

WHO CNS5 2021 classification of gliomas: a practical review and road signs for diagnosing pathologists and proper patho-clinical and neuro-oncological cooperation

Aleksandra Sejda¹, Wiesława Grajkowska², Joanna Trubicka², Ewa Szutowicz³, Tomasz Kazimierz Wojdacz⁴, Wojciech Kloc^{5,6}, Ewa Iżycka-Świeszewska^{7,8}

¹Department of Pathomorphology and Forensic Medicine, Collegium Medicum, University of Warmia and Mazury, Olsztyn, Poland, ²Department of Pathology, The Children's Memorial Health Institute, Warsaw, Poland, ³Department of Oncology and Radiotherapy, Medical University of Gdansk, Gdansk, Poland, ⁴Independent Clinical Epigenetics Laboratory, Pomeranian Medical University in Szczecin, Poland, ⁵Department of Neurosurgery, Copernicus Hospital in Gdansk, Poland, ⁶Department of Psychology, Health Sociology and Public Health School, University of Warmia and Mazury, Olsztyn, ⁷Department of Pathology and Neuropathology, Medical University of Gdansk, Gdansk, Poland, ⁸Department of Pathomorphology, Copernicus Hospital in Gdansk, Poland

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Abstract

The 5th edition of World Health Organization (WHO) Central Nervous System (CNS) tumours classification has transformed the pathological diagnosis of gliomas from purely histological to the multilayered integrated one with molecular biomarkers necessary for proper classification, risk stratification, and prognostic-predictive clinical purposes. Because of deep and important changes in taxonomy and diagnostic approach to gliomas, this manuscript is a review of WHO CNS classification 5th edition with general testing guidance for pathologists and clinicians working in neuro-oncology.

Key words: gliomas, classification, WHO, 5th edition, neuropathology.

Introduction with historical background

Glial tumours constitute a substantial and complex heterogeneous group of primary central nervous system (CNS) neoplasms. Their consecutive classifications are strongly related to histological features and clinical outcome, together with relatively recent incorporation of the molecular characteristics. The 5th edition of the World Health Organization (WHO) classification of tumours of the CNS, published at the end of 2021 has introduced new taxonomy and nomenclature of a great number of tumours including gliomas.

The story begins in 1926 when Bailey and Cushing published the book "A Classification of the Tumours of

the Glioma Group on a Histogenetic Basis with a Correlated Study of Prognosis" [4,23]. The 1st WHO classification of CNS tumours was published in 1979 [87]. The intracranial tumours were graded according to four-stage scale: grade I – the most benign neoplasms, grade II – semi-benign, grade III – relatively malignant, and grade IV – highly malignant. The widest group constituted the neuroepithelial tumours, but interestingly, glioblastoma was not included into the astrocytic neoplasm [68,87]. In the 2nd version of WHO CNS tumours classification (1993), the major progress was made by incorporation of immunohistochemistry. Glioblastoma was then put into the astrocytomas group, and the idea of glioma progression was posed [34]. In 2000,

Communicating author:

Ewa Iżycka-Świeszewska, Department of Pathology and Neuropathology, Medical University of Gdansk,
e-mail: ewa.izycka-swieszewska@gumed.edu.pl

the 3rd WHO edition has been improved by an extensive patho-clinical and radiological descriptions together with some genetic findings [18]. So-called primary glioblastomas were characterized by *EGFR* alteration and loss of chromosome 10, and 1p/19q loss in oligodendroglioma was defined [69]. In 2007, WHO 4th edition provided modifications concerning grading criteria, new entities and increasing role of molecular data [42]. Grading was presented as follows: I for low proliferation potential with radical treatment with surgery alone, II for

infiltrating lesions, prone for recurrent course, III as histological malignant neoplasms, with the need of adjuvant therapy, finally IV for highly malignant tumours with poor prognosis. Molecular alterations provided prognostic and predictive data within diagnostic categories established by conventional histological methods.

In 2016, the revised/updated 4th WHO classification of CNS tumours introduced the idea of multi-layered approach composed of integrated diagnosis, histological classification, WHO grade and molecular data [43]. The classification of diffuse gliomas was changed, a large subset was defined based on *IDH* 1/2 mutation and 1p/19q status. Thus, the oligoastrocytoma was expected to disappear, and gliomatosis became a growth pattern [45]. Diffuse gliomas included 15 entities, because different malignancy grades were assigned to different entities (e.g. anaplastic oligodendroglioma as a different type from oligodendroglioma) and because not otherwise specified (NOS) designations were assigned to distinct entities [46]. *IDH*-mutant and *IDH*-wildtype diffuse astrocytomas were divided into three types (diffuse astrocytoma, anaplastic astrocytoma and glioblastoma) depending on histology. However, due to rapidly increasing knowledge from high output molecular neuro-oncological and clinical studies, the need for the new improved system occurred. Consequently, in years 2018-2020, the international expert group Consortium to Inform Molecular and Practical Approaches to CNS Tumour Taxonomy (CIMPACT) was elaborating on new recommendations [17]. The group published seven interim updates which constitute the background of the last 5th WHO CNS classification [13,14,21,22,44,48,49].

The current 2021 WHO CNS 5th edition (WHO CNS5) reflects the use of complex histological and molecular approaches to establish a final pathological diagnosis and grading for brain tumours [11,84]. The idea is to meet the need for standards in diagnosis and to facilitate the translation of diagnostic research into practice [45]. However, the access to molecular testing is limited for many centres as well as the ability to incorporate all changes. Fortunately, there are many immunohistochemical surrogates for molecular alterations which might be successfully used to improve the diagnosis.

Currently, “glial, glioneuronal and neuronal tumours” are grouped together into a separate family and divided into six categories: 1) adult-type diffuse gliomas, 2) paediatric-type diffuse low-grade gliomas, 3) paediatric-type diffuse high-grade gliomas, 4) circumscribed astrocytic gliomas, 5) glioneuronal and neuronal tumours, and

Table I. Gliomas according to WHO 2021 classification of CNS tumours

Adult-type diffuse gliomas
Astrocytoma, <i>IDH</i> -mutant
Oligodendroglioma, <i>IDH</i> -mutant and 1p/19q-codeleted
Glioblastoma, <i>IDH</i> -wildtype
Paediatric-type diffuse low-grade gliomas
Diffuse astrocytoma, <i>MYB</i> - or <i>MYBL1</i> -altered
Angiocentric glioma
Polymorphous low-grade neuroepithelial tumour of the young
Diffuse low-grade glioma, <i>MAPK</i> pathway-altered
Paediatric-type diffuse high-grade gliomas
Diffuse midline glioma, <i>H3</i> K27-altered
Diffuse hemispheric glioma, <i>H3</i> G34-mutant
Diffuse paediatric-type high-grade glioma, <i>H3</i> -wildtype and <i>IDH</i> -wildtype
Infant-type hemispheric glioma
Circumscribed astrocytic gliomas
Pilocytic astrocytoma
High-grade astrocytoma with piloid features
Pleomorphic xanthoastrocytoma
Subependymal giant cell astrocytoma
Chordoid glioma
Astroblastoma, <i>MN1</i> -altered
Ependymal tumours
Supratentorial ependymoma
Supratentorial ependymoma, <i>ZFTA</i> fusion-positive
Supratentorial ependymoma, <i>YAP1</i> fusion-positive
Posterior fossa ependymoma
Posterior fossa group A (PFA) ependymoma
Posterior fossa group B (PFB) ependymoma
Spinal ependymoma
Spinal ependymoma, <i>MYCN</i> -amplified
Myxopapillary ependymoma
Subependymoma

6) ependymal tumours. The 2021 glial tumours WHO classification is summarized in Table I.

General changes of 5th edition CNS WHO concerning gliomas, placed into the group of “Gliomas, glioneuronal and neuronal tumours”

The major changes in the classification and grading of the diffuse gliomas comprise separation of the paediatric-type diffuse gliomas from the adult-type diffuse gliomas, being clinically and biologically distinct groups; simplification of the adult-type diffuse gliomas into three major types; and changes in the nomenclature and grading.

Several new entities have been added such as: diffuse astrocytoma *MYB*- or *MYBL1*-altered; polymorphous low-grade neuroepithelial tumour of the young; diffuse low-grade glioma, MAPK pathway-altered; diffuse hemispheric glioma, H3 G34-mutant; diffuse paediatric-type high-grade glioma, H3-wild-type and IDH-wildtype; infant-type hemispheric glioma; high-grade astrocytoma with piloid features.

The ependymomas have been reclassified by anatomical site and by the addition of genetic or epigenetic types. For some entities (diffuse paediatric-type high-grade glioma, H3-wildtype and IDH-wildtype, and for diffuse low-grade glioma, MAPK pathway-altered), histological appearance and defined molecular features must be combined within the final integrated diagnosis.

Grading is performed within (rather than across) tumour types, but linking WHO grade with clinical behaviour and prognosis is a base of criteria for most tumours. Some molecular markers connected with poorer prognosis are recommended to be added to histological diagnosis in assigning the grade. The example is homozygous deletion of *CDKN2A/B* as an indicator of grade 4 in IDH-mutant astrocytoma. Grading description has been changed from traditionally used Roman (I, II, III, IV) into Arabic numbers (grade (G) 1, 2, 3, 4) to avoid editorial errors [13,71].

The next change is the implementation of two distinct suffixes for not fully classified tumours. The designation Not Otherwise Specified (NOS) means that diagnostic testing necessary to assign a specific WHO diagnosis cannot or will not be performed. Not Elsewhere Classified (NEC) qualifier points out that all necessary used assays still do not match any entities from the classification [49]. Moreover, methylome analysis became a powerful tool in the

classification of brain tumours. Many new entities together with WHO CNS grade stratification can be identified based on the methylation profile. However, it is mostly related or obligatory for the diagnosis in rare lesions or when limited material is available [15,71].

Other minor changes include conversion of mitotic count from a denominator of 10 HPF to a defined area expressed in mm² to standardize the true area of mitotic counting, standardization of current genomic nomenclature by using Human Genome Variation Society (HGVS) notation, and the term “variant” for a specific kind of tumours has been exchanged by “subtype” [37,47].

Adult-type diffuse gliomas

The category of diffuse gliomas is simplified and distinguishes three tumour types: astrocytoma, IDH-mutant; oligodendroglioma, IDH-mutant, 1p/19q-codeleted; and glioblastoma, IDH-wildtype.

According to CNS WHO 2021 definition, “Astrocytoma, IDH-mutant, is a diffusely infiltrating *IDH1*- or *IDH2*-mutant glioma with frequent *ATRX* and/or *TP53* mutation and absence of 1p/19q codeletion (CNS WHO grade 2, 3, or 4)” [13].

