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CONTROVERSIAL REVIEW

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# On the neuronal/neuroblastic nature of medulloblastomas: a tribute to Pio del Rio Hortega and Moises Polak

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**Abstract** The concept that medulloblastomas represent cerebellar neuroblastic tumours was championed by del Rio Hortega in the 1930s and was critically reappraised in the 1960s by Moises Polak. Whereas the aetiology and molecular pathogenesis of medulloblastomas remain unresolved, there is now compelling evidence in support of a fundamentally neuronal tumour phenotype. Tumour cells express in a differentiation-dependent manner a repertoire of neuronal cytoskeletal, synaptic, and other lineage-associated proteins. Neuronal differentiation is more pronounced in the so-called nodular/desmoplastic medulloblastomas, which are typified by areas of neoplastic neuritogenesis ("pale islands") marked by the co-expression of neuronal marker proteins and neurotrophin receptors TrkA and TrkC, low proliferative indices, and apoptosis. The pale islands contain meshworks of reactive astrocytes as part of mutually inductive tumour-stromal cell interactions. However, overt glial differentiation or gliomatous transformation are uncommon. There is growing evidence to support the hypothesis that distinct subtypes of medulloblastomas may implicate transformed neuroblasts from two separate neuroepithelial sources: (a) the velum medullare for a subset of classic medulloblastomas, and (b) the external granule layer for the nodular/desmoplastic medulloblastomas as well as certain classic medulloblastomas. The nosological position of medulloblastomas is discussed in the context of the so-called embryonal central nervous system tumours with emphasis on the cerebral and cerebellar neuroblastomas. We give credence to the view that the medulloblastoma belongs to a group of central neuronal/neuroblastic tumours and call for a critical re-evaluation of its present taxonomic placement.

**Keywords** Medulloblastoma · Cerebral/cerebellar neuroblastoma · Neuroblastic tumour · Neuronal differentiation · Histogenesis

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## Introduction

"The greater part of tumours called medulloblastomata are potential or real neuroblastomata; some of them are potential or real isomorphous glioblastomata, and none offers (the elements) of bipotentiality or ambiguity that are attributed to them." Pio del Rio Hortega [82].

The origin(s) of medulloblastoma, one of the most common malignant central nervous system (CNS) tumours in children, are traceable to cerebellar embryogenesis. The anatomic predilection of medulloblastomas may be explained by the unique developmental timetable of the human cerebellum, which is governed by a temporospatial gradient of neuroblastic proliferation and neuronal migration through the end of the first postnatal year. In the onco-developmental context, the cerebellum exhibits a wide window of neoplastic vulnerability as compared to the telencephalon and other regions of the neuraxis. The mitotically active neuroblasts of the external granule layer (EGL) are potential targets of neoplastic hit(s) [86], whereas such neoplastic transformation may also involve precursor cells in the ventricular neuroepithelium of the velum medullare (VM) [42, 48, 80].

## Historical background

The histogenesis and differentiation potential of medulloblastoma have been controversial since its original description by Percival Bailey and Harvey Cushing in 1925 [3]. The view that medulloblastomas constitute distinctive cerebellar "embryonal tumours" capable of "bipotential" neuronal and glial differentiation has been historically adopted by members of the Anglo-American [3, 53, 91] and to a lesser extent, German schools [105]. This interpretation is based on the developmental concept of the 19th century German pioneer embryologist Schaper, who maintained that the EGL consists of presumptive "indifferent" precursor cells ("indifferenten Zellen") which differentiate both into glia and neurons [92]. Schaper's dogma, together with the previous claim by Obersteiner that the EGL consists of glial precursors [71], was subsequently challenged by Ramon y Cajal [81] and others (reviewed in [46]) whose work indicates that the EGL constitutes a neuroblastic matrix *sui generis* giving rise solely to granule, basket, and stellate neurons of the cerebellar cortex.

The alternative hypothesis that medulloblastomas constitute "cerebellar neuroblastomas" was championed by del Rio Hortega [82, 83] (Fig. 1), in keeping with the embryological concepts of Ramon y Cajal [81]. This view was shared by Stevenson and Echlin [99] (who even suggested the term "granuloblastoma" because of the remarkable resemblance of tumour cells to the neuroblasts of the EGL) but remained in the realm of oblivion until the introduction of immunohistochemistry (reviewed in [42]). In the 1960s, the concept of "cerebellar neuroblastoma" was critically reappraised by Moises Polak [78, 79], a protégé of del Rio Hortega in Buenos Aires, whose premature death marked the end of a passionate voice of the del Rio Hortega school on the subject. Yet, as a matter of historical interest, one wonders why was the palpable neuronal/neuroblastic nature of medulloblastoma underrated for so long.

**Fig. 1** A rare joint photograph of Don Pio and Dorothy S. Russell at The Radcliffe Infirmary, Oxford during WW II (1939/1940). Pio del Rio Hortega was the original proponent of the cerebellar neuroblastoma concept, while Dorothy Russell had subsequently assigned medulloblastoma to the group of "Tumours of the neurone series" in the first edition of the classical Russell and Rubinstein textbook [90]. Courtesy of Dr. Jennian F. Geddes and the archives of The Royal London Hospital. Reprinted from [23] by permission of the British Neuropathological Society

There is much credence in the view that the histological methods employed in the study of brain tumours, i.e. before the advent of immunohistochemistry, largely reflected the interpretation of the tumour's origin and differentiation potential. During the 1920s and 1930s, concepts of brain tumour classification were governed and dictated by national scientific traditions or "schools", which were in turn circumscribed by their own methodological approaches and attendant limitations. Thus, the German school emphasised heavily the use of reticulin stains causing an exaggerated interpretation of tumours infiltrating the leptomeninges as "meningeal sarcomas". An illustrative example in this regard is the "circumscribed arachnoidal sarcoma", a term coined in 1939 by Foerster and Gagel [21]. Rubinstein and Northfield later redefined this nosologically distinctive neoplasm that exhibits a cerebellar hemispheric predilection, as desmoplastic medulloblastoma [88]. It is noteworthy that Zülch conspicuously downplayed the issue of neuronal differentiation in medulloblastoma even in the third and last edition of his book [105].

Historically, the Anglo-American school was too heavily influenced by the use of aniline dyes, particularly phosphotungstic acid haematoxylin (PTAH), which delineate glial fibres well but have profound limitations with respect to the visualisation of immature neurons. Penfield was unable to find evidence of neuroepithelial differentiation in medulloblastoma [75]. On the other hand, Russell and Rubinstein had expressly recognised that medulloblastomas are predominantly neuronal/neuroblastic tumours that also retain a capacity for "spongioblastic" (glial) differentiation [90]. However, the authors' circumspection in this regard is reflected in the statement, "It must be admitted that histological evidence of bipotentiality in a single tumour is uncommon" [90][1]. The degree to which Dorothy S. Russell's views on the subject were influenced by her interaction with del Rio Hortega in the (pre-) World War II years at Oxford is a matter of tempting speculation (Fig. 1). It is noteworthy that in the first (1959) – but also the three subsequent editions (1963–1977) of the classical Russell and Rubinstein textbook – medulloblastoma was included under the category of "Tumours of the neurone series" together with cerebral neuroblastoma, and in this context, the synonym "neuroblastoma" was given [90]. Yet, in the extensively revised fifth

[1]Orville T. Bailey (one of Dorothy Russell's esteemed colleagues and friends in the USA) regarded these tumours as predominantly neuroblastic and incapable of spongioblastic differentiation (cited in [90]).

edition, written exclusively by the late Lucien J. Rubinstein, emphasis was reallocated to the aspect of glial differentiation so much so that the medulloblastoma was construed as a distinctive "embryonal tumour" of the cerebellum capable of "bipotential" or "divergent" glial and neuronal differentiation [91].

Conversely, the protagonists of the Spanish school, in the tradition of Ramon y Cajal, embraced the use of metallic impregnations, a far more eloquent and effective method for the elucidation of neurites in immature neurons. By utilising these histological techniques, del Rio Hortega [82, 83] and Polak [78, 79] succeeded very early on to demonstrate neoplastic neuritogenesis in medulloblastomas and hence formulate their tangible concept in favour of a "cerebellar neuroblastoma".

It should be noted that most of del Rio Hortega's contemporaries on both sides of the Atlantic had viewed with scepticism and, for the most part, dismissed a priori his claim of "cerebellar neuroblastoma" on the grounds of an adamant mistrust about the accuracy and reproducibility of his methodology. The general perception at the time was that Don Pio was intoxicated by his "methods of cookery", while morphologists were generally unenthusiastic about silver stains because of their inherently cumbersome nature and capricious performance.

During the 1970s, the subject became quiescent, but then it gradually regained impetus in the 1980s coinciding with the advent of immunohistochemistry. From this time on, a plethora of publications has emerged substantiating significant degrees of neuronal differentiation in medulloblastomas, aided by the use of antibodies against neuronal cytoskeletal, synaptic, and other lineage-associated proteins.

## The biological relevance of histogenesis and cell lineage identity in brain tumour classification

The combined histogenetic-phenotypic approach to the classification of CNS tumours has historically received wide acceptance as a scheme of histological typing, clinicopathological correlation, and nosological definition [8, 54, 61, 91, 94, 105]. Not only are histogenesis and phenotypic characterisation significant in the context of conventional tumour classification, they are also central to our biological understanding of the rules governing tumour cell growth and differentiation.

Recent advances in developmental neurobiology have generated renewed interest in brain tumour phenotypes. Similar to the haemopoietic system, CNS neoplasia is becoming increasingly understood in the context of the progenitor/stem cell properties of malignant brain tumour cells. Yet, from a historical perspective this is fundamentally the very same approach upon which Bailey and Cushing [3], but especially del Rio Hortega [82], advanced their concepts on brain tumour classification more than seven decades ago. That the phenotype of a tumour is not attributed to a differentiated cell type but determined by the developmental stage of the transformed cell (in a manner analogous to the leukaemias) [103] is not new to neuropathologists. The idea that the identity of progenitor- or precursor-like tumour cells that will dictate – or sway – the mode of differentiation of its clonal progeny has been a long-standing quest in the field of oncological neuropathology.

