGMP-dependent responses that are involved in ventral axis specification (Malbon and Wang 2006). It therefore appears that Dsh/Dvl represents a crucial branching point relatively upstream on the Wnt signaling cascade, which is capable of interpreting the variety of Wnt-generated stimuli and transmitting them downstream into physiologically appropriate signaling routes.

Activation and transcriptional properties of β-catenin

In the absence of the ligand, the cytoplasmic levels of β-catenin are constantly low via a mechanism that involves its continuous phosphorylation, ubiquitination and degradation by the 26S proteasome. Upon binding of Wnts to their receptors two critical events take place: (1) LRP5/6 becomes sequentially phosphorylated by GSK3β and CK1-γ (which is anchored in the membrane through C-terminal palmitoylation), thereby generating a high affinity docking site for recruitment of Axin to the cell membrane and its subsequent destabilization (Mao et al. 2001; Davidson et al. 2005; Zeng et al. 2005), (2) Dsh/Dvl is recruited to the membrane where it is hyperphosphorylated by CK1-ε and CK2, resulting in recruitment of Frat-1 (Kishida et al. 2001; Lee et al. 2001; Hay et al. 2005) and release of GSK3β from the scaffolding complex (Li et al. 1999; Ferkey and Kimelman 2002). The destabilization of Axin in combination with the inhibition of GSK3β activity results in the rapid accumulation of free β-catenin and its translocation into the nucleus where it potently controls transcriptional events. Upon removal of the Wnt source, β-catenin is exported from the nucleus by the APC, which is a key negative regulator of the cascade. Importantly, in addition to promoting the nuclear export of β-catenin (Bienz 2002), nuclear APC was recently shown to counteract the transcriptional activity of β-catenin and to control the periodic exchange of β-catenin-associated coregulatory complexes on the responsive promoters (Sierra et al. 2006).

In the nucleus, β-catenin regulates the expression of Tcf/LEF-dependent target genes by two distinct mechanisms. First, it displaces the Groucho/TLE repressors from the Tcf/LEF proteins via competion for overlapping binding sites, which are located at the Tcf/Lef's N-terminus (Daniels and Weis 2005; Sierra et al. 2006). Second, it recruits regulatory proteins to Tcf/LEF-bound chromatin through two potent transcription activation domains that are present at its N- and C-termini. The N-terminus interacts directly with the adaptor protein Bcl-9/Lgs (Legless) (Stadeli and Basler 2005), and in this way connects LEF-1 to the PHD finger protein Pygopus (Pygo), a transcriptional activator (Willert and Jones 2006; Hoffmans and Basler 2007). By forming this trimeric complex, Bcl-9/Lgs and Pygo proteins have also been implicated in the nuclear localization of β-catenin (Brembeck et al. 2004; Townsley et al. 2004). On the other side, the C-terminus of β-catenin recruits multiple complexes that are capable to modify and to reorganize the chromatin structure [reviewed in (Stadeli et al. 2006; Willert and Jones 2006)].

The activation of Tcf/LEFs by β-catenin can be disrupted by certain inhibitory proteins such as ICAT (inhibitor of β-catenin and Tcf) (Tago et al. 2000) or Chibby (Takemaru et al. 2003). The ICAT-mediated inhibition appears to be crucial for the specification of the fate of neural progenitor cells along the anterior-posterior axis. Indeed, in an in vitro system in which Wnt3a was

capable to redirect the fate of neural progenitors to a posterior character, ICAT could directly induce forebrain cells via its inhibitory effect on Wnt (Satoh et al. 2004). Importantly, in addition to the Tcf/LEF key DNA-binding partners, β-catenin can also interact with other DNA-binding proteins, such as members of the SRY-box containing (Sox) family of transcription factors (Zorn et al. 1999; Sinner et al. 2004), the bicoid-related protein Pitx2 (Kioussi et al. 2002) and the paired-like homeodomain transcription factor Prophet of Pit1 (Prop1), which was recently found to be important for pituitary cell fate determination (Olson et al. 2006)(see later), thereby providing additional mechanisms that might contribute to the precise control of Wnt/β-catenin-mediated cell fate decisions.

As is evident from the above, the ability of β-catenin to shuttle between the cytoplasm and the nucleus, interacting with a diverse inventory of proteins ranging from protein kinases to chromatin remodeling factors and chromatin-modifying enzymes as well as to cooperate with different DNA-binding partners in the nucleus, enables the dynamic and precise translation of the extracellular Wnt signal to a specific transcriptional output. It also reflects the competence of β-catenin to act as an additional integration point for various Wnt-induced developmental pathways downstream of Dishevelled.

The Tcf/LEF transcription factors

Another major determinant of the ability of Wnt proteins to regulate a wide range of biological processes is the remarkable diversity of the Tcf/LEF actions in the nucleus (Arce et al. 2006). This complexity is partly due to the occurrence of multiple isoforms for these transcription factors, which in combination with the diverse interactions with the DNA and a wide range of coregulatory proteins can differentially affect the expression of various target genes. Whereas nonvertebrate organisms such as flies and worms carry a single Tcf/LEF gene, higher organisms express at least four family members: Tcf-1, LEF-1, Tcf-3 and Tcf-4 (Hurlstone and Clevers 2002). Furthermore, a large number of isoforms is generated through a combination of alternative splicing and differential promoter usage (Van de Wetering et al. 1996). In addition, Tcf/LEF proteins are characterized by both redundancy and functional diversity. For example, in Xenopus, LEF-1 functions as a Wnt-dependent activator, whereas the Tcf members act both as repressors and activators (Dorsky et al. 2002; Gradl et al. 2002; Houston et al. 2002; Standley et al. 2006). In early mouse embryogenesis, Tcf-1 and LEF-1 appear to be redundant since both genes have to be deleted in order to phenocopy the Wnt3a$^{-/-}$ knockout at E9.5 (Galceran et al. 1999). In the intestine, disruption of the Tcf-4 gene was found to deplete the epithelial stem cell compartments (Korinek et al. 1998), whereas deletion of Tcf-1 resulted in tumor formation several months after birth (Roose et al. 1999), suggesting that in this system, Tcf-4 promotes stem cell maintenance, whereas Tcf-1 exerts the opposite effect. The fourth member of the family, Tcf-3, is widely expressed at early developmental stages and exhibits strong and unique actions, behaving either as an activator or a repressor. Notably, Tcf-3 inhibits the expression of genes that are activated not only by Wnt, but also by other signaling pathways (Merrill et al. 2004; Pereira et al. 2006). In vitro, Tcf-3 was recently shown to limit ES self-renewal by repressing nanog expression (Pereira et al. 2006), whereas its repression activity was found to be important for the maintenance of a stem cell niche in the hair follicles (DasGupta and Fuchs 1999; Merrill et al. 2001). Note that some of the splice variants that are produced by the Tcf/LEF loci act as natural dominant negative isoforms, and there is evidence that the relative ratio in isoform abundance may play an important role in proliferation and differentiation (Hovanes et al. 2001; Weerkamp et al. 2006; Willinger et al. 2006).

All Tcf/LEF isoforms bind β-catenin with their first 50 N-terminal amino acids. The downstream-located CRD domain that mediates the recruitment of the Groucho/TLE repressor as well as the cooperation with transcriptional activators separates the β-catenin-binding region from the high-mobility group (HMG) DNA-binding domain and the nuclear localization signal (NLS). At the C-terminus of some members, downstream of the NLS, we find the so-called “E” tail, which is amenable to alternative splicing in vertebrates and may confer selectivity in the regulation of the target genes. This region is present only in the Tcf genes (not in LEF-1) and encodes a DNA-binding domain with little sequence specificity, which can stabilize the association with certain DNA response elements, as well as a second domain that facilitates the interaction with CBP/p300. For example, two Wnt target genes, LEF-1 and Cdx-1 (caudal-related homeobox), are activated by β-catenin only when the latter is recruited to the corresponding promoters by the Tcf-1 and Tcf-4 splice variants carrying the E tail (Atcha et al. 2003; Hecht and Stemmler 2003). The E-tail has two motifs for transient binding of CtBP (C-terminal binding protein), a particularly interesting protein that can act as a transcriptional corepressor of Wnt signaling (Valenta et al. 2003; Hamada and Bienz 2004) influencing epigenetic chromatin modifications through interaction with Polycomb corepressors (Chinnadurai 2002), but also as a transcriptional co-activator of some Wnt targets (Fang et al. 2006). A rather unusual property of CtBP is that its activity is regulated by the nuclear NADH/NAD+ ratio, and therefore its impact on Wnt signaling might be controlled by the metabolic status of the cell. In

addition, CtBP, in cooperation with APC, was recently found to participate in the clearance of β-catenin from transcriptionally active Tcf/LEF/β-catenin complexes bound on the target promoter (c-Myc), a step that is thought to be a prerequisite for the subsequent recruitment of the Groucho/TLEs and Hdac1 (histone deacetylase 1) by the enhancer bound Tcf/LEFs and termination of transcription (Sierra et al. 2006). Interestingly, mouse embryos that lack the CtBP2 isoform display delayed neural development, axial truncations and reduced Brachyury (T) expression, resembling the phenotype of the Wnt3a mutants (Hildebrand and Soriano 2002).

Interactions of Tcf/LEFs with other transcriptional regulators

The relatively weak recognition of chromatin DNA by the Tcf/LEF proteins requires the cooperation with other transcription factors to increase binding affinity and specificity. This strategy allows other pathways to integrate on Tcf/LEF activity, thereby contributing further to the amazing complexity of the Wnt signaling in vertebrates. Of particular importance for the cross talk between Wnt and other major developmental pathways such as the TGF-β/BMP is the cooperation of Tcf/LEFs with the SMAD transcription factors that are bound to adjacent elements, thereby allowing the coordinated regulation of target genes (Riese et al. 1997; Labbe et al. 2000; Nishita et al. 2000), for example, Msx2 (Hussein et al. 2003) or c-Myc(Hu and Rosenblum 2005). Importantly, synergistic interactions occur also with homeodomain family proteins such as Alx4 on the NCAM promoter (Boras and Hamel 2002), Pitx2 on LEF1 (Vadlamudi et al. 2005) and Cdx1 on its own promoter (Beland et al. 2004). Another tempting Tcf/LEF-interacting partner that can significantly enhance transcriptional activation of target genes (Feng et al. 2007) is the Drosophila *split ends* (*spen*) homologue, known as *MINT* (Msx2-interacting nuclear targeting protein) in mouse or *SHARP* (SMRT/Hdac1-associated receptor protein) in human. The functional interaction between SHARP and Tcf/LEF was shown to be independent on β-catenin. Interestingly, the Drosophila homologue has been previously involved in neuronal cell fate specification and axonal guidance (Chen and Rebay 2000; Kuang et al. 2000) as well as in cell cycle regulation (Lane et al. 2000), possibly via the ability of Spen to interact with Notch and EGF (epidermal growth factor) signaling pathways (Chen and Rebay 2000; Kuang et al. 2000). Targeted ablation of the mouse homolog MINT conditionally in the brain resulted in severe reduction in hippocampal size and brain hypoplasia (Yabe et al. 2007). It is worth mentioning that SHARP was originally discovered as a corepressor for nuclear hormone receptors (Shi et al. 2001) and the DNA-binding protein RBP-Jk, a component of Notch signaling (Oswald et al. 2002). Although the underlying mechanism of SHARP action on Tcf/LEF activity is still unknown, it may involve some of the interesting properties of this large protein, such as its ability to act as a nuclear matrix-associated scaffolding factor organizing the promoter region of target genes (Sierra et al. 2004) or bind RNA coactivators (Shi et al. 2001) and participate in RNA splicing and nuclear export (Hiriart et al. 2005). Of relevance to Wnt signaling, but from a different point of view, is the ability of the aforementioned Tcf/LEF-interacting protein CtBP to increase significantly SHARP-mediated repression on Notch target genes (Nagel et al. 2005; Oswald et al. 2005) as well as to inhibit key players of BMP signaling such as Id1 in response to the SMAD6 component of TGFβ pathway (Lin et al. 2003). Considering the above observations we may infer that SHARP and CtBP may act as convergence points for most, if not all, the major developmental pathways that are known to influence the maintenance and fate determination of neural stem cells.