IDH-mutant astrocytomas range from well-differentiated, low cellular, slow-growing tumours (CNS WHO grade 2) to highly anaplastic, hypercellular and rapidly progressive one (grade 4) [71]. The average age of incidence for grade 2 and 3 IDH-mutant astrocytomas is 38 years, since grade 4 ones are typical for slightly older patients. They rarely occur in teenagers above 14 years and are uncommon in patients aged more than 55 years. There is evidence that grade 4 tumours arise rather *de novo* than in the train of progression from lower grade gliomas and frequently harbour *MGMT* promoter methylation [11]. First reported in 2008, *IDH1/2* mutations are a defining feature of this group of gliomas. The most frequent canonical (about 90% of cases) *IDH1* p.R132H pathogenic variant with arginine to histidine transition might be evaluated by immunohistochemical surrogate [73,83]. Genetic assessment of pathogenic variants in 132 and 172 codons in *IDH1* and *IDH2* genes, respectively, is recommended in the event of a negative or indeterminate result with the IDH1 p.R132H immunohistochemical stain. However, in patients older than 55 years non-canonical *IDH1* mutations are very rare, so in this group it is

not necessary to perform gene sequencing. Inactivating *ATRX* mutations (70-80% of cases) commonly co-occur with *TP53* alterations in IDH-mutant astrocytomas [26,57,81]. These often result in a truncated protein and lead to loss of nuclear *ATRX* staining. Moreover, p53 strong immunopositivity in more than 10% of tumour cell nuclei and loss of *ATRX* expression are both sufficient to diagnose IDH-mutant astrocytoma without the necessity of 1p/19q testing. Thus, fluorescence *in situ* hybridization (FISH) should be done only in cases morphologically suspicious for oligodendroglioma with retention of *ATRX* expression and detected *IDH1/2* mutation [12,44,82].

In the subgroup of so-called lower grade gliomas G2 and G3, the principal distinguishing feature is increased mitotic activity and histological anaplasia. Grade 3 tumours also often display increased cell density and greater nuclear atypia, even multinucleated tumour cells and abnormal mitoses may be seen. One mitotic figure may be sufficient for grade 3 within a very small biopsy, while more mitoses are required in a larger resection specimen. The threshold mitotic count for a CNS WHO grade 3 designation has not been established yet. By definition, microvascular proliferation and necrosis are absent. The growth fraction as determined by the Ki-67 proliferation index is usually < 4% for grade 2 IDH-mutant astrocytomas. In grade 3 tumours, the Ki-67 proliferation index is usually in the range of 4-10%, but it can overlap with values for grade 2 tumours at one end of the range and grade 4 tumours at the other side [11,13,16].

CNS WHO grade 4 tumours histologically manifest necrosis and/or microvascular proliferation in addition to the features of grade 3 lesions. Additionally, the designation of CNS WHO grade 4 IDH-mutant astrocytoma is warranted for the lower grade astrocytoma with homozygous deletion of *CDKN2A* and/or *CDKN2B*, even in the absence of G4 morphology [13,71,82]. The p16 antibody up to date was not proved to be a sufficient immunohistochemical alternative for *CDKN2A* deletion detection [62,71]. Glioblastoma IDH-wildtype has considerable histological overlap with that of CNS WHO grade 4 IDH-mutant astrocytoma, and testing for *IDH1/2* mutations is required to distinguish those tumour types. Nevertheless, some features can differentiate these two entities. Areas of ischemic zonal and/or palisading necrosis have been observed in 50% of

grade 4 IDH-mutant astrocytomas, considerably less frequently than in IDH-wildtype glioblastomas, where they are found in as many as 90% of cases. Additionally, focal oligodendroglioma-like components are more common in CNS WHO grade 4 IDH-mutant astrocytoma than in IDH-wildtype glioblastoma [11,36].

By CNS WHO 2021 definition, “Oligodendroglioma, IDH-mutant and 1p/19q codeleted, is a diffusely infiltrating glioma with *IDH1*- or *IDH2*-mutation and codeletion of chromosome arms 1p and 19q”. Two grades (WHO CNS G2 or 3) are distinguished according to histological features and to a minor degree with molecular markers. Anaplastic oligodendroglioma as a separate entity is no longer listed. The recommended criteria for grade 3 include mitotic activity ≥ 2.5 mitoses/mm², presence of microvascular proliferation and necrosis [11,24]. Other features like degree of cellularity or nuclear atypia might be also considered. An additional molecular marker is *CDKN2A/2B* homozygous deletion which was found in a subset of less than 10% oligodendrogliomas grade 3 [1,12].

The mean age of patients with oligodendroglioma grade 2 is 41-43 years, and 47-50 for grade 3 and both occur rarely in teenagers and children [55]. “Fried-egg” rounded, monotonous cells with well-defined cell membranes and perinuclear clearing, chicken wire vasculature, microcalcifications are compatible with the diagnosis of oligodendroglioma. Since WHO 2016, *IDH1/2* mutation and 1p/19q codeletion are essential for the diagnosis of oligodendroglioma, the latter examined by molecular tests usually by FISH analysis [86]. *IDH1 p.R132H* mutation appears in more than 90% of all oligodendrogliomas and can be detected by immunohistochemical equivalent (Fig. 1). The remaining ones are non-canonical mutations with *IDH2* mutation proportionally prevailing over astrocytomas [2]. Additionally, lack of p53 and retained nuclear immunohistochemical expression of *ATRX* are helpful for the proper diagnosis. Pathogenic variants in *TP53* gene and 1p/19q codeletion are nearly, but not entirely mutually exclusive in IDH-mutant gliomas. There is an inverse relationship between *ATRX* loss and presence of 1p/19q codeletion, because oligodendrogliomas, differently from astrocytomas IDH-mutant, acquire alternative lengthening of telomeres *via TERT* promoter mutation which concerns about 98% of cases [3,12]. Other genetic alterations observed in around 70% and unique for oligodendrogliomas

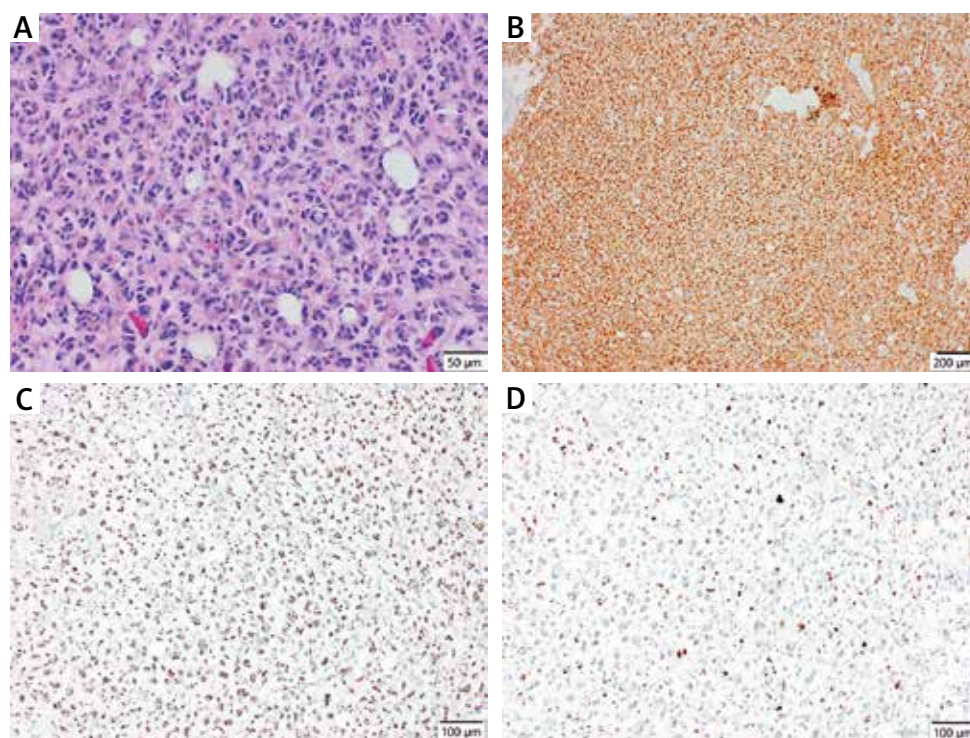


Fig. 1. Oligodendroglioma, IDH-mutant, 1p/19q-codeleted (G3 WHO CNS5). A frontal lobe glial tumour with necrosis and microvascular proliferation in a 43-year-old patient, diagnosed in 2015 as glioblastoma WHO grade IV, treated with radiotherapy after partial resection. Relapse after 5 years shows diffusely infiltrating glial tumour composed of moderately to highly anaplastic cells grouping in small clusters (**A**, 200×). IDH R132H diffusely positive (**B**, 400×), ATRX nuclear staining preserved (**C**, 100×), p53 wild type – negative in most cells (**D**, 100×). The fluorescence *in situ* hybridization showed 1p/19p codeletion, so the diagnosis was revised.

are mutations of the *CIC* gene [10]. Some previous studies found the reliability of immunohistochemical surrogates without need of 1p/ 19q status testing, but at present, molecular analysis is necessary [75].

Finally, rare tumours containing distinct regions with oligodendroglioma morphology and 1p/19q codeletion, while other regions with astrocytic morphology, ATRX loss and *TP53* pathogenic variants are reported. Also few dual-genotype IDH-mutant gliomas with uniform tumour morphology, as well as ATRX loss, *TP53* mutations, and 1p/19q codeletion throughout all tumour regions have been described. For such rare cases of IDH-mutant glioma with dual-genotype, the description NEC might be appropriate [11,49].

WHO CNS5 has changed again the definition and understanding of glioblastoma which is the most common high grade glioma in the adults. By defini-

tion: “Glioblastoma, IDH-wildtype, is a diffuse astrocytic glioma that is IDH-wildtype and H3-wildtype and has one or more of the following histological or genetic features: microvascular proliferations, necrosis, *TERT* promoter mutation, *EGFR* amplification, +7/–10 chromosome copy-number changes (CNS WHO grade 4)” [11]. The above-mentioned molecular alterations are considered to be informative and supportive of glioblastoma, IDH-wildtype diagnosis even for diffuse astrocytoma cases with not fulfilling histological criteria for grade 4 [14,54,76]. The morphological hallmarks of glioblastoma sufficient for diagnosis are invariably microvascular proliferations and necrosis either palisading or not. Other typical features are elevated mitotic index, Ki-67 range from 5% to even 50%, nuclear atypia, marked pleomorphism or high cellularity. Three morphological subtypes are now distinguished: giant cell glioblastoma,

epithelioid glioblastoma and gliosarcoma with a wide spectrum of phenotypes. Moreover, there are numerous histological patterns such as: PXA-like, small cell, glioblastoma with primitive neural component, metaplastic, lipidized, with prominent gemistocytic features, granular cell, brain abscess-like or infarction-like [6,11]. Glioblastoma has also many faces in neuroimaging [66].

