Recent advances in molecular biology have led to the development of powerful tools for the study of medulloblastoma (MB) tumorigenesis, which have revealed new insights into the molecular and genetics basis of this disease. Throughout the life of an individual, the ~3 billion base pairs of DNA that constitute the human genome are exposed to mutagens and suffer errors in DNA replication; this assault leads to the acquisition of stable mutations through rounds of clonal selection and clonal expansion. The cancer genome, as a result of this process, is profoundly different from the genome that composes the constitutional DNA. The goal of cancer genomics is the study of the human cancer genome and the collection and description of all the alterations that make up for its divergence from the constitutional DNA. The mutations, inherited or acquired, contribute in different degrees to the development of cancer and its progression from a localized cancer to one that grows uncontrolled and then metastasizes. Recent studies based on high-throughput mutation detection techniques have shown that cancer genomes harbor from 10 to over 100 mutations [1–5]. However, these studies are likely to underestimate the alterations caused by genomic rearrangements and copy number alterations (CNA); this suggests that the number of genes hit by somatic mutations is higher. Cell homeostasis can also be disrupted by mechanisms beyond the changes in DNA sequence; changes in DNA methylation status or histone acetylation can alter physiological cell processes like cell division [2, 6, 7, 8, 9]. For this reason a comprehensive description of cancer genetics has to include studies at the level of the genome, epigenome, and transcriptome, as schematized in Fig. 1.

Medulloblastoma (MB) can occur in the setting of rare genetic syndromes characterized by increased incidence of malignancies, often at young age. Studies aimed to understand the genetic basis of such rare syndromes showed that the genes so far associated with predisposition to tumors appear to act as tumor suppressors; constitutional alterations associated with somatic loss or epigenetic silencing of the wild-type (wt) allele are observed in hereditary tumors, and often, biallelic somatic loss-of-function
(LOF) mutations and epigenetic silencing can be found in the corresponding sporadic tumors. Li-Fraumeni cancer family syndrome, Gorlin syndrome, and Turcot syndrome are commonly described conditions associated with an increased risk to develop MB.

### Li-Fraumeni Cancer Family Syndrome and TP53 Mutations in MB

In the early 1990s two groups reported germ line mutations in the TP53 gene as the possible cause for the increased susceptibility to cancer observed in families affected by the Li-Fraumeni syndrome (LFS). LFS is an autosomal dominant condition characterized by the occurrence of diverse cancers of mesenchymal and epithelial origin in multiple sites; affected families show particularly high rates of malignancy and a young age at onset of malignancies such as breast cancer, brain tumors, acute leukemia, soft tissue sarcomas, bone sarcomas, and adrenal cortical carcinoma [10, 11]. In terms of relative incidence, breast carcinomas are most frequent (24.0 %) followed by bone sarcomas (12.6 %), brain tumors (12.0 %), and soft tissue sarcomas (11.6 %) [12]. Among the brain tumors histologically analyzed (N=29), 69 % were of glial origin followed by a rate of 17 % for MBs and related primitive neuroectodermal tumors of childhood [12].

TP53 is a tumor suppressor gene that codifies for a 53 KDa protein which plays a key role in regulating crucial cellular processes deregulated during malignant transformation of the cells. P53 participates in DNA damage repair by holding the cell cycle at the G1/S regulation point on DNA damage recognition, allowing DNA repair.

<table>
<thead>
<tr>
<th>Genomic Alterations</th>
<th>Gene Expression Alterations</th>
<th>Non-coding RNA Alterations</th>
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<tr>
<td>Somatic Mutations</td>
<td>Gene Overexpression or Downregulation</td>
<td>miRNA Overexpression or Downregulation</td>
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<td>Gene Amplifications</td>
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<td>Gene Deletions</td>
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**Fig. 1**
processes to take place or signaling to sensor molecules in order to start an apoptotic program. During the G1/S checkpoint activated p53 binds DNA and activates expression of several genes, including WAF1/CIP1 encoding for p21. p21 forms complexes with other proteins important for the G1/S transition in the cell cycle, inhibiting cell cycle progression when internal or external conditions are unfavorable to cell division. A decreased or aberrant activity of p53 will no longer prevent cells bearing damaged DNA from dividing, leading to uncontrolled division, genomic instability, and tumor formation.

**TP53** mutations are also observed in sporadic MBs: early studies reported an incidence of mutation in 10% of MBs [13, 14]; however, a higher frequency has been described in anaplastic MBs that show an occurrence of 27% [15, 16]. TP53 immunoreactivity accounts for one of the most reproducible molecular markers of poor outcome in MB, together with overexpression of survivin or erbB2 and amplification of the MYCC oncogene. In a recent study Tabori and colleagues correlated the incidence of TP53 mutations and common biologic prognostic markers of MB outcome. The 5-year survival rate was negatively affected by TP53 mutation; also TP53-mutated tumors recurred earlier, and 75% of the recurrent tumors classified as average risk on the basis of WHO criteria showed to be mutated in TP53 [16]. From a histological perspective in the cohort of 108 patients analyzed in the study, 50% of the severely anaplastic MBs were TP53 mutated. However, in the same study severe anaplasia was not predictive of disease outcome. In contrast with the findings reported for other molecular markers associated with negative outcome, such as **PDGFRA**, TP53-mutated tumors recurred locally, in a short time frame, with no dissemination, suggesting a possible role of TP53 mutation in therapy resistance.

