

Medulloblastoma cancer stem cells: molecular signatures and therapeutic targets

Hisham F Bahmad ,^{1,2} Robert J Poppiti^{1,3}

► Additional material is published online only. To view please visit the journal online (<http://dx.doi.org/10.1136/jclinpath-2019-206246>).

¹Arkadi M Rywlin MD
Department of Pathology and Laboratory Medicine, Mount Sinai Medical Center, Miami Beach, Florida, USA

²Department of Anatomy, Cell Biology and Physiological Sciences, Faculty of Medicine, American University of Beirut, Beirut, Lebanon

³Herbert Wertheim College of Medicine, Florida International University, Miami, Florida, USA

Correspondence to

Professor Robert J Poppiti, Arkadi M. Rywlin M.D. Department of Pathology and Laboratory Medicine, Mount Sinai Medical Center, Miami Beach FL 33140, USA; Robert.Poppiti@mmsmc.com

Received 2 October 2019
Revised 12 December 2019
Accepted 16 December 2019
Published Online First
7 February 2020

ABSTRACT

Medulloblastoma (MB) is the most common malignant primary intracranial neoplasm diagnosed in childhood. Although numerous efforts have been made during the past few years to exploit novel targeted therapies for this aggressive neoplasm, there still exist substantial hurdles hindering successful management of MB. Lately, progress in cancer biology has shown evidence that a subpopulation of cells within the tumour, namely cancer stem cells (CSCs), are thought to be responsible for the resistance to most chemotherapeutic agents and radiation therapy, accounting for cancer recurrence. Hence, it is crucial to identify the molecular signatures and genetic aberrations that characterise those CSCs and develop therapies that specifically target them. In this review, we aim to give an overview of the main genetic and molecular cues that depict MB-CSCs and provide a synopsis of the novel therapeutic approaches that specifically target this population of cells to attain enhanced antitumourous effects and therefore overcome resistance to therapy.

INTRODUCTION

Medulloblastoma (MB) is the most common malignant primary intracranial neoplasm in children, accounting for around 20% of all paediatric brain tumours.¹ It is a primitive neuroectodermal malignancy of the central nervous system that is believed to arise from neural stem cell precursors in the granular cell layer of the cerebellum.² Specifically, many studies demonstrate that it originates from the remnants of the primitive neuroectoderm in the germinal matrix of the fourth ventricle roof,^{3,4} while other studies have reported a different origin for this invasive neoplasm: the external granular layer precursor cells.^{5,6} MB is subdivided into four major entities at the molecular level⁷: MB^{WNT-activated}, MB^{SHH-activated,TP53-mutant}, MB^{SHH-activated,TP53-wildtype} and MB^{non-WNT/non-SHH} (includes MB^{non-WNT/non-SHH, Group3} and MB^{non-WNT/non-SHH, Group4}).

The incidence of MB ranges from 0.53 (children aged 0–4 years) to 0.16 (adolescents aged 15–19 years) per 100 000 population in patients up to 19 years old, and affects males more than females (1.7 times more frequently in the age group 0–14 years)⁸ and white people more than black (1.7 times).⁹ The incidence continues to decline with age, reaching 0.06 per 100 000 person-years by 55–64 years of age.⁹ The peak age at diagnosis of MB is 7 years, with more than 70% of all cases observed among children younger than 16 years of age,^{10,11} and 10%–15% of patients diagnosed in infancy.⁹ Although aggressive, the 10-year survival

for patients with MB in the USA was found to be 64.9%.⁸ This could be attributable to the multimodality treatment approach including surgical intervention, radiation therapy (RT) and chemotherapy.³

During the past decade, increased interest in understanding the molecular basis of MB has revealed new insights into the different molecular and signalling pathways that might contribute to the tumour's formation, progression and recurrence. In this regard, many articles have been published in the last few years tackling the role of cancer stem cells (CSCs) as principal drivers in MB initiation and relapse, and subsequently as potential therapeutic targets for this malignant neoplasm.¹² Indeed, the CSC concept has become increasingly prominent ever since it was first proposed four decades ago, based on their self-renewal ability, potential to differentiate into the different types of cells and uninhibited growth pattern contributing to resistance to conventional therapies.^{13,14}

CSCs were first identified in leukemias in 1973 as a distinguished population of cells, embracing specific pro-oncogenic genetic signatures, that are capable of generating malignant haematopoietic colonies.¹⁵ With time, CSCs have been gradually identified in dedicated niches of many other tumours,¹⁴ including MB in 2003.¹⁶ Indeed, CSCs had been isolated from human and mouse MBs¹⁷ and were shown to reside in a perivascular niche (PVN).¹⁸ The stem cell niche is referred to the microenvironment surrounding cancer cells, and is composed of supportive cells, extracellular matrix and factors needed to maintain cancer stemness.¹⁹ Although the niche in MB tumours is still largely undefined,²⁰ a recent report by Calabrese *et al*¹⁸ revealed that CD133-positive MB-CSCs reside near endothelial cells and small vessels, and might function as a niche.

Highly tumourigenic MB cells have been shown to display features imitating those of neural stem and progenitor cells, such as upregulation of CD133, *Nestin* and *Musashi* (*MSI1*)²¹ (*MSI1*²² and *Nestin* are evolutionally conserved markers for central nervous system (CNS) progenitor cells and neuronal stem cells), as well as developmentally related genes, such as *Ebfs*.²³ Here, we discuss the latest discoveries related to MB-CSC genetic signatures and the novel therapies that specifically target those cells based on the molecular cues they harbour.

METHODOLOGY

This review was conducted using the 'Preferred Reporting Items for Systematic Reviews and Meta-Analyses' (PRISMA) 2009 guidelines. We performed



© Author(s) (or their employer(s)) 2020. No commercial re-use. See rights and permissions. Published by BMJ.

To cite: Bahmad HF, Poppiti RJ. *J Clin Pathol* 2020;**73**:243–249.

a comprehensive search using two databases, namely OVID/Medline and PubMed, for mesh terms, keywords and combinations related to 'cancer stem cells' and 'medulloblastoma'. Complete search strategy is provided in online supplementary appendix S1. In total, 248 articles were retrieved through database search from inception to 15 September 2019. We inserted all articles into EndNote V.X8 referencing program, and excluded duplicates, abstracts, case reports, non-English articles, reviews, commentaries and editorials. As a result, a total of 105 articles were considered for full-text qualitative analysis and inclusion in the final review. Online supplementary appendix S2 illustrates the study flow chart of the review process according to PRISMA 2009 flow diagram guidelines.

