Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

# The Origins of Medulloblastoma Subtypes

Richard J. Gilbertson[1,2] and David W. Ellison[3]

Departments of [1]Developmental Neurobiology, [2]Oncology, and [3]Pathology, St. Jude Children's Research Hospital, Memphis, Tennessee 38105;
email: Richard.Gilbertson@stjude.org, David.Ellison@stjude.org

Annu. Rev. Pathol. Mech. Dis. 2008. 3:341–65

The *Annual Review of Pathology: Mechanisms of Disease* is online at pathmechdis.annualreviews.org

This article's doi:
10.1146/annurev.pathmechdis.3.121806.151518

Copyright © 2008 by Annual Reviews.
All rights reserved

1553-4006/08/0228-0341$20.00

## Key Words

PNET, cancer stem cell, development, cerebellum

## Abstract

Childhood tumors containing cells that are morphologically and functionally similar to normal progenitor cells provide fertile ground for investigating the links between development and cancer. In this respect, integrated studies of normal cerebellar development and the medulloblastoma, a malignant embryonal tumor of the cerebellum, have proven especially fruitful. Emerging evidence indicates that the different precursor cell populations that form the cerebellum and the cell signaling pathways that regulate its development likely represent distinct compartments from which the various subtypes of medulloblastoma arise. Definitive characterization of each medulloblastoma subtype will undoubtedly improve treatment of this disease and provide important insights to the origins of cancer.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

## INTRODUCTION

The medulloblastoma is an embryonal neuroepithelial tumor of the cerebellum and the most common malignant brain tumor of childhood (1, 2). Multimodality treatment regimens have significantly improved survival rates for this disease; however, approximately one-third of patients with medulloblastoma remain incurable, and current treatments significantly damage long-term survivors. To improve the outcome of those medulloblastoma patients with high-risk disease and the quality of life of all survivors, both novel therapies and an improved tumor classification are required. Novel therapies will result from a greater understanding of the disease process and are likely to involve small molecules designed to target specific pathways that become dysregulated during oncogenesis (3). An improved tumor classification will incorporate an assessment of the molecular profiles of medulloblastomas with defined biological behaviors or of the status of cellular pathways that are potential targets for novel therapies.

In creating the first classification of central nervous system (CNS) tumors, Bailey & Cushing (4, 5) used contemporary assumptions about CNS histogenesis to devise their nomenclature. They distinguished the medulloblast from the primitive spongioblast and apolar neuroblast, endowing it with the capacity to differentiate along glial and neuronal lines. A cerebellar medulloblastoma derived from this cell was considered to have the same potential, despite being composed mainly of undifferentiated cells.

The term primitive neuroectodermal tumor (PNET), which was proposed by Hart & Earle (6) in 1973, gained currency for situations in which an embryonal tumor could not be readily placed among the following categories: medulloblastoma, medulloepithelioma, ependymoblastoma, central neuroblastoma, or polar spongioblastoma. Originally, the PNET was considered a cerebral high-grade undifferentiated neuroepithelial tumor of childhood, rarely demonstrating focal differentiation along glial and neuronal lines. However, the term was soon used for a variety of undifferentiated embryonal tumors at all CNS sites and in all ages, and was championed by Rorke on the basis that CNS embryonal tumors do not differ in their essential features; they are all high-grade, undifferentiated neuroepithelial tumors with the capacity to differentiate along glial and neuronal lines (7). Importantly, Rorke emphasized an ontogenetic relationship among PNETs: that they have their origins in the primitive neuroepithelium of the subependymal ventricular matrix.

The PNET concept has been useful for the development of clinical protocols to treat the range of CNS embryonal tumors, especially those presenting in childhood. PNETs share clinical attributes, such as age at presentation and a propensity to disseminate through cerebrospinal fluid pathways, as well as many histological characteristics (1). However, from a pathological perspective, the medulloblastoma demonstrates a diversity not seen in other PNETs; for example, the distinctive architectural and cytological features of the nodular/desmoplastic medulloblastoma are unique. Such distinctive morphophenotypes have been emphasized by Rubinstein and argue for the medulloblastoma to be classified separately from other PNETs, especially if its variants exhibit different biological behaviors, which holds true for desmoplastic medulloblastomas (8, 9). The medulloblastoma's relationship to other embryonal tumors differs across editions (1993, 2000, 2007) of the World Health Organization (WHO) classification of CNS tumors, reflecting evolving perceptions of its origins and nature (**Table 1**) (10, 11). Where once it was classified alongside PNETs from other sites and regarded as the archetypal expression of this phenotype, it is now seen more as an idiosyncratic tumor defined by particular morphologic and genetic attributes.

Genetic data have further reinforced the individual nature of the medulloblastoma in tumor classifications. For example, although

most medulloblastomas are sporadic, they do occur in rare hereditary tumor syndromes such as Turcot syndrome (type 2) and nevoid basal cell carcinoma (Gorlin) syndrome (12, 13). If all PNETs were related ontogenetically, patients with these syndromes would be expected to develop such a tumor at any CNS site. However, with the exception of pineoblastomas in a very few patients with Turcot syndrome, this does not happen (14). Additionally, it is clear that medulloblastomas and supratentorial PNETs harbor distinct molecular genetic abnormalities; abnormalities of chromosome 17 are common in the former but rare in the latter (1, 15).

In this review, we consider the medulloblastoma in the context of cerebellar development and genomics. As a general principle, we propose that abnormalities at the levels of genome, transcriptome, and proteome in medulloblastoma cells reflect two fundamental processes: deregulation of signaling pathways involved in normal cerebellar development and other, more ubiquitous oncogenic events that disrupt cell proliferation, differentiation, and programmed cell death. In this way, we concatenate elements of earlier theories about the origins of medulloblastoma, proposing that, although these origins may lie in neural stem cells or progenitors that appear histologically the same as others across the CNS, these medulloblasts have unique properties that reflect their anatomic site and fate.

**Table 1 World Health Organization classification of tumors of the central nervous system**

Embryonal tumors

1993
- Medulloepithelioma
- Neuroblastoma
  - Ganglioneuroblastoma
- Ependymoblastoma
- Primitive neuroectodermal tumors (PNETs)
  - Medulloblastoma
    - Variants:
      Desmoplastic medulloblastoma
      Medullomyoblastoma
      Melanotic medulloblastoma

2000
- Medulloepithelioma
- Ependymoblastoma
- Medulloblastoma
  - Desmoplastic medulloblastoma
  - Large-cell medulloblastoma
  - Medullomyoblastoma
  - Melanotic medulloblastoma
- Supratentorial PNET
  - Neuroblastoma
  - Ganglioneuroblastoma
- Atypical teratoid/rhabdoid tumor

2007
- Medulloepithelioma
- Ependymoblastoma
- Medulloblastoma
  - Desmoplastic medulloblastoma
  - Medulloblastoma with extensive nodularity
  - Large-cell medulloblastoma
  - Anaplastic medulloblastoma
- Supratentorial PNET
  - Cerebral neuroblastoma
  - Ganglioneuroblastoma
- Atypical teratoid/rhabdoid tumor

## MEDULLOBLASTOMA HETEROGENEITY

### Morphologic Heterogeneity

The latest (2007) WHO classification of tumors of the CNS lists the classic medulloblastoma and several variants: desmoplastic, anaplastic, and large-cell medulloblastomas, and the medulloblastoma with extensive nodularity (MBEN). Of these variants, the anaplastic and large-cell medulloblastomas form a continuum; all large-cell medulloblastomas have regions of anaplasia. Large-cell and anaplastic tumors make up between 2%–4% and 10%–22% of medulloblastomas, respectively (1, 16, 17). Because these two variants form a continuum with a poor prognosis, they have been grouped as large-cell/anaplastic (LCA) medulloblastomas in several studies. Desmoplastic medulloblastomas encompass the nodular/desmoplastic medulloblastoma and the MBEN, which contribute approximately 7% and 3% of all

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

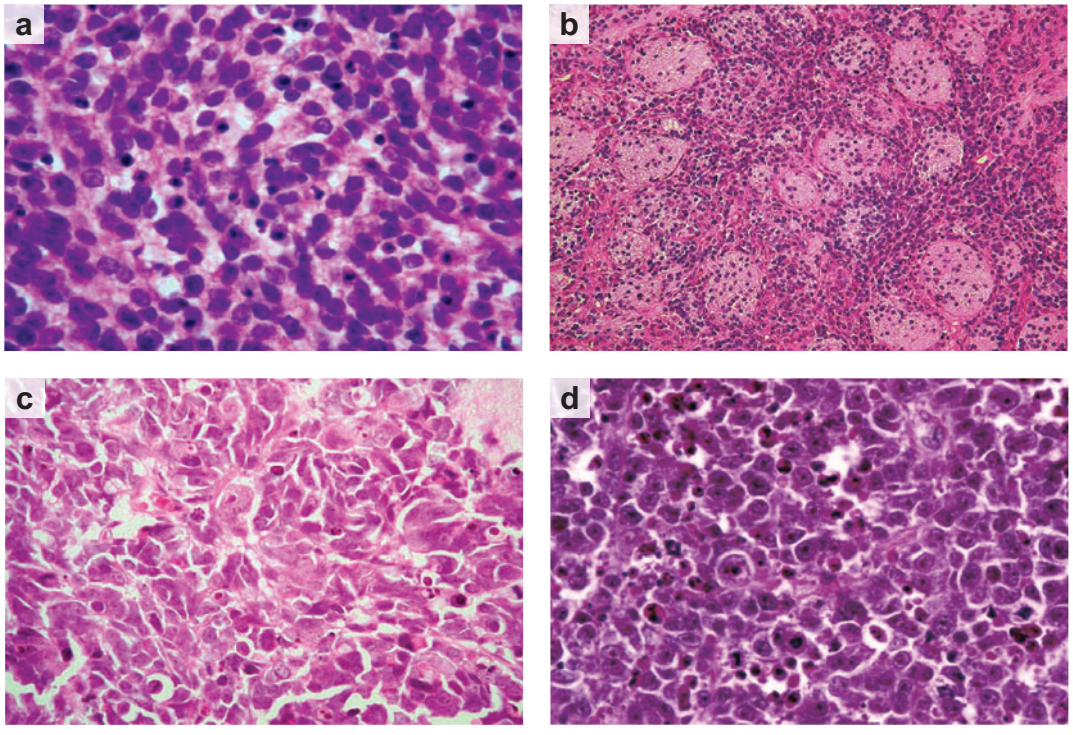


**Figure 1**

Histologic variants of medulloblastoma. (*a*) The classic medulloblastoma is composed of sheets of small uniform cells with a high nuclear-to-cytoplasmic ratio. (*b*) The nodular/desmoplastic medulloblastoma combines nodules of differentiated neurocytic cells with a low growth fraction and desmoplastic internodular zones of moderately pleomorphic cells with a high growth fraction. (*c*) The anaplastic medulloblastoma contains pleomorphic cells with polyhedral forms and a high growth fraction. Abundant apoptosis and examples of cell wrapping are evident. (*d*) The large-cell medulloblastoma contains groups of large uniform cells with vesicular nuclei and a single nucleolus. Anaplasia characterizes other regions of this variant.

medulloblastomas, respectively (1, 8). Classic tumors constitute the remainder.