The plasticity of tripotential glial precursor cells notwithstanding [70], the precursor cell's identity (i.e. neuronal versus glial) in the CNS of mammalian species may be specified before it leaves the proliferative zone [62]. This implies an early cell lineage commitment and, thus, a potentially greater genetic, as opposed to environmental, influence in determining the phenotype of the transformed cell [42]. In addition, the identity of the clonal tumour phenotype may be determined by the distinctive character of the neuroepithelium of origin, the cerebellar EGL being the quintessential example in this regard [42, 46]. Neuronal/neuroblastic tumours of the CNS, unlike their glial counterparts, embody ipso facto features of neuronal differentiation and recapitulate ancestral neuronal phenotypes unique to their neuroepithelium of origin (see below).

In contrast, the "primitive neuroectodermal tumour (PNET) concept" disregards a priori the significance of "cell of origin", claiming the existence of multidirectional phenotypic patterns in a mélange of ostensibly related neoplasms sharing "primitive"[2]-like morphological features [84, 85]. In this context, multipotentiality is construed as evidence derived from far-reaching developmental extrapolation [85]. The PNET concept assumes a tumour origin from a hypothetical common multipotent "neural stem cell" in the ventricular matrix, regardless of anatomic location within the neuraxis.

With regard to the cerebellum, the presence of such a "multipotent cell" remains an open question. In a benchmark experimental study, Sotelo et al. [39] showed that neuronal precursors in the postnatal mouse cerebellum are fully committed and, thus, are not "multipotent cells" whose adult phenotype is supposedly determined by extrinsic influences acting during and/or immediately after their last mitosis.

According to the leading PNET proponent "... many investigators remain fixed upon the notion that discovery of the "cell of origin" of these tumours will somehow enhance understanding of their nature." [85]. However, as is developed below, the premise of this assertion is open to question.

## Current concepts on the histogenesis and differentiation potential of medulloblastoma

Whereas the aetiology and pathogenesis of human medulloblastomas remain under investigation, a consensus has been reached concerning the basic cellular phenotype of these tumours (reviewed in [54, 61]).

[2]In the cellular context, the term "primitive" is problematic, for it may imply primordial or progenitor-like, precursor-like or immature (but still committed as to lineage), or morphologically "undifferentiated" and highly malignant.

**Table 1** Medulloblastoma: neuronal markers (*MAP* microtubule-associated protein)

| Medulloblastoma-neuronal markers |
|---|
| Class III β-tubulin isotype |
| MAP2 (MAP2C) |
| MAP5 (MAP1B) |
| MAPτ |
| Neurofilament protein |
| Synaptophysin |
| Neu-N protein |

The collective evidence in support of a predominantly neuronal phenotype for medulloblastoma is staggering. Tumour cells are elaborated to a variable degree with cell processes with ultrastructural features of neurites (reviewed in [43]) and express a repertoire of neuronal proteins in a differentiation-dependent manner (Table 1). These include the class III β-tubulin isotype (βIII) (Fig. 2a–f), microtubule-associated protein 2 (MAP2), τ protein, neurofilament protein, synaptophysin, Neu-N protein and calbindin-D28k (Fig. 3) [7, 18, 27, 44, 48, 49, 63, 66, 74]. Four differentiating neuronal/neuroblastic phenotypes (Nb1-Nb4) reflecting gradations of neuritic development have been identified based on combined staining patterns for βIII-tubulin and the proliferating cell nuclear antigen (PCNA)/cyclin [49] (Fig. 4).

Neuronal differentiation is more pronounced in the so-called desmoplastic medulloblastomas, hereafter referred to as nodular/desmoplastic medulloblastomas [18] (see below), which are typified by areas of neoplastic neuritogenesis ("pale islands" or "neoplastic nodules") (Fig. 2c–e) [18].

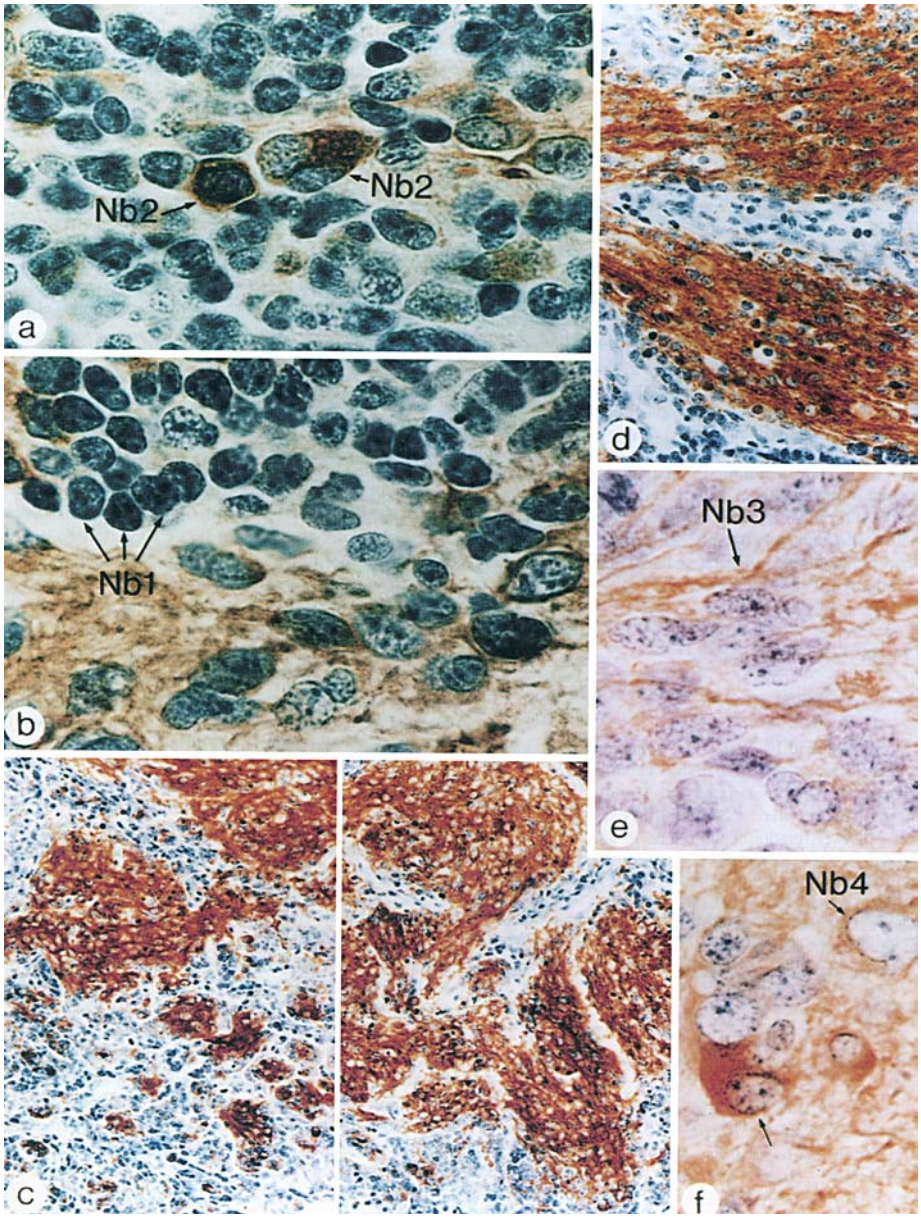


**Fig. 2** **a, b** Immunohistochemical localisation of the neuronal class III β-tubulin (*βIII*), or absence thereof, defines two neuroblastic-like tumour phenotypes in classic medulloblastoma. Nb1 identifies small, round and apolar cells, which are consistently βIII negative. Nb2 refers to somewhat larger, apolar or slightly polar or fusiform cells that exhibit βIII localisation in their "protoperikarya" or growth cone-like expansions. Nb1 and Nb2 phenotypes are depicted by *arrows* in **b** and **a**, respectively. **c–e** Immunoreactivity profile of βIII in nodular/desmoplastic medulloblastoma. Areas of neoplastic neuritogenesis referred to as the "pale islands" or "neoplastic nodules" display strong βIII staining. Individual tumour cells within the "pale islands" exhibit morphological features of immature bipolar neurons elaborating variably elongated βIII-positive neurites (Nb3 phenotype). **f** Localisation of βIII in "medulloblastoma with ganglion cells". βIII staining is present in the perikarya and neuropil of presumptively neoplastic ganglion cells (Nb4 phenotype). From [49] reproduced by permission, Dustri-Verlag Dr. Karl Feistle