Finally, another layer for regulation of Tcf/LEF activity is provided by proteins that bind on these transcription factors and inhibit their activity directly or sequester them away from their target promoters or result in their covalent modification. An example of the first category includes the Frodo proteins, which were originally identified as proteins interacting with Dishevelled. These proteins appear to act as signaling adaptors involved in both, Wnt signaling and Nodal, another major pathway operating during vertebrate development, and are also important for neural tissue specification (Hikasa and Sokol 2004; Brott and Sokol 2005). Moreover, besides the recruitment of Groucho/TLE repressors, which is a common property of all Tcf/LEF proteins, some members of the family interact also with certain co-repressors such as Kaiso, a BTB/POZ protein that acts first by antagonizing the recruitment of β-catenin on the target promoter (Siamois) and then cooperates with a Tcf/LEF member (xTcf3) to repress transcription (van Roy and McCrea 2005). Examples of the second category include the Kaiso-related protein HIC1, which together with CtBP, sequesters Tcf-4 to nuclear bodies (Valenta et al. 2006), the LIM-containing HIC5, a nuclear hormone receptor coactivator, which inhibits Tcf/LEF activity (Ghogomu et al. 2006), and the proteins I-mfa and HIC, which bind to the HMG domains of Tcf-3 and Lef-1 and prevent DNA binding (Snider et al. 2001). Lastly, covalent modifications of Tcf/LEFs include acetylation by CBP/p300 (Gay et al. 2003), sumoylation by PIASy (Sachdev et al. 2001) and phosphorylation by CK1 (Hammerlein et al. 2005), CK2 (Miravet et al. 2002) as well as the Nemo-like kinases, which decrease the DNA-binding affinity of the Tcf/LEF/β-cat complex (Ishitani et al. 2003; Lo et al. 2004). These posttranslational modifications can influence

the DNA-binding properties of Tcf/LEFs, their interaction with other partners, the nucleocytoplasmic shuttling or subnuclear localization, and while in most cases they decrease Wnt target gene expression, such modifications may also lead to increased activation. The potential intersection points between Wnt and other major developmental pathways in the nucleus that were discussed above are schematically illustrated in Fig. 1.

Wnt-signaling and patterning of the developing neural tube

Neural stem cells are considered the cellular building clocks of the developing nervous system and are characterized by their ability to undergo an extensive number of self-renewing divisions and to give rise to neurons or glia. Neural stem cells give rise to a great variety of neuronal subtypes along the entire neuraxis. Wnts and Wnt inhibitors are involved in generating this neuronal diversity by contributing to regional patterning of the neural tube in early embryogenesis. During Drosophila embryogenesis, Wnt controls the organization of neuroblasts along the anterior/posterior (A/P) axis (Kaphingst and Kunes 1994; Song et al. 2000). In Xenopus, manipulation of the canonical pathway interferes with the A/P character of neuralized animal caps (McGrew et al. 1995; Kiecker and Niehrs 2001). Moreover, application of Wnts on rostral neural plate explants or on chick embryos inhibit ventral cell fate and induce an early dorsal character in telencephalic cells (Gunhaga et al. 2003). Moreover, loss of β-catenin in the telencephalon before the onset of neurogenesis leads to the downregulation of dorsal markers and the ectopic expression of ventral markers (Backman et al. 2005). Wnt1 and Wnt3a are components of roof plate signaling (Hollyday et al. 1995; Megason and McMahon 2002), and analysis of double knockout mice revealed severe defects in dorsal interneuron specification in the developing spinal cord (Muroyama et al. 2002). Finally, an elegant study showed recently that Wnt signaling may regulate fine scale cell fate choices along the A/P axis, in part by creating an adhesion gradient, thereby contributing to the fine-scale architecture of the nervous system (Hayden et al. 2007). For further detailed discussion of the role of

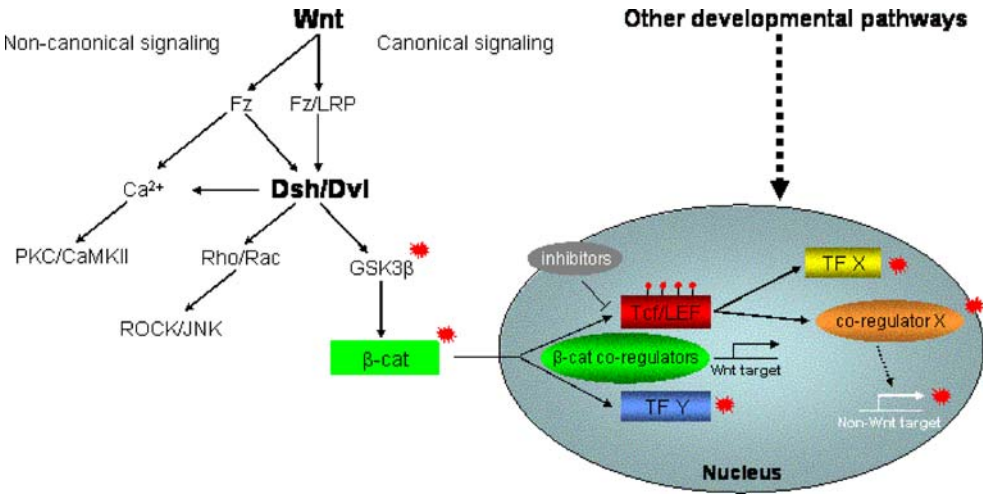

Fig. 1 Schematic representation of the potential intersection points between Wnt and other developmental pathways in the nucleus: Following the initial events that take place at the cell membrane (determined by the Wnt class, the availailable receptor/co-receptor complexes and the potential antagonists), the signal is first transmitted to Dsh/Dvl from which it is routed to different downstream effectors, resulting in the activation of non-canonical or canonical pathways. The hallmark of the canonical pathway is the nuclear translocation of β-catenin, which is also amenable to regulation by other signaling pathways (e.g., PI3K). In the nucleus, β-catenin exerts its regulatory effects on target promoters, which are recognized by Tcf/LEF (illustrated in red) or unrelated DNA-binding transcription factors (in blue; e.g., proteins of the Sox family or Prop1). On these promoters β-catenin acts by displacing available repressors (e.g., Groucho/TLE) and recruiting transcriptional activators (e.g., Pygopus) or variable chromatin remodeling complexes (in green). In the case of the Tcf/LEF proteins, the activation by β-catenin can be inhibited by other proteins such as ICAT or Chibby (in grey). On the other hand, Tcf/LEFs can also associate with coregulatory molecules that are not dependent on β-catenin (in orange; e.g., SHARP) and they may cooperate with other DNA-binding transcription factors (in yellow; e.g., Smads, Alx4, Pitx2 or Cdx1) on composite target promoters. Such β-catenin-independent coregulators and unrelated transcription factors can be regulated by different developmental pathways (e.g., EGF, Notch, TGFβ). Modulatory molecules that can sense the metabolic state of the cells such as CtBP may act as common regulators of Wnt target genes (in black) as well as of genes that are under the control of other developmental stimuli such as BMP or Notch (in white), thus allowing the coordination of their activities. Finally, covalent modifications of Tcf/LEFs such as phosphorylation, acetylation or sumoylation can influence their DNA-binding properties as well as the mode and place of interaction with their diverse partners. The potential points of intersection with other developmental pathways summarized here are illustrated with an *asterisk*. See text for details and references

Wnts in patterning of the neural tube, we would like to refer the reader to the excellent reviews by Ciani and Salinas (Ciani and Salinas 2005) and Chizhikov and Millen (Chizhikov and Millen 2005).

Wnt signaling and proliferation of neural stem cells

Tight regulation of the proliferation of neural stem and precursor cells is central for the control of the size of different CNS regions. Analysis of knockout mice has revealed a prominent function for Wnt proteins in the proliferation of neural precursor cells. During development, Wnt1 is expressed in the caudal midbrain. In the absence of Wnt1, neural precursor populations in the developing mid/hindbrain fail to expand, leading to an almost complete loss of the mid/hindbrain region (McMahon and Bradley 1990; Thomas and Capecchi 1990). Conversely, proliferation of isolated ventral midbrain precursors is stimulated by Wnt1 (Castelo-Branco et al. 2003), and increased expression of Wnt1 in the developing mid/hindbrain enhances proliferation of local precursors potentially through induction of the positive cell cycle regulator cyclin D1 and shortening of the cell cycle (Panhuysen et al. 2004). Wnt3a mutant mice display strongly reduced proliferation of precursor cells in the caudomedial cortical epithelium, which is the site from which the hippocampus arises. As a consequence, all layers of the hippocampus are greatly reduced (Lee et al. 2000). Similarly, conditional ablation of β-catenin in the developing hippocampus leads to disruption of all hippocampal subfields (Machon et al. 2003). The prominent role of Wnt signaling in the control of hippocampal development is underlined by the phenotype of the LRP6 (Zhou et al. 2004) and the LEF1 mutants (Galceran et al. 2000), which display strong hippocampal malformations. Curiously, these mutations specifically affect the proliferation of precursors to the dentate granule neurons and hence lead to loss of the dentate gyrus, but not of the pyramidal cell layers. In contrast, transgenic mice expressing a fusion gene that interferes with the transcriptional activation of all TCF/LEF family members results in the loss of all hippocampal subfields (Galceran et al. 2000). Taken together these results indicate that Wnt3a controls the proliferation of different hippocampal precursor pools through pathways that are dependent on intact β-catenin signaling, but that are distinct on the level of the receptor complex and the transcriptional complex.

Present work indicates that the proliferative action of Wnt proteins is predominantly mediated by the canonical Wnt/β-catenin pathway. Loss of the Wnt-receptor Frizzled5 in Xenopus leads to decreased proliferation of retinal precursor cells. The Frizzled5 effect can be phenocopied by inhibition of the Wnt/β-catenin pathway, indicating that the Frizzled5 mediates proliferation through the canonical Wnt-signaling pathway (Van Raay et al. 2005). In the caudomedial cortical epithelium Wnt/β-catenin signaling and the transcription factor Emx2 collaborate in a positive feedback loop to control precursor proliferation (Tole et al. 2000; Theil et al. 2002; Muzio et al. 2005). The positive cell cycle regulators CyclinD1 and c-Myc have previously been identified as direct targets of Wnt/β-catenin signaling. Indeed, mis-expression of a constitutively active form of β-catenin in neuroepithelial precursors causes a shift in the balance between cell cycle re-entrance and cell cycle exit and leads to expansion of the cortical precursor pool and enlarged cerebral cortical size at birth (Chenn and Walsh 2002). Conversely, loss of β-catenin in cortical precursors (Woodhead et al. 2006) as well as loss of the co-receptor LRP6 (Zhou et al. 2006), which leads to reduced Wnt/β-catenin signaling, result in the premature differentiation of cortical precursors and cortical hypoplasia.