The classic medulloblastoma is composed of small round or ellipsoid cells with a high nuclear-to-cytoplasmic ratio and round-to-oval or triangular hyperchromatic nuclei (**Figure 1*a***). Uniform cells with round nuclei, in which the chromatin is less condensed, are frequently intermingled with the hyperchromatic cells and occasionally form the dominant population. Several architectural features may interrupt sheets of these undifferentiated cells. Foci of necrosis and angiogenesis are variably present. The microvascular proliferation in medulloblastomas is usually more subtle than the glomeruloid angiogenesis of high-grade gliomas. Neuroblastic (Homer-Wright) rosettes, which consist of tumor cell nuclei arranged in a circular fashion around fine tangled cytoplasmic processes, are observed in less than 40% of classic tumors. Ribbons of tumor cell nuclei can also be arranged with their long axes in parallel, resembling a picket fence. Tumor cells invade the cortex, deep white matter, and nuclei of the cerebellum and frequently occupy the subarachnoid space. Cell turnover in most medulloblastomas is high; indices of proliferation and apoptosis can be as great as in any other neuroepithelial tumor. However, these measures can be unexpectedly low in some classic tumors. Overall, the mitotic index for

childhood medulloblastomas is usually in the range of 0.5% to 2% (17, 18). The growth fraction, as assessed by Ki-67 immunolabeling, is generally greater than 20% (1, 17).

Morphological differentiation in medulloblastomas can be diverse, but occurs principally along neuroepithelial lines (1, 7, 9). Neuronal differentiation is most common. Small groups of neurocytic cells or ganglion cells, which may be picked out by their larger nuclei and reduced nuclear-to-cytoplasmic ratio, are found arranged against a neuropil-like background in approximately 7% of medulloblastomas. Glial differentiation is rare and generally takes the form of scattered small groups of cells with an astrocytic phenotype. Ependymal differentiation is exceptional and would raise concerns about the diagnosis. Rarely, rhabdomyoblasts may be present as scattered single cells or in small groups. Differentiated cells that exhibit long cytoplasmic processes and a skeletal muscle phenotype, including striations, may be included in this embryonal tumor, which has been listed as a distinct entity, the medullomyoblastoma, in previous WHO classifications. However, this phenotype may occur in classic, desmoplastic, and anaplastic medulloblastomas, and thus is no longer considered a separate entity in the WHO classification. Tubules and acini, usually lined by a single layer of melanin-positive epithelioid cells, form very rarely in medulloblastomas, which have been classified previously as melanotic medulloblastomas. This and the rhabdomyoblastic phenotype have been reported to occur together in a few medulloblastomas. A very limited number of case reports attest to the medulloblastoma's capacity for differentiation beyond the neuroepithelial and mesenchymal features described above.

The desmoplastic medulloblastoma is defined as having a biphasic architecture that consists of regions with dense intercellular reticulin and nodular reticulin-free zones, in which tumor cells show a neurocytic phenotype (**Figure 1*b***) (8). Desmoplasia, a pericellular deposition of collagen in this context, and a nodular architecture may occur together or separately in medulloblastomas. Desmoplasia can be a reactive phenomenon when medulloblastoma cells invade the leptomeninges. All variants of medulloblastoma have the potential to invade the leptomeninges; thus, it is important, for accurate subclassification, to assess mass-forming intraparenchymal elements of a tumor.

The presence, extent and nature of architectural nodularity in medulloblastomas can be variable. Importantly, nodules may occur with or without internodular desmoplasia. Nodules are distinct microenvironments in which neurocytic differentiation, negligible proliferation, and scattered apoptotic cells are characteristic features. This phenotype contrasts with that in the relatively undifferentiated internodular regions, in which tumor cells show more pleomorphism and are mitotically active. Regions containing nodules with internodular desmoplasia characterize the desmoplastic (nodular/desmoplastic) medulloblastoma, so the classic or LCA medulloblastoma that produces desmoplasia when it invades the cerebellar meninges should not be classified as a desmoplastic variant. Also excluded from this diagnosis are nondesmoplastic medulloblastomas that demonstrate focal nodule formation (8). The nodules in these nondesmoplastic tumors are very similar to those in nodular/desmoplastic medulloblastomas, showing neurocytic differentiation and a negligible growth fraction. However, these medulloblastomas are aligned by their epidemiology and molecular cytogenetic profiles to the range of classic tumors. Some nodular/desmoplastic medulloblastomas contain particularly large and numerous nodules, in which columns of neurocytic cells stream across the neurofibrillary matrix. These tumors have been termed MBENs and are associated with presentation before the age of 3 years and with a good prognosis (1, 19). Other nodular/desmoplastic medulloblastomas in older children and adults are also associated with a better prognosis than that for classic tumors (8).

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

The original description of the large-cell medulloblastoma drew attention to the presence of large round cells with a prominent single nucleolus (16). These cells occupy one end of the range of medulloblastoma cell size and have an area 2–3 times greater than the mean nuclear area of small round cells in classic tumors (17). All large-cell medulloblastomas contain regions where the round cells give way to polyhedral cells that are so densely packed that they mold themselves to adjacent cells forming a paving-like pattern. Mitotic and apoptotic indices among these cells are higher than in other medulloblastomas (17). This combination of marked nuclear pleomorphism and high cell turnover constitutes the anaplastic phenotype and is present in a proportion of non–large-cell medulloblastomas (**Figure 1*c***). The designation anaplastic medulloblastoma is applied to those tumors dominated by this phenotype, and nearly all are nondesmoplastic tumors (17, 20, 21). Less-marked degrees of nuclear pleomorphism or a focus on anaplasia does not qualify for this diagnosis. Children with anaplastic medulloblastomas have a poorer outcome than those with classic tumors (17, 21). Because large-cell and anaplastic medulloblastomas share morphophenotypes and an aggressive biological behavior, they have frequently been combined, as LCA tumors, in clinical and molecular studies of medulloblastomas (**Figure 1*d***).

As embryonal CNS tumors, medulloblastomas may express markers of both neuronal and glial cell lineage, such as synaptophysin and glial fibrillary acidic protein (GFAP), respectively. Such markers aid diagnosis, as does confirmation of a high growth fraction by immunolabeling tumor cells with a Ki-67 antibody, but none of these adjuncts to histopathological diagnosis provides supplementary information about biological behavior or treatment response. Immunohistochemical detection of neuronal markers or GFAP does not appear to have independent prognostic significance, and a mitotic count or index is better at predicting outcome than a Ki-67 labeling index (1, 17, 18).

## Cytogenetic Heterogeneity

The observation that the medulloblastoma encompasses a number of distinct morphologic variants suggests that these tumors represent different entities arising through alternative mechanisms. Studies of gross chromosomal alterations in medulloblastoma support this notion and have provided the first clues that different molecular processes underlie the development of medulloblastoma subtypes (1, 3, 22, 23).

Deletions of 17p and isochromosome 17q (i17q), which combines loss of 17p and gain of 17q, have long been recognized as the most common chromosomal alterations in medulloblastoma (24). However, these alterations are not distributed equally among the histologic variants. i17q has been observed in 34% ($n$ = 20/59) and 36% ($n$ = 8/22) of classic and LCA tumors, respectively, but in only 12% ($n$ = 8/69) of desmoplastic medulloblastomas (20, 25–29). Furthermore, the presence of i17q in tumors has been associated with a poor clinical outcome, suggesting that this cytogenetic alteration may contribute to the development of aggressive variants of medulloblastoma (25, 30–32). In contrast, monosomy 6 has recently been shown to occur exclusively in favorable prognosis, mainly classic medulloblastomas that contain an intact chromosome 17 and concurrent activating mutations in the β-catenin gene (*CTNNB1*) (29, 33–35). Thus, chromosome 6 may harbor a tumor suppressor gene that cooperates with aberrant Wingless (WNT) signaling to generate an especially curable subtype of medulloblastoma. The desmoplastic and LCA variants are also associated with specific chromosomal alterations. Deletions of 9q are observed in up to 40% of desmoplastic medulloblastomas, but occur rarely in tumors of the classic variant (28, 36), and amplifications of the *MYCC* and *MYCN* oncogenes

occur predominantly in LCA tumors (1). Furthermore, a recent study showed that medulloblastoma cells transduced with *MYCC* adopt a severely anaplastic phenotype when grown as xenografts in nude mice, suggesting a causative relationship between *MYCC* expression and the LCA phenotype (37).

Although histopathologic and molecular cytogenetic assessments of medulloblastoma are useful for grouping tumors with common characteristics and can be of prognostic value, they are of limited value when attempting to determine the cellular origins of these variants. For recent progress in understanding medulloblastoma pathogenesis, we must look to studies that have considered tumorigenesis in the context of normal cerebellar development.

## DEVELOPMENT OF THE NORMAL CEREBELLUM

Since the medulloblastoma was first distinguished from other brain tumors in 1910, it has been considered to arise from neural precursor cells located in or near the cerebellum (7, 9, 38). Almost 100 years later, the precise cells of origin of the medulloblastoma and its variants remain to be determined; however, there is now considerable evidence that the origins of this disease are intimately related to the development of the normal cerebellum. Indeed, if a full understanding of the mechanisms that generate the medulloblastoma is to be achieved, we must understand first how the normal cerebellum is assembled. In particular, we should bear in mind that the cerebellum is unique among brain regions, containing both a nuclear structure, typical of subcortical regions, and an overlying laminar cortex, typical of the cerebrum. Emerging evidence indicates that oncogenic abnormalities in the different cells and signal pathways that generate the various elements of the cerebellum may drive the formation of different medulloblastoma subtypes.

### The Cells that Make the Cerebellum

The cerebellar anlage is established within the roof of the metencephalon of the mouse embryo between embryonic days (E) 10 and 11 (39). Subsequently, and in contrast to most other brain regions, the cells of the cerebellum are derived from two distinct germinal zones (**Figure 2*a***). Glutamatergic projection neurons of the deep nuclei arise from the rhombic lip at approximately E10.5 to E12.5 and migrate to their final positions via the nuclear transitory zone, which is located just below the pial surface at the rostral end of the cerebellar plate (40). GABAergic neurons that include those of the deep nuclei, Purkinje cells and Golgi neurons, arise sequentially from multipotent precursor cells of the primary germinal epithelium in the roof of the fourth ventricle (41–45). Once this neurogenic process is underway, a second germinal zone forms from cells within the rhombic lip. This germinal zone comprises granule neuron precursor cells (GNPCs) that invade rostrally across the cerebellum anlage to produce the external germinal layer (EGL). GNPCs then migrate inward, past the Purkinje cell layer, to form the mature granule cell neurons of the internal granular layer (**Figure 2*b***). The EGL persists until postnatal day (P) 15 in the mouse and into the second postnatal year in the human. Recently, a third stem cell population (**Figure 2*b***) was identified within the white matter of the postnatal cerebellum (46). These cells express the neural stem cell markers CD133 and Nestin; undergo extensive self-renewal; and are multipotent, generating astrocytes, oligodendrocytes, and neurons but not granule cell neurons, in vivo. Thus, the cerebellum includes three distinct pools of progenitor cells that might each serve as cells of origin for the medulloblastoma and its variants.