MB CLASSIFICATIONS AND CSCS

Histologically, MB is classified into four main WHO-defined subsets²⁴: classic MB, large-cell anaplastic, desmoplastic MB and MB with extensive nodularity (other variants such as medulloblastoma and melanotic do exist but are extremely rare).² At a molecular level, and according to the latest consensus nomenclature, MB is subdivided into four major entities⁷: (1) MB^{WNT-activated} (thought to originate from the lower rhombic lip),^{25 26} (2) MB^{SHH-activated,TP53-mutant} and (3) MB^{SHH-activated,TP53-wildtype} (sonic hedgehog (SHH); arises from granular neuron progenitors in the external germinal layer),^{27 28} and (4) MB^{non-WNT/non-SHH} which comprises MB^{non-WNT/non-SHH, Group3} (develops from cerebellar stem cells with high levels of MYC amplification and is considered the most aggressive among all subgroups)^{29 30} and MB^{non-WNT/non-SHH, Group4} (the most prevalent yet still of unknown origin).^{31 32}

Noteworthy, genes that are mainly deregulated in MB, such as *WNT*, *SHH* and *Notch*, as well as the proto-oncogenes *RTK* (receptor tyrosine kinase) and *MYC*, are central to molecular pathways controlling cell cycle and growth of CSCs.^{33–35} Moreover, reports from retrospective studies reveal that several molecular and genetic aberrations that correlate with MB prognosis and outcome^{36–41} are also involved in the control of CSC stemness,^{42–47} including *neurotrophin-3 receptor*,⁴⁸ *CD15*,^{49–51} *PTEN*,⁵² *MYC*,^{53–55} *ErbB2*,⁵⁶ *β-catenin*,^{57 58} *survivin*^{59 60} and *p53*.⁶¹

Further exploration showed that other markers linked to CSCs might have a pivotal role in MB tumour formation and progression.⁶² For instance, Singh *et al*¹⁷ were the first to reveal a role of CSC markers in MB, where they found that injecting a small number as low as 100 *CD133*+ cells into immunodeficient mice could yield MB tumour formation *in vivo*, whereas tumour failed to develop with *CD133*– cells even on increasing the number of injected cells to 100 000 cells. In this regard, high mRNA levels of *CD133* had been correlated with poor prognosis and increased likelihood of metastases in paediatric MB.⁶³ This demonstrates the importance of identifying novel CSC biomarkers and genetic signals and incorporating them altogether with the currently available parameters to create new stratification schemes for MB.⁶⁴ Such schemes may help to refine the management approaches for the different histological and molecular entities of this tumour.^{65–68} We will elaborate more on the various studied molecular signatures and therapeutic targets pertaining to MB-CSCs.

MB-CSCS: FROM GENES TO THERAPIES

Ever since the first MB-CSCs were isolated from human tissues in 2003 (*CD133*+*Nestin*+) showing improved proliferation, self-renewal and differentiation *in vitro*,¹⁶ subsequent research has elucidated the genetic aberrations and molecular signatures

pertaining to those CSCs, paving the way for novel therapeutic targets in this aggressive intracranial tumour.

CD133 and its relation to other molecular signatures

CD133 (prominin-1) is the most common cell surface antigen used to detect and isolate CSCs from various solid tumours.⁶⁹ Physiologically, it induces WNT/ β -catenin signalling^{70–73} and has also been described as an important regulator of PI3K/Akt signalling in CSCs.^{74 75} The use of this marker to identify MB-CSCs in paediatric tissue samples was first described by Singh *et al*,^{16 17} and the isolated *CD133*+ cells were termed brain tumour stem cells. These cells had the ability to grow into neurosphere-like clusters *in vitro* and to produce massive tumours on intracranial transplantation into NOD-SCID mice forebrains *in vivo*, expressing neural stem cell markers such as nestin.¹⁶ An alternative method for culturing MB-CSCs, other than the three-dimensional (3D) neurospheres technique, has been proposed by de la Rosa *et al*⁷⁶ using laminin-precoated flasks that enable dedifferentiation of cells and enrich the stem-like cell population. Based on their high expression of *CD133*, CSCs possess the ability to resist apoptosis as well as RT⁷⁷ and chemotherapeutic drugs.⁷⁸

A study by Annabi *et al*⁷⁹ demonstrated that members of the low-density lipoprotein receptor-related protein (LRP) family, including LRP-1, LRP-1b, LRP-5 and LRP-8, regulate the adaptive phenotype associated with *CD133*+ MB-CSCs. Another study by the same research group revealed that matrix metalloproteinase 9 and membrane type I-matrix metalloproteinase, which are major players in cancer cell invasion, metastasis and resistance to therapy, have crucial roles in maintaining the invasive phenotype of *CD133*+ neurosphere-derived MB cells, while targeting those two molecules may reduce the formation of brain tumour stem cells.⁸⁰

Inflammatory mediators such as cyclooxygenase-2 (COX-2), an enzyme that converts arachidonic acid to prostaglandins, have been shown to be overexpressed in a variety of tumours,^{81 82} including MB.⁸³ COX-2-derived prostaglandins have also been implicated in tumour growth and angiogenesis.⁸⁴ Henceforth, the role of anti-inflammatory drugs in targeting MB-CSCs has recently been assessed, whereby a study was conducted by Chen *et al*⁸⁵ and Yang *et al*⁸⁶ to assess the enhancing effects of celecoxib on ionising radiotherapy (IR) of *CD133*+ MB cells. Results demonstrated that celecoxib significantly enhanced radiosensitivity of those MB cells *in vitro* and *in vivo*.^{85 86} In the same milieu, resveratrol, a natural polyphenol derived from red wine, has been shown to inhibit proliferation and tumorigenicity of MB-CSCs and enhance radiosensitivity in treated MB-CSCs.⁸⁷

One of the mechanisms contributing to chemotherapeutic resistance in many tumours is upregulation of X linked inhibitor of apoptosis protein and cellular inhibitor of apoptosis 1/2.⁸⁸ This applies to *CD133*+ MB-CSCs, which displayed higher levels of both proteins and demonstrated hypersensitivity to treatment with small-molecule inhibitors of apoptosis proteins (IAP) inhibitors LCL161 and LBW242.⁸⁸ Another pathway that plays a role in the maintenance of *CD133*+ MB-CSCs was described by Chang *et al*,⁸⁹ who treated those cells with a potent STAT3 inhibitor, cucurbitacin I. Results revealed that the latter treatment suppressed the CSC-like properties and stemness of MB-derived *CD133*+ cells and increased the apoptotic sensitivity of those cells to RT and chemotherapeutic drugs.⁸⁹ Similarly, Garg *et al* and others showed that signal transducers and activators of the STAT3

pathway are activated in *CD133+* MB-CSCs through regulation of *c-MYC*, a key genetic driver of MB^{non-WNT/non-SHH, Group3}.^{90 91}

An orthotopic xenograft model, named MB3W1, was established using cells derived from the malignant pleural effusions from a child with MB^{non-WNT/non-SHH, Group3}.⁹² This model displayed CSC characteristics such as the ability to form neurospheres, high aldehyde dehydrogenase (ALDH) activity, expression of *CD133/CD15* stem cell markers and high tumorigenicity in NOD-SCID mice.⁹² In a similar study by Friedman *et al*,⁹³ four human paediatric MB xenografts, mainly representing group 3 tumours, were used to prove that hypoxia increases *CD133* as well as primary HSV-1 (herpes simplex virus) entry molecule nectin-1 (*CD111*) expression. Interestingly, MB cells expressing *CD111* were also found to be highly sensitive to killing by clinically relevant oncolytic HSVs (G207 and M002) in vitro and in vivo.⁹³

In a study by Lim *et al*,⁹⁴ polymeric nanoparticle formulation of curcumin was used to assess its effect as a potential therapy for MB-CSCs. This compound is derived from the Indian spice turmeric and has been proven to harbour diverse effects on human diseases: proapoptotic, antiangiogenic, anti-inflammatory, immunomodulatory and antimetastatic effects.^{95 96} Treating MB cells with curcumin decreased anchorage-independent clonogenic growth, reduced the *CD133+* stem-like population, attenuated insulin-like growth factor and STAT3 pathways, and blocked SHH signalling, but did not affect Notch signalling.⁹⁴

Hedgehog (SHH) pathway

The SHH entity of MB accounts for approximately 30% of all cases. It is driven by hedgehog ligands that undergo covalent modification by cholesterol^{97 98} and bind to the Patched (*PTCH1*) transmembrane receptor⁹⁹ to maintain tumour growth and stemness.^{100–102} In a recent study by Bell *et al*,¹⁰⁰ the authors used biomimetic high-density lipoprotein (HDL) nanoparticles to deplete cholesterol from hedgehog-driven MB and Ewing sarcoma cancer cells, via binding to the HDL receptor, scavenger receptor type B-1, and thereby targeting the CSC populations in those tumours.