Previous studies have found that Wnt signaling and PI3 kinase signaling converge on the stabilization of β-catenin to control the proliferation of stem cells in the intestine (He et al. 2004), whereas the self-renewal of hematopoietic stem cells is controlled by combinatorial Wnt and Notch signaling (Duncan et al. 2005). The proliferative effect of β-catenin on neural precursors may also be dependent on the interaction of Wnt signaling with other pathways. In the developing spinal cord BMP-signaling inhibits Wnt-induced proliferation of dorsal precursors (Ille et al. 2007). In the presence of FGF2, cortical neural precursor cells show increased proliferation in response to Wnt7a/b overexpression (Viti et al. 2003) or activated Wnt/β-catenin signaling (Israsena et al. 2004). In the absence of FGF2, however, cortical precursors will differentiate into neurons following stimulation of the Wnt/β-catenin pathway (Israsena et al. 2004). One potential mechanism through which FGF signaling modulates the biological output of Wnt/β-catenin signaling may be the cooperative activation of target genes, which control neural precursor cell proliferation. The transcription factor Sox2 prevents premature cell cycle exit of neural precursor cells in the developing spinal cord (Graham et al. 2003) and appears to be essential for proliferation of retinal precursor cells (Van Raay et al. 2005). Interestingly, Wnt and FGF signals converge to activate the expression of the transcription factor Sox2 in the neural plate of the chick (Takemoto et al. 2006). Curiously, it has been shown in other organ systems that activation of FGF signaling can increase the expression of Sox proteins (Mansukhani et al. 2005), which block the activation of canonical Wnt-signaling targets through sequestration of β-catenin (Zorn et al. 1999; Mansukhani et al. 2005) and may divert the transactivation potential of β-catenin towards different sets of downstream targets (Sinner et al. 2004). Such observations indicate that

context-dependent synergistic and inhibitory interactions among the relevant downstream components may control the biological output of Wnt and FGF signaling.

Wnt/β-catenin signaling can also control the competence of precursor cells to respond to proliferative signals. Perturbation of Wnt/β-catenin signaling in cardiac neural crest precursors leads to severe reduction of precursor proliferation and abnormalities in the cardiac outflow tract. Further analysis revealed that Wnt/β-catenin signaling induces the expression of Pitx2, a transcription factor which is specifically expressed in precursor cells of the cardiac outflow tract and of the pituitary gland. The expression of Pitx2 in turn confers the competence to precursor cells to respond to growth factor signaling with the expression of the positive cell cycle regulator cyclin D2 (Kioussi et al. 2002). An additional level of complexity is added to the Wnt/β-catenin controlled proliferation program by the ability of the Wnt/β-catenin pathway to promote proliferation through increasing the mRNA stability of cyclin D1 and cyclin D2 (Briata et al. 2003).

Wnt signaling in the maintenance of the neural precursors and the control of neural cell fate decisions

Proliferation of neural precursor cells appears to be tightly linked with maintenance of the neural precursor state. This association is illustrated best by the observations that overexpression of the negative cell cycle regulator $p27^{Kip1}$ enhances premature neuronal differentiation (Tarui et al. 2005) and that the fate of neural precursor cells, i.e., maintenance in a precursor state vs. neuronal fate commitment can be predicted from cell cycle length (Calegari et al. 2005). Therefore modulation of the expression of cell cycle regulators and of competence factors to respond to growth signaling pathways may be an eminent mechanism for maintenance of the neural precursor pool. However, observations in other organ systems strongly indicate that Wnt signaling controls stem cell maintenance also through molecular mechanisms which are independent of proliferative pathways. It has been demonstrated that β-catenin signaling is essential for the maintenance of quiescent or slowly dividing stem cells in the hair follicle (Lowry et al. 2005) and that Wnt/β-catenin signaling sustains the expression of pluripotency factors such as Oct-3/4 and Nanog in embryonic stem cells (Sato et al. 2004). In the neural crest, combinatorial Wnt and BMP signaling leads to maintenance of neural crest stem cells, whereas Wnt signaling and BMP signaling alone will induce the differentiation of stem cells into sensory ganglion cells and autonomic neurons, respectively (Kleber et al. 2005).

It has been shown that Wnt/β-catenin signaling positively controls the expression of the transcription factor neuron restrictive silencer factor/repressor element 1 transcription factor (NRSF/REST) in spinal cord progenitor cells of the developing chick (Nishihara et al. 2003). NRSF/REST is a transcription factor which negatively regulates the expression of neuronal genes through binding to a neuron restrictive silencer element (NRSE) in the promoter of neuronal genes and the recruitment of transcriptional co-repressors (Ballas and Mandel 2005). Expression of a dominant-negative NRSF/REST induces precocious differentiation of neural progenitor cells (Chen et al. 1998). Moreover, conversion of NRSF/REST in adult neural stem cells from a transcriptional repressor into a transcriptional activator through a non-coding RNA-dependent mechanism promotes neuronal differentiation (Kuwabara et al. 2004). These observations indicate that NRSF/REST may be essential for neural stem cell maintenance, and it will be interesting to examine whether the NRSF/REST activity is coordinated by Wnt/β-catenin signaling, potentially in collaboration with other signaling pathways, to control neural stem cell maintenance.

A role for Wnt/β-catenin signaling in neuronal fate determination in vivo was conclusively demonstrated for the first time in the development of neural crest derivatives. Expression of a stabilized form of β-catenin in neural crest stem cells results in increased generation of sensory neurons (Lee et al. 2004). Importantly, sensory neurogenesis in these transgenic mice occurs at the expense of other neural crest-derived lineages arguing that neural crest stem cells were induced to adopt a sensory fate.

Wnt1-induced signaling is crucial at early stages for the specification of midbrain dopaminergic neuronal precursors and for the terminal differentiation of these precursor at later developmental stages (Prakash et al. 2006). In vitro work has suggested that the terminal differentiation of the midbrain dopaminergic precursors may also involve Wnt5a-induced non-canonical signaling pathways (Castelo-Branco et al. 2006) (although the relevance of Wnt5a for dopaminergic neurogenesis in vivo awaits further confirmation). The interplay of canonical and non-canonical signaling pathways in the control of fate specification and terminal differentiation appears to be a recurring theme in stem cell biology and development. For example, stimulation of non-canonical Wnt-signaling and inhibition of Wnt/β-catenin signaling are required for the specification of ventral cell fates in developing vertebrates (Weidinger and Moon 2003; Westfall et al. 2003) and for the differentiation of chondrocyte precursors (Topol et al. 2003). Neural precursors in the developing mouse cortex on embryonic day 13.5 (E13.5) differentiate into neurons following activation of Wnt/β-catenin signaling. In these precursors Wnt/β-catenin directly activates the expression of the pro-neurogenic basic helix-loop-helix transcription factor Neurogenin1 (Hirabayashi et al. 2004), which in turn

initiates a neurogenic program. Interestingly, early cortical precursors (E10.5) show increased proliferation following stimulation of the canonical Wnt-signaling pathway in vitro and in vivo, demonstrating that the biological response of cortical precursor cells to Wnt/β-catenin signaling is stage dependent (Hirabayashi et al. 2004; Hirabayashi and Gotoh 2005). As previously discussed, it is possible that the activity of interacting pathways differs at distinct stages and determines the response of the precursor cell. The observation that isolated cortical precursors respond under the same culturing conditions with proliferation or differentiation to increased Wnt/β-catenin signaling depending on their developmental age strongly argues that extracellular cues are capable to generate steady epigenetic alterations in the nucleus of the cell precursors, thereby allowing the expression of the stage-dependent effects of Wnt/β-catenin signaling. The molecular nature of these cell intrinsic changes is presently unknown. Some clues for potential cell intrinsic changes can be deduced by the following observations. The response of neural precursor cells to signals which induce astrocytic fate determination is stage dependent (Takizawa et al. 2001; Fan et al. 2005; He et al. 2005). In vivo, neural precursors give rise to neurons at early stages and will only later on generate astrocytes. In vitro, LIF signaling induces astrocytic fate commitment only in late neural precursors, but not in early neural precursors. The failure of early neural precursors to differentiate into astrocytes appears to be the consequence of stage-specific chromatin modifications of astrocytic promoters, which inhibit the expression of astrocyte-specific genes even in the presence of strong astrocytic-fate inducers. Whether chromatin-modifications in downstream targets of the Wnt-signaling cascade are involved in the regulation of the stage-specific response of neural precursor cells to activated β-catenin signaling remains to be determined.

The outcome of Wnt/β-catenin signaling in stem cells may also be dependent on the type of transcriptional co-activator, which is recruited to the TCF/LEF/β-catenin transcriptional complex. Using a small molecule driven approach, it was shown that the interaction of TCF/LEF/β-catenin with the transcriptional co-activator p300 increases the differentiation of embryonic stem cells (Miyabayashi et al. 2007) and PC12 cells (Teo et al. 2005), while the interaction of TCF/LEF/β-catenin with the transcriptional co-activator CBP promoted the undifferentiated state of these cells (Miyabayashi et al. 2007).

Cell fate decisions of neural precursors may also involve the interaction of stabilized β-catenin with transcription factors other than those of the TCF/LEF family. The generation of several hormone-producing cell types in the pituitary gland is dependent on the presence of the transcription factor Pit1. The expression of Pit1 is positively controlled by the transcription factor Prophet of Pit1 (Prop1) and negatively regulated by the transcription factor Hesx1. Loss of β-catenin in early pituitary precursors leads to a loss of Pit1 expression and loss of Pit1-dependent cell-lineages. Interestingly, the expression pattern of TCF transcription factors and the phenotype of the LEF1 mutant mice indicate that β-catenin is not interacting with TCF/LEF transcription factors in the control of Pit1 expression and establishment of Pit1-dependent cell lineages. Instead, it was found that β-catenin interacts with Prop1 to simultaneously activate the expression of Pit1 on the one hand and to repress the expression of Hesx1 on the other hand, which together results in the strong activation of Pit1 and the establishment of Pit1-dependent cell lineages (Olson et al. 2006). Thus, the coordinated transcriptional switch of two master transcription regulators allows Wnt to provide a key signal for cell fate determination during pituitary development via the synchronized activation and repression of specific gene targets. Considering that in addition to Tcf/LEFs or Prop1, β-catenin can interact with other DNA-binding transcription factors (Kioussi et al. 2002) that might also be expressed in a tissue- and/or developmental stage-specific manner, it is tempting to speculate that an analogous mechanism may contribute on the precise control of Wnt-determined cell fate decisions and the generation of neuronal subtype diversity. Based on the experimental evidence mentioned above, the potential mechanisms, which may determine the final output of Wnt/β-catenin signaling considering (1) the link between stem cell maintenance and differentiation and (2) the interplay among Wnt and other major physiologically relevant extracellular cues, are schematically exemplified in Fig. 2. This paradigm may explain the competence of neural precursors to respond to Wnt with increased proliferation at early stages and to initiate a full neurogenic program later on.