IDH1/2 and histone H3 mutations are not present in glioblastoma and lack of *IDH1* p.R132H positivity is sufficient for the diagnosis in patients aged more than 55 years with typical histological features and

without any history of low-grade gliomas. Positivity for ATRX and negativity for p53 helps with glioblastoma diagnosis (Fig. 2) [72,82]. Molecular aberrations found in glioblastoma are diverse and include mainly point mutations, chromosomal structural changes, gene fusions as well as epigenetic changes like histone modifications or promoter methylations. The most typical fusions contain *EGFR*, *FGFR3*, *MET* or *NTRK1/2/3* genes. p14ARF-MDM2-MDM4-p53, PI3K-AKT-mTOR and CDK4/6-CDKN2A/B-RB1 pathways are highly deregulated [12,79,82]. Some experts propose that histologically lower grade IDH-wildtype

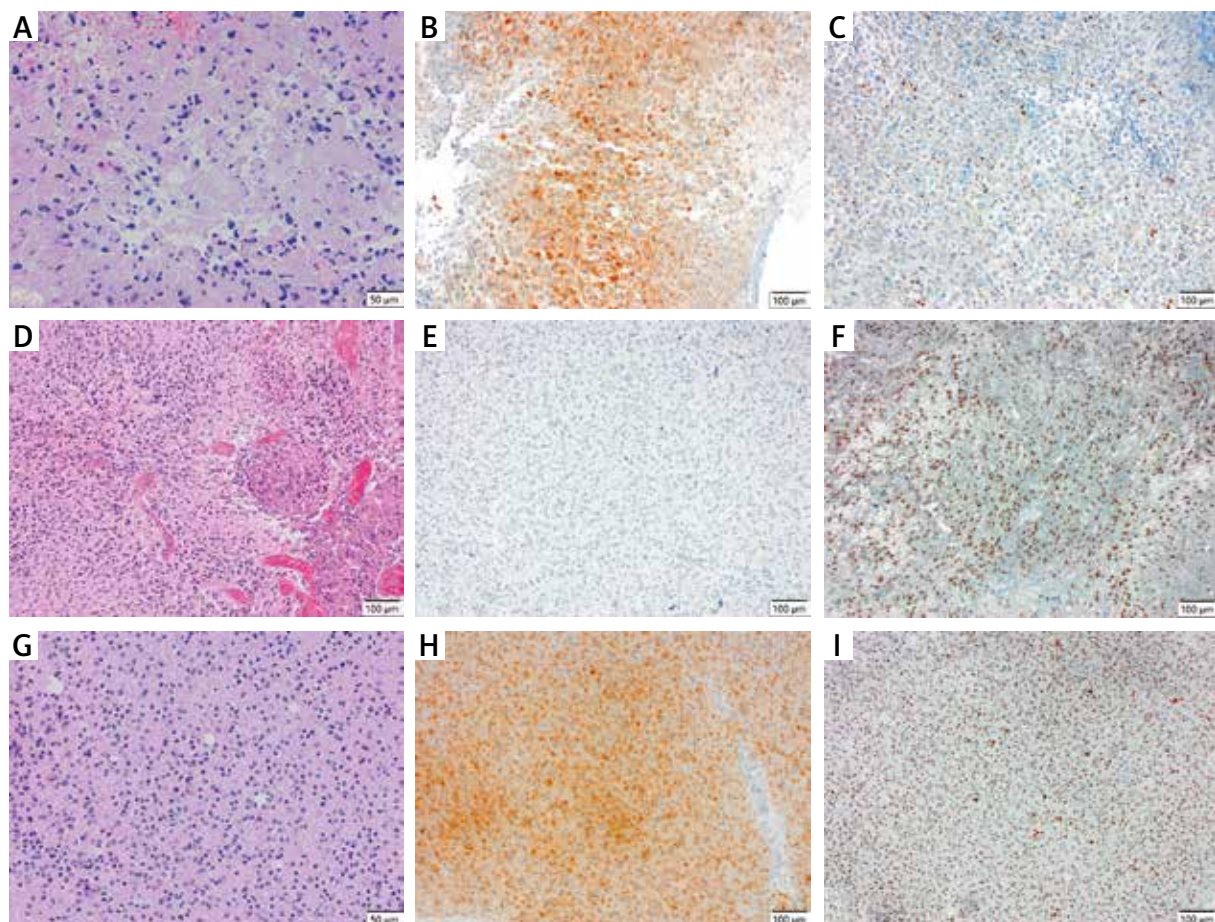


Fig. 2. Glial tumours with basic immunohistochemical molecular surrogates profile. Astrocytoma IDH-mutant, WHO CNS grade 3. Astrocytoma with gemistocytic differentiation composed of plump, eosinophilic cells with anaplastic nuclei and mitoses assigns as grade 3 (A, 200×), positive stain for IDH1 R132H (B, 100×), loss of ATRX nuclear staining with positive control in lymphocytes (C, 100×). Glioblastoma, IDH wild type, WHO CNS grade 4. Highly cellular glial tumour with necrosis and microvascular proliferation (D, 100×), IDH1 R132H negative stain (E, 100×), retained ATRX positivity (F, 100×). Oligodendroglioma, IDH-mutant, 1p/19q-codeleted, WHO CNS grade 2. Glial tumour composed of cells with uniformly round nuclei with well-defined cell membranes and clear cytoplasm (G, 200×), positive reactivity for IDH1 R132H (H, 100×), ATRX nuclear positivity (I, 100×).

astrocytomas with molecular features of glioblastoma, such as *TERT* promoter mutation, *EGFR* amplification, +7/–10 chromosome copy-number changes, might be rather G3 due to its better prognosis and lower molecular burden [9]. Finally, there is also a small subgroup of “true” low-grade IDH-wildtype astrocytomas without glioblastoma signature and with better prognosis [64].

Paediatric-type gliomas

The new classification of paediatric gliomas has been based on molecular findings and methylation signatures in these tumours. Two main groups are now distinguished: low grade and high grade tumours, but specific grades are not characterized in all entities [11,35]. The WHO grading system of adult gliomas is not enough for paediatric tumours. Each paediatric tumour type has distinctive clinical, histologic, and molecular features that are used to establish their diagnosis. In some tumour types the methylation profile is essential to establish the proper diagnosis. Multiple studies showed that some molecular aberrations are connected with the outcome and might be used to optimize the treatment and administer specific target therapies [61,67].

Paediatric-type diffuse low-grade gliomas are uncommon. They present mainly in children and sometimes in young adults and are often associated with epilepsy. Microscopically, their architecture and cytologic features are similar to other WHO grade 2 gliomas. In these tumours mitotic activity is absent or low, and necrosis or microvascular proliferation is absent. They display astrocytic, oligodendroglial or a combination of astro- and oligodendroglial differentiation. Molecularly, they present IDH-wildtype (*IDH1*, *IDH2*) and H3-wildtype (*H3-3A*, *HIST1H3B*, *HIST1H3BC*) genotype. This group includes: 1) diffuse low-grade glioma, MAPK-altered, 2) diffuse astrocytoma, *MYB*- or *MYBL1*-altered, 3) polymorphous low-grade neuroepithelial tumour of the young, and 4) angiocentric glioma.

Diffuse low-grade glioma, MAPK pathway-altered, is a low grade glioma with diffuse astrocytic- or oligodendroglial morphology, generally presenting in childhood. This tumour is characterized by alterations in genes regulated MAPK signal pathway. Typically, they have an internal tandem duplication or other alterations in the tyrosine kinase domain (TKD) of *FGFR1* as well as *BRAF p.V600E* pathogenic

variant [67,83]. Diffuse low grade (WHO G1) astrocytoma *MYB/MYBL1*-altered is a diffusely infiltrative astroglial neoplasm composed of monomorphic cells with *MYB* or *MYBL1* fusion gene. Immunohistochemical testing for the expression of MAP2 antibody or FISH analysis may help with the diagnosis [12,22]. Polymorphous low-grade neuroepithelial tumour of the young is a glial neoplasm associated with a history of epilepsy, diffuse growth patterns, calcification, presence of oligodendroglioma-like components, CD34 positivity, and MAPK pathway-activating genetic abnormalities. Almost 80% of cases are located in temporal lobes. Median age of diagnosis is about 16, but the tumour may be found also in older individuals [32,48]. The angiocentric glioma is composed of bipolar, elongated, bland cells with perivascular growth pattern, combining features of infiltrating glioma, schwannoma- and ependymoma-like areas (WHO G1). They are characterized by presence of *MYB* alterations (mainly *MYB-QKI* fusion) and co-expression of EMA and GFAP [11,27].

The paediatric-type diffuse high-grade gliomas group contains entities with a generally aggressive presentation: 1) diffuse midline glioma, H3 K27-altered, 2) diffuse hemispheric glioma, H3 G34-mutant, 3) diffuse paediatric-type high-grade glioma, H3-wildtype and IDH-wildtype; and 4) infant-type hemispheric glioma. Mutations on histone H3 were found to correlate with aggressive behaviour, p.K28M (p.K27M) and p.G35R/V (p.G34R/V) mutations are the most common. Within each of these broad clinical classes we can identify specific genetic or DNA methylation profiles that seem to correlate with clinical outcome [19,25].

Diffuse midline glioma, H3 K27-altered, is an infiltrative midline glioma with loss of H3K27 me3 and either a histone 3 c.83A>T p.Lys28Met (K27M) substitution in one of the genes coding histone H3 isoforms, aberrant overexpression of *EZH1*, or an *EGFR* mutation (WHO CNS G4) [12,44]. These tumours are located in the brain stem, thalamus, and spinal cord. Tumour cells diffusely infiltrate adjacent and distant brain structures. The histologic pattern may be different from moderately to highly anaplastic [11]. Immunohistochemical surrogates might help with the diagnosis. Loss of K27me3 expression and positive nuclear staining for H3 p.K28M (K27)-mutant protein corresponds well to this type of glioma (Fig. 3) [12,48]. Diffuse hemispheric glioma, H3 G34-mutant is a malignant, infiltrative glioma, typically of the

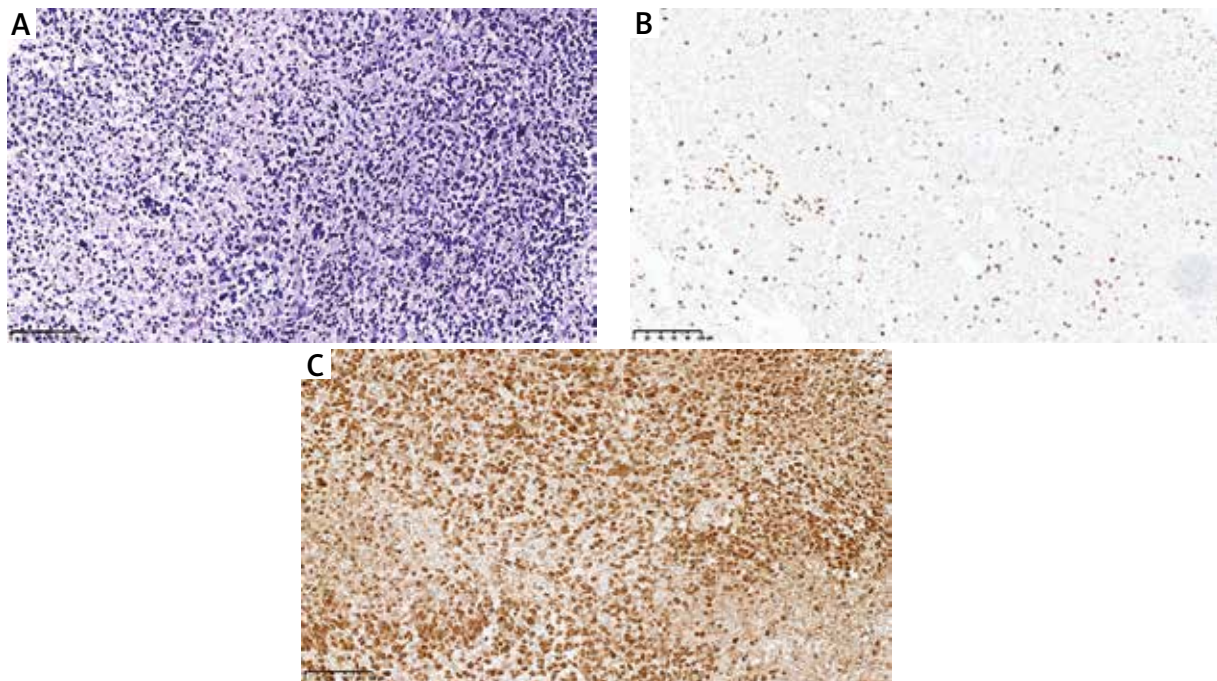


Fig. 3. Diffuse midline glioma H3 K27-altered pontine glioma. Diffusely infiltrative tumour showing monomorphic appearance with sparse pleomorphic neoplastic cells and a focus of necrosis (**A**, 100×). The loss of H3K27me3 immunostaining in tumour cells (**B**, 200×). Intense nuclear staining for H3K27-mutant protein in neoplastic cells (**C**, 200×).