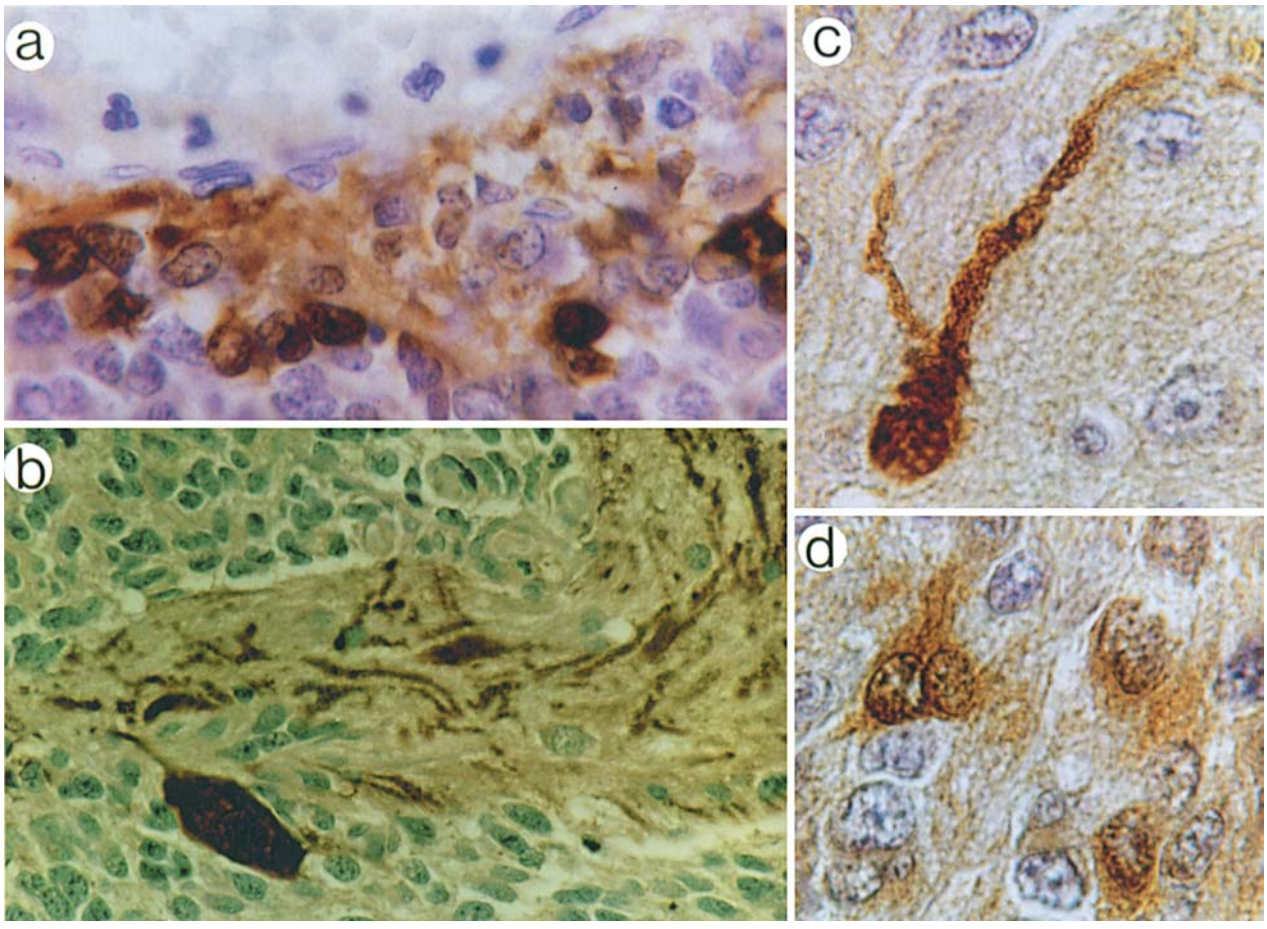


**Fig. 3** **a** Classic medulloblastoma. Robust calbindin-D28k staining of tumour cells with dual nuclear and cytoplasmic localisations. Most of the immunolabelled cells conform to the Nb2 phenotype. **b** Nodular/desmoplastic medulloblastoma. Area of cortical infiltration by tumour with an entrapped Purkinje neuron retaining calbindin-D28k staining in its soma and disrupted apical dendrites. There is lack of calbindin-D28k immunoreactivity in the adjoining neoplastic cells. **c, d** "Medulloblastoma with ganglion cells" containing calbindin-D28k-positive neuronal phenotypes. Some cells possess apical dendrite-like processes devoid of tertiary branches/branchlets, or dendritic spines (**c**), while others display two nuclei (**d**). From [48] reproduced by permission, College of American Pathologists

A subset of medulloblastomas expresses the retinal S-antigen (arrestin), a feature of photosensory differentiation, alluding to a phenotypic, and possibly ontological, kinship among certain medulloblastomas, pineal parenchymal tumours, such as pineocytomas and pineoblastomas, and retinoblastomas [5, 58, 59, 63, 76].

The morphological resemblance of tumour cells in medulloblastomas to the cells of the EGL has been long recognised in human tumours [40, 64, 93, 99] and experimental animal model systems. These include the polyoma (JC) virus-induced medulloblastoma-like neoplasms in hamsters [65, 68, 106], transgenic mice overexpressing the large T-antigen of the JC virus (reviewed in [60]), and mice heterozygous for the *PTCH* gene mutation [15, 32]. The VM has been proposed as an alternative source of origin [80].

Based on the differential developmental expression of calbindin-D28k and βIII, we have previously suggested that distinct subtypes of medulloblastomas may implicate closely related neuroblasts from two different progenitor sources: (1) the VM, for most (but not all) classic medulloblastomas, and (2) the EGL, for nodular/desmoplastic medulloblastomas [42, 48] (Table 2). Calbindin-D28k is an VM-associated neuronal calcium binding protein, which is not expressed in the EGL or its progeny [46]. Conversely, βIII is expressed metachronously in the neuronal progeny of both neuroepithelial matrix derivatives of the rhombic lip [46].

According to our hypothesis, the nodular/desmoplastic medulloblastomas, which tend to favour the cerebellar hemispheres and are typically found (albeit not invariably [44]) in an group of older patients (adolescents and young adults), originate from neoplastic transformation of EGL neuroblasts. The occurrence of these tumours in patients up to 45 years [44] and beyond (C.D.K., unpublished data) may be explained by a late neoplastic hit involving persistent orthotopic or heterotopic remnants of EGL in the adult cerebellum.

By contrast, a subset of classic medulloblastomas, which predominantly involve the vermis and are preferentially seen in children, may arise from calbindinergic neuronal precursor cells of the VM, i.e. at an earlier stage of cerebellar organogenesis [86]. In this regard, calbindin-D28k is expressed in the human medulloblastoma line D-283

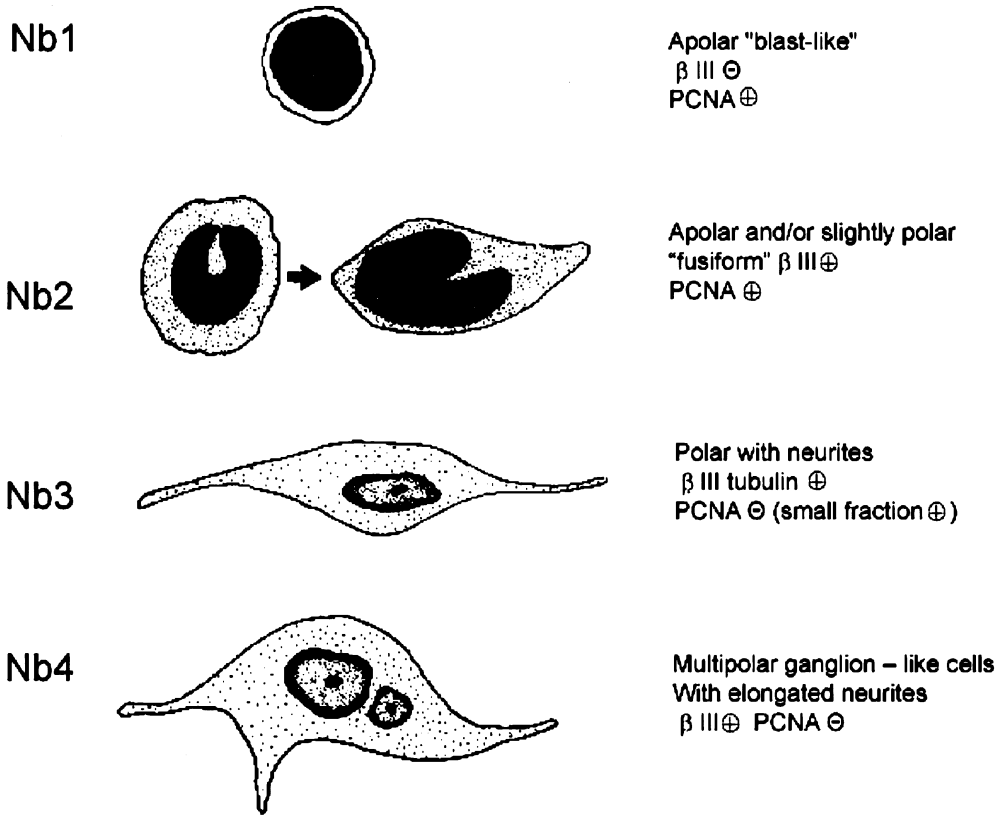


**Fig. 4** Schematic representation of the four proposed neuronal/neuroblastic tumour phenotypes in medulloblastomas according to class III β-tubulin and proliferating cell nuclear antigen/cyclin labelling. From [49] reproduced by permission, Dustri-Verlag Dr. Karl Feistle

**Table 2** Medulloblastoma: dual origin hypothesis (*βIII* class III β-tubulin isotype, *Calb* calbindin-D28 k, *EGL* external granule layer, *MB* medulloblastoma, *VM* ventricular matrix of the velum medullare – posterior medullary velum)

| External granule layer | Velum medullare |
|---|---|
| Nodular/desmoplastic MBs | Subset of classic MBs |
| Subset of classic MBs | MBs w/ganglion cells |
| EGL-like phenotype $\beta III^+$/ $p75^{NTR+}$/$Calb^-$ | VM-like phenotype $\beta III^+$/ $p75^{NTR-}$/$Calb^+$ |
| | Glial differentiation (rare) |

Med that was originally derived from metastatic implants of a classic medulloblastoma in a 6-year-old child [48, 102]. Consistent with our earlier observations, calbindin-D28k has been recently found in medulloblastomas of a group of younger patients, where it is associated with recurrence and poor prognosis [74]. That said, subsets of classic medulloblastomas may have two separate origins, the VM or the EGL [42].