### The Signals Required for Cerebellar Development

The distinct precursor cell populations that form the cerebellum require a series of

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

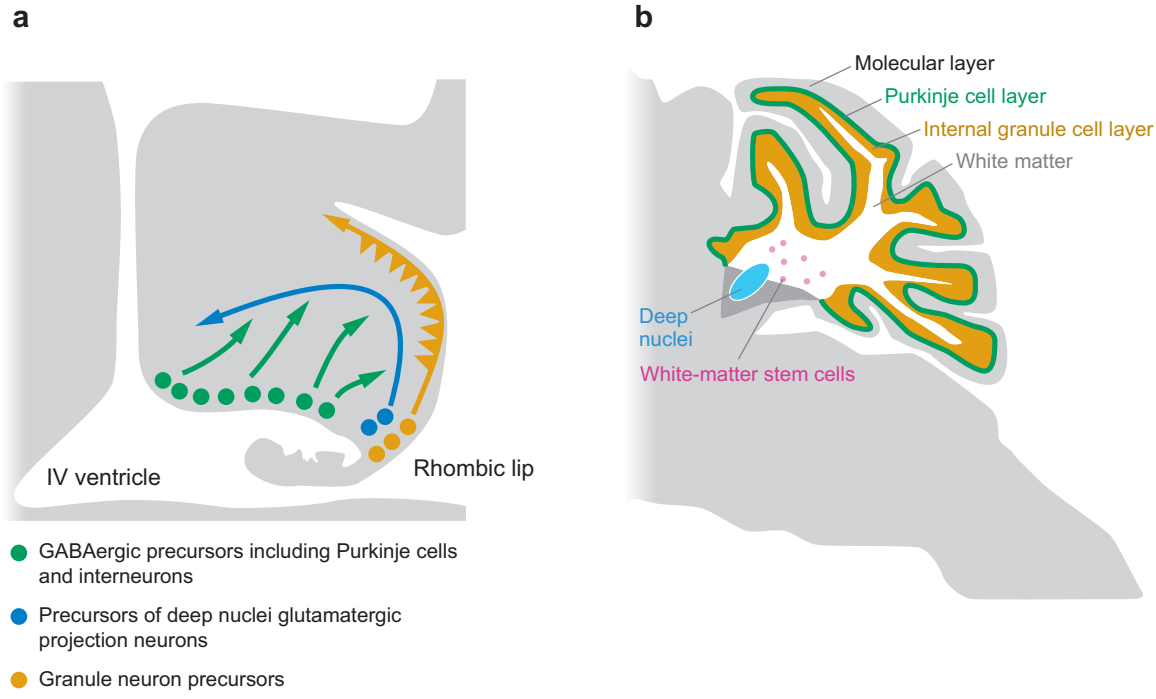


**Figure 2**

Precursor cell populations in the developing and adult cerebellum. (*a*) Embryonic cerebellum: Precursor cells of the GABAergic neurons, including Purkinje cells and interneurons, are located within the primary germinal zone in the roof of the IV ventricle. Glutamatergic neurons of the deep cerebellar nuclei arise within the rhombic lip and migrate to form the nuclei via the nuclear transitory zone. Granule neuron precursor cells also arise within the rhombic lip, but migrate across the surface of the cerebellar anlage to form the external germinal layer. Granule neuron precursor cells proliferate within the external germinal layer before migrating inward to form the glutamatergic granule neurons of the internal granule layer. (*b*) The final position in the adult cerebellum of cells generated by the precursor cells shown in panel *a*. The color convention is maintained to reflect cell lineage. A third population of stem/precursor cells has been identified in the white matter of the adult cerebellum.

coordinated cell signals to function properly (**Figure 3**). Those that regulate the formation of granule cell neurons are the best understood. The bone morphogenic proteins (BMPs) Bmp6, Bmp7, and Gdf7 produced by cells of the dorsal midline provide a signal that initiates the program of granule cell specification within cells of the rhombic lip (47). Following migration into the EGL, GNPCs proliferate in response to the mitogen sonic hedgehog (Shh), which is secreted by Purkinje cells (48, 49). Shh binds to Ptc1 receptors on the surface of GNPCs, causing Ptc1 to dissociate from another transmembrane protein, smoothened (Smo), which then acts as a positive regulator of GNPC proliferation via the Gli transcription factors (50). The transcriptional program induced in GNPCs by Shh includes expression of cyclin D1 and N-myc, which are key regulators of the Shh response (51, 52).

A variety of intrinsic and extrinsic cues modulate the response of GNPCs to Shh (**Figure 3**). Activation of the phosphatidylinositol 3-kinase pathway in GNPCs potentiates Shh signaling by inhibiting phosphorylation-dependent degradation of N-myc (53, 54) and Gli2 (55). Conversely, basic fibroblast growth factor (bFGF) signaling via extracellular signal-regulated kinase



Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

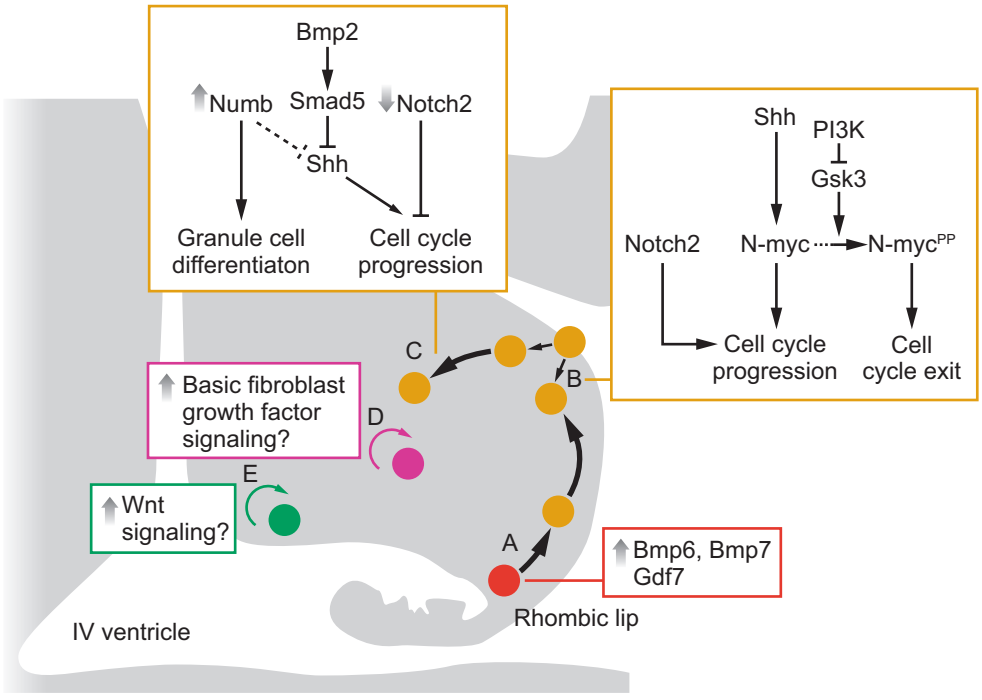


**Figure 3**

Cell signal pathways involved in cerebellar development. (*a*) Bmp6, Bmp7, and Gdf7 produced by cells of the dorsal midline provide a signal that initiates the program of granule cell specification within cells of the rhombic lip. (*b*) Granule neuron precursor cells (GNPCs) within the external germinal layer proliferate in response to Shh secreted from Purkinje cells. GNPC proliferation may be potentiated by phosphatidylinositol 3-kinase (PI3K) and Notch2 signaling. (*c*) Bmp2-mediated Smad5 signaling promotes GNPC cycle exit by suppressing the proliferative response to Shh. Increasing Numb expression and decreasing Notch2 expression may cause GNPCs to leave the cell cycle and differentiate into mature granule cells. (*d*) White-matter stem cells may be sensitive to mitogenic signals mediated by basic fibroblast growth factor, whereas (*e*) GABAergic precursor cells of the primary germinal zone may depend upon WNT signals, although this has not been shown directly.

(ERK) and c-jun N-terminal kinase (JNK) potently inhibits the proliferative response of GNPCs to Shh (56, 57). Prior to migrating from the EGL to form the internal granular layer, GNPCs exit the cell cycle and begin the process of differentiating into mature granule neurons. Bmp2-mediated Smad5 signaling appears to promote GNPC cycle exit by suppressing the proliferative response to Shh (58).

The Notch cell signaling pathway also coordinates the proliferation and differentiation of GNPCs (59). Following ligand binding, the intracellular domain of Notch receptors are released from the plasma membrane and translocate into the nucleus, where they convert the CBF1 repressor complex into an activator complex. Notch signaling is modulated by a number of proteins, including the phosphotyrosine-binding-domain-containing proteins Numb and Numblike (60), and Itch, an E3 ubiquitin ligase (61). The balance between Notch2 signaling and Numb activity contributes to the regulation of GNPC proliferation and differentiation (**Figure 3**). Notch2 signaling is downregulated as GNPCs exit the cell cycle and differentiate (62), whereas a reciprocal increase in Numb activity may promote the maturation of GNPCs into granule cell neurons

(63). Interestingly, Numb may negatively regulate GNPC proliferation in part by targeting Gli1 for Itch-dependent ubiquitination (64). In contrast to Notch2, Notch1 may play a broader role in regulating cerebellar precursors, as deletion of this gene impacts neurogenesis throughout the cerebellum (65).

In contrast to GNPCs, far less is known about the cell signaling systems that regulate the two other precursor cell pools in the cerebellum. Shh signaling is known to regulate neural stem cells throughout the developing and adult brain (66–68). Therefore, it remains possible that Shh regulates the other precursor cells in the cerebellum (**Figure 3**). However, important differences exist between GNPCs of the EGL and precursor cells of the cerebellar white matter, suggesting that these precursors are regulated by distinct mechanisms (46). Of particular note, GNPCs proliferate in response to Shh but not bFGF, whereas stem cells in the white matter proliferate in response to bFGF but not Shh (46).

WNT pathway signaling plays an important role in regulating neural stem and precursor cell proliferation in the CNS (69). Activity in the WNT pathway is related directly to the amount of free cytosolic CTNNB1 (70). In healthy cells, the level of free CTNNB1 is kept low via a multiprotein complex that includes AXIN and APC. This protein complex facilitates the phosphorylation of CTNNB1 by glycogen synthase kinase-3. Phosphorylated CTNNB1 then binds to the β-transducin repeat-containing protein, which promotes the polyubiquitination and complete proteolysis of CTNNB1. The WNT pathway is activated when WNT ligands bind to frizzled receptor proteins on the cell surface. Frizzled inactivates the AXIN and APC complexes though the activation of an intermediate protein termed Disheveled, thereby blocking CTNNB1 degradation. Consequently, levels of cytosolic CTNNB1 increase, and CTNNB1 shuttles to the nucleus to activate gene transcription. Deletion of Wnt-1 from mice completely blocks cerebellar development by preventing specification of the midbrain-hindbrain junction from which the cerebellum is derived (71, 72). Thus, early multipotent precursor cells of the cerebellum are likely to depend critically upon Wnt signals.

Thus, the cerebellum contains at least three distinct populations of neural precursor cells that display differences in both fate and function and that are regulated by different combinations of intrinsic and extrinsic cues (**Figure 2** and **Figure 3**). We propose that the different precursor cells within the cerebellum are susceptible to mutations in the signal pathways that regulate their function; these mutations subvert normal developmental programs and result in the formation of distinct subtypes of medulloblastoma.

## THE ORIGINS OF MEDULLOBLASTOMA SUBTYPES: DISORDERS OF CEREBELLAR DEVELOPMENT

### Cancer Stem Cells in Medulloblastoma

Just as normal tissues arise from multipotent self-renewing precursor cells, there is now considerable evidence that cancers arise as a consequence of aberrant developmental processes, in which the bulk of the neoplastic cells are maintained by a small fraction of stem-like cancer cells termed cancer stem cells (CSCs) (73, 74). CSCs are phenotypically similar to the normal stem cells of the corresponding tissue of origin, but they exhibit dysfunctional patterns of self-renewal and differentiation (75–80).

CSCs are defined functionally as cells that both self-renew and generate the heterogeneous lineages of cancer cells that comprise a tumor (81). CSCs are therefore defined experimentally by their ability to recapitulate all elements of a growing tumor. Singh and colleagues (79) were the first to identify a prospectively isolated population of CSCs in medulloblastoma. These cells were isolated by virtue of expressing the 120-kDa



Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

5-transmembrane cell surface protein Prominin (CD133), which marks normal human neural stem cells (82). Between 1% and 21% of cells in freshly resected medulloblastomas express CD133 (79, 80). As few as 1000 to 5000 of these $CD133^+$ cells are capable of forming tumors that recapitulate the parent tumor when injected into the brains of nonobese diabetic/severe combined immunodeficient (NOD/SCID) mice (79, 83). In contrast, similar transplants of as many as 50,000 $CD133^-$ cells do not form tumors, indicating that the capacity to recapitulate the disease resides solely in the $CD133^+$ fraction (79, 83).