Among the other drivers of the SHH pathway is the protein patched homolog 1 (*Ptch*).^{103–105} A single tumour mouse model, namely the *Ptch*^{+/-} model, has long been used to study the molecular and cellular mechanisms involved in MB formation, particularly MB^{SHH}.^{106 107} Chow *et al*¹⁰³ provided evidence that although MBs that form in *Ptch*^{+/-} MB mice are composed of three entities, all of them contain long-term, self-renewing stem cell-like cells that are responsible for tumour initiation on serial in vivo transplantations. Other studies were also published confirming this.^{107 108} Notably, high expression levels of *PTCH2* were observed in human MB tissues and correlated with a worse prognosis³⁴; although *PTCH2* is a tumour suppressor gene that inhibits SHH activity,¹⁰⁹ its role in MB might not necessarily be related to SHH pathway inhibition but to *GLI1* expression.^{34 110} This finding was further confirmed in a study by Po *et al*,¹⁰⁵ where *GLI1* and *GLI2*, the downstream effectors of SHH, were shown to bind to Nanog-specific cis-regulatory sequences in stem cells. In this regard, SHH signalling was linked to two distinct MB entities: wild-type *TP53* (MB^{SHH-activated,TP53-wildtype}) and *TP53* loss (MB^{SHH-activated,TP53-mutant}), a central event in promoting stemness, which contributes to Nanog upregulation in stem cells derived from both postnatal cerebellum and MB.¹⁰⁵

Notch pathway

Like the SHH pathway, Notch signalling is required for controlling growth and proliferation of neural stem/progenitor cells as well as embryonal brain tumours,¹¹¹ such as MB.^{20 112–115} Fan *et al*¹¹⁶ showed that MB stem-like cells exhibit higher levels of Notch signalling, which makes them more sensitive to this pathway inhibition. Indeed, Notch blockade with γ -secretase totally abolished *CD133+* MB-CSCs, leading to loss of tumour-forming capacity (due to depletion of stem-like cells).¹¹⁶ Another study by Pistollato *et al*¹¹⁷ revealed that hypoxic conditions promote *Notch1* activation with its ligand *Dll4* and lead to expansion of *CD133+* and *Nestin+* MB precursors.

PI3K pathway

The phosphoinositide-3-kinase (PI3K)/AKT signalling pathway has been reported to play an important role in the renewal of embryonic stem cells.^{118 119} Recent studies also referred to the role of PI3K/AKT/mTOR pathway in growth and maintenance of CSCs in solid tumours, such as breast and prostate cancers.^{75 120} In MB, Frasson *et al*¹¹⁹ revealed that PI3K/AKT inhibition with LY294002 yielded increased cell death of *CD133+* MB-CSCs and spared the more differentiated cells via activation of the mitochondrial apoptotic cascade. In another study by Hambardzumyan *et al*,^{121 122} the authors demonstrated that PI3K pathway plays a crucial role in regulating survival of nestin-expressing MB-CSCs residing in the PVN, and inhibition of AKT signalling sensitises PVN cells to radiation-induced apoptosis.

MYC pathway

MYC proteins have been associated with several cancers, including MB tumours^{31 123} that harbour *MYC*, *MYCN* and *MYCL1* amplifications.¹¹⁸ In this regard, somatic mutations of *TP53* had been mostly found in WNT-MB and SHH-MB and are associated with *MYCN* rather than *MYC* amplification.¹²⁴

A study from the Chesler's group reported a genetically engineered mouse model of *MYCN*-driven MB (the GTML mouse model: *Glt1-tTA* (glutamate transporter 1-tetracycline transactivator), (*TRE*)-*MYCN* (tetracycline response element) and *Luc* (luciferase)),³¹ and later used this model to establish neurosphere lines.³² Those spheres demonstrated robust proliferation and expressed neuronal markers such as *Ngn1*, *Syp*, *Olig2* and *Sox9*.¹²⁵ Besides, when transplanted orthotopically into the brains of nude mice, the neurosphere lines were able to form massive tumours with morphology that mimicked human MB, including Homer Wright (neuroblastic) rosettes.³²

Venkataraman *et al*¹²⁶ conducted a study concluding that inhibition of a member of the bromodomain and extraterminal domain family, namely BRD4, effectively suppresses *MYC*-driven MB through attenuating cancer cells self-renewal, stem cell signalling and induction of senescence in vitro and in vivo.¹²⁶

Polycomb repressive complexes

Polycomb repressive complexes 1 and 2 (PRC1 and PRC2) are evolutionarily conserved epigenetic regulators¹²⁷ implicated in cancer.¹²⁸ *BMI1* (B cell-specific Moloney murine leukaemia virus integration site), the best studied *PRC1* gene in oncology,^{129 130} is often overexpressed in cancer and has been implicated in maintaining tumour stemness by serving as a key CSC regulatory gene.^{131 132} In MB, *BMI1* is overexpressed across all subgroups, particularly MB^{non-WNT/non-SHH, Group3}.^{133 134} A study by Wang *et al*¹³⁴ revealed that SHH modulates *BMI1* to maintain MB-CSCs. Another study by Bakhshinyan *et al* used a small-molecule inhibitor to target *BMI1* in MB^{non-WNT/non-SHH, Group3} cell lines,

namely PTC-028, exemplifying significant reduction in stem cell properties in vitro and in vivo. BMI1 has also been shown to downregulate p53 in embryonal cancer precursor cells, such as neuroblastoma and MB, and subsequently promote MycN oncoprotein overexpression in those cells.¹³⁵ Manoranjan *et al*¹³⁶ studied the stem cell data gathered from genomic platforms and demonstrated that *FoxG1* interacts with *Bmi1* in MB^{non-WNT/non-SHH, Group3} to mediate stem cell self-renewal and tumour initiation.

Another protein that represents the catalytically active component of the PRC2 is enhancer of zeste homologue 2 (EZH2). This protein causes chromatin compaction and contributes to several biological processes, including differentiation, maintaining cell identity and proliferation.¹³⁷ It is shown to be highly expressed (more than twofold) in primary MB tissues and cell lines.¹³⁸ Since it was proven that EZH2 maintains glioblastoma CSCs,¹³⁹ Alimova *et al* evaluated the effects of knocking down *EZH2* expression on MB-CSCs. Results showed that neurosphere formation was attenuated after *EZH2* knockdown along with a significant decrease in Myc and Sox2 activities and G2 cell cycle arrest.¹³⁸ The authors concluded that EZH2 might be a potential therapeutic target for MB and is important for MB cell growth and transformation of neural stem cells.¹³⁸ In another study, an interaction between maternal embryonic leucine-zipper kinase and EZH2 was found in MB stem-like cells, representing an attractive therapeutic target and potential candidate for the diagnosis of MB.¹⁴⁰