cerebral hemispheres and with a missense mutation in the *H3-3A* gene that results in a p.G35R/V (G34) substitution of histone H3. This tumour is recognized mainly in children and young adults usually with glioblastoma-like or PNET-like histomorphology [60]. The H3 G34-mutant diffuse paediatric-type high-grade glioma presents with loss of ATRX immunorepression, MAP2, FOXG1, p53 positivity and Olig2 negativity [41]. Mutation specific antibody against H3.3 p.G34R/V-mutant proteins is available, but not fully specific [29,47]. Infant-type hemispheric gliomas are uncommon, cerebral, hemispheric, high grade cellular astrocytomas presenting in early childhood. They present typically with receptor tyrosine kinase (RTK) fusions, including *NTRK* family, *ROS1*, *ALK* or *MET* [12]. Diffuse paediatric-type high-grade glioma, H3-wildtype and IDH-wildtype is a diffuse glioma with histological features of malignancy, typically occurring in children, adolescents and young with IDH-wildtype (*IDH1*, *IDH2*) and H3-wildtype (*H3-3A*, *HIST1H3B*, *HIST1H3BC*) status (WHO G4). Molecular analysis distinguished three molecular subtypes of these tumours: ped RTK1, ped RTK2, ped MYCN witch might be diagnosed by methylome analysis [11,38].

Circumscribed astrocytic gliomas

This group of well-demarcated typically solid astrocytic tumours contains six entities: 1) pilocytic astrocytoma, 2) high-grade astrocytoma with piloid features, 3) pleomorphic xanthoastrocytoma, 4) subependymal giant cell astrocytoma, 5) chordoid glioma, and 6) astroblastoma MN1-altered.

Classic pilocytic astrocytomas have well defined variegate morphological forms and two subtypes are now distinguished: pilomyxoid astrocytoma and pilocytic astrocytoma with histological features of anaplasia [43]. The majority of those cases present *KIAA1549:BRAF* fusion, although pathogenic variants in *BRAF* (p.V600E), *NF1*, *KRAS*, *FGFR1*, *PTPN11*, *RAF1* as well as *NTRK* family genes fusion are also reported. All these alterations can be used as a therapeutic target in patients with inoperable tumours. Pilomyxoid astrocytomas occur mainly in infants and young children usually in hypothalamic/chiasmatic region and display more aggressive behaviour with a tendency to cerebrospinal dissemination. Histopathologically they are formed by highly isomorphic, small bipolar piloid cells with formation

of perivascular pseudorosettes in a myxoid background. Molecular studies identify MAPK pathway gene alterations similar to those in classic pilocytic astrocytoma [39,77]. Pilocytic astrocytomas with histological features of anaplasia are tumours with brisk mitotic count with or without necrosis typically located in posterior fossa in the adults [65].

High-grade astrocytoma with piloid features is a newly recognized entity defined by a specific DNA methylation profile. These tumours display histological features of anaplasia alongside with a piloid cytology, being moderately-cellular with microvascular proliferation, increased mitotic count, necrosis, but also occasional Rosenthal fibres. They present frequent MAPK pathway alterations, homozygous deletion of *CDKN2A/B*, loss of *ATRX* expression, as well as a distinct DNA methylation pattern with MGMT promoter methylation [7,63].

Pleomorphic xanthoastrocytoma (PXA) is an astrocytic tumour that primarily affects children and young adults. It involves mainly the temporal lobes, superficially with often leptomeningeal involvement. PXA is composed of pleomorphic mono or multinucleated cells with frequent nuclear inclusion and occasional cytoplasmic xanthomatous changes, usually with scattered eosinophilic granular bodies [11]. PXA can be CNS WHO G2 or 3 with a cut-off level assigned as 2.5 mitoses/mm². In grade 3 tumours, Ki67 index is approximately 15% and these tumours contain necrosis. The most frequent molecular alteration is *BRAF V600E* mutation which may be detected by immunohistochemistry. Different MAPK pathway gene mutation and homozygous deletion of *CDKN2A/2B* might be also found in those tumours [20,59,70].

Astroblastoma MN1-altered is characterized by structural rearrangement of the *MN1* gene or its fusions and a distinct DNA methylation pattern [51]. Histologic criteria for its grading and CNS WHO grade is not established. Chordoid gliomas are rare grade 2 tumours with the *p.D463H PRKCA* gene variant and consistent positivity for TTF1. Finally, no particular changes have been proposed to the classification of subependymal giant cell astrocytoma (WHO G1) [11].

Ependymal tumours

In WHO CNS5, the ependymal tumours are divided into 1) supratentorial ependymoma with two molecular subtypes 1a) supratentorial ependymoma, ZFTA fusion-positive, 1b) supratentorial ependymoma,

YAP1 fusion-positive, 2) posterior fossa ependymoma with two subtypes 2a) posterior fossa group A (PFA) 2b) posterior fossa group B (PFB) ependymoma, 3) spinal ependymoma with one subtype 3a) spinal ependymoma, *MYCN*-amplified, 4) myxopapillary ependymoma, and 5) subependymoma.

Supratentorial ependymomas constitute a one third of all ependymomas and occur in all age groups, in large part in children. They are localized in hemispheres, but not always with a connection to the ventricular system. The fourth ventricle is the most common localization of posterior fossa ependymomas, being more common in children with median age of 6 years. Spinal ependymomas are usually located in the cervical and thoracic region, most commonly in the age of 25-45 [40]. For all ependymomas, morphological and immunohistochemical features are still crucial for the diagnosis. Ependymoma is a tumour composed of uniform cells embedded in a fibrillary matrix with presence of pseudorosettes and/or ependymal rosettes with a relatively low mitotic count. Despite the molecular status and localization, two grades 2 or 3 are determined according to histological criteria such as brisk mitotic count and microvascular proliferation. Unfortunately the cut-off value of mitotic figures is undefined. The term anaplastic ependymoma is no longer recommended for grade 3 tumours [21]. In supratentorial tumours *ZFTA* and *YAP1* fusions are present, in posterior fossa methylation groups A and B are distinguished (PFA and PFB), and for spinal ependymomas, a subtype with *MYCN*-amplification is the most common alteration [21,47]. Some immunohistochemical surrogates might be used to distinguish molecular subgroups. Cytoplasmic expression of L1CAM and nuclear p65 positivity relates to *ZFTA* fusions. For *MYCN*-amplified tumours, extensive nuclear expression of *MYCN* might be detected by immunohistochemistry. *YAP1*-fused tumours usually have strong dot-like EMA immunopositivity. In most PFA ependymomas, immunoreactivity for H3 p.K28me3 (K27me3) is lost which is in contrast to the PFB group. However, in some PFA, K27me3 might be at least partially retained [48,56]. Those tumours, where the mutation analysis is not performed, should be diagnosed as ependymomas NOS.

There are no major changes to the diagnostic criteria for subependymomas (WHO CNS grade 1) and myxopapillary ependymomas (now grade 2). The provisional term “anaplastic myxopapillary ependymomas”

was introduced for cases with hypercellularity and reduced mucin in association with an increased mitotic count (≥ 2 mitoses/mm²) and Ki-67 index $\geq 10\%$ [11,47].

Methylation analysis in WHO CNS 5th edition

Covalent addition of the methyl group to cytosine within DNA strand is referred to as DNA methylation and methylation of cytosines within the gene promoter in general interferes with the gene expression. Cancer unique methylome carries the information of both somatically acquired DNA methylation changes and the level of histogenesis and the cell of origin. Locus specific changes of methylation as well as loss of methylation at the genome level are one of the hallmarks of neoplastic transformation including gliomas. In gliomas, due to the recent technological developments, the methylome analysis has become a powerful tool in the diagnostics of brain tumours that can even lead to changes of the initial histopathological diagnosis [28,30,33]. In some tumour types, the methylation profile analysis provides significant prognostic and predictive information used in clinical practice [58]. These allow to subclassify CNS tumours that were previously considered homogeneous diseases or were difficult to be categorized [33]. Moreover, the combination of artificial intelligence in analysis and a growing amount of methylation profiling data available from the particular cancer cases allowed already to establish a blueprint for new brain tumour classifiers [31]. This has given rise to new entities as well as improvement of WHO CNS grade stratification [80]. The methylome profil-

ing appears to be especially important in classification of rare cases. However, the technology used for methylation profiling is still rather cost intensive and requires high technological expertise what still limits its practical implementation in most laboratories. Nevertheless, DNA methylation profiling appears to provide a significant improvement of CNS tumour classification, but its routine application in general practice is likely to take time [33,80].

Therapeutic consideration in gliomas

Classical treatment options for gliomas include surgery, radiation therapy (RT), chemotherapy, active surveillance and supportive care. Age and performance status at the time of diagnosis are major prognostic factors. The 2021 WHO Classification of CNS tumours introduced a new diagnostic framework relying on molecular characteristics. However, most evidence on the adjuvant treatment of gliomas was generated in the clinical trials conducted when traditional histologic criteria were the basis for enrolment and cohort assessment. The results come from molecularly heterogeneous gliomas in categories that are often no longer consistent with the contemporary classification. Applying available results in patients with molecularly defined tumour types poses a significant challenge for a treating physician [52,82].