Wnt signaling in the establishment of neuronal circuitries

Wnts also play a role in neurogenesis beyond the stage of neuronal fate determination of neural stem cells. Wnts participate in the establishment and refinement of neuronal circuits by controlling axon guidance, dendritic development, synapse formation and synaptic plasticity. Intriguingly, a number of these events are controlled by β-catenin independent pathways and involve a different set of co-receptors. Wnt7a mutant mice (Hall et al. 2000) display defects in the synapses of mossy fibers on cerebellar granule cells. Further analysis demonstrated that granule cells secrete Wnt7a, which stimulates axon and growth cone remodeling in mossy fibers. In this process, Wnt7a activates a pathway,

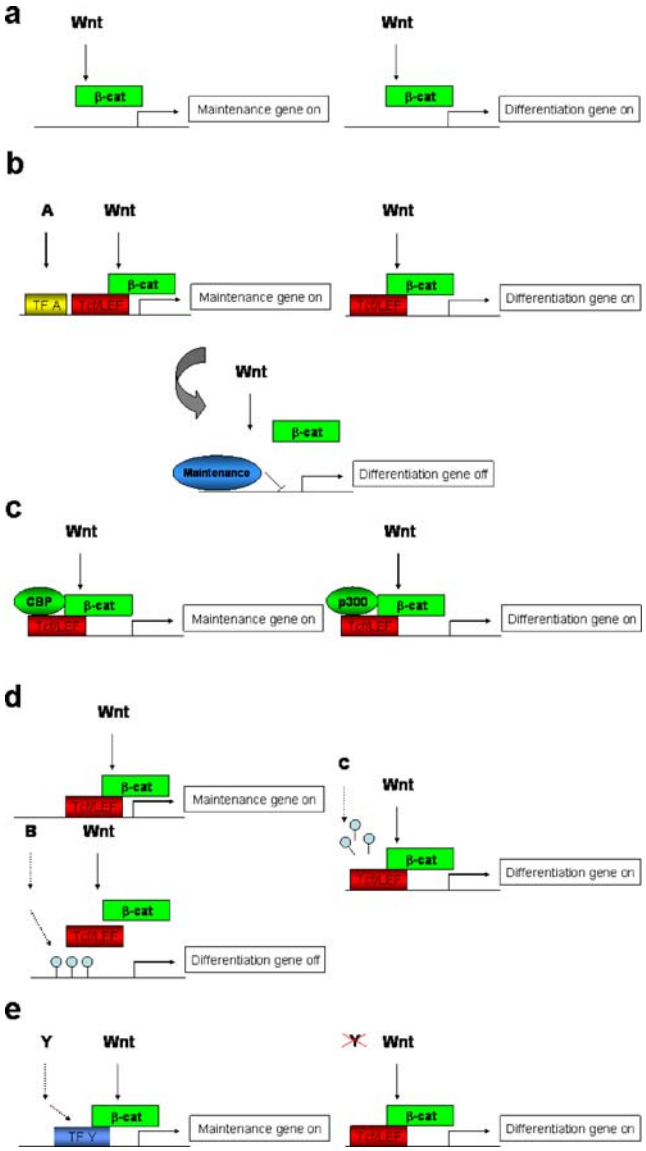

Fig. 2 Schematic representation of potential mechanisms leading to differential output of Wnt/β-catenin signaling using stem cell maintenance and differentiation as a model. **a** The expression of both maintenance and differentiation genes can require Wnt/β-catenin signaling. **b** The expression of maintenance genes is dependent on the cooperation of Wnt/β-catenin signaling with a second signal (A), which employes a second transcriptional activator (TF A) that also binds to the Wnt responsive promoter. Maintenance genes will repress the expression of differentiation genes by making their transcriptional regulatory region non-accessible to β-catenin recruitment. In the absence of A, repression will be relieved and Wnt/β-catenin signaling will predominantly lead to the expression of differentiation genes. **c** The expression of maintenance genes and differentiation genes can be differentially controlled by the selective recruitment of a specific co-activator. **d** Tcf/LEF cannot bind to the promoter of differentiation genes because of epigenetic modifications, which may potentially be induced by extrinsic signal B. In the presence of signal C the promoter becomes accessible to Tcf/LEF leading to the expression of differentiation genes. **e** Transcription factor Y (TF Y) is activated by an extrinsic signal Y. Stabilized β-catenin cooperates with TF Y to promote the expression of maintenance genes. In the absence of Y, stabilized β-catenin will associate with Tcf/LEF leading to the expression of differentiation genes

which diverges from the canonical signaling pathway at the level of GSK3β to change the phosphorylation status of the microtubule associated protein MAP1B, which in turn leads to modulation of cytoskeleton dynamics. A similar role as a target derived factor to control axonal remodeling and axonal branching has been proposed for Wnt3 in the formation of sensory-motoneuron connections in the developing spinal cord (Krylova et al. 2002).

Wnt signaling also appears to control axon guidance of long projection neurons along the rostro-caudal axis in the spinal cord. Frizzled 3 knockouts show guidance defects of sensory commissural axons in the spinal cord, which fail to project rostrally following crossing of the midline. Further analysis showed that Wnt4 is expressed in the ventral neural tube in an anterior-posterior gradient and that Wnt4 attracts commissural axons (Lyuksyutova et al. 2003). Wnt signaling also regulates the caudally directed outgrowth of the corticospinal tract. In this system, Wnts have a chemo-repulsive function. Interestingly, the repulsive activity can be blocked by antibodies to the Wnt coreceptor Ryk (Liu et al. 2005). Although in vitro assays have suggested that Ryk may be essential for Tcf/β-catenin activation, in vivo knockdown of Ryk using siRNA technology does not phenocopy Tcf/LEF or β-catenin mutants and only leads to defects in axon guidance and neurite outgrowth (Lu et al. 2004), suggesting that Ryk-dependent Wnt signaling is only involved in very specific developmental processes.

Wnt7b has been shown to increase dendritic arborization of hippocampal neurons in a GSK3β and β-catenin independent manner, indicating that dendritogenesis involves a non-canonical signaling pathway. Indeed, it has been demonstrated that Wnt7b activates JNK and that activation of JNK is sufficient to promote dendritic development (Rosso et al. 2005).

Finally, it has also been demonstrated that the expression of Wnt proteins can be enhanced by neuronal activity and that increased Wnt signaling enhance dendritic growth and facilitate long-term potentiation in the hippocampus (Chen et al. 2006; Wayman et al. 2006). Thus, Wnts appear to be activity-regulated signals which participate in the control of synaptic plasticity.

Wnt signaling in adult neurogenesis

Neural stem cells continue to generate new neurons in the adult mammalian brain in the subependymal zone (SEZ) of the lateral ventricles and the subgranular zone (SGZ) of the hippocampal dentate gyrus (Ming and Song 2005). In other regions such as the spinal cord, the striatum, and the substantia nigra neural precursor cells give rise exclusively to the glial lineage (Ming and Song 2005). In vitro analysis, however, showed that neural precursors from such non-

neurogenic regions have the potential to differentiate into neurons and glia on the clonal level (Shihabuddin et al. 2000; Lie et al. 2002). Importantly, neural precursors regardless of their origin differentiate into neurons following transplantation into the neurogenic regions and appear to adopt site-specific neuronal subtype identities; in contrast, neural precursors which have been transplanted into gliogenic regions do not differentiate into neurons, but will only give rise to glia (Suhonen et al. 1996; Shihabuddin et al. 2000; Lie et al. 2002). These results indicate that neural precursor cells in all regions of the adult central nervous system have the potential to give rise to neurons in vivo and that their fate is determined by signals provided by the environment.

Co-culture experiments have identified astrocytes in the SEZ and in the SGZ as a source for signals which promote neurogenesis. SEZ-derived astrocytes increase the formation of large neuroblast colonies from SEZ precursors in vitro (Lim and Alvarez-Buylla 1999). Similarly, hippocampus-derived astrocyte feeder layers promote the generation of neurons from adult hippocampus derived neural stem cells through increasing stem cell proliferation and induction of neuronal fate commitment (Song et al. 2002). Both membrane-bound factors as well as diffusible signals contribute to the neurogenic effect of hippocampal astrocytes. Importantly, spinal cord astrocytes do not promote neurogenesis from adult hippocampal neural stem cells, but rather promote their glial differentiation (Song et al. 2002). Thus, regional differences between astrocytes and astrocyte-derived signals appear to contribute to the spatial restriction of neurogenesis in the adult central nervous system.

Several Wnt family members are expressed in the adult dentate gyrus (Shimogori et al. 2004; Lie et al. 2005). Adult hippocampal neural stem cells express essential components of the Wnt/β-catenin signaling pathway. Moreover, Wnt/β-catenin signaling is active in proliferating cells and in a subset of newborn neurons. Interestingly, hippocampal astrocytes express at least one Wnt-family member, i.e., Wnt3, and activate Wnt/β-catenin signaling in co-cultured adult neural stem cells. Inhibition of Wnt-signaling by sFRP reduces neurogenesis in hippocampal astrocyte/neural stem cell co-cultures. Similarly, expression of a dominant negative mutant of LEF1 in adult neural stem cells significantly reduces the neurogenic effect of hippocampal astrocytes on adult neural stem cells, demonstrating that astrocyte-derived Wnts promote neurogenesis through stimulation of the Wnt/β-catenin signaling pathway (Lie et al. 2005).

Further in vitro analysis revealed that Wnt3-induced signaling is sufficient to stimulate the neuronal fate commitment of adult neural stem cells as well as the subsequent expansion of neuronally committed precursors. Overexpression of Wnt3 in the dentate gyrus of adult rats results in a two-fold increase in the number of newly generated immature neurons. In contrast, inhibition of Wnt signaling in the adult dentate gyrus abolishes the generation of new neurons almost completely, indicating that Wnt signaling is essential for adult hippocampal neurogenesis (Lie et al. 2005).

The function of Wnt signaling in adult hippocampal neurogenesis may not be limited to neuronal fate determination and neuroblast proliferation. Several recent in vitro studies have provided evidence that Wnt signaling can enhance the proliferation of neural stem cells derived from the early postnatal and adult central nervous system (Das et al. 2006; Yu et al. 2006; Hirsch et al. 2007). Moreover, analysis of the activity pattern of the Wnt/β-catenin pathway in the adult brain reveals that the pathway is not only active in the subgranular zone, i.e., the region where neural stem cells and newly generated immature neurons reside, but also strongly active in the granule cell layer (Maretto et al. 2003; Lie et al. 2005), i.e., the region where new neurons integrate into the hippocampal circuit. This activity pattern raises the possibility that Wnt/β-catenin signaling may also be involved in later events of hippocampal neurogenesis such as maturation and integration of newborn neurons.

Neurotransmitter-regulated signaling has been demonstrated to be another potent and physiological regulator of hippocampal neurogenesis (Gould et al. 1994; Cameron et al. 1995; Arvidsson et al. 2001; Tozuka et al. 2005; Wang et al. 2005; Ge et al. 2006; Overstreet-Wadiche et al. 2006; Tashiro et al. 2006). It remains to be determined if and how Wnts are cooperating with neurotransmitters in the control of hippocampal neural stem cell behavior (Jagasia et al. 2006). Complex stimuli such as exposure to an enriched environment, running, learning, and seizures, increase the rate of adult hippocampal neurogenesis [for review see (Ming and Song 2005)]. At present, it is unknown whether Wnt signaling as a central regulatory pathway in adult hippocampal neurogenesis is modulated by such stimuli and mediates the enhancement of hippocampal neurogenesis. However, it is interesting to note that two recent studies reported neuronal activity-induced expression of Wnt proteins in the hippocampus (Chen et al. 2006; Wayman et al. 2006). It will be interesting to investigate in the future whether such activity-dependent modulation of Wnt expression may mediate the neurogenic effect of behavioral stimuli.

Conclusion

In the past few years, fascinating discoveries have revealed an increasingly complex Wnt-signaling network. This remarkable Wnt web can be regulated and modulated at multiple nodes ranging from the level of ligand-receptor interaction to the level of the diversity of downstream transcriptional complexes and of the mRNA as well as protein turnover of Wnt-signaling components. The past few years have also firmly established Wnts as essential

regulators for neural stem cells and neural development. What is less clear is the question of how Wnts achieve a precise control of the multitude of biological decisions with which neural stem cells are confronted during developmental and adult neurogenesis. Incorporating the growing knowledge of the complex Wnt-signaling network into neural stem cell biology will greatly promote our understanding of how Wnts regulate neural stem cell behavior and will ultimately promote the development of novel stem cell based strategies for diseases of the nervous system.