Other genetic aberrations

Urokinase-type plasminogen activator receptor (uPAR) is a cell surface protein that drives directed extracellular proteolysis on the surface of invading cancer cells promoting invasion, migration and metastasis.¹⁴¹ It is overexpressed in the tumour-stromal invasive microenvironment in many human cancers, including MB.¹⁴² Asuthkar *et al*¹⁴³ showed that IR induces the expression of uPAR and other CSC markers, such as *MSI1* and *CD44*, and triggers WNT-7a- β -catenin signalling, which in turn promotes cancer stemness in MB. Overexpression of uPAR post-IR also negatively regulates Hand-1 activity, promoting angiogenesis via hypoxia-inducible factor-1a upregulation.¹⁴² Henceforth, targeting uPAR in patients with MB undergoing IR might overcome potential therapy resistance and prevent IR-induced tumour angiogenesis.^{142 143}

It is believed that in many tumours, drug resistance might be attributed to the efflux of chemotherapeutic drugs by key members of the ATP-binding cassette (ABC) transporter superfamily.¹⁴⁴ Since a subpopulation of cells within tumours, namely CSCs, underlie tumour progression and relapse, those cells must express ABC transporters.¹⁴⁵ Interestingly, in MB, significant correlation was found between ABCB1 expression and high-risk tumours among patients with poorer overall survival.¹⁴⁶

Polo-like kinase 1 (PLK1), a protein kinase that promotes mitosis via phosphorylating cyclin B1 and CDK1,¹⁴⁷ is shown to be overexpressed in a wide variety of cancers, including MB.¹⁴⁸ Inhibiting PLK1 by small-molecule inhibitor BI 2536 potently increased MB cellular apoptosis and sensitised cells to IR. It also reduced MB cell growth and CSC formation through decreasing the expression of SRY (sex determining region Y)-box 2 (SOX2).¹⁴⁸

Lastly, accumulating evidence demonstrates a crucial role of microRNAs in regulating and maintaining CSCs within different tumours. In MB, low expression of miR-466f-3p was found to sustain epithelial-to-mesenchymal transition in MB^{SHH-activated}

CSCs via Vegfa-Nrp2 signalling pathway.¹⁴⁹ Also, Kaid *et al*¹⁵⁰ found that miR-367 enhances stemness features of MB cells, such as proliferation, 3D tumour spheroid cell invasion and the ability to generate CD133-expressing neurosphere-like structures. Other studied microRNAs included miR-135a,¹⁵¹ miR-142-3p,¹⁵² miR-218^{153 154} and miR-34a.¹⁵⁵

CONCLUSIONS

The key for effective eradication of MB tumours and overcoming aggravating therapy resistance is the isolation of the MB-CSCs and identification of their specific molecular signatures and genetic aberrations. This eventually will lead to the development of novel therapeutic interventions and combinations to target aggressive MB stem cell-specific dysregulations.

Take home messages

- ▶ Subpopulation of cells within the tumor, named cancer stem cells, are thought to be responsible for cancer recurrence in medulloblastoma.
- ▶ The key for effective eradication of medulloblastoma tumors and overcoming aggravating therapy resistance is isolation of cancer stem cells.
- ▶ Highly tumorigenic medulloblastoma cells display features imitating those of neural stem and progenitor cells, such as upregulation of CD133 and Nestin.

Handling editor Dharendra Govender.

Acknowledgements We would like to thank all members of the Arkadi M Rywlin MD Department of Pathology and Laboratory Medicine at Mount Sinai Medical Center for their help on this work.

Contributors HFB worked on the review conception and design. HFB screened titles for relevance, abstracted the data from eligible full-text articles, analysed and interpreted the data, and drafted the manuscript. RJP critically revised the manuscript. Both authors have read and approved the final draft.

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests None declared.

Patient consent for publication Not required.

Provenance and peer review Not commissioned; externally peer reviewed.

ORCID iD

Hisham F Bahmad <http://orcid.org/0000-0003-3799-2595>

REFERENCES

- 1 Martin AM, Raabe E, Eberhart C, *et al*. Management of pediatric and adult patients with medulloblastoma. *Curr Treat Options Oncol* 2014;15:581–94.
- 2 Crawford JR, MacDonald TJ, Packer RJ. Medulloblastoma in childhood: new biological advances. *Lancet Neurol* 2007;6:1073–85.
- 3 Rossi A, Caracciolo V, Russo G, *et al*. Medulloblastoma: from molecular pathology to therapy. *Clin Cancer Res* 2008;14:971–6.
- 4 Gilbertson RJ, Ellison DW. The origins of medulloblastoma subtypes. *Annu Rev Pathol* 2008;3:341–65.
- 5 Yokota N, Aruga J, Takai S, *et al*. Predominant expression of human Zic in cerebellar granule cell lineage and medulloblastoma. *Cancer Res* 1996;56:377–83.
- 6 Behesti H, Marino S. Cerebellar granule cells: insights into proliferation, differentiation, and role in medulloblastoma pathogenesis. *Int J Biochem Cell Biol* 2009;41:435–45.
- 7 Louis DN, Ohgaki H, Wiestler OD, *et al*. *World Health organization classification of tumours of the central nervous system*. 4th edn. Lyon: IARC Press, 2016.
- 8 Ostrom QT, Gittleman H, Truitt G, *et al*. CBTRUS statistical report: primary brain and other central nervous system tumors diagnosed in the United States in 2011–2015. *Neuro Oncol* 2018;20:iv1–86.
- 9 Dolecek T, Propp J, Stroup N, *et al*. CBTRUS (central brain tumor registry of the United States) statistical report: primary brain and central nervous system tumors diagnosed in the United States in 2005–2009. *Neuro Oncol* 2012;14:v1–49.