The "dual origin hypothesis" was independently corroborated by Pietsch et al. [7], who showed that the neurotrophin receptor $p75^{NTR}$, which is expressed in EGL neuroblasts, was detected in 71% of nodular/desmoplastic, as compared to only 17% of classic medulloblastoma specimens. According to these authors, cerebellar hemispheric predilection, older patient age, positive $p75^{NTR}$ status, and negative calbindin-D28k status are features of EGL-derived nodular/desmoplastic medulloblastomas; in contrast, most classical childhood/midline medulloblastomas exhibit negative $p75^{NTR}$ and a positive calbindin-D28k profiles, thus pointing to an origin from the VM [7].

The localisation of the neurotrophin receptor TrkC and the cell cycle kinase inhibitor p27kip1 in the "pale islands", reaffirms the phenotypic kinship between the progeny of the EGL and the neuronally differentiating tumour cells in nodular/desmoplastic medulloblastomas [18]. Mutations in the sonic hedgehog pathway gene *PTCH* are considerably more common in the nodular/desmoplastic as compared to the classic subtype [77]. Mutant mice heterozygous for *PTCH* exhibit abnormal proliferations of EGL neuroblasts and have a proclivity to develop medulloblastoma-like tumours [15].

Exceptionally, mature ganglion cells are detected [8, 42, 79, 94]. Some of these cells feature a calbininergic Purkinje cell phenotype, denoting a presumptive origin from the VM for the rare subset of "medulloblastomas with ganglion cells" [42, 48] (Fig. 3c, d). However, it is unclear whether these ganglion cells are (a) neoplastic (products of terminal neuronal differentiation), (b) heterotopic (pre-existing elements of cerebellar heterotopias), or (c) in certain instances, entrapped orthotopic neurons as part of the neoplastic process [42, 48]. Evidence of post-therapy ganglionic maturation has been reported in rare instances of classic medulloblastoma [12] and cerebellar neuroblastoma [16].

Phenotypic features of a neuronal nature characterise the "large cell/anaplastic cell" variant of medulloblastoma that has emerged as a distinctive – albeit rare – subtype associated with a highly aggressive behaviour [6, 25].

In our view, the question of divergent glial differentiation in medulloblastomas should be approached with caution, for it remains to a large extent a matter of interpreta-

tion even among astute morphologists. It is our position that astrocytic proliferations in medulloblastomas are, for the most part, stromal or reactive. They are typically associated with the "pale islands" of the nodular/desmoplastic medulloblastomas, where they may play a role in cell-to-cell interactions and mutual neuronal-glial differentiation induction [43, 44, 63], analogous to the non-neoplastic Schwann cells during maturation of sympathoadrenal neuroblastomas [47].

By the same token, glial differentiation is unambiguously documented in rare instances of classic medulloblastomas, principally in the form of poorly differentiated cell populations marked by glial fibrillary acidic protein staining [4, 8, 42, 54, 61, 89, 91]. Glial-like neoplastic phenotypes may represent clonal expansions of cotransformed glial precursors in the VM, "transdifferentiation" phenomena [103], or spontaneous neoplastic transformation of astroglial stromal cells [42]. Genetic instability, mutational change and/or other events that may alter the morphological character of a tumour consequent to time and treatment are relevant considerations in this regard. As an example, delayed gliomatous transformation is a well recognised sequel of irradiation administered for the treatment of medulloblastomas [56, 97].

"Divergent" neuronal and glial differentiation has been demonstrated by Herman and Rubinstein [34] in a primary explant of medulloblastoma maintained in an organ culture system that is known to favour glial differentiation [50, 87]. However, an overgrowth of entrapped astrocytes in the primary explant cannot be ruled out based on the methods employed in that study [34]. Overall, human medulloblastoma cell lines show predominantly neuronal phenotypic features [33, 48, 52, 101].

## The position of medulloblastoma in the context of embryonal CNS tumours with special attention to cerebellar and cerebral neuroblastomas

The suffix -blastoma means different things to different people [57]. According to Bailey and Cushing [3], it denotes tumours that are basically undifferentiated and highly malignant. The common glioblastoma, a predominantly adult tumour fits into this definition (cited in [57]). Admittedly, this is an atypical use of the term in the early nomenclature of brain tumours, perhaps even a misnomer, which, however, remains deeply entrenched in clinical practice. The time-honoured exception of glioblastoma notwithstanding, "blastomas" are by strict pathological definition tumours that retain and express the potency of the embryonic primordium from which they arise [57].

In the human CNS, such neoplasms have been traditionally classified under the rubric of "embryonal tumours" [8, 54, 55, 61, 91]. Doubtless, the vast majority of them are medulloblastomas, whereas retinoblastomas, cerebral neuroblastomas, pineoblastomas, and ependymoblastomas are much less common, albeit well established entities [8, 54, 55, 61, 91]. These tumours embody a unique embryonic capacity, which is reflected in their predominant phenotype that conforms to certain reproducible patterns of lineage-dependent differentiation. In addition, the progenitor-like neoplastic phenotype may well be traceable to the neuroepithelium of origin, as suggested in medulloblastomas [42, 48, 86, 91] and retinoblastomas [45, 86, 91]. With the exception of ependymoblastoma most of the remaining "embryonal tumours" are fundamentally neuroblastic, although they retain certain signature features of the regional neuroepithelial matrix (or anlagen) from which they originate.

### The paradigm of nodular/desmoplastic medulloblastoma

The nodular/desmoplastic medulloblastoma is an unequivocal example of a cerebellar neuronal/neuroblastic tumour. Histologically, it exhibits a biphasic, nodular (follicular) architecture in which reticulin-impregnated, densely populated sheets of poorly differentiated tumour cells surround loosely textured, finely fibrillated and reticulin-free tumoral structures referred to as the "pale islands" or "neoplastic nodules". The latter constitute areas of overt neuronal differentiation marked by neoplastic neuritogenesis, as evidenced by ultrastructural features [43] and the expression of neuronal marker proteins [7, 9, 35, 43, 44, 49, 63, 66] and neurotrophin receptors (TrkA and TrkC) [18].

Aside from their uniform histology, nodular/desmoplastic medulloblastomas represent a homogeneous group with respect to their ploidy characteristics as ~90% have a near-diploid main mode and a "monomodal" DNA distribution diagram [24]. In contrast, classic medulloblastomas constitute a heterogeneous group of neoplasms exhibiting aneuploidy and heterogeneity as regards histogram type distribution [24].

The increased distribution of the pro-apoptotic p27kip1 coupled with the decreased immunoreactivity for the anti-apoptotic Bcl-2 in the "pale islands" [1, 18, 96] are consistent with the morphological features of apoptosis that are frequently encountered in areas of neuronal differentiation in medulloblastomas [18, 44, 95, 96, 98]. Apoptosis in conjunction with markedly decreased Ki-67, PCNA

**Table 3** "Pale islands" or "neoplastic nodules" of nodular/desmoplastic medulloblastomas (*GFAP* glial fibrillary acidic protein, *PCNA* proliferating cell nuclear antigen, ↑↑ markedly increased immunohistochemical staining, ↑ increased immunohistochemical staining, ↓↓ markedly decreased immunohistochemical staining, ↓ decreased immunohistochemical staining)

| | |
|---|---|
| ↑↑ | Neuronal proteins-neuritogenesis |
| ↑ | Neurotrophin receptors TrkA and TrkC |
| ↓↓ | Ki-67/MIB-1 and PCNA/cyclin |
| ↓ | p53 immunoreactivity |
| ↓ | Bcl-2 (anti-apoptotic) |
| ↑ | p27/Kip1 (pro-apoptotic) |
| ↑ | Apoptosis/apoptotic bodies |
| ↑ | $GFAP^+$ stromal astrocytes |

and p53 labelling indices, as well as increased TrkA and TrkC expression in the "pale islands" [18, 42, 49], may contribute to the more favourable prognosis associated with the nodular/desmoplastic subtype [26] (Table 3). In this regard, the nodular/desmoplastic medulloblastoma is biologically and nosologically linked to – and frequently indistinguishable from – the cerebellar neuroblastoma that is seen predominantly in infants [26, 69, 73]. The relationship between these two tumours may be viewed as a continuum reflecting gradations of neuronal differentiation, rather than as two separate tumour entities.

Because of the ambiguity of the term "desmoplastic", the designation of "PNET with desmoplasia" underscores yet another persistent clinicopathological problem. Desmoplastic reaction in a medulloblastoma owing to leptomeningeal tumour infiltration is not coequal to nodular/desmoplastic medulloblastoma [35, 42, 44, 54]. Although the relatively favourable prognosis of the latter tumour subtype was recently reappraised [26], for many years the terms "desmoplasia" and "desmoplastic medulloblastoma" were used interchangeably and were considered as poor prognostic indicators [72]. A recent study from the Pediatric Oncology Group in the United States reports that neither the increasing degrees of nodularity nor the presence of desmoplasia are associated significantly with longer survival [19]. We welcome the latest WHO classification definition of "desmoplastic medulloblastomas" on the basis of their neuronally differentiating "pale islands" [54, 55], although we believe that the term "desmoplastic" ought to be phased out in the future.

## The paradigm of cerebral neuroblastoma ("supratentorial PNET")

In spite of their cytogenetic and molecular genetic differences and their less favourable prognosis (reviewed in [54, 55]), cerebral neuroblastomas are allied – by virtue of their basic neuronal phenotype – to the cerebellar medulloblastomas/neuroblastomas [38, 91].