References

Arce L, Yokoyama NN, Waterman ML (2006) Diversity of LEF/TCF action in development and disease. Oncogene 25:7492–7504

Arvidsson A, Kokaia Z, Lindvall O (2001) N-methyl-D-aspartate receptor-mediated increase of neurogenesis in adult rat dentate gyrus following stroke. Eur J Neurosci 14:10–18

Atcha FA, Munguia JE, Li TW, Hovanes K, Waterman ML (2003) A new beta-catenin-dependent activation domain in T cell factor. J Biol Chem 278:16169–16175

Backman M, Machon O, Mygland L, van den Bout CJ, Zhong W, Taketo MM, Krauss S (2005) Effects of canonical Wnt signaling on dorso-ventral specification of the mouse telencephalon. Dev Biol 279:155–168

Ballas N, Mandel G (2005) The many faces of REST oversee epigenetic programming of neuronal genes. Curr Opin Neurobiol 15:500–506

Banziger C, Soldini D, Schutt C, Zipperlen P, Hausmann G, Basler K (2006) Wntless, a conserved membrane protein dedicated to the secretion of Wnt proteins from signaling cells. Cell 125:509–522

Barolo S, Posakony JW (2002) Three habits of highly effective signaling pathways: principles of transcriptional control by developmental cell signaling. Genes Dev 16:1167–1181

Bartscherer K, Pelte N, Ingelfinger D, Boutros M (2006) Secretion of Wnt ligands requires Evi, a conserved transmembrane protein. Cell 125:523–533

Bejsovec A (2005) Wnt pathway activation: new relations and locations. Cell 120:11–14

Beland M, Pilon N, Houle M, Oh K, Sylvestre JR, Prinos P, Lohnes D (2004) Cdx1 autoregulation is governed by a novel Cdx1-LEF1 transcription complex. Mol Cell Biol 24:5028–5038

Bienz M (2002) The subcellular destinations of APC proteins. Nat Rev Mol Cell Biol 3:328–338

Boras K, Hamel PA (2002) Alx4 binding to LEF-1 regulates N-CAM promoter activity. J Biol Chem 277:1120–1127

Brembeck FH, Schwarz-Romond T, Bakkers J, Wilhelm S, Hammerschmidt M, Birchmeier W (2004) Essential role of BCL9-2 in the switch between beta-catenin's adhesive and transcriptional functions. Genes Dev 18:2225–2230

Briata P, Ilengo C, Corte G, Moroni C, Rosenfeld MG, Chen CY, Gherzi R (2003) The Wnt/beta-catenin–>Pitx2 pathway controls the turnover of Pitx2 and other unstable mRNAs. Mol Cell 12:1201–1211

Brott BK, Sokol SY (2005) Frodo proteins: modulators of Wnt signaling in vertebrate development. Differentiation 73:323–329

Calegari F, Haubensak W, Haffner C, Huttner WB (2005) Selective lengthening of the cell cycle in the neurogenic subpopulation of neural progenitor cells during mouse brain development. J Neurosci 25:6533–6538

Cameron HA, McEwen BS, Gould E (1995) Regulation of adult neurogenesis by excitatory input and NMDA receptor activation in the dentate gyrus. J Neurosci 15:4687–4692

Castelo-Branco G, Wagner J, Rodriguez FJ, Kele J, Sousa K, Rawal N, Pasolli HA, Fuchs E, Kitajewski J, Arenas E (2003) Differential regulation of midbrain dopaminergic neuron development by Wnt-1, Wnt-3a, and Wnt-5a. Proc Natl Acad Sci USA 100:12747–12752

Castelo-Branco G, Sousa KM, Bryja V, Pinto L, Wagner J, Arenas E (2006) Ventral midbrain glia express region-specific transcription factors and regulate dopaminergic neurogenesis through Wnt-5a secretion. Mol Cell Neurosci 31:251–262

Chen F, Rebay I (2000) split ends, a new component of the Drosophila EGF receptor pathway, regulates development of midline glial cells. Curr Biol 10:943–946

Chen ZF, Paquette AJ, Anderson DJ (1998) NRSF/REST is required in vivo for repression of multiple neuronal target genes during embryogenesis. Nat Genet 20:136–142

Chen J, Park CS, Tang SJ (2006) Activity-dependent synaptic WNT release regulates hippocampal long-term potentiation. J Biol Chem

Chenn A, Walsh CA (2002) Regulation of cerebral cortical size by control of cell cycle exit in neural precursors. Science 297:365–369

Cheyette BN (2004) Ryk: another heretical Wnt receptor defies the canon. Sci STKE 2004:pe54

Chinnadurai G (2002) CtBP, an unconventional transcriptional corepressor in development and oncogenesis. Mol Cell 9:213–224

Chizhikov VV, Millen KJ (2005) Roof plate-dependent patterning of the vertebrate dorsal central nervous system. Dev Biol 277:287–295

Ciani L, Salinas PC (2005) WNTs in the vertebrate nervous system: from patterning to neuronal connectivity. Nat Rev Neurosci 6:351–362

Clevers H (2006) Wnt/beta-catenin signaling in development and disease. Cell 127:469–480

Coudreuse D, Korswagen HC (2007) The making of Wnt: new insights into Wnt maturation, sorting and secretion. Development 134:3–12

Coudreuse DY, Roel G, Betist MC, Destree O, Korswagen HC (2006) Wnt gradient formation requires retromer function in Wnt-producing cells. Science 312:921–924

Daniels DL, Weis WI (2005) Beta-catenin directly displaces Groucho/TLE repressors from Tcf/Lef in Wnt-mediated transcription activation. Nat Struct Mol Biol 12:364–371

Das AV, Zhao X, James J, Kim M, Cowan KH, Ahmad I (2006) Neural stem cells in the adult ciliary epithelium express GFAP and are regulated by Wnt signaling. Biochem Biophys Res Commun 339:708–716

DasGupta R, Fuchs E (1999) Multiple roles for activated LEF/TCF transcription complexes during hair follicle development and differentiation. Development 126:4557–4568

Davidson G, Wu W, Shen J, Bilic J, Fenger U, Stannek P, Glinka A, Niehrs C (2005) Casein kinase 1 gamma couples Wnt receptor activation to cytoplasmic signal transduction. Nature 438:867–872

Dorsky RI, Sheldahl LC, Moon RT (2002) A transgenic Lef1/beta-catenin-dependent reporter is expressed in spatially restricted domains throughout zebrafish development. Dev Biol 241:229–237

Duncan AW, Rattis FM, DiMascio LN, Congdon KL, Pazianos G, Zhao C, Yoon K, Cook JM, Willert K, Gaiano N, Reya T (2005) Integration of Notch and Wnt signaling in hematopoietic stem cell maintenance. Nat Immunol 6:314–322

Fan G, Martinowich K, Chin MH, He F, Fouse SD, Hutnick L, Hattori D, Ge W, Shen Y, Wu H, ten Hoeve J, Shuai K, Sun YE (2005) DNA methylation controls the timing of astrogliogenesis through regulation of JAK-STAT signaling. Development 132:3345–3356

Fang M, Li J, Blauwkamp T, Bhambhani C, Campbell N, Cadigan KM (2006) C-terminal-binding protein directly activates and represses Wnt transcriptional targets in Drosophila. EMBO J 25:2735–2745

Feng Y, Bommer GT, Zhai Y, Akyol A, Hinoi T, Winer I, Lin HV, Cadigan KM, Cho KR, Fearon ER (2007) Drosophila split ends homologue SHARP functions as a positive regulator of Wnt/beta-catenin/T-cell factor signaling in neoplastic transformation. Cancer Res 67:482–491

Ferkey DM, Kimelman D (2002) Glycogen synthase kinase-3 beta mutagenesis identifies a common binding domain for GBP and Axin. J Biol Chem 277:16147–16152

Finch PW, He X, Kelley MJ, Uren A, Schaudies RP, Popescu NC, Rudikoff S, Aaronson SA, Varmus HE, Rubin JS (1997) Purification and molecular cloning of a secreted, Frizzled-related antagonist of Wnt action. Proc Natl Acad Sci USA 94:6770–6775

Galceran J, Farinas I, Depew MJ, Clevers H, Grosschedl R (1999) Wnt3a-/-like phenotype and limb deficiency in Lef1(−/−)Tcf1(−/−) mice. Genes Dev 13:709–717

Galceran J, Miyashita-Lin EM, Devaney E, Rubenstein JL, Grosschedl R (2000) Hippocampus development and generation of dentate gyrus granule cells is regulated by LEF1. Development 127: 469–482

Gay F, Calvo D, Lo MC, Ceron J, Maduro M, Lin R, Shi Y (2003) Acetylation regulates subcellular localization of the Wnt signaling nuclear effector POP-1. Genes Dev 17:717–722

Ge S, Goh EL, Sailor KA, Kitabatake Y, Ming GL, Song H (2006) GABA regulates synaptic integration of newly generated neurons in the adult brain. Nature 439:589–593

Ghogomu SM, van Venrooy S, Ritthaler M, Wedlich D, Gradl D (2006) HIC-5 is a novel repressor of lymphoid enhancer factor/T-cell factor-driven transcription. J Biol Chem 281:1755–1764

Glinka A, Wu W, Delius H, Monaghan AP, Blumenstock C, Niehrs C (1998) Dickkopf-1 is a member of a new family of secreted proteins and functions in head induction. Nature 391:357–362

Gould E, Cameron HA, McEwen BS (1994) Blockade of NMDA receptors increases cell death and birth in the developing rat dentate gyrus. J Comp Neurol 340:551–565

Gradl D, Konig A, Wedlich D (2002) Functional diversity of Xenopus lymphoid enhancer factor/T-cell factor transcription factors relies on combinations of activating and repressing elements. J Biol Chem 277:14159–14171

Graham V, Khudyakov J, Ellis P, Pevny L (2003) SOX2 functions to maintain neural progenitor identity. Neuron 39:749–765

Gunhaga L, Marklund M, Sjodal M, Hsieh JC, Jessell TM, Edlund T (2003) Specification of dorsal telencephalic character by sequential Wnt and FGF signaling. Nat Neurosci 6:701–707

Habas R, Dawid IB (2005) Dishevelled and Wnt signaling: is the nucleus the final frontier? J Biol 4:2

Habas R, Dawid IB, He X (2003) Coactivation of Rac and Rho by Wnt/Frizzled signaling is required for vertebrate gastrulation. Genes Dev 17:295–309

Hall AC, Lucas FR, Salinas PC (2000) Axonal remodeling and synaptic differentiation in the cerebellum is regulated by WNT-7a signaling. Cell 100:525–535

Hamada F, Bienz M (2004) The APC tumor suppressor binds to C-terminal binding protein to divert nuclear beta-catenin from TCF. Dev Cell 7:677–685

Hammerlein A, Weiske J, Huber O (2005) A second protein kinase CK1-mediated step negatively regulates Wnt signalling by disrupting the lymphocyte enhancer factor-1/beta-catenin complex. Cell Mol Life Sci 62:606–618

Hay E, Faucheu C, Suc-Royer I, Touitou R, Stiot V, Vayssiere B, Baron R, Roman-Roman S, Rawadi G (2005) Interaction between LRP5 and Frat1 mediates the activation of the Wnt canonical pathway. J Biol Chem 280:13616–13623

Hayden MA, Akong K, Peifer M (2007) Novel roles for APC family members and Wingless/Wnt signaling during Drosophila brain development. Dev Biol 305:358–376

He F, Ge W, Martinowich K, Becker-Catania S, Coskun V, Zhu W, Wu H, Castro D, Guillemot F, Fan G, de Vellis J, Sun YE (2005) A positive autoregulatory loop of Jak-STAT signaling controls the onset of astrogliogenesis. Nat Neurosci 8:616–625

He X, Axelrod JD (2006) A WNTer wonderland in Snowbird. Development 133:2597–2603