**Gorlin Syndrome and Mutations in the PTCH (Patched) Gene**

In 1960 Robert J. Gorlin and Robert W. Goltz described a genetic syndrome inherited in an autosomal dominant manner that greatly predispose to a skin cancer known as basal cell carcinoma. Patients affected by the Gorlin syndrome also present several abnormalities of the skin, skeleton, and nervous system, but more importantly they develop multiple cancers at an early age. About 90% of the patients are affected by multiple basal cell carcinomas, while MB occurs less frequently in 3–5% of affected patients [17, 18]. The peak incidence of MB in Gorlin syndrome-affected individuals is at approximately 2 years of age, while it is 7 years of age in the sporadic form of MB [19, 20]. The desmoplastic subtype is the only histopathologic subtype of MB reported in the current Gorlin syndrome population, while it accounts for no more than 20% of sporadic MB [19]. The locus containing the causative gene was mapped to chromosome 9q22.3. A combination of genetic linkage in five families affected by Gorlin syndrome supported by a loss-of-heterozygosity (LOH) study on sporadic basal cell carcinomas [21–23]. Fine mutational analysis of the locus 9q22.3 revealed alterations in the human homolog of the *Drosophila patched (ptc)* gene, whose reduced or absent expression was linked, respectively, to developmental abnormalities and tumorigenesis [24, 25]. The function of the *Drosophila* patched protein is well-studied: it acts as an inhibitor of the Hedgehog (HH) signaling pathway in the context of development. The HH signaling pathway is named after the family of extracellular HH ligands, of which there are three in mammals: Sonic Hedgehog (SHH), Indian Hedgehog (IHH), and Desert Hedgehog (DHH); **ptc** encodes for the receptor of such ligands and, in absence of ligand, it associates and represses the function of the seven-pass membrane protein Smoothened (SMOH) [26]. When PTCH1 is ligand-bound, repression of SMOH is relieved and one or more of the **GLI** transcription factors are activated leading to the modulation of the transcription of downstream target genes. Given the well-known role of the *Drosophila* homolog, human **PTCH1** was predicted and demonstrated to produce constitutive activation of **SHH** signaling after its biallelic inactivation; **PTCH1** fulfills the conditions to be defined as a classic tumor suppressor gene [27]. In sporadic MBs where no evidence of a familial predisposition can be asserted, there is a fraction...
of cases where mutations of the genes \textit{PTCH1} and \textit{SMO\textsubscript{H}} occur \cite{28-32}; \textit{GLI} gene family members, downstream effectors of the pathway, are instead amplified in glioblastoma \cite{33}. These observations from human dataset, fortified with data from loss-of-function studies in mice (\textit{Ptc\textsuperscript{\textast}} mice carrying one inactivated allele developed MB with a 30\% penetrance), helped to establish a role for aberrant SHH signaling in MB; more recently a whole transcriptome analysis on larger cohorts set around 25–35\% the fraction of human MBs characterized by SHH pathway activation \cite{34,35}.

\textbf{Turcot Syndrome and \textit{Wnt} Pathway}

The Canadian surgeon Jacques Turcot in 1959 described a recessive hereditary condition defined by the association between familial polyposis of the colon and brain tumors. Nowadays this rare genetic condition is counted under the category of constitutional mismatch repair-deficiency (CMMR-D) as it is caused by homozygous mutations or compound heterozygosity of the mismatch repair (MMR) genes \textit{MLH1}, \textit{MSH2}, \textit{MSH6}, or \textit{PMS2}. Similarly families affected by the autosomal dominant disorder, familial adenomatous polyposis (FAP), also develop brain tumors associated with other neoplasms \cite{36,37}. These two conditions show enough similarities to support the proposed re-denomination of brain tumor-polyposis syndrome type 1 and brain tumor-polyposis syndrome type 2 (BTP-1 and BTP-2) to indicate, respectively, the Turcot and the FAP syndrome \cite{38}. Early studies indicated that FAP is linked to Chr:5q15-q22 \cite{39,40}; in parallel LOF analysis on tumor tissues indicated how 20\% of sporadic colorectal adenocarcinomas lose one of the alleles on chromosome 5q \cite{41}, and extracolonic neoplasms also confirmed the role of hemizygous or homozygously mutated \textit{APC} gene in tumorigenesis \cite{42-44}. \textit{APC} gene mutations were also identified in 10 of 12 families with FAP in which at least one patient developed a central nervous system tumor, mainly MB (79\%), as an extracolonic manifestation of FAP \cite{45}. In a recent study on 28 BTP subjects, MB was the most common brain tumor, with young females under the age of 20 most frequently affected \cite{46}. \textit{APC} exerts its tumor suppression function by inhibiting the activation of the \textit{Wnt} signaling pathway preventing the uncontrolled growth of cells and, in turn, the tumorigenic process. The \textit{Wnt} signaling pathway was at first described in Drosophila, where its components regulate embryonic development, cell differentiation, and cell polarity generation. Core elements of the signaling pathway include the \textit{\textbeta}-catenin destruction complex” composed of scaffold proteins like \textit{APC} and \textit{AXIN}, whose role is to cage \textit{\textbeta}-catenin in the cytoplasm and direct it to proteasomal degradation; the interaction between \textit{\textbeta}-catenin and the complex is regulated by a protein kinase like GSK3-\textbeta and CKI. In the presence of Wnt ligands, the destruction complex is destabilized, releasing \textit{\textbeta}-catenin to enter the nucleus where it binds TCF/LEF family transcription factors to promote specific gene expression regulating cell cycle progression, apoptosis, and differentiation \cite{47,48}. A hint about the role of \textit{Wnt} and its signaling pathway in cancer came from the discovery of the first \textit{Wnt} gene: mice infected with mouse mammary tumor virus (MMTV) developed tumors; the provirus insertion sites were clustered around a locus on Chr:15 containing a putative oncogene, \textit{Int1} (integration 1), that was activated as consequence of viral integration \cite{49}. The putative oncogene \textit{Int1} was then named \textit{Wnt1} after its sequence homology with the Drosophila \textit{Wg} gene. The high incidence of MB in BTP-2 patients has pointed toward the presence of abnormalities in the \textit{Wnt} pathway; also in sporadic MB, about 15\% harbor mutations of pathway components \textit{APC} and \textit{CTNNB1} (encoding for b-catenin) \cite{50-53}. A study conducted on 109 MB patients in 2005 showed that about 25\% of the tumors displayed \textit{\textbeta}-catenin nuclear staining, indicative of \textit{Wnt} pathway activation. Nucleo-positive tumors were associated with a better overall survival and event-free survival (OS and EFS) than negative tumors; interestingly \textit{\textbeta}-catenin nucleo-positivity was evident among classic and anaplastic MBs. Mutations in the GSK-3\textbeta phosphorylation domain of \textit{CTNNB1} were associated with aberrant nuclear accumulation.
of β-catenin; notably these missense mutations frequently occurred in residues altered also in other cancers. In the majority of non-MB cancers, APC is inactivated by the effect of truncating mutations or deletions, but only missense mutations have been found in MB; if these mutations are sufficient to induce nuclear localization of β-catenin, with activation of the pathway, is debatable [52, 53].

The study of MB associated with cancer-predisposing genetic syndromes has shed light on the genetic pathways involved in MB development, confirming the early assumption, derived from histological observation of the tumor tissue, that MB is a disease of development. As other signaling pathways crucial for cerebellar development were screened for their role in MB, in a gene-candidate approach, it became evident that unbiased whole genome strategies would complement and broaden the pool of MB cancer genes, dramatically increasing the understanding of MB pathogenesis.