The cell of origin of most CSCs has yet to be determined. Some CSCs may arise from differentiated cells, which acquire stem cell properties as a result of mutation (84, 85). However, there is increasing evidence, especially in the CNS, that CSCs can arise directly from precursor cells. For example, two recent studies found that subsets of ependymoma and astrocytoma exhibit distinct patterns of gene expression that correlate closely with the anatomic location of the tumor, and specifically with those of precursor cells in the corresponding region of the CNS (80, 86). Furthermore, similar to normal neural stem cells, brain CSCs reside in perivascular niches that maintain the stem-like properties of these cancer cells (83).

If medulloblastoma CSCs arise from cerebellar precursor cells, then this raises the important question of whether CSCs in different medulloblastoma variants originate from the different precursor cell populations in the cerebellum. $CD133^+$ CSCs have been isolated from both classic and desmoplastic variants of medulloblastoma, and xenografts of these cells recapitulate, at least in part, some of the morphologic features of these variants (79). Thus, CSCs in medulloblastoma variants may contain intrinsic differences that reflect distinct cellular origins. Evidence derived from immunohistochemical studies support the notion that medulloblastoma variants might arise from distinct populations of precursor cells in the cerebellum. For example, subsets of medulloblastoma have been shown to express markers of cells in either the primary germinal zone (subventricular zone of the cerebellar anlage), for example, calbindin-D28K, parvalbumin, nestin, vimentin, and GFAP, or the secondary germinal zone (GNPC), for example, $p75^{NTR}$, TrkC, Zic1, and Math1, but generally not to express both (87–90). Further comparative studies of normal and neoplastic precursor cell populations in the cerebellum should advance our understanding of the origins of medulloblastoma CSCs and the contribution of these cells to the formation of different disease subtypes.

## Disrupted Signal Pathways and the Origins of Medulloblastoma Subtypes

In addition to studies of medulloblastoma CSCs, the detection of abnormalities in the signal pathways that regulate normal cerebellar development has provided compelling evidence that the different medulloblastoma subtypes arise from distinct cell lineages.

SHH signaling that critically regulates the proliferation of GNPCs was first implicated in medulloblastoma formation following the discovery of inactivating *PTCH1* mutations in the germ line of kindreds with the nevoid basal-cell carcinoma syndrome (NBCCS, also known as Gorlin syndrome) (91, 92). Patients with NBCCS develop characteristic bone cysts and multiple basal cell carcinomas, and are predisposed to a variety of other tumor types, including medulloblastoma (12, 93). For these patients, the lifetime risk of developing medulloblastoma is approximately 4%. Subsequent studies have identified *PTCH1* mutations in 9% of sporadic medulloblastomas ($n = 22/233$ sequences in the literature) (29, 94–99). A minority of medulloblastomas with *PTCH1* mutations retain the wild-type allele, suggesting *PTCH1* may be haploinsufficient for medulloblastoma formation. Definitive evidence that *PTCH1*

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

functions as a tumor suppressor gene was provided when 15% of mice with heterozygous deletion of *Ptch1* (the mouse ortholog of *PTCH1*) developed a medulloblastoma (100). These mouse tumors display a very similar gene expression signature to those of human *PTCH1* mutant tumors, which includes target genes of the SHH pathway (29, 101).

Mutations in other members of the SHH pathway have since been implicated in medulloblastoma. Suppressor of fused (SUFU) operates as a potent repressor of Gli activation in mammalian cells via an unknown mechanism. Deletion of *Sufu* renders the Shh pathway constitutively active and no longer dependent upon upstream components, including Smo and Shh. Mice bearing heterozygous deletion of *Sufu* develop an NBCCS-like skin phenotype, and humans with germ-line mutations in *SUFU* are predisposed to medulloblastoma (102, 103). Although inactivating mutations in *SUFU* have been identified in sporadic medulloblastomas (2%, $n = 5/224$), these are far less common than *PTCH1* mutations (29, 103, 104). A truncating mutation in *PTCH2* was observed in a single medulloblastoma, implicating this second SHH receptor in tumor formation (105). Deletion of *Ptch2* is insufficient to cause medulloblastoma in mice, but loss of this receptor may enhance tumorigenesis in the context of *Ptch1* heterozygosity (106). Recently, *RENKCTD11* that maps to 17p13.2, the most frequently deleted locus in medulloblastoma, was proposed as a further suppressor of Gli-mediated transactivation and medulloblastoma tumorigenesis, although this remains to be confirmed (107).

Because SHH signaling is especially important in GNPC biology (**Figure 3**), is there evidence that tumors in which this pathway is mutated constitute a definable subset that arises from these precursor cells? Four lines of evidence suggest that this hypothesis may be correct. First, medulloblastomas with mutations or deletions of *PTCH1*, *SUFU*, or *RENKDCT11* are frequently, but not exclusively, desmoplastic variants (1, 29, 107). As noted above, deletions of 9q that probably target *PTCH1* are also observed predominantly in desmoplastic medulloblastomas (28, 36). Second, although 15% of $Ptch1^{+/-}$ mice develop medulloblastoma between 3–6 months of age, by 4–6 weeks of age more than half have ectopic collections of GNPC-like cells at the surface of the cerebellum (100, 108, 109). These cells express markers of granule cell lineage, exhibit activation of Shh target genes, and proliferate extensively in vitro. Furthermore, these cells are not merely GNPCs that have failed to migrate, as they lack expression of the wild-type *Ptch1* allele and display a unique gene expression profile that more closely resembles $Ptch1^{+/-}$ tumor cells than GNPCs (109). Similar effects have been observed following the direct injection of a Shh-producing retrovirus into the cerebella of E13.5 embryonic mice (110). Third, targeted expression of a constitutively active form of *Smo* to GNPCs of transgenic mice results in hyperplasia of the EGL and in medulloblastoma formation (111). Similar to mouse and human *PTCH1* mutant medulloblastomas, the tumors that develop in these transgenic mice express high levels of Shh target genes; however, they also show evidence of Notch signal activation that is not seen in *Ptch1* mutant medulloblastomas (29). Thus, constitutive activation of *Smo* may invoke additional effects outside the Shh pathway. Finally, enforced expression of N-myc or cyclin D1 in primary $Ink4c^{-/-}$, $p53^{-/-}$ GNPCs initiates medulloblastomas when these cells are injected back into the brains of immunocompromised recipient animals. These engineered tumors exhibit gene expression profiles indistinguishable from medulloblastomas that arise spontaneously in mice and that delete *Ptch1* (111a). Together, these data suggest that GNPCs are susceptible to transformation by mutations in the SHH signal pathway, and that this transformation leads to the development of a unique type of medulloblastoma, with CSCs that give rise to desmoplastic tumors (**Figure 4**).

As the observation that medulloblastoma occurs in NBCCS led to the discovery of Shh



Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

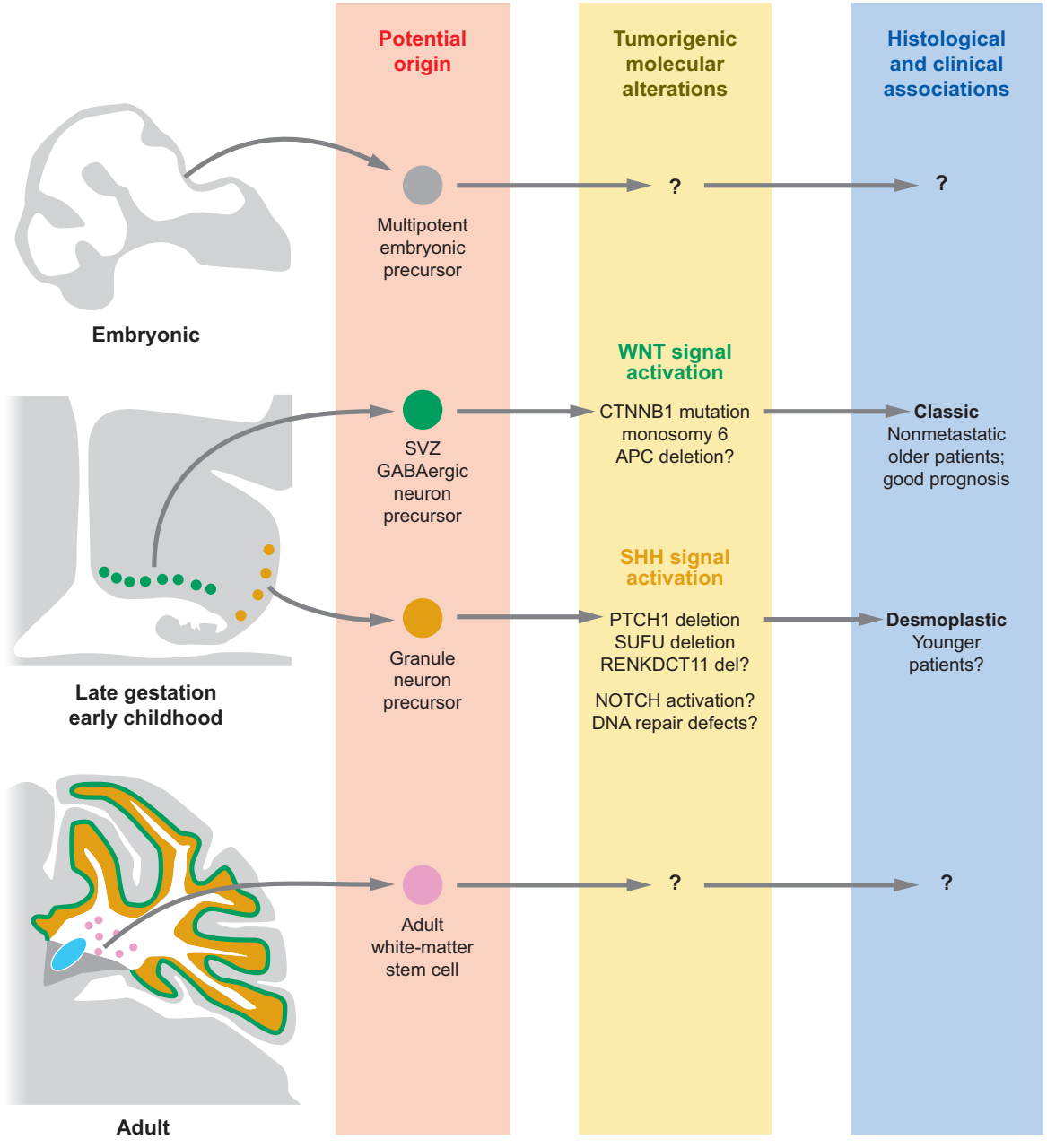


**Figure 4**

Potential cellular and molecular origins of medulloblastoma subgroups. Cancer stem cells of medulloblastoma may arise from multipotent precursor cells of the developing embryo, although the types of mutations to which these cells may be susceptible and the forms of the disease to which these tumors may give rise remain unclear. Medulloblastomas that develop with activating mutations in the WNT and SHH pathways are mutually exclusive and are predominantly classic, good prognosis and desmoplastic, mixed prognosis tumors, respectively (see text). White-matter stem cells may be cells of origin of some adult medulloblastomas, but this remains to be shown definitively.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

pathway abnormalities in sporadic tumors, the WNT pathway has been implicated by the occurrence of medulloblastoma in another familial brain tumor syndrome: Turcot syndrome, which is characterized clinically by the association of a primary brain tumor and multiple colorectal adenomas (112). Since it was first described, Turcot syndrome has been refined to include brain tumor–polyposis (BTP) syndrome Type 1, in which kindreds harbor germ-line mutations in the DNA mismatch-repair gene *hMLH1*, and Type 2, which is associated with mutations in *APC*. The spectrum of brain tumors differs between BTPS1 and BTPS2. Individuals with BTPS1 most commonly develop gliomas, whereas BTPS2 kindreds mainly develop medulloblastomas (13, 112, 113).