In the nosological context, some of the supratentorial neoplasms originally designated as PNET by Hart and Earle [32] may in fact represent poorly differentiated neu-

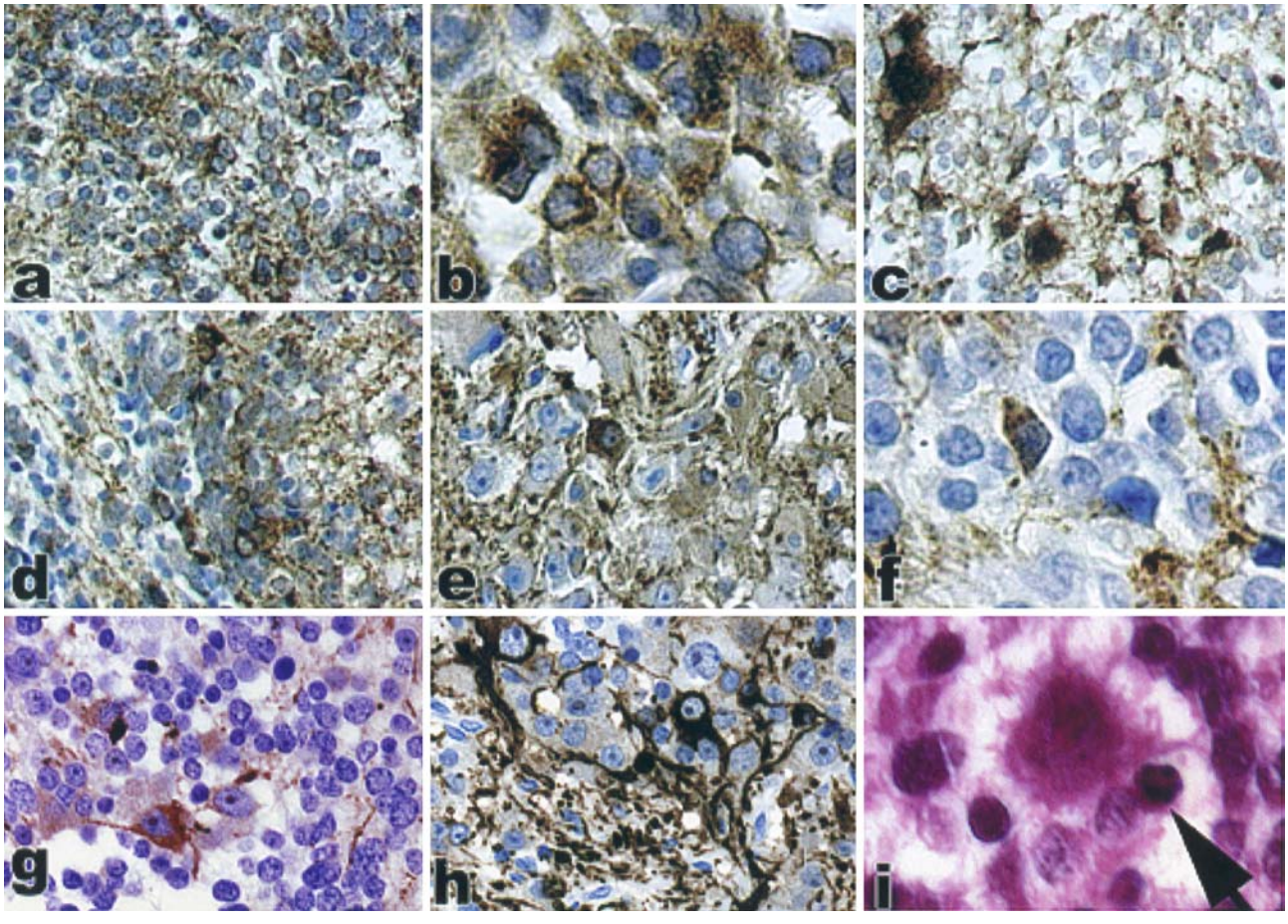


**Fig. 5a–i** Immunohistochemical profile of a cerebral ganglioneuroblastoma using a panel of antibodies to neuron-associated marker proteins. **a–c** Widespread tumour cell immunoreactivity for class III β-tubulin highlights a morphological gamut of neuronal differentiation, which recapitulates the phenotypic gradient seen in cerebellar medulloblastomas. **d** Staining of tumour cells using a monoclonal antibody to phosphorylated τ protein. **e**, **f** Synaptophysin labelling of both mature ganglion cells and less well-differentiated neoplastic neurons; note "immature pattern" of perikaryal localisation. **g**, **h** Phosphoneurofilament staining is present predominantly in neoplastic ganglion cells (**h**) but also in occasional less differentiated tumour cells. **i** Apoptotic body (*arrow*) in an area of neuronal/"ganglionic" differentiation

roblastic tumours without overt morphological features of neuronal differentiation. In support of this contention is the placement of the cerebral neuroblastomas/ganglioneuroblastomas under the category of "supratentorial PNET" in the latest WHO classification of tumours of the nervous system [54].

According to Horten and Rubinstein [38], "the cellular nature and biological behaviour of cerebral neuroblastoma recall those of the cerebellar medulloblastoma". As exemplified in a case of cerebral ganglioneuroblastoma arising in the parietal region in a 2-year-old girl, there are striking phenotypic similarities between the latter tumour and medulloblastomas, which exhibit neuritogenesis and ganglionic maturation. Early neuronal differentiation coincides with widespread protoperikaryal and neuritic localisation of the neuronal class III β-tubulin (Fig. 5a–c), and focal detection of τ protein (Fig. 5d) and synaptophysin (Fig. 5e, f). Phosphoneurofilament immunoreactivity is present predominantly, but not exclusively, in tumour cells exhibiting ganglionic maturation (Fig. 5g, h). Thus, the cellular distribution of neuronal marker proteins in cerebral (ganglio) neuroblastomas is differentiation dependent, recapitulating a temporospatial gradient of expression seen in neuronal development [46, 51] and neoplasia [22, 36, 44, 45, 47]. Apoptotic cells in areas of neuronal differentiation (Fig. 5i) are analogous to those encountered in medulloblastomas (see above).

## Central versus peripheral neuroblastic tumours

Cerebellar medulloblastomas, cerebral neuroblastomas/ganglioneuroblastomas, pineoblastomas, and retinoblastomas represent neoplasms related to the transformation of presumptive CNS neuronal restricted precursor cells (neural tube-derived neuroblasts), while sympathoadrenal neuroblastomas/ganglioneuroblastomas/ganglioneuromas are the neoplastic analogues of sympathoadrenal neuroblasts (neural crest-derived cells)[3]. The heterogeneity of neuronal/neuroblastic tumours rests with the distinctive progenitor cell properties of neoplastic cells that recapitulate ancestral features of the regional neuroepithelial matrix from which they arise (e.g. medulloblastomas from the EGL or VM; retinoblastomas from the outer neuroblastic zone of the retina; and sympathoadrenal neuroblastomas from sympathoadrenal neuroblasts, the precursors of adrenergic neurons).

The recognition of the predominantly neuronal character of medulloblastoma notwithstanding, one of the legitimate concerns pertaining to the classification of this tumour is that the introduction of the terms "neuroblastic" or "neuroblastoma" might actually confuse rather than help. This is because (a) neuroblastoma is a well-established clinicopathological entity outside the CNS with characteristic chromosomal and molecular genetic features (that are absent from medulloblastoma), and (b) a variety of brain tumours have been already designated "neuroblastoma" such as the "supratentorial PNET" and central neurocytoma.

To address this problem of classification one should recognise from the outset that central and peripheral neuroblastic tumours are not identical. Central neuroblastic tumours never secrete catecholamines nor do they exhibit the same chromosomal alterations as their peripheral counterparts [54]. Also, there are cytogenetic and molecular genetic differences amongst tumours of either the central (e.g. medulloblastoma versus cerebral neuroblastoma) or the peripheral neuroblastic tumour categories (e.g. sympathoadrenal versus olfactory neuroblastomas) [54]. Furthermore, individual tumour entities do not always exhibit certain cytogenetic or molecular genetic defining characteristics. Thus, isochrome 17 q, the most common specific cytogenetic abnormality in medulloblastomas is present only in ~50% of cases [54]. Involvement of the *Wnt* signalling pathway by mutually exclusive *adenomatous polyposis coli* or *β-catenin* gene mutations occurs in ~13% of sporadic medulloblastomas, while inactivating mutations of the *PTCH* gene are even rarer [54].

For classification purposes the only unifying feature between medulloblastomas and other allied tumours of the central and peripheral nervous systems is their common neuronal/neuroblastic phenotype and their capacity to undergo overt neuronal differentiation in vivo and in vitro. In this regard, neurotrophins and their cognate receptors TrkA, TrkB and TrkC, which regulate proliferation, differentiation and apoptotic cell death of neuronal progenitor cells/neuroblasts, appear to play a central role in the differentiation and apoptosis of both central and peripheral neuroblastic tumours [13, 14, 20]. Nerve growth factor induces massive apoptosis in human medulloblastoma cell lines expressing the TrkA receptor [67]. It is noteworthy that the expression of high levels of TrkC mRNA is associated with a more favourable prognosis in both patients with medulloblastomas [30, 31] and peripheral neuroblastomas [100].

Collectively, the existence of histogenetic, cytogenetic and molecular genetic differences between medulloblastomas and other central and peripheral neuroblastic tumours do not negate their fundamental phenotypic kinship or the appropriateness of their general classification under distinct/specific entities of central and peripheral neuroblastic tumours. In this connection, the biological diversity and nomenclature of gliomas may be viewed as an analogous case in point. By virtue of their common glial phenotype and despite their molecular genetic differences both "de novo" and "secondary" subtypes of glioblastoma multiforme are classified as astrocytic tumours [54].

[3]The occurrence of primary, poorly differentiated intraspinal neoplasms – some with neural-like features – is exceptionally rare (~18 reported cases in the English literature). Such tumours are currently classified as "spinal PNET" and may be dural, intrathecal, intramedullary, or arise from the spinal roots of the cauda equina [104]. It is presently unclear whether they comprise a single biological entity. In particular, the disseminating lesions originating in the cauda equina have a presumptive peripheral nervous system origin and thus warrant nosological distinction from the sympathetic nervous system-derived "dumbbell" neuroblastomas and the metastatic sympathoadrenal neuroblastomas.