He XC, Zhang J, Tong WG, Tawfik O, Ross J, Scoville DH, Tian Q, Zeng X, He X, Wiedemann LM, Mishina Y, Li L (2004) BMP signaling inhibits intestinal stem cell self-renewal through suppression of Wnt-beta-catenin signaling. Nat Genet 36:1117–1121

Hecht A, Stemmler MP (2003) Identification of a promoter-specific transcriptional activation domain at the C terminus of the Wnt effector protein T-cell factor 4. J Biol Chem 278:3776–3785

Hikasa H, Sokol SY (2004) The involvement of Frodo in TCF-dependent signaling and neural tissue development. Development 131:4725–4734

Hildebrand JD, Soriano P (2002) Overlapping and unique roles for C-terminal binding protein 1 (CtBP1) and CtBP2 during mouse development. Mol Cell Biol 22:5296–5307

Hirabayashi Y, Gotoh Y (2005) Stage-dependent fate determination of neural precursor cells in mouse forebrain. Neurosci Res 51:331–336

Hirabayashi Y, Itoh Y, Tabata H, Nakajima K, Akiyama T, Masuyama N, Gotoh Y (2004) The Wnt/beta-catenin pathway directs neuronal differentiation of cortical neural precursor cells. Development 131:2791–2801

Hiriart E, Gruffat H, Buisson M, Mikaelian I, Keppler S, Meresse P, Mercher T, Bernard OA, Sergeant A, Manet E (2005) Interaction of the Epstein-Barr virus mRNA export factor EB2 with human Spen proteins SHARP, OTT1, and a novel member of the family, OTT3, links Spen proteins with splicing regulation and mRNA export. J Biol Chem 280:36935–36945

Hirsch C, Campano LM, Wohrle S, Hecht A (2007) Canonical Wnt signaling transiently stimulates proliferation and enhances neurogenesis in neonatal neural progenitor cultures. Exp Cell Res 313:572–587

Hoang B, Moos M Jr, Vukicevic S, Luyten FP (1996) Primary structure and tissue distribution of FRZB, a novel protein related to Drosophila frizzled, suggest a role in skeletal morphogenesis. J Biol Chem 271:26131–26137

Hoffmans R, Basler K (2007) BCL9-2 binds Arm/beta-catenin in a Tyr142-independent manner and requires Pygopus for its function in Wg/Wnt signaling. Mech Dev 124:59–67

Hofmann K (2000) A superfamily of membrane-bound O-acyltransferases with implications for wnt signaling. Trends Biochem Sci 25:111–112

Hollyday M, McMahon JA, McMahon AP (1995) Wnt expression patterns in chick embryo nervous system. Mech Dev 52:9–25

Houston DW, Kofron M, Resnik E, Langland R, Destree O, Wylie C, Heasman J (2002) Repression of organizer genes in dorsal and ventral Xenopus cells mediated by maternal XTcf3. Development 129:4015–4025

Hovanes K, Li TW, Munguia JE, Truong T, Milovanovic T, Lawrence Marsh J, Holcombe RF, Waterman ML (2001) Beta-catenin-sensitive isoforms of lymphoid enhancer factor-1 are selectively expressed in colon cancer. Nat Genet 28:53–57

Hsieh JC (2004) Specificity of WNT-receptor interactions. Front Biosci 9:1333–1338

Hsieh JC, Kodjabachian L, Rebbert ML, Rattner A, Smallwood PM, Samos CH, Nusse R, Dawid IB, Nathans J (1999) A new secreted protein that binds to Wnt proteins and inhibits their activities. Nature 398:431–436

Hsieh JC, Lee L, Zhang L, Wefer S, Brown K, DeRossi C, Wines ME, Rosenquist T, Holdener BC (2003) Mesd encodes an LRP5/6 chaperone essential for specification of mouse embryonic polarity. Cell 112:355–367

Hu MC, Rosenblum ND (2005) Smad1, beta-catenin and Tcf4 associate in a molecular complex with the Myc promoter in dysplastic renal tissue and cooperate to control Myc transcription. Development 132:215–225

Hurlstone A, Clevers H (2002) T-cell factors: turn-ons and turn-offs. EMBO J 21:2303–2311

Hussein SM, Duff EK, Sirard C (2003) Smad4 and beta-catenin co-activators functionally interact with lymphoid-enhancing factor to regulate graded expression of Msx2. J Biol Chem 278:48805–48814

Ille F, Atanasoski S, Falk S, Ittner LM, Marki D, Buchmann-Moller S, Wurdak H, Suter U, Taketo MM, Sommer L (2007) Wnt/BMP signal integration regulates the balance between proliferation and differentiation of neuroepithelial cells in the dorsal spinal cord. Dev Biol 304:394–408

Ilyas M (2005) Wnt signalling and the mechanistic basis of tumour development. J Pathol 205:130–144

Ishitani T, Ninomiya-Tsuji J, Matsumoto K (2003) Regulation of lymphoid enhancer factor 1/T-cell factor by mitogen-activated protein kinase-related Nemo-like kinase-dependent phosphorylation in Wnt/beta-catenin signaling. Mol Cell Biol 23:1379–1389

Israsena N, Hu M, Fu W, Kan L, Kessler JA (2004) The presence of FGF2 signaling determines whether beta-catenin exerts effects on proliferation or neuronal differentiation of neural stem cells. Dev Biol 268:220–231

Itoh K, Brott BK, Bae GU, Ratcliffe MJ, Sokol SY (2005) Nuclear localization is required for Dishevelled function in Wnt/beta-catenin signaling. J Biol 4:3

Jagasia R, Song H, Gage FH, Lie DC (2006) New regulators in adult neurogenesis and their potential role for repair. Trends Mol Med 12:400–405

Kaphingst K, Kunes S (1994) Pattern formation in the visual centers of the Drosophila brain: wingless acts via decapentaplegic to specify the dorsoventral axis. Cell 78:437–448

Kawano Y, Kypta R (2003) Secreted antagonists of the Wnt signalling pathway. J Cell Sci 116:2627–2634

Kazanskaya O, Glinka A, del Barco Barrantes I, Stannek P, Niehrs C, Wu W (2004) R-Spondin2 is a secreted activator of Wnt/beta-catenin signaling and is required for Xenopus myogenesis. Dev Cell 7:525–534

Kiecker C, Niehrs C (2001) A morphogen gradient of Wnt/beta-catenin signalling regulates anteroposterior neural patterning in Xenopus. Development 128:4189–4201

Kikuchi A, Yamamoto H, Kishida S (2007) Multiplicity of the interactions of Wnt proteins and their receptors. Cell Signal 19:659–671

Kim KA, Kakitani M, Zhao J, Oshima T, Tang T, Binnerts M, Liu Y, Boyle B, Park E, Emtage P, Funk WD, Tomizuka K (2005) Mitogenic influence of human R-spondin1 on the intestinal epithelium. Science 309:1256–1259

Kioussi C, Briata P, Baek SH, Rose DW, Hamblet NS, Herman T, Ohgi KA, Lin C, Gleiberman A, Wang J, Brault V, Ruiz-Lozano P, Nguyen HD, Kemler R, Glass CK, Wynshaw-Boris A, Rosenfeld MG (2002) Identification of a Wnt/Dvl/beta-Catenin –> Pitx2 pathway mediating cell-type-specific proliferation during development. Cell 111:673–685

Kishida M, Hino S, Michiue T, Yamamoto H, Kishida S, Fukui A, Asashima M, Kikuchi A (2001) Synergistic activation of the Wnt signaling pathway by Dvl and casein kinase Iepsilon. J Biol Chem 276:33147–33155

Kleber M, Lee HY, Wurdak H, Buchstaller J, Riccomagno MM, Ittner LM, Suter U, Epstein DJ, Sommer L (2005) Neural crest stem cell maintenance by combinatorial Wnt and BMP signaling. J Cell Biol

Klein TJ, Mlodzik M (2005) Planar cell polarization: an emerging model points in the right direction. Annu Rev Cell Dev Biol 21:155–176

Kohn AD, Moon RT (2005) Wnt and calcium signaling: beta-catenin-independent pathways. Cell Calcium 38:439–446

Korinek V, Barker N, Moerer P, van Donselaar E, Huls G, Peters PJ, Clevers H (1998) Depletion of epithelial stem-cell compartments in the small intestine of mice lacking Tcf-4. Nat Genet 19:379–383

Krupnik VE, Sharp JD, Jiang C, Robison K, Chickering TW, Amaravadi L, Brown DE, Guyot D, Mays G, Leiby K, Chang B, Duong T, Goodearl AD, Gearing DP, Sokol SY, McCarthy SA (1999) Functional and structural diversity of the human Dickkopf gene family. Gene 238:301–313

Krylova O, Herreros J, Cleverley KE, Ehler E, Henriquez JP, Hughes SM, Salinas PC (2002) WNT-3, expressed by motoneurons, regulates terminal arborization of neurotrophin-3-responsive spinal sensory neurons. Neuron 35:1043–1056

Kuang B, Wu SC, Shin Y, Luo L, Kolodziej P (2000) split ends encodes large nuclear proteins that regulate neuronal cell fate and axon extension in the Drosophila embryo. Development 127:1517–1529

Kuhl M (2004) The WNT/calcium pathway: biochemical mediators, tools and future requirements. Front Biosci 9:967–974

Kuwabara T, Hsieh J, Nakashima K, Taira K, Gage FH (2004) A small modulatory dsRNA specifies the fate of adult neural stem cells. Cell 116:779–793

Labbe E, Letamendia A, Attisano L (2000) Association of Smads with lymphoid enhancer binding factor 1/T cell-specific factor mediates cooperative signaling by the transforming growth factor-beta and wnt pathways. Proc Natl Acad Sci USA 97:8358–8363

Lane ME, Elend M, Heidmann D, Herr A, Marzodko S, Herzig A, Lehner CF (2000) A screen for modifiers of cyclin E function in Drosophila melanogaster identifies Cdk2 mutations, revealing the insignificance of putative phosphorylation sites in Cdk2. Genetics 155:233–244

Lee E, Salic A, Kirschner MW (2001) Physiological regulation of [beta]-catenin stability by Tcf3 and CK1epsilon. J Cell Biol 154:983–993

Lee HY, Kleber M, Hari L, Brault V, Suter U, Taketo MM, Kemler R, Sommer L (2004) Instructive role of Wnt/beta-catenin in sensory fate specification in neural crest stem cells. Science 303:1020–1023

Lee SM, Tole S, Grove E, McMahon AP (2000) A local Wnt-3a signal is required for development of the mammalian hippocampus. Development 127:457–467

Leyns L, Bouwmeester T, Kim SH, Piccolo S, De Robertis EM (1997) Frzb-1 is a secreted antagonist of Wnt signaling expressed in the Spemann organizer. Cell 88:747–756

Li L, Mao J, Sun L, Liu W, Wu D (2002) Second cysteine-rich domain of Dickkopf-2 activates canonical Wnt signaling pathway via LRP-6 independently of dishevelled. J Biol Chem 277:5977–5981

Li L, Yuan H, Weaver CD, Mao J, Farr GH 3rd, Sussman DJ, Jonkers J, Kimelman D, Wu D (1999) Axin and Frat1 interact with dvl and GSK, bridging Dvl to GSK in Wnt-mediated regulation of LEF-1. EMBO J 18:4233–4240

Lie DC, Dziewczapolski G, Willhoite AR, Kaspar BK, Shults CW, Gage FH (2002) The adult substantia nigra contains progenitor cells with neurogenic potential. J Neurosci 22:6639–6649

Lie DC, Colamarino SA, Song HJ, Desire L, Mira H, Consiglio A, Lein ES, Jessberger S, Lansford H, Dearie AR, Gage FH (2005) Wnt signalling regulates adult hippocampal neurogenesis. Nature 437:1370–1375