Mutations in a variety of WNT pathway members have since been identified in sporadic medulloblastomas. These include activating mutations in *CTNNB1* (8%, $n = 30/383$) and rare inactivating mutations in *APC* (3%, $n = 7/186$) and *AXIN* (2%, $n = 2/81$) (29, 114–119). In contrast to tumors that harbor mutations in the Shh pathway, 75% ($n = 25/33$) of tumors containing *CTNNB1* mutations are of the classic histologic type (29, 34, 115, 117). Mutant *CTNNB1* medulloblastomas tend to arise in an older subset of children, and two independent studies of prospectively treated patients have shown that these tumors are eminently curable (34, 35). In an analysis of medulloblastomas from children entered in the International Society for Pediatric Oncology/United Kingdom Children's Cancer Study Group PNET3 trial ($n = 109$), children with medulloblastomas that showed a nucleopositive CTNNB1 immunophenotype had a five-year event-free survival of 89% (95% CI, 77–100) versus 60% (95% CI, 49–70) for patients with β-catenin nucleonegative tumors, $P = 0.003$ (34). Similarly, in a study of 69 children treated on the St. Jude Medulloblastoma 96 clinical trial, 100% with CTNNB1 nucleopositive tumors were event free at five years versus the 70% of those with nucleonegative disease ($P = 0.03$) (35). Importantly, this study also demonstrated that CTNNB1 nucleopositive tumors and tumors displaying evidence of Shh pathway activation (immunopositive for Gli1 and SFRP1) are mutually exclusive and have significantly different survival rates in favor of the CTNNB1 nucleopositive tumors. Thus, mutations in the WNT and SHH pathways appear to generate different types of medulloblastoma that display distinct patterns of clinical behavior and morphology.

If *SHH*-mutant tumors arise from GNPCs susceptible to mutations in the SHH pathway, is there evidence that *WNT*-mutant tumors arise from a different population of precursor cells? Although no mouse models of WNT pathway mutant medulloblastoma have been generated so far, there is evidence that GNPCs are not susceptible to *Ctnnb1* mutations. We have found that targeted, high-level expression of constitutively activated *Ctnnb1* in GNPCs of transgenic mice (using the Math1 1.7 kb enhancer) does not alter cerebellar development or cause tumors (M. Thompson, P. Gibson & R. Gilbertson, unpublished observations). Furthermore, transduction of P7 GNPCs with mutant *Ctnnb1* ex vivo does not alter the proliferation or Shh responsiveness of these cells (Y. Pei & R. Wechsler-Reya, personal communication). Transgenic expression of *Ctnnb1* in postmitotic granule cell neurons is also nontumorigenic (120). Thus, GNPCs are unlikely to be cells of origin of *WNT*-mutant medulloblastomas; rather, we propose these tumors arise from a separate population of precursor cells, possibly those of the primary germinal zone (**Figure 4**).

Although *WNT* and *SHH* mutations appear mutually exclusive, these alterations affect only 30% to 40% of medulloblastomas (29). Therefore, it is highly likely that genes regulating other cell signaling pathways and processes are mutated to form other variants of medulloblastoma (**Figure 4**). Consistent with the notion that signaling pathways important in cerebellar development might be mutated in subtypes of medulloblastoma,

evidence has linked activation of the NOTCH pathway to medulloblastoma tumorigenesis. Expression of truncated, constitutively active Notch2 in embryonal brain tumor cell lines accelerates cell proliferation, soft agar colony formation, and xenograft growth (121). This study demonstrated that *NOTCH2* is expressed to high levels in some medulloblastomas and is occasionally amplified. It remains to be determined if NOTCH2 contributes to the formation of *SHH*-mutant, *WNT*-mutant, or a different subset of medulloblastoma. Interestingly, NOTCH signal blockade causes the cell cycle exit, apoptosis, and differentiation of some medulloblastoma cell lines and appears to deplete $CD133^+$ CSCs in these tumor cell populations (122). Thus, because NOTCH2 is thought to maintain the proliferation of GNPCs (**Figure 3**), this cell signal pathway may promote the formation of medulloblastoma CSCs from GNPCs.

Genes that have recently emerged as important suppressors of medulloblastoma tumorigenesis are those that regulate the DNA damage response. The DNA repair pathway includes proteins such as poly-(ADP-ribose) polymerase (PARP-1) that sense DNA damage (123) and proteins that repair the damage. DNA double-strand breaks activate two major repair pathways, homologous recombination repair and nonhomologous end joining. The breast/ovarian cancer susceptibility protein BRCA2 is a DNA repair protein with a key role in homologous recombination repair. BRCA2 colocalizes with Partner and localizer of BRCA2 (PALB2) protein, which stabilizes BRCA2 within key nuclear structures, facilitating DNA repair. Proteins involved in nonhomologous end joining include the nuclear ligase Lig4 and XRCC4.

Inactivating mutations in *BRCA2* and *PALB2* cause Fanconi anemia (FA) types D1 and N, respectively (124). FA includes a collection of disorders characterized by chromosomal instability, growth retardation, congenital malformations, progressive bone marrow failure, cancer predisposition, and cellular hypersensitivity to DNA cross-linking agents (125). Mutations in 12 genes have been identified in families with the various forms of the disease (126, 127). FA-D1 and -N each carry a high risk of childhood solid tumors, including medulloblastoma. Indeed, at least five of seven childhood brain tumors diagnosed in six FA-D1 kindreds were medulloblastomas (124, 128), and seven patients identified with FA-N5 had medulloblastoma (126).

Although suppression of human medulloblastoma by other components of the DNA damage response pathway remains to be established, mice null for either *Lig4*, *XRCC4*, or *Parp1* and *p53* develop medulloblastoma with high penetrance (129–131). Interestingly, most *XRCC4*-null/p53-deficient medulloblastomas delete *Ptch1* and, in *Parp-1* deficient mice, develop within the EGL. These tumors also express markers of GNPC and Shh pathway activation (130, 131). Thus, defects in DNA repair may cooperate with mutations in the Shh pathway to cause medulloblastoma (**Figure 4**). Further work will be required to determine the extent to which human sporadic medulloblastomas acquire mutations in the DNA repair machinery, and if these mutations cause a distinct subset of tumors or cooperate with mutations in the Shh pathway.

Further work will also be required to determine exactly how many medulloblastoma molecular subtypes exist and what metrics should be used to define them. These metrics may ultimately include a combination of tumor histology, markers of specific precursor cell populations, and pathway mutations. However, as we refine our classification of medulloblastoma, it will be important to bear in mind that one or more of these metrics may be shared by different disease subgroups.

## GENOMICS: SEEING THE WOOD AND THE TREES

Our current understanding of the degree to which subtypes of medulloblastoma share patterns of histology, gene expression profiles, and genetic alterations has evolved through

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

the comparison of parallel studies that have interrogated only one or two of these characteristics in any given set of tumors. The sum of this research suggests that medulloblastoma comprises several distinct disease entities. However, if we are to generate a definitive classification system for medulloblastoma that accounts for disease origins, molecular mechanisms, and clinical behavior, then it is imperative that we take a global view of the disease. This will dictate that we catalog comprehensively and concurrently the clinical, histologic, and molecular characteristics of large cohorts of prospectively treated patients. These data should enable us to develop a classification scheme for medulloblastoma with great clinical utility, as well as provide unprecedented data with which to solve the cellular and molecular origins of the disease.

The sequencing of the human genome and the development of microarray technologies that measure genome-wide patterns of gene dose and expression have made possible the generation of comprehensive molecular fingerprints of cancer. These technologies have unmasked novel tumor subgroups associated with specific molecular alterations (80, 132–137). The full power of this approach is realized when these comprehensive data sets are integrated with details of tumor histology, gene mutation, and clinical behavior. In short, the genome revolution has placed within our grasp the capacity to definitively characterize and classify cancer. Significant strides have already been made in medulloblastoma, and we estimate that ongoing studies will lead to a novel holistic classification system for this disease within the next five years.

Microarray technology has matured exponentially over the past 10 years, with the rapid introduction of ever more reliable and inclusive tools for measuring gene dosage and expression. The first studies of medulloblastoma gene expression profiles were published more than five years ago and employed microarrays that contained probes for less than 20% of the genes represented on array platforms today. Nevertheless, these pioneering studies provided proof-of-principle that subgroups of medulloblastoma can be identified using genomics (138, 139). MacDonald and colleagues (138) identified 85 genes that were significantly and differentially expressed between 10 metastatic and 13 nonmetastatic tumors. Subsequently, Pomeroy and colleagues (139) showed that desmoplastic and classic medulloblastoma are distinguishable by gene expression and that prediction of prognosis might be possible on the basis of gene expression signatures. Studies using more comprehensive microarrays have confirmed that medulloblastoma includes distinct subgroups. Note that one of the most comprehensive analyses of medulloblastoma conducted to date profiled 46 cases of the disease on the basis of clinical history, tumor histology, U133A gene expression profile (including 33,000 probe sets), direct gene sequencing (*CTNNB1*, *APC*, *AXIN*, *PTCH*, *SUFU*, *TP53*), and fluorescent in situ hybridization (*MYCC*, 17p/17q, 6p/6q) (29). This study confirmed that tumors harboring mutations in the WNT and SHH pathways are mutually exclusive and display distinct patterns of gene expression driven in large part by the presence of subgroup-specific signal pathway mutations and chromosomal alterations. Similar studies of much larger cohorts of tumors derived from prospectively treated patients are now underway within two ongoing North American trials, under the auspices of St. Jude Children's Research Hospital and the Children's Oncology Group. The inclusion of additional microarray technologies within these studies will allow us also to excavate other characteristics of the medulloblastoma genome that have yet to be investigated comprehensively. For example, changes in gene dosage (deletion or amplification) and methylation that cause aberrant expression of genes in medulloblastoma can now be comprehensively measured using array technologies (140–143). The concurrent measurement of these changes alongside profiles of gene expression should prove especially powerful for understanding the molecular basis



Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

of medulloblastoma subgroups. The recent discovery that micro-RNAs are involved in the initiation and progression of human cancer presents another layer of complexity that may contribute to the formation of tumor subgroups. Indeed, expression profiling of human tumors has identified micro-RNA signatures associated with clinical and molecular subgroups of cancer (133, 144). Similar assays for proteomic profiling may also prove useful.

The hope is that, like contours on a map, the successive layering of clinical, histologic, genomic, and proteomic profiles will build a comprehensive view of medulloblastoma, resulting ultimately in a definitive and clinically useful classification system. There are challenges associated with this approach. First, although efforts are underway to conduct these studies prospectively, it is likely that larger collaborative studies will be needed to capture and to analyze all disease subgroups satisfactorily. Second, the computational, biostatistical, and bioinformatic challenges associated with archiving and analyzing these immense data sets are considerable. Comparing data from different array platforms represents a particular challenge. However, successful approaches to achieve this are already under development. Third, although it will be important to capture whole-genome data to define individual subgroups, once established, much of these data will be redundant and it should prove possible to differentiate medulloblastoma subtypes on the basis of a few characteristics. It will be important to determine precisely which characteristics should be selected to define medulloblastoma subgroups and the methods that will be used to assess these in the diagnostic laboratory. Finally, the use of these data to understand fully the origins of medulloblastoma represents a unique challenge. However, integration of gene expression data derived from human medulloblastomas and the developing mouse cerebellum has already provided insights into the origins of the disease.

## CONCLUSIONS

We believe that childhood medulloblastoma presents a paradigm of aberrant tissue development; neoplastic transformation occurs in the setting of signaling pathways and molecular processes that normally control formation of the cerebellum. Molecular signatures of dysfunction in the WNT and SHH pathways can be found in up to 50% of medulloblastomas, and it is likely that other developmental mechanisms are dysregulated in remaining tumors. The existence of significant associations between WNT and SHH pathway signatures and specific histopathological features and biological behavior reinforces the clinicopathological relevance of determining the origins of medulloblastoma variants. Uncovering the origins of this tumor not only reveals potential novel therapeutic approaches, but allows improved therapeutic stratification of patients using existing adjuvant therapies. Thus, a refined classification of medulloblastoma based on a combination of histopathological and molecular assessments will allow a more tailored approach to therapy, with maximal therapy for high-risk disease and optimized therapy aimed at avoiding long-term adverse effects in those with low-risk disease.