- 10 Karajannis M, Allen JC, Newcomb EW. Treatment of pediatric brain tumors. *J Cell Physiol* 2008;217:584–9.
- 11 Packer RJ, Rood BR, MacDonald TJ. Medulloblastoma: present concepts of stratification into risk groups. *Pediatr Neurosurg* 2003;39:60–7.
- 12 Azzarelli R, Simons BD, Philpott A. The developmental origin of brain tumours: a cellular and molecular framework. *Development* 2018;145. doi:10.1242/dev.162693
- 13 Bahmad HF, Chamaa F, Assi S, et al. Cancer stem cells in neuroblastoma: expanding the therapeutic frontier. *Front Mol Neurosci* 2019;12.
- 14 Batlle E, Clevers H. Cancer stem cells revisited. *Nat Med* 2017;23:1124–34.
- 15 Moore MAS, Williams N, Metcalf D. In vitro colony formation by normal and leukemic human hematopoietic cells: characterization of the colony-forming cells 2. *J Natl Cancer Inst* 1973;50:603–23.
- 16 Singh SK, Clarke ID, Terasaki M, et al. Identification of a cancer stem cell in human brain tumors. *Cancer Res* 2003;63:5821–8.
- 17 Singh SK, Hawkins C, Clarke ID, et al. Identification of human brain tumour initiating cells. *Nature* 2004;432:396–401.
- 18 Calabrese C, Poppleton H, Kocak M, et al. A perivascular niche for brain tumor stem cells. *Cancer Cell* 2007;11:69–82.
- 19 Fuchs E, Tumber T, Guasch G. Socializing with the neighbors: stem cells and their niche. *Cell* 2004;116:769–78.
- 20 Fan X, Eberhart CG. Medulloblastoma stem cells. *J Clin Oncol* 2008;26:2821–7.
- 21 Ahmad Z, Jasnos L, Gil V, et al. Molecular and in vivo characterization of cancer-propagating cells derived from MYCN-dependent medulloblastoma. *PLoS One* 2015;10:e0119834.
- 22 Kaneko Y, Sakakibara S, Imai T, et al. Musashi1: an evolutionally conserved marker for CNS progenitor cells including neural stem cells. *Dev Neurosci* 2000;22:139–53.
- 23 Corno D, Pala M, Cominelli M, et al. Gene signatures associated with mouse postnatal hindbrain neural stem cells and medulloblastoma cancer stem cells identify novel molecular mediators and predict human medulloblastoma molecular classification. *Cancer Discov* 2012;2:554–68.
- 24 Louis DN, Ohgaki H, Wiestler OD, et al. The 2007 WHO classification of tumours of the central nervous system. *Acta Neuropathol* 2007;114:97–109.
- 25 Vriend J, Ghavami S, Marzban H. The role of the ubiquitin proteasome system in cerebellar development and medulloblastoma. *Mol Brain* 2015;8:64.
- 26 Gibson P, Tong Y, Robinson G, et al. Subtypes of medulloblastoma have distinct developmental origins. *Nature* 2010;468:1095–9.
- 27 Schüller U, Heine VM, Mao J, et al. Acquisition of granule neuron precursor identity is a critical determinant of progenitor cell competence to form Shh-induced medulloblastoma. *Cancer Cell* 2008;14:123–34.
- 28 Yang Z-J, Ellis T, Markant SL, et al. Medulloblastoma can be initiated by deletion of patched in lineage-restricted progenitors or stem cells. *Cancer Cell* 2008;14:135–45.
- 29 Pei Y, Moore CE, Wang J, et al. An animal model of Myc-driven medulloblastoma. *Cancer Cell* 2012;21:155–67.
- 30 Kawauchi D, Robinson G, Uziel T, et al. A mouse model of the most aggressive subgroup of human medulloblastoma. *Cancer Cell* 2012;21:168–80.
- 31 Swartling FJ, Grimmer MR, Hackett CS, et al. Pleiotropic role for MYCN in medulloblastoma. *Genes Dev* 2010;24:1059–72.
- 32 Swartling FJ, Savov V, Persson AI, et al. Distinct neural stem cell populations give rise to disparate brain tumors in response to N-myc. *Cancer Cell* 2012;21:601–13.
- 33 Varnum-Finney B, Xu L, Brashem-Stein C, et al. Pluripotent, cytokine-dependent, hematopoietic stem cells are immortalized by constitutive Notch1 signaling. *Nat Med* 2000;6:1278–81.
- 34 Cordeiro BM, Oliveira ID, Alves MTdeS, et al. SHH, WNT, and NOTCH pathways in medulloblastoma: when cancer stem cells maintain self-renewal and differentiation properties. *Childs Nerv Syst* 2014;30:1165–72.
- 35 Guessous F, Li Y, Abounader R. Signaling pathways in medulloblastoma. *J Cell Physiol* 2008;217:577–83.
- 36 Korshunov A, Savostikova M, Ozerov S. Immunohistochemical markers for prognosis of average-risk pediatric medulloblastomas. The effect of apoptotic index, TrkC, and c-myc expression. *J Neurooncol* 2002;58:271–9.
- 37 Aldosari N, Bigner SH, Burger PC, et al. Mycc and MYCN oncogene amplification in medulloblastoma. A fluorescence in situ hybridization study on paraffin sections from the children's Oncology Group. *Arch Pathol Lab Med* 2002;126:540–4.
- 38 Grotzer MA, Hogarty MD, Janss AJ, et al. Myc messenger RNA expression predicts survival outcome in childhood primitive neuroectodermal tumor/medulloblastoma. *Clin Cancer Res* 2001;7:2425–33.
- 39 Min HS, Lee YJ, Park K, et al. Medulloblastoma: histopathologic and molecular markers of anaplasia and biologic behavior. *Acta Neuropathol* 2006;112:13–20.
- 40 Ray A, Ho M, Ma J, et al. A clinicobiological model predicting survival in medulloblastoma. *Clin Cancer Res* 2004;10:7613–20.
- 41 Haberler C, Slavc I, Czech T, et al. Histopathological prognostic factors in medulloblastoma: high expression of survivin is related to unfavourable outcome. *Eur J Cancer* 2006;42:2996–3003.
- 42 Huang G-H, Xu Q-F, Cui Y-H, et al. Medulloblastoma stem cells: promising targets in medulloblastoma therapy. *Cancer Sci* 2016;107:583–9.
- 43 Huang X, Ketova T, Ltingtung Y, et al. Isolation, enrichment, and maintenance of medulloblastoma stem cells. *Jove* 2010.
- 44 Zanini C, Ercole E, Mandili G, et al. Medullospheres from DAOY, UW228 and ONS-76 cells: increased stem cell population and proteomic modifications. *PLoS One* 2013;8:e63748.
- 45 Srivastava VK, Nalbantoglu J. Flow cytometric characterization of the DAOY medulloblastoma cell line for the cancer stem-like phenotype. *Cytometry Part A* 2008;73A:940–8.
- 46 Srivastava VK, Nalbantoglu J. The cellular and developmental biology of medulloblastoma: current perspectives on experimental therapeutics. *Cancer Biol Ther* 2010;9:843–52.
- 47 Sümer-Turanligil NC, Cetin Emel Öykü, Uyanıkgil Y. A contemporary review of molecular candidates for the development and treatment of childhood medulloblastoma. *Childs Nerv Syst* 2013;29:381–8.
- 48 Chopin V, Lagadec C, Toillon R-A, et al. Neurotrophin signaling in cancer stem cells. *Cell Mol Life Sci* 2016;73:1859–70.
- 49 Lowry NA, Temple S. Identifying the perpetrator in medulloblastoma: Dorian gray versus Benjamin button. *Cancer Cell* 2009;15:83–5.
- 50 Read T-A, Fogarty MP, Markant SL, et al. Identification of CD15 as a marker for Tumor-Propagating cells in a mouse model of medulloblastoma. *Cancer Cell* 2009;15:135–47.
- 51 Ward RJ, Lee L, Graham K, et al. Multipotent CD15+ cancer stem cells in patched-1-deficient mouse medulloblastoma. *Cancer Res* 2009;69:4682–90.
- 52 Zhu G, Rankin SL, Larson JD, et al. Pten signaling in the postnatal perivascular progenitor niche drives medulloblastoma formation. *Cancer Res* 2017;77:123–33.
- 53 Elbadawy M, Usui T, Yamawaki H, et al. Emerging roles of c-myc in cancer stem cell-related signaling and resistance to cancer chemotherapy: a potential therapeutic target against colorectal cancer. *Int J Mol Sci* 2019;20:2340.
- 54 Zhang H-L, Wang P, M-Z L, et al. C-Myc maintains the self-renewal and chemoresistance properties of colon cancer stem cells. *Oncol Lett* 2019;17:4487–93.
- 55 Yang A, Qin S, Schulte BA, et al. Myc inhibition depletes cancer stem-like cells in triple-negative breast cancer. *Cancer Res* 2017;77:6641–50.
- 56 Wang X, Sun Y, Wong J, et al. Ppary maintains ErbB2-positive breast cancer stem cells. *Oncogene* 2013;32:5512–21.
- 57 Pandit H, Li Y, Li X, et al. Enrichment of cancer stem cells via β -catenin contributing to the tumorigenesis of hepatocellular carcinoma. *BMC Cancer* 2018;18:783–83.
- 58 Valkenburg KC, Graveel CR, Zylstra-Diegel CR, et al. Wnt/ β -catenin Signaling in Normal and Cancer Stem Cells. *Cancers* 2011;3:2050–79.
- 59 Yamamoto M, Suzuki S, Togashi K, et al. AS602801, an anticancer stem cell candidate drug, reduces survivin expression and sensitizes A2780 ovarian cancer stem cells to carboplatin and paclitaxel. *Anticancer Res* 2018;38:6699–706.
- 60 Ejarque M, Ceperuelo-Mallfré V, Serena C, et al. Survivin, a key player in cancer progression, increases in obesity and protects adipose tissue stem cells from apoptosis. *Cell Death Dis* 2017;8:e2802.
- 61 Chen X, Guan H, Liu X-D, et al. P53 positively regulates the expression of cancer stem cell marker CD133 in HCT116 colon cancer cells. *Oncol Lett* 2018;16:431–8.
- 62 Flores DG, Ledur PF, Abujamra AL, et al. Cancer stem cells and the biology of brain tumors. *Curr Stem Cell Res Ther* 2009;4:306–13.
- 63 Raso A, Mascelli S, Biondini R, et al. High levels of PROM1 (CD133) transcript are a potential predictor of poor prognosis in medulloblastoma. *Neuro Oncol* 2011;13:500–8.
- 64 Rodini CO, Suzuki DE, Nakahata AM, et al. Aberrant signaling pathways in medulloblastomas: a stem cell connection. *Arq Neuropsiquiatr* 2010;68:947–52.
- 65 Kumar V, Kumar V, McGuire T, et al. Challenges and recent advances in medulloblastoma therapy. *Trends Pharmacol Sci* 2017;38:1061–84.
- 66 Manoranjan B, Garg N, Bakhshinyan D, et al. The role of stem cells in pediatric central nervous system malignancies. *Adv Exp Med Biol* 2015;853:49–68.
- 67 Manoranjan B, Venugopal C, McFarlane N, et al. Medulloblastoma stem cells: where development and cancer cross pathways. *Pediatr Res* 2012;71:516–22.
- 68 Manoranjan B, Venugopal C, McFarlane N, et al. Medulloblastoma stem cells: modeling tumor heterogeneity. *Cancer Lett* 2013;338:23–31.
- 69 Glumac PM, LeBeau AM. The role of CD133 in cancer: a Concise review. *Clin Transl Med* 2018;7:18.
- 70 Bisson I, Prowse DM. Wnt signaling regulates self-renewal and differentiation of prostate cancer cells with stem cell characteristics. *Cell Res* 2009;19:683–97.
- 71 Mak AB, Nixon AML, Kittanakom S, et al. Regulation of CD133 by HDAC6 promotes β -catenin signaling to suppress cancer cell differentiation. *Cell Rep* 2012;2:951–63.
- 72 Zhukova N, Ramaswamy V, Remke M, et al. Wnt activation by lithium abrogates TP53 mutation associated radiation resistance in medulloblastoma. *Acta Neuropathologica Communications* 2014;2.
- 73 Bassani B, Bartolini D, Pagani A, et al. Fenretinide (4-HPR) targets caspase-9, ERK 1/2 and the Wnt3a/ β -Catenin pathway in medulloblastoma cells and medulloblastoma cell spheroids. *PLoS One* 2016;11:e0154111.
- 74 Wei Y, Jiang Y, Zou F, et al. Activation of PI3K/Akt pathway by CD133-p85 interaction promotes tumorigenic capacity of glioma stem cells. *Proc Natl Acad Sci U S A* 2013;110:6829–34.
- 75 Dubrovskaya A, Kim S, Salamone RJ, et al. The role of PTEN/Akt/PI3K signaling in the maintenance and viability of prostate cancer stem-like cell populations. *Proc Natl Acad Sci U S A* 2009;106:268–73.