Lim DA, Alvarez-Buylla A (1999) Interaction between astrocytes and adult subventricular zone precursors stimulates neurogenesis. Proc Natl Acad Sci USA 96:7526–7531

Lin X, Liang YY, Sun B, Liang M, Shi Y, Brunicardi FC, Shi Y, Feng XH (2003) Smad6 recruits transcription corepressor CtBP to repress bone morphogenetic protein-induced transcription. Mol Cell Biol 23:9081–9093

Liu Y, Shi J, Lu CC, Wang ZB, Lyuksyutova AI, Song XJ, Zou Y (2005) Ryk-mediated Wnt repulsion regulates posterior-directed growth of corticospinal tract. Nat Neurosci 8:1151–1159

Lo MC, Gay F, Odom R, Shi Y, Lin R (2004) Phosphorylation by the beta-catenin/MAPK complex promotes 14-3-3-mediated nuclear export of TCF/POP-1 in signal-responsive cells in *C. elegans*. Cell 117:95–106

Logan CY, Nusse R (2004) The Wnt signaling pathway in development and disease. Annu Rev Cell Dev Biol 20:781–810

Lowry WE, Blanpain C, Nowak JA, Guasch G, Lewis L, Fuchs E (2005) Defining the impact of beta-catenin/Tcf transactivation on epithelial stem cells. Genes Dev 19:1596–1611

Lu W, Yamamoto V, Ortega B, Baltimore D (2004) Mammalian Ryk is a Wnt coreceptor required for stimulation of neurite outgrowth. Cell 119:97–108

Lyuksyutova AI, Lu CC, Milanesio N, King LA, Guo N, Wang Y, Nathans J, Tessier-Lavigne M, Zou Y (2003) Anterior-posterior guidance of commissural axons by Wnt-frizzled signaling. Science 302:1984–1988

Machon O, van den Bout CJ, Backman M, Kemler R, Krauss S (2003) Role of beta-catenin in the developing cortical and hippocampal neuroepithelium. Neuroscience 122:129–143

Malbon CC, Wang HY (2006) Dishevelled: a mobile scaffold catalyzing development. Curr Top Dev Biol 72:153–166

Mansukhani A, Ambrosetti D, Holmes G, Cornivelli L, Basilico C (2005) Sox2 induction by FGF and FGFR2 activating mutations inhibits Wnt signaling and osteoblast differentiation. J Cell Biol 168:1065–1076

Mao J, Wang J, Liu B, Pan W, Farr GH 3rd, Flynn C, Yuan H, Takada S, Kimelman D, Li L, Wu D (2001) Low-density lipoprotein receptor-related protein-5 binds to Axin and regulates the canonical Wnt signaling pathway. Mol Cell 7:801–809

Mao B, Wu W, Davidson G, Marhold J, Li M, Mechler BM, Delius H, Hoppe D, Stannek P, Walter C, Glinka A, Niehrs C (2002) Kremen proteins are Dickkopf receptors that regulate Wnt/beta-catenin signalling. Nature 417:664–667

Maretto S, Cordenonsi M, Dupont S, Braghetta P, Broccoli V, Hassan AB, Volpin D, Bressan GM, Piccolo S (2003) Mapping Wnt/beta-catenin signaling during mouse development and in colorectal tumors. Proc Natl Acad Sci USA 100:3299–3304

McGrew LL, Lai CJ, Moon RT (1995) Specification of the anteroposterior neural axis through synergistic interaction of the Wnt signaling cascade with noggin and follistatin. Dev Biol 172:337–342

McMahon AP, Bradley A (1990) The Wnt-1 (int-1) proto-oncogene is required for development of a large region of the mouse brain. Cell 62:1073–1085

Megason SG, McMahon AP (2002) A mitogen gradient of dorsal midline Wnts organizes growth in the CNS. Development 129:2087–2098

Merrill BJ, Gat U, DasGupta R, Fuchs E (2001) Tcf3 and Lef1 regulate lineage differentiation of multipotent stem cells in skin. Genes Dev 15:1688–1705

Merrill BJ, Pasolli HA, Polak L, Rendl M, Garcia-Garcia MJ, Anderson KV, Fuchs E (2004) Tcf3: a transcriptional regulator of axis induction in the early embryo. Development 131:263–274

Mikels AJ, Nusse R (2006) Purified Wnt5a protein activates or inhibits beta-catenin-TCF signaling depending on receptor context. PLoS Biol 4:e115

Ming GL, Song H (2005) Adult neurogenesis in the mammalian central nervous system. Annu Rev Neurosci 28:223–250

Miravet S, Piedra J, Miro F, Itarte E, Garcia de Herreros A, Dunach M (2002) The transcriptional factor Tcf-4 contains different binding sites for beta-catenin and plakoglobin. J Biol Chem 277:1884–1891

Miyabayashi T, Teo JL, Yamamoto M, McMillan M, Nguyen C, Kahn M (2007) Wnt/beta-catenin/CBP signaling maintains long-term murine embryonic stem cell pluripotency. Proc Natl Acad Sci USA 104:5668–5673

Moriguchi T, Kawachi K, Kamakura S, Masuyama N, Yamanaka H, Matsumoto K, Kikuchi A, Nishida E (1999) Distinct domains of mouse dishevelled are responsible for the c-Jun N-terminal kinase/stress-activated protein kinase activation and the axis formation in vertebrates. J Biol Chem 274:30957–30962

Muroyama Y, Fujihara M, Ikeya M, Kondoh H, Takada S (2002) Wnt signaling plays an essential role in neuronal specification of the dorsal spinal cord. Genes Dev 16:548–553

Muzio L, Soria JM, Pannese M, Piccolo S, Mallamaci A (2005) A mutually stimulating loop involving emx2 and canonical wnt signalling specifically promotes expansion of occipital cortex and hippocampus. Cereb Cortex 15:2021–2028

Nagel AC, Krejci A, Tenin G, Bravo-Patino A, Bray S, Maier D, Preiss A (2005) Hairless-mediated repression of notch target genes requires the combined activity of Groucho and CtBP corepressors. Mol Cell Biol 25:10433–10441

Nam JS, Turcotte TJ, Smith PF, Choi S, Yoon JK (2006) Mouse cristin/R-spondin family proteins are novel ligands for the Frizzled 8 and LRP6 receptors and activate beta-catenin-dependent gene expression. J Biol Chem 281:13247–13257

Nishihara S, Tsuda L, Ogura T (2003) The canonical Wnt pathway directly regulates NRSF/REST expression in chick spinal cord. Biochem Biophys Res Commun 311:55–63

Nishita M, Hashimoto MK, Ogata S, Laurent MN, Ueno N, Shibuya H, Cho KW (2000) Interaction between Wnt and TGF-beta signalling pathways during formation of Spemann's organizer. Nature 403:781–785

Nusse R, Varmus HE (1982) Many tumors induced by the mouse mammary tumor virus contain a provirus integrated in the same region of the host genome. Cell 31:99–109

Olson LE, Tollkuhn J, Scafoglio C, Krones A, Zhang J, Ohgi KA, Wu W, Taketo MM, Kemler R, Grosschedl R, Rose D, Li X, Rosenfeld MG (2006) Homeodomain-mediated beta-catenin-dependent switching events dictate cell-lineage determination. Cell 125:593–605

Oswald F, Kostezka U, Astrahantseff K, Bourteele S, Dillinger K, Zechner U, Ludwig L, Wilda M, Hameister H, Knochel W, Liptay S, Schmid RM (2002) SHARP is a novel component of the Notch/RBP-Jkappa signalling pathway. EMBO J 21:5417–5426

Oswald F, Winkler M, Cao Y, Astrahantseff K, Bourteele S, Knochel W, Borggrefe T (2005) RBP-Jkappa/SHARP recruits CtIP/CtBP corepressors to silence Notch target genes. Mol Cell Biol 25:10379–10390

Overstreet-Wadiche LS, Bensen AL, Westbrook GL (2006) Delayed development of adult-generated granule cells in dentate gyrus. J Neurosci 26:2326–2334

Panakova D, Sprong H, Marois E, Thiele C, Eaton S (2005) Lipoprotein particles are required for Hedgehog and Wingless signalling. Nature 435:58–65

Panhuysen M, Vogt Weisenhorn DM, Blanquet V, Brodski C, Heinzmann U, Beisker W, Wurst W (2004) Effects of Wnt1 signaling on proliferation in the developing mid-/hindbrain region. Mol Cell Neurosci 26:101–111

Pereira L, Yi F, Merrill BJ (2006) Repression of Nanog gene transcription by Tcf3 limits embryonic stem cell self-renewal. Mol Cell Biol 26:7479–7491

Piccolo S, Agius E, Leyns L, Bhattacharyya S, Grunz H, Bouwmeester T, De Robertis EM (1999) The head inducer Cerberus is a multifunctional antagonist of Nodal, BMP and Wnt signals. Nature 397:707–710

Pinson KI, Brennan J, Monkley S, Avery BJ, Skarnes WC (2000) An LDL-receptor-related protein mediates Wnt signalling in mice. Nature 407:535–538

Prakash N, Brodski C, Naserke T, Puelles E, Gogoi R, Hall A, Panhuysen M, Echevarria D, Sussel L, Weisenhorn DM, Martinez S, Arenas E, Simeone A, Wurst W (2006) A Wnt1-regulated genetic network controls the identity and fate of midbrain-dopaminergic progenitors in vivo. Development 133:89–98

Prasad BC, Clark SG (2006) Wnt signaling establishes anteroposterior neuronal polarity and requires retromer in *C. elegans*. Development 133:1757–1766

Riese J, Yu X, Munnerlyn A, Eresh S, Hsu SC, Grosschedl R, Bienz M (1997) LEF-1, a nuclear factor coordinating signaling inputs from wingless and decapentaplegic. Cell 88:777–787

Roose J, Huls G, van Beest M, Moerer P, van der Horn K, Goldschmeding R, Logtenberg T, Clevers H (1999) Synergy between tumor suppressor APC and the beta-catenin-Tcf4 target Tcf1. Science 285:1923–1926

Rosso SB, Sussman D, Wynshaw-Boris A, Salinas PC (2005) Wnt signaling through Dishevelled, Rac and JNK regulates dendritic development. Nat Neurosci 8:34–42

Sachdev S, Bruhn L, Sieber H, Pichler A, Melchior F, Grosschedl R (2001) PIASy, a nuclear matrix-associated SUMO E3 ligase, represses LEF1 activity by sequestration into nuclear bodies. Genes Dev 15:3088–3103

Sato N, Meijer L, Skaltsounis L, Greengard P, Brivanlou AH (2004) Maintenance of pluripotency in human and mouse embryonic stem cells through activation of Wnt signaling by a pharmacological GSK-3-specific inhibitor. Nat Med 10:55–63

Satoh K, Kasai M, Ishidao T, Tago K, Ohwada S, Hasegawa Y, Senda T, Takada S, Nada S, Nakamura T, Akiyama T (2004) Anteriorization of neural fate by inhibitor of beta-catenin and T cell factor (ICAT), a negative regulator of Wnt signaling. Proc Natl Acad Sci USA 101:8017–8021

Schwarz-Romond T, Merrifield C, Nichols BJ, Bienz M (2005) The Wnt signalling effector Dishevelled forms dynamic protein assemblies rather than stable associations with cytoplasmic vesicles. J Cell Sci 118:5269–5277

Shi Y, Downes M, Xie W, Kao HY, Ordentlich P, Tsai CC, Hon M, Evans RM (2001) Sharp, an inducible cofactor that integrates nuclear receptor repression and activation. Genes Dev 15:1140–1151