The primary goal of surgery in gliomas is to establish a histological and molecular diagnosis and, as the extent of the resection is a major prognostic factor, to remove as much tumour tissue as feasible without compromising neurological function [53]. Postoperative neurological deficits are a negative prognostic factor and should be avoided, especially

Table II. Treatment options according to the tumour type in the adult type diffuse gliomas

Tumour type	Treatment
Oligodendroglioma, IDH-mutant, 1p19q codeleted, CNS WHO grade 2	RT in combination with PCV (or TMZ if toxicity is a concern) Wait and see – in patients with positive prognostic factors (complete resection, younger age)
Oligodendroglioma, IDH-mutant, 1p19q codeleted, CNS WHO grade 3	Radiation in combination with PCV (or TMZ if toxicity is a concern)
Astrocytoma, IDH-mutant, CNS WHO grade 2	RT with adjuvant chemotherapy (PCV or TMZ) Wait and see – in patients with favourable prognostic factors (complete resection, younger age, no neurological deficits)
Astrocytoma, IDH-mutant, CNS WHO grade 3	Adjuvant RT followed by TMZ
Astrocytoma, IDH-mutant, CNS WHO grade 4	Adjuvant RT followed by TMZ concurrent TMZ and RT with six months of adjuvant TMZ
Glioblastoma, IDH-wildtype, CNS WHO grade 4	Concurrent TMZ and RT with six months of adjuvant TMZ

CNS – central nervous system, WHO – World Health Organization, TMZ – temozolomide, PCV – procarbazine, lomustine, and vincristine, RT – radiotherapy, IDH – isocitrate dehydrogenase

Table III. Molecular markers of glial tumours and immunohistochemical surrogates

Type of tumour	Molecular markers/characteristics of molecular profile, detecting methods	Immunohistochemical surrogate
Adult-type diffuse gliomas		
Astrocytoma, IDH-mutant	<i>IDH1</i> (mostly p.R132H), <i>IDH2</i> <i>ATRX</i> , <i>TP53</i> sequencing	IDH R132H positive Loss of <i>ATRX</i> p53 positive (mutation pattern)
Oligodendroglioma, IDH-mutant and 1p/19q-codeleted	1p/19q-codeletion, FISH <i>IDH1</i> (mostly p.R132H), <i>IDH2</i> <i>TERT</i> , <i>CIC</i> sequencing	IDH R132H positive <i>ATRX</i> positive p53 (no mutation pattern)
Glioblastoma, IDH-wildtype	IDH-wildtype (<i>IDH1/2</i>) <i>TERT</i> , <i>EGFR</i> amplification, +7/–10 chromosome copy-number changes sequencing, FISH	IDH R132H negative <i>ATRX</i> positive p53 (no mutation pattern)
Paediatric-type diffuse low-grade gliomas		
Diffuse astrocytoma, <i>MYB</i> - or <i>MYBL1</i> -altered	<i>MYB</i> , <i>MYBL1</i> FISH IDH-wildtype (<i>IDH2</i> , <i>IDH2</i>) H3-wildtype (<i>H3-3A</i> , <i>HIST1H3B</i> , <i>HISTH3BC</i>) sequencing	MAP2 IDH1 R132H negative
Angiocentric glioma	<i>MYB</i> (mostly <i>MYB:QKI</i>) FISH	
Polymorphous low-grade neuroepithelial tumour of the young	<i>BRAF</i> (mostly <i>BRAF p.V600E</i>) sequencing FGFR fusions, FISH	<i>BRAF</i> V600E IDH1 R132H negative
Diffuse low-grade glioma, MAPK pathway-altered	<i>FGFR1</i> <i>BRAF p.V600E</i> sequencing	<i>BRAF</i> V600E IDH1 R132H negative
Paediatric-type diffuse high-grade gliomas		
Diffuse midline glioma, H3 K27-altered	H3-3A, <i>HIST1H3B</i> , <i>HIST1H3BC</i> (p.K28M) <i>TP53</i> , <i>ACVR1</i> , <i>PDGFRA</i> , <i>EGFR</i> , <i>EZH1P</i> sequencing	Loss of H3 K27me3 H3 p.K28M positive <i>EZH1P</i> positive IDH R132H negative
Diffuse hemispheric glioma, H3 G34-mutant	H3-3A (p.G35R/V) <i>TP53</i> , <i>ATRX</i> , <i>MGMT</i> sequencing	H3.3 p.G34R/V positive Loss of <i>ATRX</i> p53 positive (mutation pattern) IDH R132H negative
Diffuse paediatric-type high-grade glioma (pHGG), H3-wildtype and IDH-wildtype	IDH-wildtype (<i>IDH1</i> , <i>IDH2</i>) H3-wildtype (<i>H3-3A</i> , <i>HIST1H3B</i> , <i>HIST1H3BC</i>) <i>PDGFRA</i> , <i>MYCN</i> , <i>EGFR</i> sequencing	IDH1 R132H negative H3.3 p.G34R/V negative H3 K27me3 positive <i>ATRX</i> positive
Infant-type hemispheric glioma	<i>NTRK 1/2/3</i> , <i>ALK</i> , <i>ROS</i> , <i>MET</i> FISH, sequencing	<i>ALK</i> , <i>ROS</i> positive
Circumscribed astrocytic gliomas		
Pilocytic astrocytoma	<i>KIAA1549:BRAF</i> gene fusions, <i>NTRK1</i> , <i>NTRK2</i> gene fusions FISH <i>BRAF</i> , <i>NF1</i> , <i>IDH1/2</i> wildtype sequencing	<i>BRAF</i> V600E
High-grade astrocytoma with piloid features	<i>ATRX</i> , <i>NF1</i> , <i>CDKN2A/B</i> sequencing	Loss of <i>ATRX</i> IDH R132H negative
Pleomorphic xanthoastrocytoma	<i>BRAF p.V600E</i> , <i>CDKN2A/B</i> sequencing	<i>BRAF</i> V600E IDH R132H negative <i>ATRX</i> positive
Astroblastoma, <i>MN1</i> -altered	<i>MN1</i> (mainly <i>BEND2</i> fusion) FISH	
Ependymal tumour		
Supratentorial ependymoma		
Supratentorial ependymoma, <i>ZFTA</i> fusion-positive	<i>ZFTA-RELA</i> FISH	L1CAM positive p65 positive
Supratentorial ependymoma, <i>YAP1</i> fusion-positive	<i>YAP1-MAMLD1</i> FISH	
Posterior fossa ependymoma		
Posterior fossa group A (PFA) ependymoma	Reduction in H3 p.K28me3 <i>EZH1P</i> sequencing	Loss of H3 K27me3
Posterior fossa group B (PFB) ependymoma	Retained H3 p.K28me3	
Spinal ependymoma		
Spinal ependymoma, <i>MYCN</i> -amplified	<i>NF2</i> , <i>MYCN</i> sequencing, FISH	<i>MYCN</i> positive

in younger patients. In order to achieve the maximal safe resection, multiple technical developments were introduced including surgical navigation systems with multiparametric magnetic resonance imaging (MRI), tumour tissue visualization with 5-aminolevulinic acid and intraoperative neuro-monitoring. To assess the completeness of surgical resection contrast enhanced MRI performed within 24-48 hours of surgery is recommended. Due to the infiltrative growth pattern of many types of diffuse gliomas despite the efforts to obtain radiologically complete resection, most patients eventually experience recurrence. Adjuvant therapeutic approaches are used to delay tumour progression and improve survival. In order to help clinicians in decision making and address problems with applicability of the available clinical trial results in the recently defined tumour types, EANO, ASCO and the Society for Neuro-Oncology (SNO) issued updated guidelines for the treatment of glial tumours in adults [52,82]. Table II shows basic treatment options according to the tumour type [5,58,74,78,85]. Decision making based on MGMT promoter methylation status is limited to people of older age or poor performance status [58,85]. At recurrence, standards of care are not defined. Surgery and radiotherapy might be considered, especially in patients with longer time to recurrence from the completion of the first-line treatment. Nitrosourea regimens, temozolomide and, with consideration of the country-specific label, bevacizumab are options of pharmacotherapy with no proven impact on overall survival. Multiple trials designed for molecularly defined disease entities are ongoing but currently only minority of the adults patients can benefit from molecularly targeted therapies.

Currently overall 5-year survival rate in children with brain tumours exceeds 70% but it strongly depends on the tumour type. Majority of patients experience long-term sequelae related to the presence of tumour and its treatment. Survival in many tumour types is dependent on the use of radiotherapy which has debilitating effects on growth and neurologic development, induction of secondary tumours has been observed in long-term survivors. Different dedicated treatment protocols concern the specific paediatric gliomas, where the re-stratification and development of molecularly targeted therapies can significantly affect overall survival, side effects and quality of life [8,50].

Conclusions

Every glioma patient needs the individual neuro-oncological approach, since brain tumour diagnostics and therapy have some immanent limitations and difficulties on different levels. The final diagnosis of gliomas depends on a complex clinical, neuro-imaging, pathological and molecular approach, as well as experience and continuous cooperation. The classification of central nervous system tumours in 2021 was updated with continuation of previously established conceptions and is constantly evolving. However, histopathological examination together with immunohistochemistry remains the main fundamentals of the diagnosis, especially for those working in the centres without access to broad molecular diagnostics tools. All specialists who handle CNS tumours have to be aware of new trends, actual diagnostic approaches and potential of new molecular tests. Table III shows the most common molecular alterations found in tumours, as well as available immunohistochemical surrogates which should be implemented and available in neurooncological diagnostic centers. In pediatric tumours and some unequivocal cases the methylation profiling may be necessary. It is an open question how well this new classification will stand the test of time and permanent progress, become widely implemented in everyday practice, and first of all – bring therapeutic innovations for the patients.

Testing algorithms of molecular biomarkers and workflows differ across pathologists, institutions and countries, but crucial ancillary tests are needed to classify gliomas and optimize clinical management.

Disclosure

The authors report no conflict of interest.

References

1. Appay R, Dehais C, Maurage CA, Alentorn A, Carpentier C, Colin C, Ducray F, Escande F, Idbaih A, Kamoun A, Marie Y, Mokhtari K, Tabouret E, Trabelsi N, Uro-Coste E, Delattre J-Y, Figarella-Branger D, Desenclos C, Sevestre H, Menei P, Rousseau A, Cruel T, Lopez S, Mihai MI, Petit A, Adam C, Parker F, Dam-Hieu P, Quintin-Roue I, Eimer S, Loiseau H, Bekaert L, Chapon F, Ricard D, Godfraind C, Khalil T, Cazals-Hatem D, Faillot T, Gaultier C, Tortel MC, Carpiuc I, Richard P, Lahiani W, Aubriot-Lorton H, Ghiringhelli F, Maurage CA, Ramirez C, Gueye EM, Labrousse F, Chinot O, Bauchet L, Rigau V, Beauchesne P, Gauchotte G, Campone M, Loussouarn D, Fontaine D, Vanden-