Our comprehension of the genetic base of MB has indeed progressed hand in hand with the refinement of high-throughput technologies, which have reached unprecedented resolution and sensitivity in the past 10 years.

**MB Genome: A Low-Resolution Picture**

The first report of G-banded karyotypes of MBs covered seven tumors and sought to determine if specific chromosomal abnormalities characterized the tumors; the predominant findings in the karyotyped samples were structural abnormalities consisting of both deletions and unbalanced translocations in what appeared a near-diploid karyotype [54]. Thus, even at a low-resolution analysis, human MB karyotypes were found to be profoundly different from malignant gliomas, as MB contain mainly structural alterations of chromosomes 1, 3, 17, and 20, usually resulting in partial trisomy, whereas the most frequent abnormalities in malignant gliomas are gains of whole copies of chromosome #7 and losses of 10 [55, 56]. The most common abnormality found consistently in MB by several groups was isochromosome 17q [i(17q)]; this rearrangement resulted in trisomy for 1q and monosomy for 17p [54, 57, 58]. An i(17q) is also commonly found in chronic myeloid leukemia; a translocation of genetic material between chromosomes 15 and 17, t(15;17), is instead typical of acute promyelocytic leukemia; this alteration fuses part of the PML gene from chr15 with part of the RARA gene from chr17. The complex structure of chromosome 17 predisposes it to a form of homologous recombination that occurs between two segments of DNA that display high sequence homology but are not on two different alleles; this results in DNA rearrangements that cause several disorders. Chromosome 17 contains between 2.5% and 3% of the total DNA in cells and about 1,200 protein-coding genes, among them are genes involved in early onset breast cancer (BRCA1), neurofibromatosis (NF1), and the DNA damage response (TP53 encoding the p53 protein) [59, 60]. Even though i(17q) is the most common MB abnormality, unique in certain tumor samples, the gene or gene combination driving tumorigenesis has remained elusive. Cytogenetic studies can also detect gene amplification; frequently these events appear as small fragments of extra-chromosomal DNA called double minutes (DM), and often these DNA lengths harbor oncogenes or gene related to drug resistance [61]. Human MB-derived cell lines analyzed at early passages as well as xenografts harbored amplification of the MYC oncogene [62–64]. The MYC gene is part of a gene family (MYC, MYCN, and MYCLI) that accounts for the most prevalent target of gene amplification in MB; the MYC gene maps to chr8q24 and encodes for a transcription factor that regulates genes involved in cell division, cell growth, and apoptosis [65–67]. The advent of comparative genomic hybridization (CGH), a molecular-cytogenetic method for the evaluation of copy number changes (gains/losses) in the DNA content of tumor cells, has revealed previously uncharacterized alterations in MB genome. Consistent gains on chromosomes 7 and 17q as well as losses on 10q, 11, 16q, 17p, and 8p were reported, together with the well-known i(17q) and gross losses of chromosome 1, 3, and 20. When stringent methods of statistical analysis
were applied, CGH data suggested a much higher degree of genomic imbalance in MB than has been possible to observe with lower-resolution techniques [68, 69]. However, an intrinsic limitation of CGH is that it will detect only chromosomal changes that alter the copy number; aberrations such as balanced reciprocal translocations or inversions cannot be detected, as they do not change the copy number. Although a technique that fuses fluorescence in situ hybridization (FISH) and CGH, known as spectral karyotyping, allows the detection of subtle genomic rearrangements like balanced translocation, it was not until the advent of high-resolution techniques that the full extent of MB genome aberrations was shown in detail [70].

**MB Genome: Lessons from High-Resolution Studies**

The most advanced tools available to omics scientists to detect genome abnormalities in the submegabase range are digital karyotyping, array comparative genomic hybridization (aCGH), single nucleotide polymorphism (SNP) arrays, and next-generation sequencing (NGS). Three independent groups have applied digital karyotyping to MB genomics; two cell lines D487Med and D425Med were analyzed, and both showed amplification of the OTX2 gene. The analysis of human tumors confirmed that OTX2 was overexpressed in anaplastic MB [71, 72]. More recently, OTX2 was found to be focally amplified in a large cohort of MB samples analyzed by high-density SNP array. OTX2 protein acts as a transcription factor and plays a role in forebrain development; in the cerebellum OTX2 is expressed in proliferating granule cell precursors [73]. Nakahara and colleagues instead applied digital karyotyping and high-density SNP arrays to identify a rare, recurrent homozygous deletion of Kruppel-like factor 4 (KLF4); KLF is a well-characterized tumor suppressor gene in colonic, gastric, and pancreatic carcinoma [74–77]. Several groups have pursued a genome-wide survey of the oncogenes aberrantly expressed in MB through aCGH; gains of several candidate oncogenes as MET and CDK6 have been found to be amplified in 38.5 % and 30 % of the analyzed cases [78, 79]. Although, in light of the recent advances in MB molecular classification, the traditional picture based on five distinct histological subgroups (classic, desmoplastic, anaplastic, large-cell, and MB with extensive nodularity) has lost a prime role in patient stratification, it is still worth of notice that genomics approaches have shed some light on the basis of the different histological presentations of MB. In a 2006 study, Ehrbrecht and colleagues investigated the genomics of desmoplastic MBs by CGH on a cohort of 22 sporadic cases followed by aCGH on a subset of samples that showed 9p and 17q22-q24 amplification. Among others, JMJD2C amplified on 9p24 was subsequently confirmed in an independent cohort [80, 81]; this gene encodes for a histone lysine demethylase that plays a key role in the posttranslational modification of histones and thus regulating the functional chromatin organization, a critical step in epigenetic regulation. As highlighted in the first part of this chapter, many developmental pathways are potentially involved in the MB pathogenesis; however, the genetic determinants of the deregulation still remain poorly characterized. The Wnt pathway is one of the signaling cascades that regulate cerebellar development; in the effort to understand the genetic base of the Wnt pathway alteration in MB, two independent studies have used genomics approaches to identify potential mechanisms [35, 82]. Monosomy of chr6 stood out as the hallmark of MBs harboring nuclear β-catenin, CTNNB1, or APC mutations; the alteration has been consistently observed in following studies making monosomy of chr6/CTNNB1 a genomic marker of Wnt pathway tumors that are consistently associated with a highly favorable prognosis [83, 84]. When the power of molecular techniques is applied to cohorts of an adequate number of samples with robust clinical follow-up, genomic data can be used to stratify patients in risk class based on their molecular profile, beyond the classic histological classification. The study by Pfister and colleagues pursued this goal with a cohort of 80 primary MBs and a validation set of 296 independent tumor samples; gain of chr6q, amplification
of MYC and MYCN, and an isolated gain of 17q and i(17q) were all associated with a poor clinical outcome; the work also confirmed loss of 6q as indicator of good prognosis. The proposed molecular strategy was based on four markers: MYC/MYC amplification and 6q loss as indicators of worse and of best outcome, respectively, and 6q gain, 17q gain, and 6q/17q balanced to further stratify the patients from poor to good prognosis [67]. As the study of the MB genome progressed, in the last decade it became clear how the bottleneck for a comprehensive picture of MB aberrations depended on the cohort size and the resolution of the platform utilized in the study. In their 2009 study, Northcott and colleagues collected an unprecedented number of tumor samples, 201 primary MB and 11 MB cell lines, and analyzed their genomes using high-resolution SNPs genotyping arrays [85]. The innovative technology allowed a resolution of at least an order of magnitude higher than any previous array-based study of the MB genome. This led to the identification of hundreds of high-level amplifications and homozygous deletions, but unexpectedly only a few amplifications and homozygous deletions were found in more than one sample. Narrowing down the analysis on one of the most interesting deletions, chr 9q34 which is among the more common alterations in MB, a single gene mapped to the minimal deleted region: EHMT1. The gene encodes for a euchromatic histone (H3K9) methyltransferase which is part of a protein complex that mediates epigenetic silencing by demethylation of H3K9; further functional data in the study confirmed the key role of EHMT1 as a tumor suppressor gene in MB [86]. In early 2011 a seminal study focusing on the genome-wide analysis of MB was published; the authors used a combination of next-generation sequencing on protein-coding exons and high-density SNP array on a discovery cohort of 22 MBs, 17 primary tumors, 4 tumors propagated as xenograft in nude mice, and one cell line. When analyzed at this unprecedented depth, the MB genome revealed an average of 11 alterations. PTCH1, CTNNB1, MYC, PTEN, TP53, and OTX were commonly mutated across different samples confirming the role of the SHH and Wnt pathway in MB pathogenesis; other genes as MLL2, SMARCA4, and MLL3 indicated that mechanisms controlling chromatin remodeling and transcriptional regulation are frequently deregulated in MB [4] (Table 1).