Shihabuddin LS, Horner PJ, Ray J, Gage FH (2000) Adult spinal cord stem cells generate neurons after transplantation in the adult dentate gyrus. J Neurosci 20:8727–8735

Shimogori T, VanSant J, Paik E, Grove EA (2004) Members of the Wnt, Fz, and Frp gene families expressed in postnatal mouse cerebral cortex. J Comp Neurol 473:496–510

Sierra OL, Cheng SL, Loewy AP, Charlton-Kachigian N, Towler DA (2004) MINT, the Msx2 interacting nuclear matrix target, enhances Runx2-dependent activation of the osteocalcin fibroblast growth factor response element. J Biol Chem 279:32913–32923

Sierra J, Yoshida T, Joazeiro CA, Jones KA (2006) The APC tumor suppressor counteracts beta-catenin activation and H3K4 methylation at Wnt target genes. Genes Dev 20:586–600

Sinner D, Rankin S, Lee M, Zorn AM (2004) Sox17 and beta-catenin cooperate to regulate the transcription of endodermal genes. Development 131:3069–3080

Snider L, Thirlwell H, Miller JR, Moon RT, Groudine M, Tapscott SJ (2001) Inhibition of Tcf3 binding by I-mfa domain proteins. Mol Cell Biol 21:1866–1873

Song Y, Chung S, Kunes S (2000) Combgap relays wingless signal reception to the determination of cortical cell fate in the Drosophila visual system. Mol Cell 6:1143–1154

Song H, Stevens CF, Gage FH (2002) Astroglia induce neurogenesis from adult neural stem cells. Nature 417:39–44

Stadeli R, Basler K (2005) Dissecting nuclear Wingless signalling: recruitment of the transcriptional co-activator Pygopus by a chain of adaptor proteins. Mech Dev 122:1171–1182

Stadeli R, Hoffmans R, Basler K (2006) Transcription under the control of nuclear Arm/beta-catenin. Curr Biol 16:R378–R385

Standley HJ, Destree O, Kofron M, Wylie C, Heasman J (2006) Maternal XTcf1 and XTcf4 have distinct roles in regulating Wnt target genes. Dev Biol 289:318–328

Suhonen JO, Peterson DA, Ray J, Gage FH (1996) Differentiation of adult hippocampus-derived progenitors into olfactory neurons in vivo. Nature 383:624–627

Tago K, Nakamura T, Nishita M, Hyodo J, Nagai S, Murata Y, Adachi S, Ohwada S, Morishita Y, Shibuya H, Akiyama T (2000) Inhibition of Wnt signaling by ICAT, a novel beta-catenin-interacting protein. Genes Dev 14:1741–1749

Takemaru K, Yamaguchi S, Lee YS, Zhang Y, Carthew RW, Moon RT (2003) Chibby, a nuclear beta-catenin-associated antagonist of the Wnt/Wingless pathway. Nature 422:905–909

Takemoto T, Uchikawa M, Kamachi Y, Kondoh H (2006) Convergence of Wnt and FGF signals in the genesis of posterior neural plate through activation of the Sox2 enhancer N-1. Development 133:297–306

Takizawa T, Nakashima K, Namihira M, Ochiai W, Uemura A, Yanagisawa M, Fujita N, Nakao M, Taga T (2001) DNA methylation is a critical cell-intrinsic determinant of astrocyte differentiation in the fetal brain. Dev Cell 1:749–758

Tamai K, Semenov M, Kato Y, Spokony R, Liu C, Katsuyama Y, Hess F, Saint-Jeannet JP, He X (2000) LDL-receptor-related proteins in Wnt signal transduction. Nature 407:530–535

Tarui T, Takahashi T, Nowakowski RS, Hayes NL, Bhide PG, Caviness VS (2005) Overexpression of p27 Kip 1, probability of cell cycle exit, and laminar destination of neocortical neurons. Cereb Cortex 15:1343–1355

Tashiro A, Sandler VM, Toni N, Zhao C, Gage FH (2006) NMDA-receptor-mediated, cell-specific integration of new neurons in adult dentate gyrus. Nature 442:929–933

Teo JL, Ma H, Nguyen C, Lam C, Kahn M (2005) Specific inhibition of CBP/beta-catenin interaction rescues defects in neuronal differentiation caused by a presenilin-1 mutation. Proc Natl Acad Sci USA 102:12171–12176

Theil T, Aydin S, Koch S, Grotewold L, Ruther U (2002) Wnt and Bmp signalling cooperatively regulate graded Emx2 expression in the dorsal telencephalon. Development 129:3045–3054

Thomas KR, Capecchi MR (1990) Targeted disruption of the murine int-1 proto-oncogene resulting in severe abnormalities in midbrain and cerebellar development. Nature 346:847–850

Tole S, Goudreau G, Assimacopoulos S, Grove EA (2000) Emx2 is required for growth of the hippocampus but not for hippocampal field specification. J Neurosci 20:2618–2625

Topol L, Jiang X, Choi H, Garrett-Beal L, Carolan PJ, Yang Y (2003) Wnt-5a inhibits the canonical Wnt pathway by promoting GSK-3-independent beta-catenin degradation. J Cell Biol 162:899–908

Townsley FM, Cliffe A, Bienz M (2004) Pygopus and Legless target Armadillo/beta-catenin to the nucleus to enable its transcriptional co-activator function. Nat Cell Biol 6:626–633

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

Vadlamudi U, Espinoza HM, Ganga M, Martin DM, Liu X, Engelhardt JF, Amendt BA (2005) PITX2, beta-catenin and LEF-1 interact to synergistically regulate the LEF-1 promoter. J Cell Sci 118:1129–1137

Valenta T, Lukas J, Korinek V (2003) HMG box transcription factor TCF-4's interaction with CtBP1 controls the expression of the Wnt target Axin2/Conductin in human embryonic kidney cells. Nucleic Acids Res 31:2369–2380

Valenta T, Lukas J, Doubravska L, Fafilek B, Korinek V (2006) HIC1 attenuates Wnt signaling by recruitment of TCF-4 and beta-catenin to the nuclear bodies. EMBO J 25:2326–2337

Van de Wetering M, Castrop J, Korinek V, Clevers H (1996) Extensive alternative splicing and dual promoter usage generate Tcf-1 protein isoforms with differential transcription control properties. Mol Cell Biol 16:745–752

van Es JH, Barker N, Clevers H (2003) You Wnt some, you lose some: oncogenes in the Wnt signaling pathway. Curr Opin Genet Dev 13:28–33

Van Raay TJ, Moore KB, Iordanova I, Steele M, Jamrich M, Harris WA, Vetter ML (2005) Frizzled 5 signaling governs the neural potential of progenitors in the developing Xenopus retina. Neuron 46:23–36

van Roy FM, McCrea PD (2005) A role for Kaiso-p120ctn complexes in cancer? Nat Rev Cancer 5:956–964

Veeman MT, Axelrod JD, Moon RT (2003) A second canon. Functions and mechanisms of beta-catenin-independent Wnt signaling. Dev Cell 5:367–377

Viti J, Gulacsi A, Lillien L (2003) Wnt regulation of progenitor maturation in the cortex depends on Shh or fibroblast growth factor 2. J Neurosci 23:5919–5927

Wallingford JB, Habas R (2005) The developmental biology of Dishevelled: an enigmatic protein governing cell fate and cell polarity. Development 132:4421–4436

Wang HY, Malbon CC (2003) Wnt signaling, Ca2+, and cyclic GMP: visualizing Frizzled functions. Science 300:1529–1530

Wang LP, 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

Wayman GA, Impey S, Marks D, Saneyoshi T, Grant WF, Derkach V, Soderling TR (2006) Activity-dependent dendritic arborization mediated by CaM-kinase I activation and enhanced CREB-dependent transcription of Wnt-2. Neuron 50:897–909

Weerkamp F, Baert MR, Naber BA, Koster EE, de Haas EF, Atkuri KR, van Dongen JJ, Herzenberg LA, Staal FJ (2006) Wnt signaling in the thymus is regulated by differential expression of intracellular signaling molecules. Proc Natl Acad Sci USA 103:3322–3326

Wehrli M, Dougan ST, Caldwell K, O'Keefe L, Schwartz S, Vaizel-Ohayon D, Schejter E, Tomlinson A, DiNardo S (2000) arrow encodes an LDL-receptor-related protein essential for Wingless signalling. Nature 407:527–530

Weidinger G, Moon RT (2003) When Wnts antagonize Wnts. J Cell Biol 162:753–755

Weitzman JB (2005) Dishevelled nuclear shuttling. J Biol 4:1

Westfall TA, Brimeyer R, Twedt J, Gladon J, Olberding A, Furutani-Seiki M, Slusarski DC (2003) Wnt-5/pipetail functions in vertebrate axis formation as a negative regulator of Wnt/beta-catenin activity. J Cell Biol 162:889–898

Wharton KA Jr (2003) Runnin' with the Dvl: proteins that associate with Dsh/Dvl and their significance to Wnt signal transduction. Dev Biol 253:1–17

Willert K, Jones KA (2006) Wnt signaling: is the party in the nucleus? Genes Dev 20:1394–1404

Willert K, Brown JD, Danenberg E, Duncan AW, Weissman IL, Reya T, Yates JR 3rd, Nusse R (2003) Wnt proteins are lipid-modified and can act as stem cell growth factors. Nature 423:448–452

Willinger T, Freeman T, Herbert M, Hasegawa H, McMichael AJ, Callan MF (2006) Human naive CD8 T cells down-regulate expression of the WNT pathway transcription factors lymphoid enhancer binding factor 1 and transcription factor 7 (T cell factor-1) following antigen encounter in vitro and in vivo. J Immunol 176:1439–1446

Woodhead GJ, Mutch CA, Olson EC, Chenn A (2006) Cell-autonomous beta-catenin signaling regulates cortical precursor proliferation. J Neurosci 26:12620–12630

Xu Q, Wang Y, Dabdoub A, Smallwood PM, Williams J, Woods C, Kelley MW, Jiang L, Tasman W, Zhang K, Nathans J (2004) Vascular development in the retina and inner ear: control by Norrin and Frizzled-4, a high-affinity ligand-receptor pair. Cell 116:883–895

Yabe D, Fukuda H, Aoki M, Yamada S, Takebayashi S, Shinkura R, Yamamoto N, Honjo T (2007) Generation of a conditional knockout allele for mammalian Spen protein Mint/SHARP. Genesis 45:300–306

Yu JM, Kim JH, Song GS, Jung JS (2006) Increase in proliferation and differentiation of neural progenitor cells isolated from postnatal and adult mice brain by Wnt-3a and Wnt-5a. Mol Cell Biochem 288:17–28

Zeng X, Tamai K, Doble B, Li S, Huang H, Habas R, Okamura H, Woodgett J, He X (2005) A dual-kinase mechanism for Wnt co-receptor phosphorylation and activation. Nature 438:873–877

Zhai L, Chaturvedi D, Cumberledge S (2004) Drosophila wnt-1 undergoes a hydrophobic modification and is targeted to lipid rafts, a process that requires porcupine. J Biol Chem 279:33220–33227

Zhou CJ, Zhao C, Pleasure SJ (2004) Wnt signaling mutants have decreased dentate granule cell production and radial glial scaffolding abnormalities. J Neurosci 24:121–126

Zhou CJ, Borello U, Rubenstein JL, Pleasure SJ (2006) Neuronal production and precursor proliferation defects in the neocortex of mice with loss of function in the canonical Wnt signaling pathway. Neuroscience 142:1119–1131

Zorn AM, Barish GD, Williams BO, Lavender P, Klymkowsky MW, Varmus HE (1999) Regulation of Wnt signaling by Sox proteins: XSox17 alpha/beta and XSox3 physically interact with beta-catenin. Mol Cell 4:487–498