- 76 de la Rosa J, Sáenz Antoñanzas A, Shahi MH, *et al.* Laminin-Adherent versus suspension-non-adherent cell culture conditions for the isolation of cancer stem cells in the DAOY medulloblastoma cell line. *Tumour Biology* 2016;37:12359–70.
- 77 Blazek ER, Foutch JL, Maki G. Daoy medulloblastoma cells that express CD133 are radiosensitive relative to CD133– cells, and the CD133+ sector is enlarged by hypoxia. *Int J Radiat Oncol Biol Phys* 2007;67:1–5.
- 78 Dean M, Fojo T, Bates S. Tumour stem cells and drug resistance. *Nat Rev Cancer* 2005;5:275–84.
- 79 Annabi B, Doumit J, Plouffe K, *et al.* Members of the low-density lipoprotein receptor-related proteins provide a differential molecular signature between parental and CD133(+) DAOY medulloblastoma cells. *Mol Carcinog* 2010;218:n/a–7.
- 80 Annabi B, Rojas-Sutterlin S, Laflamme C, *et al.* Tumor environment dictates medulloblastoma cancer stem cell expression and invasive phenotype. *Molecular Cancer Research* 2008;6:907–16.
- 81 Greenhough A, Smartt HJM, Moore AE, *et al.* The COX-2/PGE2 pathway: key roles in the hallmarks of cancer and adaptation to the tumour microenvironment. *Carcinogenesis* 2009;30:377–86.
- 82 Dannenberg AJ, Altorki NK, Boyle JO, *et al.* Cyclo-Oxygenase 2: a pharmacological target for the prevention of cancer. *Lancet Oncol* 2001;2:544–51.
- 83 Baryawno N, Sveinbjörnsson B, Eksborg S, *et al.* Tumor-growth-promoting cyclooxygenase-2 prostaglandin E2 pathway provides medulloblastoma therapeutic targets. *Neuro Oncol* 2008;10:661–74.
- 84 Masferrer JL, Leahy KM, Koki AT, *et al.* Antiangiogenic and antitumor activities of cyclooxygenase-2 inhibitors. *Cancer Res* 2000;60:1306–11.
- 85 Chen K-H, Hsu C-C, Song W-S, *et al.* Celecoxib enhances radiosensitivity in medulloblastoma-derived CD133-positive cells. *Child's Nervous System* 2010;26:1605–12.
- 86 Yang M-Y, Lee H-T, Chen C-M, *et al.* Celecoxib suppresses the phosphorylation of STAT3 protein and can enhance the radiosensitivity of medulloblastoma-derived cancer stem-like cells. *Int J Mol Sci* 2014;15:11013–29.
- 87 KH L, Chen YW, Tsai PH, *et al.* Evaluation of radiotherapy effect in resveratrol-treated medulloblastoma cancer stem-like cells child's nervous system. *Child's Nervous System* 2009;25:543–50.
- 88 Chen S-M, Li Y-Y, Tu C-H, *et al.* Blockade of inhibitors of apoptosis proteins in combination with conventional chemotherapy leads to synergistic antitumor activity in medulloblastoma and cancer stem-like cells. *PLoS One* 2016;11:e0161299.
- 89 Chang C-J, Chiang C-H, Song W-S, *et al.* Inhibition of phosphorylated STAT3 by curcubatin I enhances chemoradiosensitivity in medulloblastoma-derived cancer stem cells. *Child's Nervous System* 2012;28:363–73.
- 90 Garg N, Bakshshinyan D, Venugopal C, *et al.* Cd133+ brain tumor-initiating cells are dependent on STAT3 signaling to drive medulloblastoma recurrence. *Oncogene* 2017;36:606–17.
- 91 Ray S, Coulter DW, Gray SD, *et al.* Suppression of STAT3 NH₂-terminal domain chemosensitizes medulloblastoma cells by activation of protein inhibitor of activated STAT3 via de-repression by microRNA-21. *Mol Carcinog* 2018;57:536–48.
- 92 Dietl S, Schwinn S, Dietl S, *et al.* MB3W1 is an orthotopic xenograft model for anaplastic medulloblastoma displaying cancer stem cell- and group 3-properties. *BMC Cancer* 2016;16:115.
- 93 Friedman GK, Moore BP, Nan L, *et al.* Pediatric medulloblastoma xenografts including molecular subgroup 3 and CD133+ and CD15+ cells are sensitive to killing by oncolytic herpes simplex viruses. *Neuro Oncol* 2016;18:227–35.
- 94 Lim KJ, Bisht S, Bar EE, *et al.* A polymeric nanoparticle formulation of curcumin inhibits growth, clonogenicity and stem-like fraction in malignant brain tumors. *Cancer Biol Ther* 2011;11:464–73.
- 95 Hatcher H, Planalp R, Cho J, *et al.* Curcumin: from ancient medicine to current clinical trials. *Cell Mol Life Sci* 2008;65:1631–52.
- 96 Maheshwari RK, Singh AK, Gaddipati J, *et al.* Multiple biological activities of curcumin: a short review. *Life Sci* 2006;78:2081–7.
- 97 Xiao X, Tang J-J, Peng C, *et al.* Cholesterol modification of smoothed is required for hedgehog signaling. *Mol Cell* 2017;66:154–62.
- 98 Hentschel A, Zahedi RP, Ahrends R. Protein lipid modifications—More than just a greasy ballast. *Proteomics* 2016;16:759–82.
- 99 Wu F, Zhang Y, Sun B, *et al.* Hedgehog signaling: from basic biology to cancer therapy. *Cell Chemical Biology* 2017;24:252–80.
- 100 Bell JB, Rink JS, Eckerdt F, *et al.* HdI nanoparticles targeting sonic hedgehog subtype medulloblastoma. *Sci Rep* 2018;8:1211.
- 101 Enguita-Germán M, Schiapparelli P, Rey JA, *et al.* Cd133+ cells from medulloblastoma and PNET cell lines are more resistant to cyclopamine inhibition of the sonic hedgehog signaling pathway than CD133– cells. *Tumour Biol* 2010;31:381–90.
- 102 Ronci M, Catanzaro G, Pieroni L, *et al.* Proteomic analysis of human sonic hedgehog (Shh) medulloblastoma stem-like cells. *Mol Biosyst* 2015;11:1603–11.
- 103 Chow K-H, Shin D-M, Jenkins MH, *et al.* Epigenetic states of cells of origin and tumor evolution drive tumor-initiating cell phenotype and tumor heterogeneity. *Cancer Res* 2014;74:4864–74.
- 104 Po A, Abballe L, Sabato C, *et al.* Sonic hedgehog medulloblastoma cancer stem cells Mimome and transcriptome highlight novel functional networks. *Int J Mol Sci* 2018;19:2326.