- bos-Burel F, Le Floch A, Roger P, Blechet C, Fesneau M, Carpentier A, Delattre JY, Elouadhani-Hamdi S, Polivka M, Larrieu-Ciron D, Milin S, Colin P, Diebold MD, Chiforeanu D, Vauleon E, Langlois O, Laquerriere A, Forest F, Motso-Fotso MJ, Andraud M, Runavot G, Lhermitte B, Noel G, Gaillard S, Villa C, Desse N, Rousselot-Denis C, Zemmoura I, Cohen-Moyal E, Uro-Coste E, Dhermain F, Network P. CDKN2A homozygous deletion is a strong adverse prognosis factor in diffuse malignant IDH-mutant gliomas. *Neuro Oncol* 2019; 21: 1519-1528.
2. Appay R, Tabouret E, Macagno N, Touat M, Carpentier C, Colin C, Ducray F, Idhah A, Mokhtari K, Uro-Coste E, Dehais C, Figarella-Branger D, Network P. IDH2 mutations are commonly associated with 1p/19q codeletion in diffuse adult gliomas. *Neuro Oncol* 2018; 20: 716-718.
 3. Arita H, Narita Y, Fukushima S, Tateishi K, Matsushita Y, Yoshida A, Miyakita Y, Ohno M, Collins VP, Kawahara N, Shibui S, Ichimura K. Upregulating mutations in the TERT promoter commonly occur in adult malignant gliomas and are strongly associated with total 1p19q loss. *Acta Neuropathol* 2013; 126: 267-276.
 4. Bailey OT. Genesis of the Percival-Bailey-Cushing Classification of gliomas. *Pediatr Neurosci* 1986; 12: 261-265.
 5. Baumert BG, Hegi ME, van den Bent MJ, von Deimling A, Golia T, Hoang-Xuan K, Brandes AA, Kantor G, Taphoorn MJB, Ben Hassel M, Hartmann C, Ryan G, Capper D, Kros JM, Kurscheid S, Wick W, Enting R, Reni M, Thiessen B, Dhermain F, Bromberg JE, Feuvret L, Reijneveld JC, Chinot O, Gijtenbeek JMM, Rossiter JP, Dif N, Balana C, Bravo-Marques J, Clement PM, Marosi C, Tzuk-Shina T, Nordal RA, Rees J, Lacombe D, Mason WP, Stupp R. Temozolomide chemotherapy versus radiotherapy in high-risk low-grade glioma (EORTC 22033-26033): a randomised, open-label, phase 3 intergroup study. *Lancet Oncol* 2016; 17: 1521-1532.
 6. Becker AP, Sells BE, Haque SJ, Chakravarti A. Tumor Heterogeneity in Glioblastomas: From Light Microscopy to Molecular Pathology. *Cancers (Basel)* 2021; 13: 761.
 7. Bender K, Perez E, Chirica M, Onken J, Kahn J, Brenner W, Ehret F, Euskirchen P, Koch A, Capper D, Kaul D. High-grade astrocytoma with piloid features (HGAP): the Charite experience with a new central nervous system tumor entity. *J Neurooncol* 2021; 153: 109-120.
 8. Bennett J, Erker C, Lafay-Cousin L, Ramaswamy V, Hukin J, Vanan MI, Cheng S, Coltin H, Fonseca A, Johnston D, Lo A, Zeller S, Alvi S, Bowes L, Brossard J, Charlebois J, Eisenstat D, Felton K, Fleming A, Jabado N, Larouche V, Legault G, Mpofu C, Perreault S, Silva M, Sinha R, Strother D, Tsang DS, Wilson B, Crooks B, Bartels U. Canadian pediatric neuro-oncology standards of practice. *Front Oncol* 2020; 10: 593192.
 9. Berzero G, Di Stefano AL, Ronchi S, Bielle F, Villa C, Guillerme E, Capelle L, Mathon B, Laurence A, Giry M, Schmitt Y, Marie Y, Idhah A, Hoang-Xuan K, Delattre J-Y, Mokhtari K, Sanson M. IDH-wildtype lower-grade diffuse gliomas: the importance of histological grade and molecular assessment for prognostic stratification. *Neuro Oncol* 2021; 23: 955-966.
 10. Bettegowda C, Agrawal N, Jiao Y, Sausen M, Wood LD, Hruban RH, Rodriguez FJ, Cahill DP, McLendon R, Riggins G, Velculescu VE, Oba-Shinjo SM, Marie SKN, Vogelstein B, Bigner D, Yan H, Papadopoulos N, Kinzler KW. Mutations in CIC and FUBP1 contribute to human oligodendroglioma. *Science* 2021; 333: 1453-1455.
 11. Board Editorial. WHO Classification of Tumours. Central nervous system tumours. Fifth Edition. International Agency for Research on Cancer, Lyon 2021.
 12. Brat DJ, Aldape K, Bridge JA, Canoll P, Colman H, Hameed MR, Harris BT, Hattab EM, Huse JT, Jenkins RB, Lopez-Terrada DH, McDonald WC, Rodriguez FJ, Souter LH, Colasacco C, Thomas NE, Yount MH, van den Bent MJ, Perry A. Molecular biomarker testing for the diagnosis of diffuse gliomas. *Arch Pathol Lab Med* 2022; 146: 547-574.
 13. Brat DJ, Aldape K, Colman H, Figarella-Branger D, Fuller GN, Giannini C, Holland EC, Jenkins RB, Kleinschmidt-DeMasters B, Komori T, Kros JM, Louis DN, McLean C, Perry A, Reifenberger G, Sarkar C, Stupp R, van den Bent MJ, von Deimling A, Weller M. cIMPACT-NOW update 5: recommended grading criteria and terminologies for IDH-mutant astrocytomas. *Acta Neuropathologica* 2020; 139: 603-608.
 14. Brat DJ, Aldape K, Colman H, Holland EC, Louis DN, Jenkins RB, Kleinschmidt-DeMasters BK, Perry A, Reifenberger G, Stupp R, von Deimling A, Weller M. cIMPACT-NOW update 3: recommended diagnostic criteria for "Diffuse astrocytic glioma, IDH-wildtype, with molecular features of glioblastoma, WHO grade IV". *Acta Neuropathol* 2018; 136: 805-810.
 15. Capper D, Jones DTW, Sill M, Hovestadt V, Schrimpf D, Sturm D, Koelsche C, Sahm F, Chavez L, Reuss DE, Kratz A, Wefers AK, Huang K, Pajtler KW, Schweizer L, Stichel D, Olar A, Engel NW, Lindenberg K, Harter PN, Braczynski AK, Plate KH, Dohmen H, Garvalov BK, Coras R, Holsken A, Hewer E, Bewerunge-Hudler M, Schick M, Fischer R, Beschoner R, Schittenhelm J, Staszewski O, Wani K, Varlet P, Pages M, Temming P, Lohmann D, Selt F, Witt H, Milde T, Witt O, Aronica E, Giangaspero F, Rushing E, Scheurlen W, Geisenberger C, Rodriguez FJ, Becker A, Preusser M, Haberler C, Bjerkvig R, Cryan J, Farrell M, Deckert M, Hench J, Frank S, Serrano J, Kannan K, Tsigos A, Bruck W, Hofer S, Brehmer S, Seiz-Rosenhagen M, Hanggi D, Hans V, Rozsnoki S, Hansford JR, Kohlhof P, Kristensen BW, Lechner M, Lopes B, Mawrin C, Ketter R, Kulozik A, Khatib Z, Heppner F, Koch A, Juvet A, Keohane C, Muhleisen H, Mueller W, Poh U, Prinz M, Benner A, Zaparka M, Gottardo NG, Driever PH, Kramm CM, Muller HL, Rutkowski S, von Hoff K, Fruhwald MC, Gnekow A, Fleischhack G, Tippelt S, Calaminus G, Monoranu CM, Perry A, Jones C, Jacques TS, Radlwimmer B, Gessi M, Pietsch T, Schramm J, Schackert G, Westphal M, Reifenberger G, Wesseling P, Weller M, Collins VP, Blumcke I, Bendszus M, Debus J, Huang A, Jabado N, Northcott PA, Paulus W, Gajjar A, Robinson GW, Taylor MD, Jaunmuktane Z, Ryzhova M, Platten M, Unterberg A, Wick W, Karajannis MA, Mittelbronn M, Acker T, Hartmann C, Aldape K, Schuller U, Buslei R, Lichter P, Kool M, Herold-Mende C, Ellison DW, Hasselblatt M, Snuder M, Brandner S, Korshunov A, von Deimling A, Pfister SM. DNA methylation-based classification of central nervous system tumours. *Nature* 2018; 555: 469-474.
 16. Carstam L, Corell A, Smits A, Denes A, Barcheus H, Modin K, Sjogren H, Ferreyra Vega S, Bontell TO, Caren H, Jakola AS. WHO grade loses its prognostic value in molecularly defined diffuse lower-grade gliomas. *Front Oncol* 2022; 11: 803975.
 17. Castro LNG, Wesseling P. The cIMPACT-NOW updates and their significance to current neuro-oncology practice. *Neurooncol Pract* 2021; 8: 4-10.