## Other so-called embryonal tumours of the CNS

In contrast to the aforementioned neuronal/neuroblastic tumours, the precise nature of such neoplasms as "medulloepithelioma", "melanotic neuroectodermal tumour of infancy", "medullomyoblastoma", "melanotic medulloblastoma" and atypical teratoid/rhabdoid tumour is less clear. It is possible that some of these exceptionally rare neoplasms may fundamentally represent germ cell tumours with embryonic differentiation potential and thus, may be construed as examples of immature teratomas or teratoids of the CNS. Also, in our experience, the so-called medullomyoblastoma may not be a single entity, but a feature of several small cell neoplasms of the posterior fossa. These include instances of (a) neuroblastic tumours (medulloblastomas) containing rare myoblasts or myotubes as an expression of stromal metaplasia; (b) glioblastomas with rhabdomyoblastic differentiation, (c) embryonal rhabdomyosarcomas mimicking medulloblastomas, and (d) immature teratomas/teratoid with divergent neural and myogenous differentiation. The teratomatous or malformative nature of cerebellar medullomyoblastomas has been previously appraised in this regard [37]. Also, the unusual case of a midcerebellar neoplasm in a 7-year-old boy reported by Dickson et al. [17] illustrates this challenging nosological problem. Although clinically consistent with a medulloblastoma, immunohistochemistry and electron microscopy demonstrated glial and rhabdomyoblastic differentiation [17]. Similarly, atypical teratoid/rhabdoid tumours of the CNS, which are typified by somatic mutations of the gene *hSNF/INI1* (reviewed in [55]), may well represent teratoma variants, given their potential for multidirectional neuroepithelial, mesenchymal and epithelial (squamous and glandular) differentiation.

The so-called lipomatous medulloblastoma (cerebellar liponeurocytoma) is an exceptionally rare tumour encountered predominantly in adults and typified by areas of lipomatous differentiation, evidence of advanced neuronal differentiation, low proliferative potential and a favourable prognosis [54]. In our view, the indolent natural history of this lesion and its relationship to the "lipidised neuroectodermal tumour of the cerebellum with myoid differentiation" [28] point in the direction of a low-grade teratoid or a malformative/dysplastic tumour rather than a true medulloblastoma variant.

Although the neuroepithelial nature of the "primitive medullary epithelium", as well as the presence of divergent neuroblastic, ganglionic, astrocytic, ependymoblastic and ependymal features in the so-called medulloepithelioma are unequivocal [10, 41], similar patterns have also been described in the neuroepithelial component of a spontaneous murine ovarian teratoma [11]. This raises the conjecture of a teratomatous nature for "medulloepithelioma" particularly given its potential for mesenchymal differentiation [2].

## Epilogue

After many decades of heated debate and controversy, the original postulate of del Rio Hortega has proven to be tenable insofar as the existence of immature/precursor-like or differentiating neuronal phenotypes in the so-called medulloblastomas is indisputable. Importantly, in these tumours neuronal differentiation emerges as a major biological attribute with potential clinical implications. Yet, despite the acceptance of the tumour's predominantly neuronal nature by most authorities nowadays [8, 54, 61, 94], medulloblastoma remains ingrained under the taxonomic ordinance of "embryonal tumours" together with biologically disparate neoplasms, such as the atypical teratoid/rhabdoid tumour [54, 55]. Arguably, in the absence of a definable "multipotent cell" in the cerebellum (i.e. a "medulloblast") [46], the term medulloblastoma represents a time-honoured misnomer. In recognition of the fact that this nomenclature is deeply routed in clinical

**Table 4** Working classification of neuronal/neuroblastic tumours of the CNS (modified after Russell and Rubinstein [90]) (*NA* not assigned, *WHO* World Health Organisation)

| Central neuronal/neuroblastic tumours |
|---|
| **I. "Embryonal-type" neuroblastic tumours**[a] |
| 1. Cerebellar medulloblastoma (WHO grade IV)[b] |
| Classic medulloblastoma |
| Nodular/desmoplastic medulloblastoma/cerebellar neuroblastoma |
| "Large-cell/anaplastic" medulloblastoma |
| "Medulloblastoma with ganglion cells" |
| 2. Cerebral neuroblastoma (telencephalon) (WHO grade IV)[b] |
| Classic neuroblastoma |
| Ganglioneuroblastoma |
| 3. Pineoblastoma (pineal region) (WHO grade IV)[b] |
| 4. Retinoblastoma (neural retina) (WHO grade NA) |
| **II. Predominantly "adult-type" neuronal or mixed neuronal-glial tumours**[c] |
| 1. Central neurocytoma (intraventricular) (WHO grade II)[b] |
| 2. Gangliocytoma (WHO grade I)[b] |
| Cerebellar dysplastic gangliocytoma /Lhermitte-Duclos disease |
| Hypothalamic/diencephalic gangliocytoma/hamartoma |
| 3. Ganglioglioma (anywhere in the neuraxis) (WHO grades I, II, III)[b] |
| Desmoplastic infantile ganglioglioma (WHO grade I) |
| 4. Pineocytoma (pineal region) (WHO grade II)[b] |
| Pineal parenchymal tumours with intermediate differentiation |

[a]The nosology of poorly-differentiated primary spinal cord tumours with neural-like features is uncertain. These exceptionally rare neoplasms are currently referred to as primary 'spinal PNETs'. They may be dural, intrathecal or intramedullary, or arise in the cauda equina region. Accordingly, they warrant distinction from sympathoadrenal neuroblastomas metastatic to the spinal cord
[b]WHO classification 2000 [54]
[c]The nosology of the following neoplasms is currently uncertain: extraventricular neurocytoma (including 'intramedullary' and 'cauda equina' neurocytomas), 'cerebellar liponeurocytoma' and 'papillary glioneuronal tumour'

practice, perhaps even to the extent of preserving the tumour's nosological identity [42], we subscribe to the prevailing view that the term medulloblastoma should be retained as a designation *sui generis*. However, we suggest that the latter should be part of a group of central neuronal/neuroblastic tumours (together with the cerebral neuroblastoma, pineoblastoma and retinoblastoma) based on the original classification framework of Russell and Rubinstein [90] (Table 4).

By emphasising the predominantly neuronal/neuroblastic nature of medulloblastoma it is envisaged that this tumour will be explored increasingly in the context of signal transduction pathways governing embryonic neuronal growth, differentiation and apoptosis.

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## References

1. Adesina AM, Dunn ST, Moore WE, Nalbantoglu J (2000) Expression of p27kip1 and p53 in medulloblastoma: relationship with cell proliferation and survival. Pathol Res Pract 196: 243–250
2. Auer RN, Becker LE (1983) Cerebral medulloepithelioma with bone, cartilage, and striated muscle. Light microscopic and immunohistochemical study. J Neuropathol Exp Neurol 42:256–261
3. Bailey P, Cushing H (1925) Medulloblastoma cerebelli: a common type of midcerebellar glioma of childhood. Arch Neurol Psychiatry 14:192–223
4. Barnard RO, Panbakian H (1980) Astrocytic differentiation in medulloblastoma. J Neurol Neurosurg Psychiatry 43:1041–1044
5. Bonnin JM, Perentes E (1988) Retinal S-antigen immunoreactivity in medulloblastomas. Acta Neuropathol 76:204–207
6. Brown HG, Kepner JL, Perlman EJ, Friedman HS, Strother DR, Duffner PK, Kun LE, Goldthwaite PT, Burger PC (2000) "Large cell/anaplastic" medulloblastomas: a Pediatric Oncology Group Study. J Neuropathol Exp Neurol 59:857–865
7. Bühren J, Christoph AH, Buslei R, Albrecht S, Wiestler OD, Pietsch T (2000) Expression of the neurotrophin receptor $p75^{NTR}$ in medulloblastomas is correlated with distinct histological and clinical features: evidence for a medulloblastoma subtype derived from the external granule cell layer. J Neuropathol Exp Neurol. 59:229–240
8. Burger PC, Scheithauer BW (1994) Tumors of the central nervous system. Atlas of tumor pathology. Third Series, Fascicle 10, Armed Forces Institute of Pathology, Washington, DC, pp 193–225
9. Burger PC, Grahmann FC, Bliestle A, Kleihues P (1987) Differentiation in medulloblastoma. A histological and immunohistochemical study. Acta Neuropathol (Berl) 73:115–123
10. Caccamo DV, Herman MM, Rubinstein LJ (1989) An immunohistochemical study of the primitive and maturing elements of human cerebral medulloepitheliomas. Acta Neuropathol 79:248–254
11. Caccamo D, Katsetos CD, Herman MM, Frankfurter A, Collins VP, Rubinstein LJ (1989) Immunohistochemistry of a spontaneous murine ovarian teratoma with neuroepithelial differentiation. Neuron-associated β-tubulin as a marker for primitive neuroepithelium. Lab Invest 60:390–398
12. Cai DX, Mafra M, Schmidt RE, Scheithauer BW, Park TS, Perry A (2000) Medulloblastomas with extensive posttherapy neuronal maturation. Report of two cases. J Neurosurg 93: 330–334
13. Chou TT, Trojanowski JQ, Lee VM (1997) Neurotrophin signal transduction in medulloblastoma. J Neurosci Res 49:522–527
14. Chou TT, Trojanowski JQ, Lee VM (2000) A novel apoptotic pathway induced by nerve growth factor-mediated TrkA activation in medulloblastoma. J Biol Chem 275:565–570
15. Corcoran RB, Scott MP (2001) A mouse model for medulloblastoma and basal cell nevus syndrome. J Neurooncol 53: 307–318
16. De Chadarevian JP, Montes JL, O'Gorman AM, Freeman CR (1987) Maturation of cerebellar neuroblastoma into ganglioneuroma with melanosis. A histologic, immunocytochemical, and ultrastructural study. Cancer 59:69–76
17. Dickson DW, Hart MN, Menezes A, Cancilla PA (1983) Medulloblastoma with glial and rhabdomyoblastic differentiation. A myoglobin and glial fibrillary acidic protein immunohistochemical and ultrastructural study. J Neuropathol Exp Neurol 42:639–647
18. Eberhart CG, Kaufman WE, Tihan T, Burger PC (2001) Apoptosis, neuronal maturation, and neurotrophin expression within medulloblastoma nodules. J Neuropathol Exp Neurol 60:462–469
19. Eberhart CG, Kepner JL, Goldthwaite PT, Kun LE, Duffner PK, Friedman HS, Strother DR, Burger PC (2002) Histopathologic grading of medulloblastomas: a Pediatric Oncology Group study. Cancer 94:552–560
20. Eggert A, Ikegaki N, Liu X, Chou TT, Lee VM, Trojanowski JQ, Brodeur GM (2000) Molecular dissection of TrkA signal transduction pathways mediating differentiation in human neuroblastoma cells. Oncogene 19:2043–2051
21. Foerster O, Gagel O (1939) Das umschriebene Arachoidalsarkom des Kleinhirns. Z Gesamte Neurol Psychiat 164:565–580
22. Frierson HF, Ross GW, Mills SE, Frankfurter A (1990) Olfactory neuroblastomas: additional immunohistochemical characterization. Am J Clin Pathol 94:547–553
23. Geddes JF (1998) Why do we remember Dorothy Russell? Neuropathol Appl Neurobiol 24:268–270
24. Giangaspero F, Chieco P, Ceccarelli C, Lisignoli G, Pozzuoli R, Gambacorta M, Rossi G, Burger PC (1991) "Desmoplastic" versus "classic" medulloblastoma: comparison of DNA content, histopathology and differentiation. Virchows Arch [A] 418:207–214
25. Giangaspero F, Rigobello L, Badiali M, Loda M, Andreini L, Basso G, Zorzi F, Montaldi A (1992) Large-cell medulloblastomas. A distinct variant with highly aggressive behavior. Am J Surg Pathol 16:687–693
26. Giangaspero F, Perilongo G, Fondelli MP, Brisigotti M, Carollo C, Burnelli R, Burger PC, Garre ML (1999) Medulloblastoma with extensive nodularity: a variant with favorable prognosis. J Neurosurg 91:971–977
27. Giordana MT, Cavala P, Duto A, Borsotti L, Chio A, Schiffer D (1997) Is medulloblastoma the same tumor in children and adults? J Neurooncol 35:169–176
28. Gonzalez-Campora R, Weller RO (1998) Lipidized mature neuroectodermal tumour of the cerebellum with myoid differentiation. Neuropathol Appl Neurobiol 24:397–402
29. Goodrich LV, Milenkovic L, Higgins KM, Scott MP (1997) Altered neural cell fates and medulloblastoma in mouse patched mutants. Science 277:1109–1113