In the future, we predict that diagnostic evaluation of medulloblastomas will involve a multifaceted approach, requiring assessments of histopathological features, molecular abnormalities related to developmental pathways, other genetic abnormalities with independent prognostic significance, and potential therapeutic molecular targets, as well as clinical variables and neuroimaging.

## DISCLOSURE STATEMENT

The authors are not aware of any biases that might be perceived as affecting the objectivity of this review.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

## ACKNOWLEDGMENTS

We apologize to those authors whose work we could not cite owing to space limitations. R.J.G. holds the Sydney Schlobohm Leadership Chair of Research from the Brain Tumor Society and is supported by National Institutes of Health grants CA096832 and CA081457. R.J.G. and D.W.E. are supported by the American Lebanese Syrian Associated Charities.

## LITERATURE CITED

1. Ellison D. 2002. Classifying the medulloblastoma: insights from morphology and molecular genetics. *Neuropathol. Appl. Neurobiol.* 28:257–82
2. Gajjar A, Hernan R, Kocak M, Fuller C, Lee Y, et al. 2004. Clinical, histopathologic, and molecular markers of prognosis: toward a new disease risk stratification system for medulloblastoma. *J. Clin. Oncol.* 22:984–93
3. Ellison DW, Clifford SC, Gajjar A, Gilbertson RJ. 2003. What's new in neuro-oncology? Recent advances in medulloblastoma. *Eur. J. Paediatr. Neurol.* 7:53–66
4. Bailey P, Cushing H. 1925. Medulloblastoma cerebelli. *Arch. Neurol. Psychiatry* 14:192–223
5. Bailey P, Cushing H. 1926. *A classification of tumors of the glioma group on a histogenetic basis with a correlation study of prognosis*. Philadelphia: Lippincott
6. Hart MN, Earle KM. 1973. Primitive neuroectodermal tumors of the brain in children. *Cancer* 32:890–7
7. Rorke LB. 1983. The cerebellar medulloblastoma and its relationship to primitive neuroectodermal tumors. *J. Neuropathol. Exp. Neurol.* 42:1–15
8. McManamy CS, Pears J, Weston CL, Hanzely Z, Ironside JW, et al. 2007. Nodule formation and desmoplasia in medulloblastomas: defining the nodular/desmoplastic variant and its biological behaviour. *Brain Pathol.* 17:151–64
9. Rubinstein LJ. 1985. Embryonal central neuroepithelial tumors and their differentiating potential. A cytogenetic view of a complex neuro-oncological problem. *J. Neurosurg.* 62:795–805
10. Kleihues P, Burger PC, Scheithauer BW. 1993. The new WHO classification of brain tumours. *Brain Pathol.* 3:255–68
11. Kleihues P, Cavenee W. 2000. *Tumours of the Nervous System. World Health Organization Classification of Tumours.* Lyon, France: IARC
12. Evans DG, Farndon PA, Burnell LD, Gattamaneni HR, Birch JM. 1991. The incidence of Gorlin syndrome in 173 consecutive cases of medulloblastoma. *Br. J. Cancer* 64:959–61
13. Paraf F, Jothy S, Van Meir EG. 1997. Brain tumor-polyposis syndrome: two genetic diseases? *J. Clin. Oncol.* 15:2744–58
14. Ikeda J, Sawamura Y, van Meir EG. 1998. Pineoblastoma presenting in familial adenomatous polyposis (FAP): random association, FAP variant or Turcot syndrome? *Br. J. Neurosurg.* 12:576–78
15. Russo C, Pellarin M, Tingby O, Bollen AW, Lamborn KR, et al. 1999. Comparative genomic hybridization in patients with supratentorial and infratentorial primitive neuroectodermal tumors. *Cancer* 86:331–39
16. Giangaspero F, Rigobello L, Badiali M, Loda M, Andreini L, et al. 1992. Large-cell medulloblastomas. A distinct variant with highly aggressive behavior. *Am. J. Surg. Pathol.* 16:687–93

17. McManamy CS, Lamont JM, Taylor RE, Cole M, Pearson AD, et al. 2003. Morphophenotypic variation predicts clinical behavior in childhood nondesmoplastic medulloblastomas. *J. Neuropathol. Exp. Neurol.* 62:627–32
18. Gilbertson RJ, Jaros E, Perry RH, Kelly PJ, Lunec J, et al. 1997. Mitotic percentage index: a new prognostic factor for childhood medulloblastoma. *Eur. J. Cancer* 33:609–15
19. Giangaspero F, Perilongo G, Fondelli MP, Brisigotti M, Carollo C, et al. 1999. Medulloblastoma with extensive nodularity: a variant with favorable prognosis. *J. Neurosurg.* 91:971–77
20. Brown HG, Kepner JL, Perlman EJ, Friedman HS, Strother DR, et al. 2000. "Large cell/anaplastic" medulloblastomas: a Pediatric Oncology Group Study. *J. Neuropathol. Exp. Neurol.* 59:857–65
21. Eberhart CG, Kepner JL, Goldthwaite PT, Kun LE, Duffner PK, et al. 2002. Histopathologic grading of medulloblastomas: a Pediatric Oncology Group study. *Cancer* 94:552–60
22. Gilbertson R. 2002. Paediatric embryonic brain tumours. Biological and clinical relevance of molecular genetic abnormalities. *Eur. J. Cancer* 38:675–85
23. Goussia AC, Bruner JM, Kyritsis AP, Agnantis NJ, Fuller GN. 2000. Cytogenetic and molecular genetic abnormalities in primitive neuroectodermal tumors of the central nervous system. *Anticancer Res.* 20:65–73
24. Griffin CA, Hawkins AL, Packer RJ, Rorke LB, Emanuel BS. 1988. Chromosome abnormalities in pediatric brain tumors. *Cancer Res.* 48:175–80
25. Gilbertson R, Wickramasinghe C, Hernan R, Balaji V, Hunt D, et al. 2001. Clinical and molecular stratification of disease risk in medulloblastoma. *Br. J. Cancer* 85:705–12
26. Lamont JM, McManamy CS, Pearson AD, Clifford SC, Ellison DW. 2004. Combined histopathological and molecular cytogenetic stratification of medulloblastoma patients. *Clin. Cancer Res.* 10:5482–93
27. Leonard JR, Cai DX, Rivet DJ, Kaufman BA, Park TS, et al. 2001. Large cell/anaplastic medulloblastomas and medullomyoblastomas: clinicopathological and genetic features. *J. Neurosurg.* 95:82–88
28. Nicholson JC, Ross FM, Kohler JA, Ellison DW. 1999. Comparative genomic hybridization and histological variation in primitive neuroectodermal tumours. *Br. J. Cancer* 80:1322–31
29. Thompson MC, Fuller C, Hogg TL, Dalton J, Finkelstein D, et al. 2006. Genomics identifies medulloblastoma subgroups that are enriched for specific genetic alterations. *J. Clin. Oncol.* 24:1924–31
30. Batra SK, McLendon RE, Koo JS, Castelino-Prabhu S, Fuchs HE, et al. 1995. Prognostic implications of chromosome 17p deletions in human medulloblastomas. *J. Neuro Oncol.* 24:39–45
31. Cogen PH. 1991. Prognostic significance of molecular genetic markers in childhood brain tumors. *Pediatr. Neurosurg.* 17:245–50
32. Scheurlen WG, Schwabe GC, Joos S, Mollenhauer J, Sorensen N, et al. 1998. Molecular analysis of childhood primitive neuroectodermal tumors defines markers associated with poor outcome. *J. Clin. Oncol.* 16:2478–85
33. Clifford SC, Lusher ME, Lindsey JC, Langdon JA, Gilbertson RJ, et al. 2006. Wnt/Wingless pathway activation and chromosome 6 loss characterize a distinct molecular subgroup of medulloblastomas associated with a favorable prognosis. *Cell Cycle* 5:2666–70

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

34. Ellison DW, Onilude OE, Lindsey JC, Lusher ME, Weston CL, et al. 2005. β-Catenin status predicts a favorable outcome in childhood medulloblastoma. *J. Clin. Oncol.* 23:7951–57
35. Gajjar A, Chintagumpala M, Ashley D, Kellie S, Kun LE, et al. 2006. Risk-adapted craniospinal radiotherapy followed by high-dose chemotherapy and stem-cell rescue in children with newly diagnosed medulloblastoma (St Jude Medulloblastoma-96): long-term results from a prospective, multicentre trial. *Lancet Oncol.* 7:813–20
36. Schofield D, West DC, Anthony DC, Marshal R, Sklar J. 1995. Correlation of loss of heterozygosity at chromosome 9q with histological subtype in medulloblastomas. *Am. J. Pathol.* 146:472–80
37. Stearns D, Chaudhry A, Abel TW, Burger PC, Dang CV, et al. 2006. c-myc overexpression causes anaplasia in medulloblastoma. *Cancer Res.* 66:673–81
38. Wright JS. 1910. Neurocytoma or neuroblastoma, a kind of tumour not generally recognised. *J. Exp. Med.* 12:556–61
39. Miale IL, Sidman RL. 1961. An autoradiographic analysis of histogenesis in the mouse cerebellum. *Exp. Neurol.* 4:277–96
40. Fink AJ, Englund C, Daza RA, Pham D, Lau C, et al. 2006. Development of the deep cerebellar nuclei: transcription factors and cell migration from the rhombic lip. *J. Neurosci.* 26:3066–76
41. Altman J, Bayer SA. 1985. Embryonic development of the rat cerebellum. I. Delineation of the cerebellar primordium and early cell movements. *J. Comp. Neurol.* 231:1–26
42. Lin JC, Cepko CL. 1999. Biphasic dispersion of clones containing Purkinje cells and glia in the developing chick cerebellum. *Dev. Biol.* 211:177–97
43. Mathis L, Bonnerot C, Puelles L, Nicolas JF. 1997. Retrospective clonal analysis of the cerebellum using genetic laacZ/lacZ mouse mosaics. *Development* 124:4089–104
44. Morales D, Hatten ME. 2006. Molecular markers of neuronal progenitors in the embryonic cerebellar anlage. *J. Neurosci.* 26:12226–36
45. Zhang L, Goldman JE. 1996. Generation of cerebellar interneurons from dividing progenitors in white matter. *Neuron* 16:47–54
46. Lee A, Kessler JD, Read TA, Kaiser C, Corbeil D, et al. 2005. Isolation of neural stem cells from the postnatal cerebellum. *Nat. Neurosci.* 8:723–29
47. Alder J, Lee KJ, Jessell TM, Hatten ME. 1999. Generation of cerebellar granule neurons in vivo by transplantation of BMP-treated neural progenitor cells. *Nat. Neurosci.* 2:535–40
48. Dahmane N, Ruiz-i-Altaba A. 1999. Sonic hedgehog regulates the growth and patterning of the cerebellum. *Development* 126:3089–100
49. Lewis PM, Gritli-Linde A, Smeyne R, Kottmann A, McMahon AP. 2004. Sonic hedgehog signaling is required for expansion of granule neuron precursors and patterning of the mouse cerebellum. *Dev. Biol.* 270:393–410
50. Ruiz i Altaba A, Palma V, Dahmane N. 2002. Hedgehog-Gli signalling and the growth of the brain. *Nat. Rev. Neurosci.* 3:24–33
51. Kenney AM, Cole MD, Rowitch DH. 2003. Nmyc upregulation by sonic hedgehog signaling promotes proliferation in developing cerebellar granule neuron precursors. *Development* 130:15–28
52. Oliver TG, Grasfeder LL, Carroll AL, Kaiser C, Gillingham CL, et al. 2003. Transcriptional profiling of the Sonic hedgehog response: a critical role for N-myc in proliferation of neuronal precursors. *Proc. Natl. Acad. Sci. USA* 100:7331–36