18. Cavenee K, Kleihues P. Pathology and Genetics of Tumours of the Nervous System. WHO Classification of Tumours. Third edition. International Agency for Research on Cancer, Lyon 2000.
19. Chatwin HV, Cruz JC, Green AL. Pediatric high-grade glioma: moving toward subtype-specific multimodal therapy. *FEBS J* 2021; 288: 6127-6141.
20. Dias-Santagata D, Lam Q, Vernovsky K, Vena N, Lennerz JK, Borger DR, Batchelor TT, Ligon KL, Iafrate AJ, Ligon AH, Louis DN, Santagata S. BRAF V600E mutations are common in pleomorphic xanthoastrocytoma: diagnostic and therapeutic implications. *PLoS One* 2011; 6: e17948.
21. Ellison DW, Aldape KD, Capper D, Fouladi M, Gilbert MR, Gilbertson RJ, Hawkins C, Merchant T, Pajtler K, Venneti S, Louis DN. cIMPACT-NOW update 7: advancing the molecular classification of ependymal tumors. *Brain Pathol* 2020; 30: 863-866.
22. Ellison DW, Hawkins C, Jones DTW, Onar-Thomas A, Pfister SM, Reifenberger G, Louis DN. cIMPACT-NOW update 4: diffuse gliomas characterized by MYB, MYBL1, or FGFR1 alterations or BRAF(V600E) mutation. *Acta Neuropathol* 2019; 137: 683-687.
23. Ferguson S, Lesniak MS. Percival Bailey and the classification of brain tumors. *Neurosurg Focus* 2005; 18: 1-6.
24. Figarella-Branger D, Mokhtari K, Dehais C, Carpentier C, Colin C, Jouve A, Uro-Coste E, Forest F, Maurage CA, Vignaud JM, Polivka M, Lechapt-Zalcman E, Eimer S, Viennet G, Quintin-Roue I, Aubriot-Lorton MH, Diebold M-D, Loussouarn D, Lacroix C, Rigau V, Laquerriere A, Vandenbos F, Michalak S, Sevestre H, Peoch M, Labrousse F, Christov C, Kemery JL, Chenard MP, Chiforeanu D, Ducray F, Idbah A, Delattre JY, Network P. Mitotic index, microvascular proliferation, and necrosis define 3 pathological subgroups of prognostic relevance among 1p/19q co-deleted anaplastic oligodendrogliomas. *Neuro Oncol* 2016; 18: 888-890.
25. Funakoshi Y, Hata N, Kuga D, Hatae R, Sangatsuda Y, Fujioka Y, Takigawa K, Mizoguchi M. Pediatric glioma: an update of diagnosis, biology, and treatment. *Cancers* 2021; 13: 758.
26. Gondim DD, Gener MA, Curless KL, Cohen-Gadol AA, Hattab EM, Cheng L. Determining IDH-mutational status in gliomas using IDH1-R132H antibody and polymerase chain reaction. *Appl Immunohistochem Mol Morphol* 2019; 27: 722-725.
27. Han GQ, Zhang JS, Ma Y, Gui QP, Yin S. Clinical characteristics, treatment and prognosis of angiocentric glioma. *Oncol Lett* 2020; 20: 1641-1648.
28. Hanahan D. Hallmarks of cancer: new dimensions. *Cancer Discov* 2022; 12: 31-46.
29. Haque F, Varlet P, Puntinet J, Storer L, Bountali A, Rahman R, Grill J, Carcaboso AM, Jones C, Layfield R, Grundy RG. Evaluation of a novel antibody to define histone 3.3 G34R mutant brain tumours. *Acta Neuropathol Commun* 2017; 5: 45.
30. Hegi ME, Diserens A, Gorlia T, Hamou M, de Tribolet N, Weller M, Kros JM, Hainfellner JA, Mason W, Mariani L, Bromberg JEC, Hau P, Mirimanoff RO, Cairncross JG, Janzer RC, Stupp R. MGMT gene silencing and benefit from temozolomide in glioblastoma. *N Engl J Med* 2005; 352: 997-1003.
31. Hegi ME, Kleihues P, Wen PY, Suva ML. Toward methylation-based classification of central nervous system tumors. *Neuro Oncol* 2018; 20: 579-581.
32. Huse JT, Snuderl M, Jones DTW, Brathwaite CD, Altman N, Lavi E, Saffery R, Sexton-Oates A, Blumcke I, Capper D, Karajannis MA, Benayed R, Chavez L, Thomas C, Serrano J, Borsu L, Ladanyi M, Rosenblum MK. Polymorphous low-grade neuroepithelial tumor of the young (PLNTY): an epileptogenic neoplasm with oligodendroglioma-like components, aberrant CD34 expression, and genetic alterations involving the MAP kinase pathway. *Acta Neuropathol* 2017; 133: 417-429.
33. Karimi S, Zuccato JA, Mamatjan Y, Mansouri S, Suppiah S, Nasiri F, Diamandis P, Munoz DG, Aldape KD, Zadeh G. The central nervous system tumor methylation classifier changes neurooncology practice for challenging brain tumor diagnoses and directly impacts patient care. *Clin Epigenetics* 2019; 11: 185.
34. Kleihues P, Burger PC, Scheithauer BW. Histological Typing of Tumours of the Central Nervous System. 2nd ed. Springer-Verlag, Berlin 1993.
35. Komori T. The molecular framework of pediatric-type diffuse gliomas: shifting toward the revision of the WHO classification of tumors of the central nervous system. *Brain Tumor Pathol* 2021; 38: 1-3.
36. Komori T. Grading of adult diffuse gliomas according to the 2021 WHO Classification of Tumors of the Central Nervous System. *Lab Invest* 2022; 102: 126-133.
37. Komori T. The 2021 WHO classification of tumors, 5th edition, central nervous system tumors: the 10 basic principles. *Brain Tumor Pathol* 2022; 39: 47-50.
38. Korshunov A, Schrimpf D, Ryzhova M, Sturm D, Chavez L, Hovestadt V, Sharma T, Habel A, Burford A, Jones C, Zheludkova O, Kumirova E, Kramm CM, Golanov A, Capper D, von Deimling A, Pfister SM, Jones DTW. H3-/IDH-wild type pediatric glioblastoma is comprised of molecularly and prognostically distinct subtypes with associated oncogenic drivers. *Acta Neuropathol* 2017; 134: 507-516.
39. Kulac I, Tihan T. Pilocytic astrocytomas: a short review. *Brain Tumor Pathol* 2019; 36: 52-55.
40. Larrew T, Saway BF, Lowe SR, Olar A. Molecular classification and therapeutic targets in ependymoma. *Cancers (Basel)* 2021; 13: 6218.
41. Lim KY, Won JK, Park CK, Kim SK, Choi SH, Kim T, Yun H, Park SH. H3 G34-mutant high-grade glioma. *Brain Tumor Pathol* 2021; 38: 4-13.
42. Louis DN, Ohgaki H, Wiestler OD, Cavenee WK. WHO Classification of Tumours of the Central Nervous System. 4th ed. International Agency for Research on Cancer, Lyon 2007.
43. Louis DN, Ohgaki H, Wiestler O, Cavenee W. WHO Classification of Tumours of the Central Nervous System. Revised 4th ed. International Agency for Research on Cancer, Lyon 2016.
44. Louis DN, Giannini C, Capper D, Paulus W, Figarella-Branger D, Lopes MB, Batchelor TT, Cairncross JG, van den Bent M, Wick W, Wesseling P. cIMPACT-NOW update 2: diagnostic clarifications for diffuse midline glioma, H3 K27M-mutant and diffuse astrocytoma/anaplastic astrocytoma, IDH-mutant. *Acta Neuropathol* 2018; 135: 639-642.
45. Louis DN, Perry A, Burger P, Ellison DW, Reifenberger G, von Deimling A, Aldape K, Brat D, Collins VP, Eberhart C, Figarella-Branger D, Fuller GN, Giangaspero F, Giannini C, Hawkins C, Kleihues P, Korshunov A, Kros JM, Lopes MB, Ng HK, Ohgaki H, Paulus W, Pietsch T, Rosenblum M, Rushing E, Soylemezoglu F, Wiestler O, Wesseling P. International Society of Neuropathol-

- ogy-Haarlem consensus guidelines for nervous system tumor classification and grading. *Brain Pathol* 2014; 24: 429-435.
46. Louis DN, Perry A, Reifenberger G, von Deimling A, Figarella-Branger D, Cavenee WK, Ohgaki H, Wiestler OD, Kleihues P, Ellison DW. The 2016 World Health Organization Classification of Tumors of the Central Nervous System: a summary. *Acta Neuropathol* 2016; 131: 803-820.
 47. Louis DN, Perry A, Wesseling P, Brat DJ, Cree IA, Figarella-Branger D, Hawkins C, Ng HK, Pfister SM, Reifenberger G, Soffietti R, von Deimling A, Ellison DW. The 2021 WHO Classification of Tumors of the Central Nervous System: a summary. *Neuro Oncol* 2021; 23: 1231-1251.
 48. Louis DN, Wesseling P, Aldape K, Brat DJ, Capper D, Cree IA, Eberhart C, Figarella-Branger D, Fouladi M, Fuller GN, Giannini C, Haberler C, Hawkins C, Komori T, Kros JM, Ng HK, Orr BA, Park SH, Paulus W, Perry A, Pietsch T, Reifenberger G, Rosenblum M, Rous B, Sahm F, Sarkar C, Solomon DA, Tabori U, van den Bent MJ, von Deimling A, Weller M, White VA, Ellison DW. cIMPACT-NOW update 6: new entity and diagnostic principle recommendations of the cIMPACT-Utrecht meeting on future CNS tumor classification and grading. *Brain Pathol* 2020; 30: 844-856.
 49. Louis DN, Wesseling P, Paulus W, Giannini C, Batchelor TT, Cairncross JG, Capper D, Figarella-Branger D, Lopes MB, Wick W, van den Bent M. cIMPACT-NOW update 1: not otherwise specified (NOS) and not elsewhere classified (NEC). *Acta Neuropathol* 2018; 135: 481-484.
 50. Lutz K, Juenger ST, Messing-Juenger M. Essential management of pediatric brain tumors. *Children (Basel)* 2022; 9: 498.
 51. Mhatre R, Sugur HS, Nandeesh BN, Chickabasaviah Y, Saini J, Santosh V (2019) MN1 rearrangement in astroblastoma: study of eight cases and review of literature. *Brain Tumor Pathol* 2019; 36: 112-120.
 52. Mohile NA, Messersmith H, Gatson NT, Hottinger AF, Lassman A, Morton J, Ney D, Nghiemphu PL, Olar A, Olson J, Perry J, Portnow J, Schiff D, Shannon A, Shih HA, Strowd R, van den Bent M, Ziu M, Blakeley J. Therapy for diffuse astrocytic and oligodendroglial tumors in adults: ASCO-SNO guideline. *J Clin Oncol* 2022; 40: 403-426.
 53. Molinaro A, Hervey-Jumper S, Morshed RA. Association of maximal extent of resection of contrast-enhanced and non-contrast-enhanced tumor with survival within molecular subgroups of patients with newly diagnosed glioblastoma. *JAMA Oncol* 2020; 6: 444-503.
 54. Onizuka H, Masui K, Komori T. Diffuse gliomas to date and beyond 2016 WHO Classification of Tumours of the Central Nervous System. *Int J Clin Oncol* 2020; 25: 997-1003.
 55. Ostrom QT, Cioffi G, Gittleman H, Patil N, Waite K, Kruchko C, Barnholtz-Sloan JS. CBTRUS statistical report: primary brain and other central nervous system tumors diagnosed in the United States in 2012-2016. *Neuro Oncol* 2019; 21 (Suppl 5): v1-v100.
 56. Panwalkar P, Clark J, Ramaswamy V, Hawes D, Yang FS, Dunham C, Yip S, Hukin J, Sun YL, Schipper MJ, Chavez L, Margol A, Pekmezci M, Chung C, Banda A, Bayliss JM, Curry SJ, Santi M, Rodriguez FJ, Snuderl M, Karajannis MA, Saratsis AM, Horbinski CM, Carret AS, Wilson B, Johnston D, Lafay-Cousin L, Zelcer S, Eisenstat D, Silva M, Scheinemann K, Jabado N, McNeely PD, Kool M, Pfister SM, Taylor MD, Hawkins C, Korshunov A, Judkins AR, Venneti S. Immunohistochemical analysis of H3K27me3 demonstrates global reduction in group-A childhood posterior fossa ependymoma and is a powerful predictor of outcome. *Acta Neuropathol* 2017; 134: 705-714.
 57. Parsons DW, Jones S, Zhang XS, Lin JCH, Leary RJ, Angenendt P, Mankoo P, Carter H, Siu IM, Gallia GL, Olivi A, McLendon R, Rasheed BA, Keir S, Nikolskaya T, Nikolsky Y, Busam DA, Tekleab H, Diaz LA, Hartigan J, Smith DR, Strausberg RL, Marie SKN, Shinjo SMO, Yan H, Riggins GJ, Bigner DD, Karchin R, Papadopoulos N, Parmigiani G, Vogelstein B, Velculescu VE, Kinzler KW. An integrated genomic analysis of human glioblastoma Multiforme. *Science* 2008; 321: 1807-1812.
 58. Perry JR, Laperriere N, O'Callaghan CJ, Brandes AA, Menten J, Phillips C, Fay M, Nishikawa R, Cairncross JG, Roa W, Osoba D, Rossiter JP, Sahgal A, Hirte H, Laigle-Donadey F, Franceschi E, Chinot O, Golfinopoulos V, Fariselli L, Wick A, Feuvret L, Back M, Tills M, Winch C, Baumert BG, Wick W, Ding K, Mason WP, Trial I. Short-course radiation plus temozolomide in elderly patients with glioblastoma. *N Engl J Med* 2017; 376: 1027-1037.
 59. Phillips JJ, Gong H, Chen K, Joseph NM, van Ziffle J, Bastian BC, Grenert JP, Kline CN, Mueller S, Banerjee A, Nicolaides T, Gupta N, Berger MS, Lee HS, Pekmezci M, Tihan T, Bollen AW, Perry A, Shieh JTC, Solomon DA. The genetic landscape of anaplastic pleomorphic xanthoastrocytoma. *Brain Pathol* 2019; 29: 85-96.
 60. Picart T, Barritault M, Poncet D, Berner L, Izquierdo C, Tabouret E, Figarella-Branger D, Idbaih A, Bielle F, Bourg V, Vandenbos FB, Moyal EC-J, Uro-Coste E, Guyotat J, Honnorat J, Gabut M, Meyronet D, Ducray F. Characteristics of diffuse hemispheric gliomas, H3 G34-mutant in adults. *Neurooncol Adv* 2021; 3: v000061.
 61. Plant-Fox AS, O'Halloran K, Goldman S. Pediatric brain tumors: the era of molecular diagnostics, targeted and immune-based therapeutics, and a focus on long term neurologic sequelae. *Curr Probl Cancer* 2021; 45: 100777.
 62. Purkait S, Jha P, Sharma MC, Suri V, Sharma M, Kale SS, Sarkar C. CDKN2A deletion in pediatric versus adult glioblastomas and predictive value of p16 immunohistochemistry. *Neuropathol* 2013; 33: 405-412.
 63. Reinhardt A, Stichel D, Schrimpf D, Sahm F, Korshunov A, Reuss DE, Koelsche C, Huang K, Wefers AK, Hovestadt V, Sill M, Gramatzki D, Felsberg J, Reifenberger G, Koch A, Thomale UW, Becker A, Hans VH, Prinz M, Staszewski O, Acker T, Dohmen H, Hartmann C, Mueller W, Tuffaha MSA, Paulus W, Hess K, Brokinkel B, Schittenhelm J, Monoranu CM, Kessler AF, Loehr M, Buslei R, Deckert M, Mawrin C, Kohlhof P, Hewer E, Olar A, Rodriguez FJ, Giannini C, NageswaraRao AA, Tabori U, Nunes NM, Weller M, Pohl U, Jaunmuktane Z, Brandner S, Unterberg A, Haenggi D, Platten M, Pfister SM, Wick W, Herold-Mende C, Jones DTW, Deimling A, Capper D. Anaplastic astrocytoma with piloid features, a novel molecular class of IDH wildtype glioma with recurrent MAPK pathway, CDKN2A/B and ATRX alterations. *Acta Neuropathol* 2018; 136: 273-291.
 64. Richardson TE, Hatanpaa KJ, Walker JM. Molecular characterization of "true" low-grade IDH-wild-type astrocytomas. *J Neuropathol Exp Neurol* 2021; 80: 431-435.