### Table 1: Most prominent CNA in MB genome

<table>
<thead>
<tr>
<th>Gains</th>
<th>Losses</th>
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<tr>
<td>Chromosome</td>
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<tr>
<td>17q</td>
<td>10q</td>
</tr>
<tr>
<td>17p</td>
<td>11p</td>
</tr>
<tr>
<td>i(17q)</td>
<td>8p</td>
</tr>
<tr>
<td>7</td>
<td>16q</td>
</tr>
<tr>
<td>2p</td>
<td>9q</td>
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<td>1q</td>
<td>17p</td>
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<td></td>
<td>6</td>
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**MB Transcriptome**

The collection of all mRNA transcripts in the cell, which are products of genes that are being actively expressed at any given time, constitutes the transcriptome. Several strategies have been employed to analyze comprehensively the MB transcriptome, from early serial analysis of gene expression (SAGE) approaches to more recent expression profile platforms. The first study which focused on the comparison of the MB transcriptome versus its normal counterpart, fetal brain at 24.5 weeks, was accomplished in 1999 by Michiels and colleagues. The authors compared mRNA from the two sources and found 138 genes with significant differential expression, two of these differentially expressed genes, ZIC1 and OTX2, are usually expressed in the external granular layer of the cerebellum as well as in the subventricular zone, thus confirming the early assumptions that MB arises in pluripotent proliferating cells in the germinai areas of the cerebellum [87]. About 30% of MB cases are metastatic right at the moment of diagnosis, and metastases has been a well-characterized negative prognostic factor, indicative of a high-risk patient. In 2001 an expression profile study offered the scientific community with some molecular actors distinctive of metastatic MB. The study comprised 23 primary MBs (M+ or M0) whose analysis made...
recently offered also a comparison of the genome aberrations that discriminate MBs from PNETs [92, 93]. Following the McDonald study in using molecular profiling to understand the differences across MBs with distinctive clinical features, Pomeroy et al. also compared the profiles of desmoplastic MBs versus classic MBs, confirming that the SHH signaling pathway plays a crucial role in desmoplastic MBs. The study was also pivotal in supporting the use of molecular profiling of human tumors to obtain indications about disease prognosis; the authors extrapolated a gene signature based on eight genes that successfully predicted the survival status for 47 of the 60 patients profiled. Many studies followed along the lines indicated by Pomeroy and McDonald studies; among them is worth to mention, the informative expression profile of 35 MBs that were generated by Neben and colleagues, to identify transcripts associated with patient outcome. Of note, the authors performed an mRNA expression analysis followed by validation on tissue macroarray (TMA), defining a STK154 as a negative prognostic marker of overall survival [94].

**MB Molecular Subgroups: More Than One Disease**

MB is the most common pediatric brain tumor, but it is still a rare disease compared to other childhood cancers. The necessity to collect large cohorts of samples has hindered the understanding of subtle molecular differences among different MB tumor samples for a long time; these molecular differences are now offering new and exciting perspectives on MB pathogenesis and progress. In their study from 2006 Thompson and colleagues were the first to apply unsupervised hierarchical clustering to a dataset of 46 MBs. They analyzed the tumor’s expression profile and used the most differentially expressed genes to define five molecular subgroups of MB. Notably, the authors also showed that tumors harboring a CTNNB1 mutation or monosomy of Chr 6 were also showing altered expression of Wnt pathway genes; the same can be said for mutations in PTCH1, which always correlate with alterations in SHH pathway genes [35]. Along the same line as the Thompson work, Kool and colleagues further explored the potential of molecular classification of MB in a dataset of 52 cases. The authors confirmed the five molecular subgroups previously identified but also made some unprecedented correlations between molecular subgroups and clinical features. The non-SHH/-Wnt subgroups were found to be correlated with the presence of metastatic disease, supporting the potential of patient stratification based on molecular signatures [84]. Ultimately the understanding of MB genomics increases proportionally with the size of the tumor cohorts analyzed; a glimpse of the future potential of such studies come from two recent works from
two different groups. Northcott and colleagues recently contributed to the definition of MB sub-

groups performing gene expression profiles and DNA copy number aberrations for 103 primary MBs. Through a combination of different bio-