- 105 Po A, Ferretti E, Miele E, *et al.* Hedgehog controls neural stem cells through p53-regulation of Nanog. *Embo J* 2010;29:2646–58.
- 106 Corcoran RB, Scott MP. A mouse model for medulloblastoma and basal cell nevus syndrome. *J Neurooncol* 2001;53:307–18.
- 107 Tanori M, Pannicelli A, Pasquali E, *et al.* Cancer risk from low dose radiation in Ptch1/ mice with inactive DNA repair systems: therapeutic implications for medulloblastoma. *DNA Repair* 2019;74:70–9.
- 108 Tanno B, Leonardi S, Babini G, *et al.* Nanog-driven cell-reprogramming and self-renewal maintenance in PTCH1 +/- granule cell precursors after radiation injury. *Sci Rep* 2017;7:14238.
- 109 Rahnama F, Toftgård R, Zaphiropoulos PG. Distinct roles of PTCH2 splice variants in hedgehog signalling. *Biochem J* 2004;378:325–34.
- 110 Miele E, Po A, Begalli F, *et al.* β -arrestin1-mediated acetylation of Gli1 regulates Hedgehog/Gli signaling and modulates self-renewal of SHH medulloblastoma cancer stem cells. *BMC Cancer* 2017;17:488.
- 111 Pierfelice TJ, Schreck KC, Eberhart CG, *et al.* Notch, neural stem cells, and brain tumors. *Cold Spring Harb Symp Quant Biol* 2008;73:367–75.
- 112 Raffel C, Jenkins RB, Frederick L, *et al.* Sporadic medulloblastomas contain PTCH mutations. *Cancer Res* 1997;57:842–5.
- 113 Reifenberger J, Wolter M, Weber RG, *et al.* Missense mutations in SMOH in sporadic basal cell carcinomas of the skin and primitive neuroectodermal tumors of the central nervous system. *Cancer Res* 1998;58:1798–803.
- 114 Eberhart CG, Tihan T, Burger PC. Nuclear localization and mutation of β -catenin in medulloblastomas. *J Neuropathol Exp Neurol* 2000;59:333–7.
- 115 Wang Y, Wang Y, Chen H, *et al.* Endothelial cells promote formation of medulloblastoma stem-like cells via Notch pathway activation. *J Mol Neurosci* 2017;63:152–8.
- 116 Fan X, Matsui W, Khaki L, *et al.* Notch pathway inhibition depletes stem-like cells and blocks engraftment in embryonal brain tumors. *Cancer Res* 2006;66:7445–52.
- 117 Pistollato F, Rampazzo E, Persano L, *et al.* Interaction of hypoxia-inducible factor-1 α and Notch signaling regulates medulloblastoma precursor proliferation and fate. *Stem Cells* 2010;28:1918–29.
- 118 Northcott PA, Shih DJH, Peacock J, *et al.* Subgroup-specific structural variation across 1,000 medulloblastoma genomes. *Nature* 2012;488:49–56.
- 119 Frasson C, Rampazzo E, Accordi B, *et al.* Inhibition of PI3K signalling selectively affects medulloblastoma cancer stem cells. *Biomed Res Int* 2015;2015:1–11.
- 120 Zhou J, Wulfkühle J, Zhang H, *et al.* Activation of the PTEN/mTOR/STAT3 pathway in breast cancer stem-like cells is required for viability and maintenance. *Proc Natl Acad Sci U S A* 2007;104:16158–63.
- 121 Hambardzumyan D, Becher OJ, Rosenblum MK, *et al.* PI3K pathway regulates survival of cancer stem cells residing in the perivascular niche following radiation in medulloblastoma in vivo. *Genes Dev* 2008;22:436–48.
- 122 Hambardzumyan D, Becher OJ, Holland EC. Cancer stem cells and survival pathways. *Cell Cycle* 2008;7:1371–8.
- 123 Roussel MF, Robinson GW. Role of MYC in medulloblastoma. *Cold Spring Harb Perspect Med* 2013;3:a014308.
- 124 Pfaff E, Remke M, Sturm D, *et al.* Tp53 mutation is frequently associated with *CTNMB1* mutation or *MYCN* amplification and is compatible with long-term survival in medulloblastoma. *J Clin Oncol* 2010;28:5188–96.
- 125 Becher OJ, Holland EC. Sox2, a marker for stem-like tumor cells in skin squamous cell carcinoma and hedgehog subgroup medulloblastoma. *Embo J* 2014;33:1984–6.
- 126 Venkataraman S, Alimova I, Balakrishnan I, *et al.* Inhibition of BRD4 attenuates tumor cell self-renewal and suppresses stem cell signaling in Myc driven medulloblastoma. *Oncotarget* 2014;5:2355–71.
- 127 Schuettengruber B, Bourbon H-M, Di Croce L, *et al.* Genome regulation by polycomb and trithorax: 70 years and counting. *Cell* 2017;171:34–57.
- 128 Chan LH, Beckedorff F, Zhang Y, *et al.* Polycomb complexes associate with enhancers and promote oncogenic transcriptional programs in cancer through multiple mechanisms. *Nat Commun* 2018;9:3377.
- 129 Chowdhury M, Mihara K, Yasunaga S, *et al.* Expression of polycomb-group (PcG) protein Bmi-1 predicts prognosis in patients with acute myeloid leukemia. *Leukemia* 2007;21:1116–22.
- 130 Nacerddine K, Baudry J-B, Ginjala V, *et al.* Akt-Mediated phosphorylation of BMI1 modulates its oncogenic potential, E3 ligase activity, and DNA damage repair activity in mouse prostate cancer. *J Clin Invest* 2012;122:1920–32.
- 131 Sauvageau M, Sauvageau G. Polycomb group proteins: multi-faceted regulators of somatic stem cells and cancer. *Cell Stem Cell* 2010;7:299–313.
- 132 Spemann A, van Lohuizen M. Polycomb silencers control cell fate, development and cancer. *Nat Rev Cancer* 2006;6:846–56.
- 133 Leung C, Lingbeek M, Shakhova O, *et al.* Bmi1 is essential for cerebellar development and is overexpressed in human medulloblastomas. *Nature* 2004;428:337–41.
- 134 Wang X, Venugopal C, Manoranjan B, *et al.* Sonic hedgehog regulates Bmi1 in human medulloblastoma brain tumor-initiating cells. *Oncogene* 2012;31:187–99.
- 135 Calao M, Sekyere EO, Cui HJ, *et al.* Direct effects of BMI1 on p53 protein stability inactivates oncoprotein stress responses in embryonal cancer precursor cells at tumor initiation. *Oncogene* 2013;32:3616–26.