65. Rodriguez FJ, Scheithauer BW, Burger PC, Jenkins S, Giannini C. Anaplasia in pilocytic astrocytoma predicts aggressive behavior. *Am J Surg Pathol* 2010; 34: 147-160.
66. Roux A, Roca P, Edjlali M, Sato K, Zanello M, Dezamis E, Gori P, Lion S, Fleury A, Dhermain F, Meder JF, Chretien F, Lechapt E, Varlet P, Oppenheim C, Pallud J. MRI atlas of IDH wild-type supratentorial glioblastoma: probabilistic maps of phenotype, management, and outcomes. *Radiology* 2019; 293: 633-643.
67. Ryall S, Tabori U, Hawkins C. Pediatric low-grade glioma in the era of molecular diagnostics. *Acta Neuropathol Commun* 2020; 8: 30.
68. Scheithauer BW. Development of the WHO classification of tumors of the central nervous system: a historical perspective. *Brain Pathol* 2009; 19: 551-564.
69. Sejda A, Grajkowska W, Iżycka-Świeszewska E. The history of the classification of central nervous system tumors. In: *History of pathology: a meeting point between medicine, art, museum, and a look at the future*. Medical University of Gdańsk, Gdańsk 2018; 71-79.
70. Shaikh N, Brahmabhatt N, Kruser TJ, Kam KL, Appin CL, Wadhvani N, Chandler J, Kumthekar P, Lukas RV. Pleomorphic xanthoastrocytoma: a brief review. *CNS Oncol* 2019; 8: CNS39.
71. Shirahata M, Ono T, Stichel D, Schrimpf D, Reuss DE, Sahm F, Koelsche C, Wefers A, Reinhardt A, Huang K, Sievers P, Shimizu H, Nanjo H, Kobayashi Y, Miyake Y, Suzuki T, Adachi J, Mishima K, Sasaki A, Nishikawa R, Bewerunge-Hudler M, Ryzhova M, Absalyamova O, Golanov A, Sinn P, Platten M, Jungk C, Winkler F, Wick A, Hanggi D, Unterberg A, Pfister SM, Jones DTW, van den Bent M, Hegi M, French P, Baumert BG, Stupp R, Gorlia T, Weller M, Capper D, Korshunov A, Herold-Mende C, Wick W, Louis DN, von Deimling A. Novel, improved grading system(s) for IDH-mutant astrocytic gliomas. *Acta Neuropathol* 2018; 136: 153-166.
72. Sonoda Y, Yokoo H, Tanaka S, Kinoshita M, Nakada M, Nishihara H, Comm Mol Diag Japan Soc B. Practical procedures for the integrated diagnosis of astrocytic and oligodendroglial tumors. *Brain Tumor Pathol* 2019; 36: 56-62.
73. Sporikova Z, Slavkovsky R, Tuckova L, Kalita O, Houdova MM, Ehrmann J, Hajduch M, Hrabalek L, Vaverka M. IDH1/2 mutations in patients with diffuse gliomas: a single centre retrospective massively parallel sequencing analysis. *Appl Immunohistochem Mol Morphol* 2022; 30: 178-183.
74. Stupp R, Hegi ME, Mason WP, van den Bent MJ, Taphoorn MJB, Janzer RC, Ludwin SK, Allgeier A, Fisher B, Belanger K, Hau P, Brandes AA, Gijtenbeek J, Marosi C, Vecht CJ, Mokhtari K, Wesseling P, Villa S, Eisenhauer E, Gorlia T, Weller M, Lacombe D, Cairncross JG, Mirimanoff RO, European Org R, Treatment, Canc Brain Tumour G, Radiat Oncol G, Natl Canc Inst Canada Clin T. Effects of radiotherapy with concomitant and adjuvant temozolomide versus radiotherapy alone on survival in glioblastoma in a randomised phase III study: 5-year analysis of the EORTC-NCIC trial. *Lancet Oncol* 2009; 10: 459-466.
75. Tanboon J, Williams EA, Louis DN. The diagnostic use of immunohistochemical surrogates for signature molecular genetic alterations in gliomas. *J Neuropathol Exp Neurol* 2016; 75: 4-18.
76. Tesileanu CMS, Dirven L, Wijnenga MMJ, Koekkoek JAF, Vincent A, Dubbink HJ, Atmodimedjo PN, Kros JM, van Duinen SG, Smits M, Taphoorn MJB, French PJ, van den Bent MJ. Survival of diffuse astrocytic glioma, IDH1/2 wildtype, with molecular features of glioblastoma, WHO grade IV: a confirmation of the cIMPACT-NOW criteria. *Neuro Oncol* 2020; 22: 515-523.
77. Tihan T, Fisher PG, Kepner JL, Godfraind C, McComb RD, Goldthwaite PT, Burger PC. Pediatric astrocytomas with monomorphous pilomyxoid features and a less favorable outcome. *J Neuropathol Exp Neurol* 1999; 58: 1061-1068.
78. van den Bent MJ, Tesileanu CMS, Wick W, Sanson M, Brandes AA, Clement PM, Erridge S, Vogelbaum MA, Nowak AK, Baurain JF, Mason WP, Wheeler H, Chinot OL, Gill S, Griffin M, Rogers L, Taal W, Ruda R, Weller M, McBain C, Reijneveld J, Enting RH, Caparrotti F, Lesimple T, Clenton S, Gijtenbeek A, Lim E, Herrlinger U, Hau P, Dhermain F, de Heer I, Aldape K, Jenkins RB, Dubbink HJ, Kros JM, Wesseling P, Nuyens S, Golfopoulos V, Gorlia T, French P, Baumert BG. Adjuvant and concurrent temozolomide for 1p/19q non-co-deleted anaplastic glioma (CATNON; EORTC study 26053-22054): second interim analysis of a randomised, open-label, phase 3 study. *Lancet Oncol* 2021; 22: 813-823.
79. Velazquez Vega JE, Brat DJ, Ryken TC, Olson JJ. The role of neuropathology in the management of newly diagnosed glioblastoma: a systematic review and evidence-based clinical practice guideline. *J Neurooncol* 2020; 150: 143-164.
80. Verburg N, Barthel FP, Anderson KJ, Johnson KC, Koopman T, Yaqub MM, Hoekstra OS, Lammertsma AA, Barkhof F, Pouwels PJW, Reijneveld JC, Rozemuller AJM, Belien JAM, Boellaard R, Taylor MD, Das S, Costello JF, Vandertop WP, Wesseling P, Hamer PCdW, Verhaak RGW. Spatial concordance of DNA methylation classification in diffuse glioma. *Neuro Oncol* 2021; 23: 2054-2065.
81. Watanabe T, Nobusawa S, Kleihues P, Ohgaki H. IDH1 mutations are early events in the development of astrocytomas and oligodendrogliomas. *Am J Pathol* 2009; 174: 1149-1153.
82. Weller M, van den Bent M, Preusser M, Le Rhun E, Tonn JC, Minniti G, Bendszus M, Balana C, Chinot O, Dirven L, French P, Hegi ME, Jakola AS, Platten M, Roth P, Ruda R, Short S, Smits M, Taphoorn MJB, von Deimling A, Westphal M, Soffietti R, Reifenberger G, Wick W. EANO guidelines on the diagnosis and treatment of diffuse gliomas of adulthood. *Nat Rev Clin Oncol* 2022; 19: 357-358.
83. Wesseling P. Neurooncology: 2021 update. *Free Neuropathol* 2022; 2: 5.
84. Wesseling P, Rozowsky JS. Neurooncology: 2022 update. *Free Neuropathol* 2022; 3: 4.
85. Wick W, Platten M, Meisner C, Felsberg J, Tabatabai G, Simon M, Nikkhah G, Papsdorf K, Steinbach JP, Sabel M, Combs SE, Vesper J, Braun C, Meixensberger J, Ketter R, Mayer-Steinacker R, Reifenberger G, Weller M, German Canc S. Temozolomide chemotherapy alone versus radiotherapy alone for malignant astrocytoma in the elderly: the NOA-08 randomised, phase 3 trial. *Lancet Oncol* 2012; 13: 707-715.
86. Woehrer A, Sander P, Habeder C, Kern S, Maier H, Preusser M, Hartmann C, Kros JM, Hainfellner JA. FISH-based detection of 1p 19q codeletion in oligodendroglial tumors: procedures and protocols for neuropathological practice – a publication under the auspices of the Research Committee of the European Confederation of Neuropathological Societies (Euro-CNS). *Clin Neuropathol* 2011; 30: 47-55.
87. Zulch K. Histological typing of tumours of the central nervous system. World Health Organization, Geneva 1979.