informatics approaches based on the most informa-
tive genes in the dataset, the authors defined four different molecular subgroups, Wnt, SHH, C, and D. These findings were confirmed by the way of immunohistochemistry in a large dataset of 294 MBs. The molecular subgroups showed specific demographics, histology, metastatic status, and DNA copy number aberrations. To translate the results in a flexible tool that could be applied rou-
tinely in pathology laboratories, the authors also characterized and refined a protocol based on four antibodies: DKK1 (WNT), SFRP1 (SHH), NPR3 (group C), and KCNA1 (group D) [34]. The vast majority of the tested samples stained uniquely for one of the four antibodies, allowing the reliable classification of formalin-fixed MBs. One of the most important finding of the study was the revision of clinical features and their relation with patients’ prognosis. The metastatic status at the moment of diagnosis, which has been a well-established hallmark of poor outcome, was found to be not reliable in group C patients (NPR3-positive tumors) who exhibited a significantly diminished progression-free and overall survival irrespective of their metastatic status. Cho and colleagues, analyzing a dataset of 194 MBs by high-density SNPs array and by expression profile, further honed the molecular classification proposed by Northcott et al. They showed how the less-resolved molecular subgroups (C and D) could be further subtyped; the especially bad prognosis of the newly defined molecular group characterized by MYC amplification and transcriptionally by enrichment of photoreceptor pathways clearly indicates that MB molecular classification is necessary to develop new therapeu-
tic strategies [65].

In 2012 a genetic molecular profiling on 1,000 MBs samples identified four prominent subgroups [95]. These pioneer studies driven by a clonal genetic selection describe each group origin. Group 1 tumors show mutations in SHH and its receptors; group 2 tumors are driven by changes in Wnt signaling, generally through its main effector gene, β-catenin; group 3 tumors are driven by changes in TGFβ–OTX2 signaling; and group 4 tumors are driven by mutations in MYC and MYCN [96]. The molecular signature followed by next-generation sequencing (NGS) studies now are undergoing at international level effort, being prepared to be integrated with path-

ological classifications thus gathering clinical prognostication with medulloblastoma molecular subgrouping [97].

### Profiling MB Transcriptome: Noncoding RNA Alterations

A new class of genes producing small noncoding RNAs, the microRNAs (miRNAs), has been dis-

covered over the last recent years. These short RNAs (18–24 nucleotides) bind to cis-regulatory elements mainly present in the 3’ UTR of mRNAs, resulting in translational inhibition or mRNA degradation [98, 99]. In mammals, miRN-\n
As predicted to control the activity of ~50% of all protein-coding genes. Functional studies indicate that miRNAs participate in the regulation of almost every cellular process investigated so far and that changes in their expression are associated with many human pathologies [100, 101]. MicroRNAs could be useful tools to control cancers; in fact, they might be able to fulfill this task through their simultaneous control of multiple target genes. MiRNAs have been linked to the initiation, progression, and metastasis of human malignancies, with some species display-
ing oncogenic and others tumor suppressive potential [102, 103]. They are often expressed aberrantly in tumors as compared to normal tis-

sues and are likely to contribute to tumorigenesis by deregulating critical target genes [104–106]. Several miRNAs have been studied in MB, as listed in Table 2; the entries on this list are continuously growing with the number of studies published.

The miR-17/92 polycistron has been found recurrently amplified in 6% of pediatric MBs by the way of profiling the expression of 427 mature miRNAs in a series of 90 primary human
Table 2  Noncoding RNA alterations in MB

<table>
<thead>
<tr>
<th>Noncoding RNAs</th>
<th>Noncoding RNA alterations and their functions in MB</th>
<th>References</th>
</tr>
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<tbody>
<tr>
<td>miR-124</td>
<td>Targets CKD6 and regulation of cell cycle, its expression is inhibited in MB</td>
<td>[118]</td>
</tr>
<tr>
<td>miR-324-5p, miR-326, miR-19a, miR-20, miR-92</td>
<td>Hedgehog-dependent proliferation, downregulated in MB</td>
<td>[112]</td>
</tr>
<tr>
<td>miR-199b-5p</td>
<td>Targets Hes1, impairment of cancer stem cells CD133+, silenced in MB</td>
<td>[109]</td>
</tr>
<tr>
<td>miR-let7g, miR-9, miR-106b, miR-125a-b, miR-191</td>
<td>Regulation of proliferation and apoptosis of MB cells, found upregulated in MB</td>
<td>[117]</td>
</tr>
<tr>
<td>miR-17-92</td>
<td>Overexpressed in the Shh subgroup of MBs and elevated levels of MYC/MYCN expression</td>
<td>[66, 107]</td>
</tr>
<tr>
<td>miR30b, miR30d</td>
<td>Targets of a novel recurrent MB amplification at 8q24.22-q24.23</td>
<td>[120]</td>
</tr>
<tr>
<td>miR128</td>
<td>Targets Bmi-1 and inhibition of MB cell growth</td>
<td>[121]</td>
</tr>
<tr>
<td>miR34a</td>
<td>Targets Notch ligand Delta-like 1(Dll1), impairment of cancer stem cell compartments</td>
<td>[111]</td>
</tr>
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</table>

MBs [107, 108]. The components of this cluster (miR-92, miR-19a, and miR-20) are the most highly upregulated miRNAs in MB; of note their expression was highest in the subgroup of MBs associated with the activation of the SHH signaling pathway. In the subset of MBs with miR-17/92 upregulation, the authors also noted elevated levels of MYC/MYCN. Consistently with Shh regulation, the Shh treatment of primary murine cerebellar granule neuron precursors (CGNP), cells of origin of the SHH-associated MBs, resulted in increased miR-17/92 expression. In CGNPs, the Shh effector N-myc, but not Gli1, induced miR-17/92 expression. Furthermore, ectopic miR-17/92 expression in CGNPs synergized with exogenous Shh to increase proliferation and also enabled proliferation in the absence of Shh. MiR-17/92 is a positive effector of Shh-mediated proliferation, and aberrant expression/amplification of this miRNA confers a growth advantage to MBs [107]. Northcott and colleagues identified a focal amplification on chromosome 13q31.3, which mapped to the same miRNA cluster. The expression of miR-17/92 was confirmed to be most elevated in MBs of the Shh subgroup and was also associated with elevated MYC/MYCN levels. These studies suggest that aberrant expression of miRNAs, encoded by the miR-17/92, enhances the growth potential of MB and that miRNA-mediated modulation of Hedgehog signaling may be an important contributing factor to MB pathogenesis [108].