30. Grotzer MA, Janss AJ, Fung K, Biegel JA, Sutton LN, Rorke LB, Zhao H, Cnaan A, Phillips PC, Lee VM, Trojanowski JQ (2000) TrkC expression predicts good clinical outcome in primitive neuroectodermal brain tumors. J Clin Oncol 18: 1027–1035
31. Grotzer MA, Hogarty MD, Janss AJ, Liu X, Zhao H, Eggert A, Sutton LN, Rorke LB, Brodeur GM, Phillips PC (2001) MYC messenger RNA expression predicts survival outcome in childhood primitive neuroectodermal tumor/medulloblastoma. Clin Cancer Res 7:2425–2433
32. Hart MN, Earle KM (1973) Primitive neuroectodermal tumours of the brain in children. Cancer 32:890–897
33. He XM, Wikstrand CJ, Friedman HS, Bigner SH, Pleasure S, Trojanowski JQ, Bigner DD (1991) Differentiation characteristics of newly established medulloblastoma cell lines (D384 Med, D425 Med, and D458 Med) and their transplantable xenografts. Lab Invest 64:833–843
34. Herman MM, Rubinstein LJ (1984) Divergent glial and neuronal differentiation in a cerebellar medulloblastoma in an organ culture system: in vitro occurrence of synaptic ribbons. Acta Neuropathol (Berl) 65:10–24
35. Herpers MJHM, Budka H (1985) Primitive neuroectodermal tumours including the medulloblastoma: glial differentiation signaled by immunoreactivity for GFAP is restricted to the pure desmoplastic medulloblastoma ('arachnoidal sarcoma of the cerebellum'). Clin Neuropathol 4:12–18
36. Hessler RB, Lopes MBS, Frankfurter A, Reidy J, VandenBerg SR (1992) Cytoskeletal immunohistochemistry of central neurocytomas. Am J Surg Pathol 16:1031–1038
37. Holl T, Kleihues P, Yasargil MG, Wiestler OD (1991) Cerebellar medullomyoblastoma with advanced neuronal differentiation and hamartomatous component. Acta Neuropathol 82:408–413
38. Horten BC, Rubinstein LJ (1976) Primary cerebral neuroblastoma. A clinicopathological study of 35 cases. Brain 99:735–756
39. Jankovski A, Rossi F, Sotelo C (1996) Neuronal precursors in the postnatal mouse cerebellum are fully committed cells: evidence from heterochronic transplantations. Eur J Neurosci 8:2308–2319
40. Kadin ME, Rubinstein LJ, Nelson JS (1970) Neonatal cerebellar medulloblastoma originating from the fetal external granular layer. J Neuropathol Exp Neurol 29: 583–600
41. Karch SB, Urich H (1972) Medulloepithelioma: definition of an entity. J Neuropathol Exp Neurol 31:27–53
42. Katsetos CD, Burger PC (1994) Medulloblastoma. Semin Diagn Pathol 11:85–97
43. Katsetos CD, Liu HM, Zacks SI (1988) Immunohistochemical and ultrastructural observations on Homer Wright (neuroblastic) rosettes and the "pale islands" of human cerebellar medulloblastomas. Hum Pathol 19:1219–1227
44. Katsetos CD, Herman MM, Frankfurter A, Gass P, Collins VP, Walker CC, Rosemberg S, Barnard RO, Rubinstein LJ (1989) Cerebellar desmoplastic medulloblastomas: a further immunohistochemical characterization of the reticulin-free pale islands. Arch Pathol Lab Med 113:1019–1029
45. Katsetos CD, Herman MM, Frankfurter A, Uffer S, Perentes E, Rubinstein LJ (1991) Neuron-associated class III β-tubulin isotype, microtubule-associated protein 2, and synaptophysin in human retinoblastomas in situ. Further immunohistochemical observations on the Flexner-Wintersteiner rosettes. Lab Invest 64:45–54
46. Katsetos CD, Frankfurter A, Christakos S, Mancall EL, Vlachos I, Urich H (1993) Differential localization of class III β-tubulin isotype (βIII) and calbindin-D28k defines distinct neuronal types in the developing human cerebellar cortex. J Neuropathol Exp Neurol 52: 655–666
47. Katsetos CD, Karkavelas G, Frankfurter A, Vlachos I, Vogeley K, Schober R, Wechsler W, Urich H (1994) The stromal Schwann cell during maturation of peripheral neuroblastomas: immunohistochemical observations with antibodies to the neuronal class III β-tubulin isotype (βIII), and S-100 protein. Clin Neuropathol 13 :171–180
48. Katsetos CD, Herman MM, Krishna L, Vender J, Vinores SA, Agamanolis DP, Schiffer D, Burger PC, Urich H (1995) Calbindin-D28k in medulloblastoma subsets and in the human medulloblastoma cell line D283 Med. Arch Pathol Lab Med 119:735–743
49. Katsetos CD, Krishna L, Frankfurter A, Karkavelas G, Wolfe DE, Valsamis MP, Schiffer D, Vlachos I, Urich H (1995) Cytomorphological scheme of differentiating neuronal phenotypes in cerebellar medulloblastomas based on immunolocalization of class III β-tubulin isotype (βIII) and the proliferating cell nuclear antigen (PCNA)/cyclin. Clin Neuropathol 14: 172–181
50. Katsetos CD, Herman MM, Balin BJ, Vinores SA, Hessler RM, Arking EJ, Karkavelas G, Frankfurter A (1998) Class III β-tubulin isotype (βIII) in the adrenal medulla. III. Differential expression of neuronal and glial antigens identifies two distinct populations of neuronal and glial-like (sustentacular) cells in the PC12 rat pheochromocytoma cell line maintained in a Gelfoam matrix system. Anat Rec 250:351–366
51. Katsetos CD, Karkavelas G, Herman MM, Vinores SA, Provencio J, Spano A, Frankfurter A (1998) Class III β-tubulin isotype in the adrenal medulla. I. Localization in the developing human adrenal medulla. Anat Rec 250:343–349
52. Kenigsberg RL, Hong Y, Yao H, Lemieux N, Michaud J, Tautu C, Theoret Y (1997) Effects of basic fibroblast growth factor on the differentiation, growth, and viability of a new human medulloblastoma cell line (UM-MB1). Am J Pathol 151:867–881
53. Kershman J (1938) The medulloblast and the medulloblastoma: a study of human embryos. Arch Neurol Psychiatr 40: 937–967
54. Kleihues P, Cavenee WK (2000) Pathology and genetics of tumours of the nervous system. IARC Press, Lyon
55. Kleihues P, Louis DN, Scheithauer BW, Rorke LB, Reifenberger G, Burger PC, Cavenee WK (2002) The WHO classification of tumors of the nervous system. J Neuropathol Exp Neurol 61:215–225
56. Klériga E, Sher JH, Nallainathan S-K, Stein SC, Sacher M (1978) Development of cerebellar malignant astrocytoma at site of a medulloblastoma treated 11 years earlier. Case report. J Neurosurg 49:445–449
57. Kissane JM (1994) Blastomas in infancy and childhood. Semin Diagn Pathol 11:83–84
58. Korf HW, Korf B, Schachenmayr W, Chader GJ, Wiggert B (1992) Immunocytochemical demonstration of interphotoreceptor retinoid-binding protein in cerebellar medulloblastoma. Acta Neuropathol 83:482–487
59. Kramm CM, Korf HW, Czerwionka M, Schachenmayr W, Grip WJ de (1991) Photoreceptor differentiation in cerebellar medulloblastoma: evidence for a functional photopigment and authentic S-antigen (arrestin). Acta Neuropathol 81:296–302
60. Krynska B, Otte J, Franks R, Khalili K, Croul S (1999) Human ubiquitous JCV-CY T-antigen gene induces brain tumours in experimental animals. Oncogene 18:39–46
61. Lantos PL, Kleihues P, VandenBerg SV, Lopes MBS (1997) Tumours of the central nervous system. In: Graham DI, Lantos PL (eds) Greenfield's neuropathology. 6th edn. Arnold, London, pp 583–879
62. Luskin MB, Parnavelas JG, Barfield J (1993) Neurons, astrocytes, and oligodendrocytes of the rat cerebral cortex originate from separate progenitor cells: an ultrastructural analysis of closely related cells. J Neurosci 13:1730–1750
63. Maraziotis T, Perentes E, Karamitopoulou E, Nakagawa Y, Gessaga EC, Probst A, Frankfurter A (1992) Neuron-associated class III β-tubulin isotype, retinal S-antigen, synaptophysin and glial fibrillary acidic protein in human medulloblastomas: a clinicopathological analysis of 36 cases. Acta Neuropathol 84:355–363
64. Marburg O (1931) Zur Kenntnis der sog. Medulloblastome (Sphäroblastoma polymorph). Dtsch Z Nervenheilk 117/119: 289–308