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

53. Kenney AM, Widlund HR, Rowitch DH. 2004. Hedgehog and PI-3 kinase signaling converge on Nmyc1 to promote cell cycle progression in cerebellar neuronal precursors. *Development* 131:217–28
54. Sjostrom SK, Finn G, Hahn WC, Rowitch DH, Kenney AM. 2005. The Cdk1 complex plays a prime role in regulating N-myc phosphorylation and turnover in neural precursors. *Dev. Cell* 9:327–38
55. Riobo NA, Lu K, Ai X, Haines GM, Emerson CP Jr. 2006. Phosphoinositide 3-kinase and Akt are essential for Sonic Hedgehog signaling. *Proc. Natl. Acad. Sci. USA* 103:4505–10
56. Fogarty MP, Emmenegger BA, Grasfeder LL, Oliver TG, Wechsler-Reya RJ. 2007. Fibroblast growth factor blocks Sonic hedgehog signaling in neuronal precursors and tumor cells. *Proc. Natl. Acad. Sci. USA*. 104:2973–78
57. Wechsler-Reya RJ, Scott MP. 1999. Control of neuronal precursor proliferation in the cerebellum by Sonic Hedgehog. *Neuron* 22:103–14
58. Rios I, Alvarez-Rodriguez R, Marti E, Pons S. 2004. Bmp2 antagonizes sonic hedgehog-mediated proliferation of cerebellar granule neurones through Smad5 signalling. *Development* 131:3159–68
59. Yoon K, Gaiano N. 2005. Notch signaling in the mammalian central nervous system: insights from mouse mutants. *Nat. Neurosci.* 8:709–15
60. Cayouette M, Raff M. 2002. Asymmetric segregation of Numb: a mechanism for neural specification from *Drosophila* to mammals. *Nat. Neurosci.* 5:1265–69
61. Lai EC. 2002. Protein degradation: four E3s for the notch pathway. *Curr. Biol.* 12: R74–78
62. Solecki DJ, Liu XL, Tomoda T, Fang Y, Hatten ME. 2001. Activated Notch2 signaling inhibits differentiation of cerebellar granule neuron precursors by maintaining proliferation. *Neuron* 31:557–68
63. Klein AL, Zilian O, Suter U, Taylor V. 2004. Murine numb regulates granule cell maturation in the cerebellum. *Dev. Biol.* 266:161–77
64. Di Marcotullio L, Ferretti E, Greco A, De Smaele E, Po A, et al. 2006. Numb is a suppressor of Hedgehog signalling and targets Gli1 for Itch-dependent ubiquitination. *Nat. Cell Biol.* 8:1415–23
65. Lutolf S, Radtke F, Aguet M, Suter U, Taylor V. 2002. Notch1 is required for neuronal and glial differentiation in the cerebellum. *Development* 129:373–85
66. Lai K, Kaspar BK, Gage FH, Schaffer DV. 2003. Sonic hedgehog regulates adult neural progenitor proliferation in vitro and in vivo. *Nat. Neurosci.* 6:21–27
67. Machold R, Hayashi S, Rutlin M, Muzumdar MD, Nery S, et al. 2003. Sonic hedgehog is required for progenitor cell maintenance in telencephalic stem cell niches. *Neuron* 39:937–50
68. Palma V, Lim DA, Dahmane N, Sanchez P, Brionne TC, et al. 2005. Sonic hedgehog controls stem cell behavior in the postnatal and adult brain. *Development* 132:335–44
69. Ciani L, Salinas PC. 2005. WNTs in the vertebrate nervous system: from patterning to neuronal connectivity. *Nat. Rev. Neurosci.* 6:351–62
70. Henderson BR, Fagotto F. 2002. The ins and outs of APC and β-catenin nuclear transport. *EMBO Rep.* 3:834–39
71. 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* 62:1073–85
72. Thomas KR, Capecchi MR. 1990. Targeted disruption of the murine int-1 proto-oncogene resulting in severe abnormalities in midbrain and cerebellar development. *Nature* 346:847–50

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

73. Clarke MF, Fuller M. 2006. Stem cells and cancer: two faces of Eve. *Cell* 124:1111–15
74. Pardal R, Clarke MF, Morrison SJ. 2003. Applying the principles of stem-cell biology to cancer. *Nat. Rev. Cancer* 3:895–902
75. Bonnet D, Dick JE. 1997. Human acute myeloid leukemia is organized as a hierarchy that originates from a primitive hematopoietic cell. *Nat. Med.* 3:730–37
76. Hemmati HD, Nakano I, Lazareff JA, Masterman-Smith M, Geschwind DH, et al. 2003. Cancerous stem cells can arise from pediatric brain tumors. *Proc. Natl. Acad. Sci. USA* 100:15178–83
77. Ignatova TN, Kukekov VG, Laywell ED, Suslov ON, Vrionis FD, et al. 2002. Human cortical glial tumors contain neural stem-like cells expressing astroglial and neuronal markers in vitro. *Glia* 39:193–206
78. Lapidot T, Sirard C, Vormoor J, Murdoch B, Hoang T, et al. 1994. A cell initiating human acute myeloid leukaemia after transplantation into SCID mice. *Nature* 367:645–48
79. Singh SK, Hawkins C, Clarke ID, Squire JA, Bayani J, et al. 2004. Identification of human brain tumour initiating cells. *Nature* 432:396–401
80. Taylor MD, Poppleton H, Fuller C, Su X, Liu Y, et al. 2005. Radial glia cells are candidate stem cells of ependymoma. *Cancer Cell* 8:323–35
81. Clarke MF, Dick JE, Dirks PB, Eaves CJ, Jamieson CH, et al. 2006. Cancer stem cells: perspectives on current status and future directions: AACR Workshop on Cancer Stem Cells. *Cancer Res.* 66:9339–44
82. Uchida N, Buck DW, He D, Reitsma MJ, Masek M, et al. 2000. Direct isolation of human central nervous system stem cells. *Proc. Natl. Acad. Sci. USA* 97:14720–25
83. Calabrese C, Poppleton H, Kocak M, Hogg TL, Fuller C, et al. 2007. A perivascular niche for brain tumor stem cells. *Cancer Cell* 11:69–82
84. Bachoo RM, Maher EA, Ligon KL, Sharpless NE, Chan SS, et al. 2002. Epidermal growth factor receptor and Ink4a/Arf: convergent mechanisms governing terminal differentiation and transformation along the neural stem cell to astrocyte axis. *Cancer Cell* 1:269–77
85. Krivtsov AV, Twomey D, Feng Z, Stubbs MC, Wang Y, et al. 2006. Transformation from committed progenitor to leukaemia stem cell initiated by MLL-AF9. *Nature* 442:818–22
86. Sharma MK, Mansur DB, Reifenberger G, Perry A, Leonard JR, et al. 2007. Distinct genetic signatures among pilocytic astrocytomas relate to their brain region origin. *Cancer Res.* 67:890–900
87. Buhren J, Christoph AH, Buslei R, Albrecht S, Wiestler OD, et al. 2000. Expression of the neurotrophin receptor p75NTR in medulloblastomas is correlated with distinct histological and clinical features: evidence for a medulloblastoma subtype derived from the external granule cell layer. *J. Neuropathol. Exp. Neurol.* 59:229–40
88. Pomeroy SL, Sutton ME, Goumnerova LC, Segal RA. 1997. Neurotrophins in cerebellar granule cell development and medulloblastoma. *J. Neurooncol.* 35:347–52
89. Salsano E, Pollo B, Eoli M, Giordana MT, Finocchiaro G. 2004. Expression of MATH1, a marker of cerebellar granule cell progenitors, identifies different medulloblastoma subtypes. *Neurosci. Lett.* 370:180–85
90. Yokota N, Aruga J, Takai S, Yamada K, Hamazaki M, et al. 1996. Predominant expression of human zic in cerebellar granule cell lineage and medulloblastoma. *Cancer Res.* 56:377–83
91. Gailani MR, Bale SJ, Leffell DJ, DiGiovanna JJ, Peck GL, et al. 1992. Developmental defects in Gorlin syndrome related to a putative tumor suppressor gene on chromosome 9. *Cell* 69:111–17

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

92. Hahn H, Wicking C, Zaphiropoulous PG, Gailani MR, Shanley S, et al. 1996. Mutations of the human homolog of *Drosophila* patched in the nevoid basal cell carcinoma syndrome. *Cell* 85:841–51
93. Kimonis VE, Goldstein AM, Pastakia B, Yang ML, Kase R, et al. 1997. Clinical manifestations in 105 persons with nevoid basal cell carcinoma syndrome. *Am. J. Med. Genet.* 69:299–308
94. Pietsch T, Waha A, Koch A, Kraus J, Albrecht S, et al. 1997. Medulloblastomas of the desmoplastic variant carry mutations of the human homologue of *Drosophila* patched. *Cancer Res.* 57:2085–88
95. Raffel C, Jenkins RB, Frederick L, Hebrink D, Alderete B, et al. 1997. Sporadic medulloblastomas contain PTCH mutations. *Cancer Res.* 57:842–45
96. Vorechovsky I, Tingby O, Hartman M, Stromberg B, Nister M, et al. 1997. Somatic mutations in the human homologue of *Drosophila* patched in primitive neuroectodermal tumours. *Oncogene* 15:361–66
97. Wolter M, Reifenberger J, Sommer C, Ruzicka T, Reifenberger G. 1997. Mutations in the human homologue of the *Drosophila* segment polarity gene patched (PTCH) in sporadic basal cell carcinomas of the skin and primitive neuroectodermal tumors of the central nervous system. *Cancer Res.* 57:2581–85
98. Xie J, Johnson RL, Zhang X, Bare JW, Waldman FM, et al. 1997. Mutations of the PATCHED gene in several types of sporadic extracutaneous tumors. *Cancer Res.* 57:2369–72
99. Zurawel RH, Allen C, Chiappa S, Cato W, Biegel J, et al. 2000. Analysis of PTCH/SMO/SHH pathway genes in medulloblastoma. *Genes Chromos. Cancer* 27:44–51
100. Goodrich LV, Milenkovic L, Higgins KM, Scott MP. 1997. Altered neural cell fates and medulloblastoma in mouse patched mutants. *Science* 277:1109–13
101. Lee Y, Miller HL, Jensen P, Hernan R, Connelly M, et al. 2003. A molecular fingerprint for medulloblastoma. *Cancer Res.* 63:5428–37
102. Svard J, Heby-Henricson K, Persson-Lek M, Rozell B, Lauth M, et al. 2006. Genetic elimination of Suppressor of fused reveals an essential repressor function in the mammalian Hedgehog signaling pathway. *Dev. Cell* 10:187–97
103. Taylor MD, Liu L, Raffel C, Hui CC, Mainprize TG, et al. 2002. Mutations in SUFU predispose to medulloblastoma. *Nat. Genet.* 31:306–10
104. Koch A, Waha A, Hartmann W, Milde U, Goodyer CG, et al. 2004. No evidence for mutations or altered expression of the Suppressor of Fused gene (SUFU) in primitive neuroectodermal tumours. *Neuropathol. Appl. Neurobiol.* 30:532–39
105. Smyth I, Narang MA, Evans T, Heimann C, Nakamura Y, et al. 1999. Isolation and characterization of human patched 2 (PTCH2), a putative tumour suppressor gene inbasal cell carcinoma and medulloblastoma on chromosome 1p32. *Hum. Mol. Genet.* 8:291–97
106. Lee Y, Miller HL, Russell HR, Boyd K, Curran T, et al. 2006. Patched2 modulates tumorigenesis in patched1 heterozygous mice. *Cancer Res.* 66:6964–71
107. Di Marcotullio L, Ferretti E, De Smaele E, Argenti B, Mincione C, et al. 2004. REN(KCTD11) is a suppressor of Hedgehog signaling and is deleted in human medulloblastoma. *Proc. Natl. Acad. Sci. USA* 101:10833–38
108. Kim JYH, Nelson AL, Algon SA, Graves O, Sturla LM, et al. 2003. Medulloblastoma tumorigenesis diverges from cerebellar granule cell differentiation in patched heterozygous mice. *Dev. Biol.* 263:50–66