Our group discovered for the first time an miRNA deregulated in MB that targets the Notch pathway and depletes the tumor stem cells quota [109]. We identified miR199b-5p as targeting HES1, the principal Notch effector, demonstrating a new mechanism of regulation of the Notch pathway. The miRNA 199b-5p inhibits HES1 expression by binding to its 3'UTR. It is known that HES1 plays a crucial role in MB biology because high levels of HES1 protein expression correlate with negative outcome in MB patients [110]. Furthermore, the miRNA 199b-5p affects MB cancer stem cells by decreasing the side population (SP) quota as well as the CD133+ cells compartment. In MB, the expression of the neural stem cell marker CD133 has been associated with both tumor initiation capacity and radioreistance. MiRNA 199b-5p was found downregulated in MB patients’ tissues compared to healthy control cerebellum tissues. The analysis of 61 patients with MB showed that the expression of miR-199b-5p in the nonmetastatic cases was significantly higher than in the metastatic cases. The correlation with survival for the patients with high levels of miR-199b expression showed a positive trend to better overall survival than for the low-expressing patients. We further showed that in a xenograft mouse model, MB tumor burden is reduced by miRNA 199b-5p, indicating the use of this miRNA as an adjuvant therapy after surgery, in combination with radiation and chemotherapy, for the improvement of anticancer MB therapies and patient quality of life. To date,
The interest in miRNAs role in MB pathogenesis has been increasing over the last few years; the availability of high-throughput approaches to comprehensively profile miRNA expressions has allowed to investigate miRNAs involvement in MB carcinogenesis and their prognostic relevance. Specific miRNA expression patterns were correlated to MB histotypes (anaplastic, classic, and desmoplastic), to specific sets of molecular alterations (ErbB2- or MYC-overexpressing tumors), and to disease-risk stratification [111].

The group headed by Gulino studied miRNAs in the context of SHH signaling; miR-125b, miR-326, and miR-324-5p expression were found decreased in MB; the altered expression of these miRNAs led to tumor cell proliferation through an SHH-dependent mechanism. They identified 78 miRNAs with altered expression in MB compared with normal adult and fetal cerebellar cells. Several of the identified miRNAs have been implicated in other cancer types including glioblastoma [113]. The majority of these miRNAs were found downregulated in MB, supporting a role for miRNAs as tumor suppressors. Additionally, they found increased expression of miR-9 and miR-125a whose increased expression was capable of decreasing proliferation, augmenting apoptosis, and ultimately promoting arrest of tumor growth. The pro-apoptotic effect was mediated by miR-9 and miR-125a targeting of the TrkC receptor, which was found in this study to be upregulated in MB cells. They also found that miR-let-7g, miR-19a, miR-106b, and miR-191 were significantly upregulated in anaplastic compared with desmoplastic MBs; miR-let-7g and miR-106b were differentially expressed in desmoplastic compared with classic MBs. Changes in the expression of Her2 (ErbB2) and MYC have been demonstrated to impact biological activity and clinical features of MB [114–116]. The Gulino team additionally examined miRNAs expression from MBs overexpressing either Her2 or MYC and identified an miRNA signature in the two groups of MBs. The expression of miR-10b, miR-135a, miR-135b, miR-125b, and miR-153 was altered in Her2-overexpressing tumors, whereas MYC-overexpressing MBs had expression changes in miR-181b, miR-128a, and miR-128b. Additionally, the amount of expression change of two miRNAs correlated with disease risk. miR-31 and miR-153 were found downregulated in all MBs; the degree of change was directly proportional to disease severity [117]. Although these data are of interest, the identified signature would be of further importance as validation experiments in an independent cohort will be performed.

An additional study was related to miR-124, which is preferentially expressed in differentiating neurons and in CGNPs which are thought to be the cells of origin of MBs [118]. MiR-124 deregulation is common in MBs, and the restoration of its function inhibits cell proliferation, suggesting that it may act as a growth suppressor. Two targets of miR-124 have been identified: cyclin-dependent kinase 6 (CDK6) that was identified as an adverse prognostic marker in MB and SLC16A1 that may represent a novel therapeutic target for the treatment of malignant MBs [118]. SLC16A1, solute carrier family 16, functions to efflux lactic acid during aerobic glycolysis, and its inhibition resulted in a decrease of intracellular pH to a lethal level. This study demonstrates that miR-124 deregulation is common in MB, and the restoration of its function inhibits cell proliferation, suggesting that miR-124 may act as a growth suppressor, raising the possibility that the miR-124/SLC16A1 pathway may represent a novel therapeutic target for the treatment of malignant MBs [119].

Further recent work demonstrated that miR-30b and miR-30d are amplified in MB and are putative oncogenic targets of a novel recurrent MB amplicon at 8q24.22-q24.23 [120]. miR-128a inhibits the growth of MB cells by targeting the Bmi-1 oncogene. This miRNA alters the
intracellular redox state of the tumor cells and promotes cellular senescence. MiR-128a has growth suppression activity in MB cell lines, and this activity is partially mediated by targeting Bmi-1. This finding has implications for the modulation of redox states in cancer stem cells, which are thought to be resistant to therapy due to their low ROS states [121].

At this time, we can conclude that despite the amount of investigation related to miRNAs in MB genetics is still at the beginning stage.

### Profiling MB Transcriptome: Epigenetic Alterations

Epigenetic changes have been shown to be key players in tumorigenesis; they are defined as heritable changes in gene expression that are not accompanied by modifications in primary DNA sequence. Epigenetic modifications include DNA methylation on cytosine residues, most often in the context of CpG dinucleotides, as well as post-translational modification of histone proteins, such as methylation, acetylation, and phosphorylation [122–124]. Hypermethylation of CpG islands located at the 5′ end of genes has been reported in most cancers, either alone or in combination with genetic alterations (as gene deletions or mutations); hypermethylation of CpG islands overall can contribute to tumor suppressor gene silencing [123]. The study of epigenetic changes in MB offers significant potential for an improved understanding of its genetics [125, 126]. Several studies have suggested that multiple loci undergo changes in their methylation status during MB pathogenesis [85, 125, 126]. Among the earliest studies to implicate aberrant promoter methylation in MB on a global scale is the study by Frühwald and colleagues who used restriction landmark genomic scanning to analyze DNA methylation patterns in 17 primary MBs and 5 MB cell lines. Using this method, the authors identified methylation in up to 1% of all CpG islands in primary tumors and up to 6% in MB cell lines [125]. In addition, an association between hypermethylated sequences in MB and a poor prognosis were implied. Collectively, these findings provided early evidence that epigenetic events are likely to play a role in MB.