65. Matsuda M, Yasui K, Nagashima K, Mori W (1987) Origin of the medulloblastoma experimentally induced by human polyomavirus JC. J Natl Cancer Inst 79:585–591
66. Molenaar WM, Jansson DS, Gould VE, Rorke LB, Franke WW, Lee VM, Packer RJ, Trojanowski JQ (1989) Molecular markers of primitive neuroectodermal tumours and other pediatric central nervous system tumours. Lab Invest 61:635–643
67. Muragaki Y, Chou TT, Kaplan DR, Trojanowski JQ, Lee VM (1997) Nerve growth factor induces apoptosis in human medulloblastoma cell lines that express TrkA receptors. J Neurosci 17:530–542
68. Nagashima K, Yahui K, Kimura J, Washizu M, Yamaguchi K, Mori W (1984) Induction of brain tumours by a newly isolated JC virus (Tokyo-1 strain). Am J Pathol 116:455–463
69. Nishio S, Inamura T, Morioka T, Ishihara S, Hirano K, Murakami N, Fukui M (2000) Cerebellar neuroblastoma in an infant. Clin Neurol Neurosurg 102:52–57
70. Noble M, Mayer-Proschel M (1997) Growth factors, glia and gliomas. J Neurooncol 35:193–209
71. Obersteiner H (1883) Der feinere Bau der Kleinhirnrinde bei Menschen und Tieren. Biologisches Zentralblatt 3:145–155
72. Park TS, Hoffman HJ, Hendrick EB, Humphreys RP, Becker LE (1983) Medulloblastoma: clinical presentation and management. Experience at the Hospital for Sick Children, Toronto, 1950–1980. J Neurosurg 58:543–552
73. Pearl GS, Takei Y (1981) Cerebellar "neuroblastoma": nosology as it relates to medulloblastoma. Cancer 47:772–779
74. Pelc K, Vincent S, Ruchoux MM, Kiss R, Pochet R, Sariban E, Decaestecker C, Heizmann CW (2002) Calbindin-d28k. A marker of recurrence in medulloblastomas. Cancer 95:410–419
75. Penfield W (1931) The classification of gliomas and neuroglial cell types. Arch Neurol Psychiatry 26:745–753
76. Perentes E, Rubinstein LJ, Herman MM, Donoso LA (1986) S-antigen immunoreactivity in human pineal glands and pineal parenchymal tumors. A monoclonal antibody study. Acta Neuropathol (Berl) 71:224–227
77. Pietsch T, Waha A, Koch A, Kraus J, Albrecht S, Tonn J, Sorensen N, Berthold F, Henk B, Schmandt N, Wolf HK, Deimling A von, Wainwright B, Chenevix-Trench G, Wiestler OD, Wicking C (1997) Medulloblastomas of the desmoplastic variant carry mutations of the human homologue of *Drosophila* patched. Cancer Res 57:2085–2088
78. Polak M (1966) Blastomas del sistema nervioso central y periferico. Patologia y ordenacion histogenética. Lopez Libreros Editores, Buenos Aires
79. Polak M (1967) On the true nature of the so-called medulloblastoma. Acta Neuropathol (Berl) 8:84–95
80. Raaf J, Kernohan JW (1944) Relation of abnormal collections of cells in posterior medullary velum of the cerebellum to the origin of medulloblastoma. Arch Neurol Psychiatry 52:163–169
81. Ramon y Cajal S (1911) Histologie du système nervaux de l' homme et des vertébrés. Institute Ramon y Cajal, Madrid, II, pp 80–106
82. Rio Hortega P del (1932) Estructura y systematisacion de los gliomas y paragliomas. Arch Esp Oncol 2:411–678
83. Rio Hortega P del (1945) Nomenclatura y clasificacion de los tumores del sistema nervioso. Arch Histol (B. Aires) 3:5–63
84. Rorke LB (1983) The cerebellar medulloblastoma and its relationship to primitive neuroectodermal tumors. J Neuropathol Exp Neurol 42:1–15
85. Rorke LB (1994) Experimental production of primitive neuroectodermal tumors and its relevance to human neuro-oncology. Am J Pathol 144:444–448
86. Rubinstein LJ (1991) Glioma cytogeny and differentiation viewed through the window of neoplastic vulnerability. In: Salcman M (ed) Neurobiology of brain tumors, vol 4: Concepts in neurosurgery. Williams & Wilkins, Baltimore, pp 35–52
87. Rubinstein LJ, Herman MM (1975) Studies on the differentiation of human and experimental gliomas in organ culture systems. Recent Results Cancer Res 51:35–51
88. Rubinstein LJ, Northfield DWG (1964) The medulloblastoma and the so-called cerebellar arachnoidal sarcoma: a reappraisal of an old nosological problem. Brain 87:379–410
89. Rubinstein LJ, Herman MM, Hanbery JW (1974) The relationship between differentiating medulloblastoma and dedifferentiating cerebellar astrocytoma. Light, electron microscopic, trissue and organ culture observations. Cancer 33: 675–690
90. Russell DS, Rubinstein LJ (1959) Pathology of tumours of the nervous system, 1st edn. Edward Arnold, London, pp 155–159
91. Russell DS, Rubinstein LJ (1989) Pathology of tumours of the nervous system, 5th edn. Edward Arnold, London, pp 251–274
92. Schaper A (1897) Die frühesten Differenzierunggänge im ZNS. Arch Entwickl Mech Org 5:81–130
93. Scheinker IM (1939) Zur Frage der Pathogenese und Pathologie der Medulloblastome. Monatsschr Psychiatr Neurol 101: 103–121
94. Schiffer D, Giordana MT, Mauro A, Soffietti R (1993) Brain tumors. Pathology and its biological correlates. Springer, Berlin Heidelberg New York, pp 241–260
95. Schiffer D, Cavalla P, Chio A, Giordana MT, Marino S, Mauro A, Migheli A (1994) Tumor cell proliferation and apoptosis in medulloblastoma. Acta Neuropathol 87:362–370
96. Schiffer D, Bortolotto S, Bosone L, Cancelli I, Cavalla P, Schiffer P, Piva P (1999) Cell cycle inhibitor p27/Kip-1 expression in non-astrocytic and non-oligodendrocytic human nervous system tumours. Neurosci Lett 264:29–32
97. Schmidbauer M, Budka H, Bruckner R, Vorkapic P (1987) Glioblastoma developing at the site of a cerebellar medulloblastoma treated 6 years earlier. J Neurosurg 67:915–918
98. Schubert TE, Cervos-Navarro J (1998) The histopathologic and clinical relevance of apoptotic cell death in medulloblastomas. J Neuropathol Exp Neurol 57:10–15
99. Stevenson L, Echlin R (1934) The nature and origin of some tumors of the cerebellum (medulloblastoma). Arch Neurol Psychiatry 31:93–109
100. Svensson T, Ryden M, Schilling FH, Dominici C, Sehgal R, Ibanez CF, Kogner P (1997) Coexpression of mRNA for the full-length neurotrophin receptor trk-C and trk-A in favourable neuroblastoma. Eur J Cancer 33:2058–2063
101. Vinores SA, Herman MM, Katsetos CD, May EE, Frankfurter A (1994) Neuron-associated class III β-tubulin, tau and MAP2 in the D-283 Med cell line and in primary explants of human medulloblastoma. Histochem J 26:678–685
102. Wang YZ, Christakos S (1995) Retinoic acid regulates the expression of the calcium binding protein, calbindin-D28k. Mol Endocrinol 9:1510–1521
103. Wernig M, Brustle O (2002) Fifty ways to make a neuron: shifts in stem cell hierarchy and their implications for neuropathology and CNS repair. J Neuropathol Exp Neurol 61: 101–110
104. Yavuz AA, Yaris N, Yavuz MN, Sari A, Reis AK, Aydin F (2002) Primary intraspinal primitive neuroectodermal tumor: case report of a tumor arising from the sacral spinal nerve root and review of the literature. Am J Clin Oncol 25:135–139
105. Zülch K-J (1986) Brain tumors. Their biology and pathology, 3rd edn. Springer, Berlin Heidelberg New York, pp 324–337
106. Zu Rhein GM, Varakis JN (1979) Perinatal induction of medulloblastomas in Syrian golden hamsters by JC virus, a human papova virus. Nat Cancer Inst Monogr 51:204–208