- 136 Manoranjan B, Wang X, Hallett RM, *et al.* Foxg1 interacts with BMI1 to regulate self-renewal and tumorigenicity of medulloblastoma stem cells. *Stem Cells* 2013;31:1266–77.
- 137 Margueron R, Reinberg D. The polycomb complex PRC2 and its mark in life. *Nature* 2011;469:343–9.
- 138 Alimova I, Venkataraman S, Harris P, *et al.* Targeting the enhancer of zeste homologue 2 in medulloblastoma. *Int J Cancer* 2012;131:1800–9.
- 139 Suva M-L, Riggi N, Janiszewska M, *et al.* Ezh2 is essential for glioblastoma cancer stem cell maintenance. *Cancer Res* 2009;69:9211–8.
- 140 Liu H, Sun Q, Sun Y, *et al.* Melk and EZH2 cooperate to regulate medulloblastoma cancer stem-like cell proliferation and differentiation. *Molecular Cancer Research* 2017;15:1275–86.
- 141 Waltz DA, Natkin LR, Fujita RM, *et al.* Plasmin and plasminogen activator inhibitor type 1 promote cellular motility by regulating the interaction between the urokinase receptor and vitronectin. *J Clin Invest* 1997;100:58–67.
- 142 Asuthkar S, Gogineni VR, Rao JS, *et al.* Nuclear translocation of Hand-1 acts as a molecular switch to regulate vascular radiosensitivity in medulloblastoma tumors: the protein uPAR is a cytoplasmic sequestration factor for Hand-1. *Mol Cancer Ther* 2014;13:1309–22.
- 143 Asuthkar S, Gondi CS, Nalla AK, *et al.* Urokinase-type plasminogen activator receptor (uPAR)-mediated regulation of WNT/ β -catenin signaling is enhanced in irradiated medulloblastoma cells. *J Biol Chem* 2012;287:20576–89.
- 144 Szakács G, Paterson JK, Ludwig JA, *et al.* Targeting multidrug resistance in cancer. *Nat Rev Drug Discov* 2006;5:219–34.
- 145 Coyle B, Kessler M, Sabnis DH, *et al.* Abcb1 in children's brain tumours. *Biochem Soc Trans* 2015;43:1018–22.
- 146 Othman RT, Kimishi I, Bradshaw TD, *et al.* Overcoming multiple drug resistance mechanisms in medulloblastoma. *Acta Neuropathol Commun* 2014;2.
- 147 Strebhardt K. Multifaceted polo-like kinases: drug targets and antitargets for cancer therapy. *Nat Rev Drug Discov* 2010;9:643–60.
- 148 Harris PS, Venkataraman S, Alimova I, *et al.* Polo-Like kinase 1 (Plk1) inhibition suppresses cell growth and enhances radiation sensitivity in medulloblastoma cells. *BMC Cancer* 2012;12:80.
- 149 Besharat ZM, Sabato C, Po A, *et al.* Low expression of miR-466f-3p sustains epithelial to mesenchymal transition in sonic hedgehog medulloblastoma stem cells through Vegfa-Nrp2 signaling pathway. *Front Pharmacol* 2018;9:1281–81.
- 150 Kaid C, Silva PBG, Cortez BA, *et al.* miR-367 promotes proliferation and stem-like traits in medulloblastoma cells. *Cancer Sci* 2015;106:1188–95.
- 151 Hemmesi K, Squadrito ML, Mestdagh P, *et al.* miR-135a Inhibits Cancer Stem Cell-Driven Medulloblastoma Development by Directly Repressing *Arhgef6* Expression. *Stem Cells* 2015;33:1377–89.
- 152 Lee Y-Y, Yang Y-P, Huang M-C, *et al.* MicroRNA142-3p promotes tumor-initiating and radioresistant properties in malignant pediatric brain tumors. *Cell Transplant* 2014;23:669–90.
- 153 Venkataraman S, Birks DK, Balakrishnan I, *et al.* MicroRNA 218 acts as a tumor suppressor by targeting multiple cancer phenotype-associated genes in medulloblastoma. *J Biol Chem* 2013;288:1918–28.
- 154 Venkataraman S, Alimova I, Fan R, *et al.* MicroRNA 128a increases intracellular ROS level by targeting Bmi-1 and inhibits medulloblastoma cancer cell growth by promoting senescence. *PLoS One* 2010;5:e10748.
- 155 de Antonellis P, Medaglia C, Cusanelli E, *et al.* Mir-34A targeting of Notch ligand delta-like 1 impairs CD15+/CD133+ tumor-propagating cells and supports neural differentiation in medulloblastoma. *PLoS One* 2011;6:e24584.

Appendix S1. Detailed search strategy**PubMed Search Strategy** (last searched on September 16, 2019)

Search	Query	Items found
#7	Search #5 AND #6	121
#6	Search medulloblastoma[MeSH Terms]	6707
#5	Search #1 OR #2 OR #3 OR #4	100519
#4	Search cancer stem cell[MeSH Terms]	19486
#3	Search (cancer stem cell*[tw] OR csc*[tw] OR cancer stem-like cell[tw])	25150
#2	Search tumor-initiating cell[MeSH Terms]	19486
#1	Search (tumor-initiating cell*[tw] OR tic*[tw] OR tumor initiating cell[tw])	66276



**Appendix S2. PRISMA 2009 Flow Diagram:
Medulloblastoma Cancer Stem Cells: Molecular Signatures and
Therapeutic Targets**