In another study, using microarray-based differential methylation hybridization, Waha and colleagues identified hypermethylation of the SCG5 (secretory granule, neuroendocrine protein 1) gene in 16 (~70%) of 23 primary MBs and 7 (~87%) of 8 MB cell lines [127].

The expression of SCG5 was found to be downregulated in the majority of primary samples and cell lines as compared with normal cerebellar controls, and SCG5 transcription was restored in cell lines treated with the demethylating agent, 5-aza (5-aza-2′-deoxycytidine). Furthermore, the reexpression of SCG5 in the D283Med cell line resulted in growth suppression and reduced colony formation, suggesting that SCG5 may be a putative tumor suppressor gene in MB. A link between histone methylation genes and MB has also previously been hypothesized based on the observation that copy number alterations affecting chromosomal regions containing histone methyltransferases or demethylases occur in a subset of MBs [85, 128]. It is known that deregulation of Wnt signaling occurs in up to 20% of MB. Kongkham et al., using a genome-wide approach, identified the secreted frizzled-related protein 1, 2, and 3 (SFRP1, SFRP2, and SFRP3) family of Wnt inhibitors as putative tumor suppressor genes silenced by promoter region methylation in MB. SFRP1, SFRP2, and SFRP3 expression increased after 5-aza-2′-deoxycytidine treatment. SFRP1, SFRP2, and SFRP3 methylation was identified in 23.5%, 3.9%, and 15.7% of primary MB specimens, respectively, by methylation-specific PCR; SFRP1 was expressed at levels twofold lower than that in the normal cerebellum. In MB, the loss of Wnt pathway inhibition due to SFRP gene silencing demonstrates an additional mechanism that may contribute to the deregulation of Wnt signaling [129]. RASSF1A (RAS association
domain gene) regulates cyclin D1 expression, which is important in controlling the cell cycle. In contrast to other malignancies, hypermethylation of RASSFIA in MB is not accompanied by allelic loss of 3p21.3 or mutation, indicating that biallelic loss is the primary mechanism of inactivation of RASSFIA [130].

CASP8 is a cysteine protease involved in death-receptor-mediated apoptosis [131]. Promoter hypermethylation of CASP8 leading to loss of CASP8 mRNA expression induces resistance to apoptosis induced by tumor necrosis factor-inducing ligand (TRAIL). In primary tumors, aberrant promoter methylation of CASP8 was seen most frequently in classic and anaplastic MBs and is an independent prognostic factor of poor outcome.

Transcriptional silencing of SGNE1/7B2, a gene located on 15q11-15, occurs predominantly in classic MB. SGNE1 is a calcium-dependent serine protease that inhibits tumor cell proliferation. Another gene, ZIC2, a zinc-finger transcription factor essential for the development of the central nervous system was found downregulated in MBs [132]. Pfister and colleagues developed and applied a technique known as array-based profiling of reference-independent methylation status (aPRIMES) to globally survey DNA methylation patterns in the MB genome. They showed a striking association between samples classified as either “low methylators” or “high methylators” and patient outcome, with the high-methylator group exhibiting reduced overall survival. In addition, the C2H2-type zinc-finger protein ZIC2 was identified as a hypermethylated candidate using aPRIMES and was subsequently confirmed to be epigenetically silenced in a panel of primary MB by using a combination of pyrosequencing and quantitative RT-PCR analysis. In total, three other members of the S100 gene family were found to be aberrantly methylated in 10–20% of MBs [133]. Hypermethylation and silencing of S100A6 are associated with the large-cell anaplastic subtype of MB. The pro-metastatic gene S100A4 is a direct target of ErbB2 signaling and is associated with a poor prognosis in MB. MCJ, a member of the DNAJ protein family that influences chemotherapy resistance, was found inactivated by biallelic hypermethylation, but hypermethylation of one allele also occurs in combination with genetic loss of the second allele.

The 17p13.3 locus, the most common genetic defect in MB, contains the gene hypermethylated in cancer 1 (HIC1), a transcriptional repressor that is a frequent target of epigenetic gene silencing in MB [134]. The Ptc1 gene is a well-characterized tumor suppressor in MB; its compound Ptc1/Hic1 heterozygotes display a fourfold increased incidence of MB. Hic1 is a direct transcriptional repressor of Atonal Homolog 1 (Atoh1), a pro-neural transcription factor essential for cerebellar development [134].

An epigenome-wide screen in MB cell lines, using 5-aza-2 deoxycytidine to find genes aberrantly silenced by promoter hypermethylation, identified an inhibitor of HGF/MET signaling, serine protease inhibitor Kunitz-type 2 (SPINT2/HAI-2), as a putative tumor suppressor silenced by promoter methylation in MB [129]. SPINT2 gene expression was downregulated, and MET expression was upregulated in 73.2% and 45.5% of tumors, respectively. SPINT2 promoter methylation was detected in 34.3% of primary MBs examined by methylation-specific PCR. SPINT2 reexpression in MB cell lines reduced proliferative capacity, anchorage-independent growth, cell motility in vitro, and increased overall survival times in vivo in a xenograft model. These data support the role of SPINT2 as another putative tumor suppressor gene methylated in MB [129]. The crucial role of epigenetic changes in MB was further underlined by Ecke and colleagues who showed reduced tumor incidence in heterozygous Ptc1−/− mice after reduction of endogenous DNA methyltransferase 1 (Dnmt1) activity. This reduction was achieved by a combined treatment with the Dnmt inhibitor 5-aza-2 deoxycytidine (5-aza-dC) and the histone deacetylase (HDAC) inhibitor valproic acid (VPA). The combination of the two drugs
efficiently prevented MB formation, whereas monotherapies with either drug were less effective. Wild-type Ptc1 expression was efficiently reactivated in tumors by 5-aza-dC/VPA combination therapy. This was associated with reduced methylation of the Ptc promoter and induction of histone hyperacetylation suggesting the inhibition of HDACs in vivo. However, the treatment was not effective in advanced-stage tumors. This was the first in vivo demonstration that showed that targeting of Dnmt and HDAC activities is highly effective in preventing the formation of Ptc-associated tumors [135].