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

109. Oliver TG, Read TA, Kessler JD, Mehmeti A, Wells JF, et al. 2005. Loss of patched and disruption of granule cell development in a preneoplastic stage of medulloblastoma. *Development* 132:2425–39
110. Weiner HL, Bakst R, Hurlbert MS, Ruggiero J, Ahn E, et al. 2002. Induction of medulloblastomas in mice by sonic hedgehog, independent of Gli1. *Cancer Res.* 62:6385–89
111. Hallahan AR, Pritchard JI, Hansen S, Benson M, Stoeck J, et al. 2004. The SmoA1 mouse model reveals that notch signaling is critical for the growth and survival of sonic hedgehog-induced medulloblastomas. *Cancer Res.* 64:7794–800
111a. Zindy F, Uziel T, Ayrault O, Calabrese C, Valentine M, et al. 2007. Genetic alterations in mouse medulloblastomas and generation of tumors *de novo* from primary cerebellar granule neuron precursors. *Cancer Res.* 67:2676–84
112. Hamilton SR, Liu B, Parsons RE, Papadopoulos N, Jen J, et al. 1995. The molecular basis of Turcot's syndrome. *N. Engl. J. Med.* 332:839–47
113. Attard TM, Giglio P, Koppula S, Snyder C, Lynch HT. 2007. Brain tumors in individuals with familial adenomatous polyposis: a cancer registry experience and pooled case report analysis. *Cancer* 109:761–66
114. Baeza N, Masuoka J, Kleihues P, Ohgaki H. 2003. AXIN1 mutations but not deletions in cerebellar medulloblastomas. *Oncogene* 22:632–36
115. Eberhart CG, Tihan T, Burger PC. 2000. Nuclear localization and mutation of β-catenin in medulloblastomas. *J. Neuropathol. Exp. Neurol.* 59:333–37
116. Huang H, Mahler-Araujo BM, Sankila A, Chimelli L, Yonekawa Y, et al. 2000. APC mutations in sporadic medulloblastomas. *Am. J. Pathol.* 156:433–37
117. Koch A, Waha A, Tonn JC, Sorensen N, Berthold F, et al. 2001. Somatic mutations of WNT/wingless signaling pathway components in primitive neuroectodermal tumors. *Int. J. Cancer* 93:445–49
118. Yokota N, Nishizawa S, Ohta S, Date H, Sugimura H, et al. 2002. Role of Wnt pathway in medulloblastoma oncogenesis. *Int. J. Cancer* 101:198–201
119. Zurawel RH, Chiappa SA, Allen C, Raffel C. 1998. Sporadic medulloblastomas contain oncogenic β-catenin mutations. *Cancer Res.* 58:896–99
120. Kratz JE, Stearns D, Huso DL, Slunt HH, Price DL, et al. 2002. Expression of stabilized beta-catenin in differentiated neurons of transgenic mice does not result in tumor formation. *BMC Cancer* 2:33
121. Fan X, Mikolaenko I, Elhassan I, Ni X, Wang Y, et al. 2004. Notch1 and notch2 have opposite effects on embryonal brain tumor growth. *Cancer Res.* 64:7787–93
122. Fan X, Matsui W, Khaki L, Stearns D, Chun J, et al. 2006. Notch pathway inhibition depletes stem-like cells and blocks engraftment in embryonal brain tumors. *Cancer Res.* 66:7445–52
123. Jagtap P, Szabo C. 2005. Poly(ADP-ribose) polymerase and the therapeutic effects of its inhibitors. *Nat. Rev. Drug Discov.* 4:421–40
124. Offit K, Levran O, Mullaney B, Mah K, Nafa K, et al. 2003. Shared genetic susceptibility to breast cancer, brain tumors, and Fanconi anemia. *J. Natl. Cancer Inst.* 95:1548–51
125. Kutler DI, Singh B, Satagopan J, Batish SD, Berwick M, et al. 2003. A 20-year perspective on the International Fanconi Anemia Registry (IFAR). *Blood* 101:1249–56
126. Reid S, Schindler D, Hanenberg H, Barker K, Hanks S, et al. 2007. Biallelic mutations in PALB2 cause Fanconi anemia subtype FA-N and predispose to childhood cancer. *Nat. Genet.* 39:162–64
127. Taniguchi T, D'Andrea AD. 2006. Molecular pathogenesis of Fanconi anemia: recent progress. *Blood* 107:4223–33

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

128. Hirsch B, Shimamura A, Moreau L, Baldinger S, Hag-alshiekh M, et al. 2004. Association of biallelic BRCA2/FANCD1 mutations with spontaneous chromosomal instability and solid tumors of childhood. *Blood* 103:2554–59
129. Lee Y, McKinnon PJ. 2002. DNA ligase IV suppresses medulloblastoma formation. *Cancer Res.* 62:6395–99
130. Tong WM, Ohgaki H, Huang H, Granier C, Kleihues P, et al. 2003. Null mutation of DNA strand break-binding molecule poly(ADP-ribose) polymerase causes medulloblastomas in $p53^{-/-}$ mice. *Am. J. Pathol.* 162:343–52
131. Yan CT, Kaushal D, Murphy M, Zhang Y, Datta A, et al. 2006. XRCC4 suppresses medulloblastomas with recurrent translocations in p53-deficient mice. *Proc. Natl. Acad. Sci. USA* 103:7378–83
132. Bild AH, Yao G, Chang JT, Wang Q, Potti A, et al. 2006. Oncogenic pathway signatures in human cancers as a guide to targeted therapies. *Nature* 439:353–57
133. Calin GA, Croce CM. 2006. MicroRNA signatures in human cancers. *Nat. Rev. Cancer* 6:857–66
134. Lu J, Getz G, Miska EA, Alvarez-Saavedra E, Lamb J, et al. 2005. MicroRNA expression profiles classify human cancers. *Nature* 435:834–38
135. Phillips HS, Kharbanda S, Chen R, Forrest WF, Soriano RH, et al. 2006. Molecular subclasses of high-grade glioma predict prognosis, delineate a pattern of disease progression, and resemble stages in neurogenesis. *Cancer Cell* 9:157–73
136. Quackenbush J. 2006. Microarray analysis and tumor classification. *N. Engl. J. Med.* 354:2463–72
137. Sotiriou C, Neo SY, McShane LM, Korn EL, Long PM, et al. 2003. Breast cancer classification and prognosis based on gene expression profiles from a population-based study. *Proc. Natl. Acad. Sci. USA* 100:10393–98
138. MacDonald TJ, Brown KM, LaFleur B, Peterson K, Lawlor C, et al. 2001. Expression profiling of medulloblastoma: PDGFRA and the RAS/MAPK pathway as therapeutic targets for metastatic disease. *Nat. Genet.* 29:143–52
139. Pomeroy SL, Tamayo P, Gaasenbeek M, Sturla LM, Angelo M, et al. 2002. Prediction of central nervous system embryonal tumour outcome based on gene expression. *Nature* 415:436–42
140. Dutt A, Beroukhim R. 2007. Single nucleotide polymorphism array analysis of cancer. *Curr. Opin. Oncol.* 19:43–49
141. Engle LJ, Simpson CL, Landers JE. 2006. Using high-throughput SNP technologies to study cancer. *Oncogene* 25:1594–601
142. Huebert DJ, Bernstein BE. 2005. Genomic views of chromatin. *Curr. Opin. Genet. Dev.* 15:476–81
143. van Steensel B. 2005. Mapping of genetic and epigenetic regulatory networks using microarrays. *Nat. Genet.* 37(Suppl):18–24
144. Cummins JM, He Y, Leary RJ, Pagliarini R, Diaz LA Jr, et al. 2006. The colorectal microRNAome. *Proc. Natl. Acad. Sci. USA* 103:3687–92

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

Annual Review of Pathology: Mechanisms of Disease

Volume 3, 2008

# Contents



Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

**Indexes**



**Errata**

An online log of corrections to *Annual Review of Pathology: Mechanisms of Disease* articles may be found at http://pathol.annualreviews.org

Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.

# ANNUAL REVIEWS

**It's about time. Your time. It's time well spent.**

## New From Annual Reviews:

## *Annual Review of Statistics and Its Application*

Volume 1 • Online January 2014 • http://statistics.annualreviews.org

Editor: **Stephen E. Fienberg,** *Carnegie Mellon University*

Associate Editors: **Nancy Reid,** *University of Toronto*

**Stephen M. Stigler,** *University of Chicago*

The *Annual Review of Statistics and Its Application* aims to inform statisticians and quantitative methodologists, as well as all scientists and users of statistics about major methodological advances and the computational tools that allow for their implementation. It will include developments in the field of statistics, including theoretical statistical underpinnings of new methodology, as well as developments in specific application domains such as biostatistics and bioinformatics, economics, machine learning, psychology, sociology, and aspects of the physical sciences.

**Complimentary online access to the first volume will be available until January 2015.**

TABLE OF CONTENTS:

- *What Is Statistics?* Stephen E. Fienberg
- *A Systematic Statistical Approach to Evaluating Evidence from Observational Studies,* David Madigan, Paul E. Stang, Jesse A. Berlin, Martijn Schuemie, J. Marc Overhage, Marc A. Suchard, Bill Dumouchel, Abraham G. Hartzema, Patrick B. Ryan
- *The Role of Statistics in the Discovery of a Higgs Boson,* David A. van Dyk
- *Brain Imaging Analysis,* F. DuBois Bowman
- *Statistics and Climate,* Peter Guttorp
- *Climate Simulators and Climate Projections,* Jonathan Rougier, Michael Goldstein
- *Probabilistic Forecasting,* Tilmann Gneiting, Matthias Katzfuss
- *Bayesian Computational Tools,* Christian P. Robert
- *Bayesian Computation Via Markov Chain Monte Carlo,* Radu V. Craiu, Jeffrey S. Rosenthal
- *Build, Compute, Critique, Repeat: Data Analysis with Latent Variable Models,* David M. Blei
- *Structured Regularizers for High-Dimensional Problems: Statistical and Computational Issues,* Martin J. Wainwright
- *High-Dimensional Statistics with a View Toward Applications in Biology,* Peter Bühlmann, Markus Kalisch, Lukas Meier
- *Next-Generation Statistical Genetics: Modeling, Penalization, and Optimization in High-Dimensional Data,* Kenneth Lange, Jeanette C. Papp, Janet S. Sinsheimer, Eric M. Sobel
- *Breaking Bad: Two Decades of Life-Course Data Analysis in Criminology, Developmental Psychology, and Beyond,* Elena A. Erosheva, Ross L. Matsueda, Donatello Telesca
- *Event History Analysis,* Niels Keiding
- *Statistical Evaluation of Forensic DNA Profile Evidence,* Christopher D. Steele, David J. Balding
- *Using League Table Rankings in Public Policy Formation: Statistical Issues,* Harvey Goldstein
- *Statistical Ecology,* Ruth King
- *Estimating the Number of Species in Microbial Diversity Studies,* John Bunge, Amy Willis, Fiona Walsh
- *Dynamic Treatment Regimes,* Bibhas Chakraborty, Susan A. Murphy
- *Statistics and Related Topics in Single-Molecule Biophysics,* Hong Qian, S.C. Kou
- *Statistics and Quantitative Risk Management for Banking and Insurance,* Paul Embrechts, Marius Hofert

**Access this and all other Annual Reviews journals via your institution at www.annualreviews.org.**

**ANNUAL REVIEWS | Connect With Our Experts**

Tel: 800.523.8635 (US/CAN) | Tel: 650.493.4400 | Fax: 650.424.0910 | Email: service@annualreviews.org


Annu. Rev. Pathol. Mech. Dis. 2008.3:341-365. Downloaded from www.annualreviews.org
by University of Melbourne on 10/10/14. For personal use only.