Although loss of 17p13 has been often associated with p53 genetic alteration or HIC1 gene hypermethylation, other tumor suppressor genes located in this region such as KCTD11REN (KCTD11) are potentially lost in human MB, in part by LOH and in part by uncharacterized epigenetic events [136]. Using a panel of 177 human tumor samples and their normal matching samples representing 18 different types of cancer, it was showed that the downregulation of KCTD11 protein level is a specific and a diffusely common event in tumorigenesis. Additionally, it was identified that a CpG island and several Sp1 binding sites on KCTD11 promoter, Sp1 transcription factor, and DNA methylation contribute, at least in part, in the regulation of KCTD11 expression [136]. KLF4 expression is also lost in more than 40% of primary MBs both at the RNA and protein levels. MB cell lines drastically increase the expression of KLF4 in response to the demethylating agent 5-azacytidine and demonstrate demethylation of the promoter CpG islands by bisulfite sequencing. Methylation-specific PCR, targeting the KLF4 promoter, demonstrates CpG methylation in approximately 16% of primary MBs. KLF4 is inactivated by either genetic or epigenetic mechanisms in a large subset of MBs, and it likely functions as a tumor suppressor gene in the pathogenesis of MB [75].

A genome-wide method for detecting regions of CpG methylation on the basis of the increased melting temperature of methylated DNA, termed denaturation analysis of methylation differences (DAMD), was developed by Diede S. J. and colleagues. They found common regions of cancer-specific methylation changes in primary MBs in critical developmental regulatory pathways, including Shh, Wnt, retinoic acid receptor (RAR), and bone morphogenetic protein (BMP). One of the commonly methylated loci is the PTCH1-1C promoter that is methylated in both primary patient samples and human MB cell lines. Treatment with the DNA methyltransferase inhibitor 5-aza-2′-deoxycytidine (5-aza-dC) increases the expression of PTCH1 and other methylated loci. This data demonstrate that epigenetic silencing by DNA methylation of PTCH1 contributes to the formation of this childhood cancer and suggests the use of DNA demethylating agents as a potential strategy for therapy [137].

Of particular interest are the findings of Northcott and colleagues that, using high-resolution SNP genotyping of 212 MBs, identified previously unknown amplifications and homozygous deletions, including recurrent, mutually exclusive, highly focal genetic events in genes targeting histone lysine methylation, particularly that of histone 3 lysine 9 (H3K9).

Restoration of the expression of genes controlling H3K9 methylation greatly decreased the proliferation of MB in vitro. Copy number aberrations of genes with critical roles in writing, reading, removing, and blocking the state of histone lysine methylation, particularly at H3K9, suggest that defective control of the histone code contributes to the pathogenesis of MB [85]. A detailed summary of genes and literature describing their involvement in epigenetic changes of expression are shown in Table 3.

The recent epigenome studies of MB have demonstrated the implication of epigenetic gene silencing in this pediatric tumor as a crucial mechanism of tumor suppressor gene inactivation. The integration of epigenetic profiles and genome aberrations in the context of MB molecular subgrouping will be of invaluable significance to address important questions in MB pathogenesis and recurrence, paving the way for new therapeutic strategies.
Conclusions

The genomic landscape of MB has been shown, in all its complexity, by several intensive studies including genome-, transcriptome-, and epigenome-wide profiling of tumor cohorts. Genomics projects of unprecedented dimension have revealed new insight into molecular aberrations of the disease and have implicated new mechanisms leading to deregulation of the MB transcriptome. The molecular classification of MBs outperforms the classical histological stratification and is likely to revolutionize MB patient management and MB therapy. Although the understanding of the molecular genetics underlying MB pathogenesis has improved greatly, much has to be done to correlate the different aspects of MB genomics to clinical features of the disease: to achieve this goal large sample cohorts are necessary, and given the relative rarity of the disease, only integrative approaches based on the collaboration of several groups across the globe are likely to provide the comprehensive knowledge of MB that we crave for. Next-generation sequencing technologies recently revolutionized the approach to cancer genomics: The Cancer Genome Atlas project is performing intensive studies to chart the genomic alterations involved in more than 20 types of cancer. As genomic studies proceed and produce a large amount of data, discriminating between so-called driver and passenger mutations will be the next challenge. Not all the alterations found in cancer genomes are thought to critically contribute to the progression of the disease, some are merely a bystander effect of generalized genome instability; to distinguishing mutations that confer a clonal advantage (drivers) in tumorigenesis from those that do not, a robust bioinformatics strategy will have to be implemented and candidates validated in relevant models. The future suggests to be an exciting time for MB researchers with a comprehensive catalogue of the key genomic changes to mine and explore in their relation to MB; cancer genomics will support advances in developing more effective ways to diagnose, treat, and prevent cancer.

<table>
<thead>
<tr>
<th>Table 3</th>
<th>Genes epigenetically silenced in MB</th>
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<tr>
<td>Gene</td>
<td>Function</td>
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<tr>
<td>SCG5</td>
<td>Hypermethylation in 70% of 23 primary MBs and 87% of 8 MB cell lines</td>
</tr>
<tr>
<td>SFRP1, SFRP2, SFRP3</td>
<td>Wnt inhibitors, silenced by promoter region methylation in MB. Methylation was identified in 23.5%, 3.9%, and 15.7% of primary MB</td>
</tr>
<tr>
<td>RASSF1A</td>
<td>Regulates cyclin D1 expression; hypermethylation in MB</td>
</tr>
<tr>
<td>CASP8</td>
<td>Cysteine protease involved in death-receptor-mediated apoptosis. In primary tumors, aberrant promoter methylation of CASP8 was seen most frequently in classic and anaplastic MB</td>
</tr>
<tr>
<td>SGNE1/7B2 and ZIC2</td>
<td>Calcium-dependent serine protease and zinc-finger transcription factor essential for the development of the central nervous system; silenced in MB</td>
</tr>
<tr>
<td>S100 gene family</td>
<td>S100A6 is hypermethylated and associated with large-cell anaplastic MB subtype while S100A4, a pro-metastatic gene, target of erbB2 signaling is hypomethylated</td>
</tr>
<tr>
<td>HIC1</td>
<td>Transcriptional repressor, direct inhibitor of Atonal Homolog 1 (Atoh1), a pro-neural transcription factor essential for cerebellar development. Epigenetic silencing in MB</td>
</tr>
<tr>
<td>SPINT2</td>
<td>Downregulated by promoter methylation in 34.3% of primary MBs</td>
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The miR-17/92 polycistron is up-regulated in sonic hedgehog-driven medulloblastomas and induced by hedgehog activators. Cancer Res 69(8):2349–2355


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### Author Queries

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