

Update on molecular findings, management and outcome in low-grade gliomas

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Abstract | Low-grade infiltrating gliomas in adults include diffuse astrocytoma, oligoastrocytoma and oligodendroglioma. The current gold standard diagnosis of these tumors relies on histological classification; however, emerging molecular abnormalities discovered in these tumors are playing an increasingly prominent part in the process of tumor diagnosis and, consequently, patient management. The frequency and clinical importance of tumor protein p53 (*TP53*) abnormalities, deletions involving chromosomes 1p and 19q, *O*⁶-methylguanine-DNA methyltransferase (*MGMT*) promoter methylation status, abnormalities in the *PTEN* tumor suppressor gene and the *BRAF* oncogene, and isocitrate dehydrogenase (*IDH*) mutations have become better defined. Molecular markers have not, historically, had an important role in determining the course of treatment for patients with low-grade gliomas, but ongoing phase III clinical trials incorporate 1p deletion or 1p19q codeletion status—and future trials plan to incorporate *MGMT* promoter methylation status—as stratification factors. Future trials will need to incorporate *IDH* mutational status in addition to these factors. Ultimately, molecular marker assessment will, hopefully, improve the accuracy of tumor diagnosis and enhance the effectiveness of treatment to achieve improved patient outcomes.

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Introduction

The current gold standard method for the diagnosis of low-grade infiltrating gliomas relies on histological assessment of neoplastic brain tissue. Discoveries related to underlying molecular abnormalities in these tumors, however, have begun to generate new paradigms for understanding how these neoplasms develop, how they can be diagnosed, and how they might be more effectively treated. The prevalence rate of primary gliomas is ≈34 per 100,000 individuals,¹ while the incidence is ≈6 per 100,000 person-years.¹ Among the subtypes of low-grade infiltrating gliomas, incidence rates of 0.14, 0.30 and 0.10 new cases per 100,000 individuals have been estimated for diffuse astrocytomas, oligodendrogliomas and mixed oligoastrocytomas, respectively.²

The first section of this Review highlights the known functions, frequencies of occurrence, and methods of detecting tumor protein p53 (*TP53*) pathway abnormalities, chromosomal deletions involving 1p and 19q, *O*⁶-methylguanine-DNA methyltransferase (*MGMT*) promoter methylation status, phosphatase and tensin homolog (*PTEN*) and v-raf murine sarcoma viral oncogene homolog B1 (*BRAF*) abnormalities, and isocitrate dehydrogenase (*IDH*) mutations in low-grade gliomas. Some of the advantages and disadvantages of various detection methods are considered. Issues related to specific molecular methods of analysis and topics such as amplification have been covered elsewhere,^{3–5} and will not be discussed in detail here. The second section

of the Review discusses how incorporation of selected molecular data in ongoing phase III clinical trials influences patient management and outcomes. Here, the discussion primarily focuses on the use of 1p deletion or 1p19q codeletion status, *MGMT* promoter methylation status, and the future use of *IDH* mutational status as stratification factors in such trials.

Low-grade gliomas defined

The current WHO classification of primary CNS tumors recognizes four separate tumor grades (I–IV), which can be grouped into low-grade (I and II) or high-grade (III and IV) categories depending on the presence or absence of high-grade features, such as microvascular proliferation and necrosis.² Low-grade gliomas are tumors that exhibit glial differentiation and which, by definition, lack high-grade findings. Low-grade glioma categories include subependymal giant cell astrocytoma, pilocytic astrocytoma, pilomyxoid astrocytoma, diffuse astrocytoma, pleomorphic xanthoastrocytoma, oligodendroglioma, oligoastrocytoma, and the various ependymomas. The category of low-grade glioma excludes the so-called neuroepithelial and mixed glial–neuronal tumors.

The current Review focuses on diffuse astrocytomas, oligodendrogliomas and oligoastrocytomas, all of which are characterized by their infiltration into surrounding brain tissue. Diffuse astrocytomas are infiltrating, hypercellular tumors composed of atypical cells that show astrocytic differentiation and mildly increased mitotic activity. Oligodendrogliomas are infiltrating tumors composed of cells with scant cytoplasm, round

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Competing interests

The authors declare no competing interests.

Key points

- Isocitrate dehydrogenase (*IDH*) mutations seem to occur earlier than tumor protein p53 (*TP53*) mutations or deletions of chromosomes 1p and/or 19q among a subset of diffuse astrocytomas and oligodendroglial tumors
- Tumors with 1p and/or 19q chromosomal deletions usually also have *IDH* mutations
- 1p19q codeletion and *IDH* mutation each have favorable prognostic value, with *IDH*-mutated, 1p19q-intact tumors having an outcome intermediate between 1p19q-codeleted tumors and tumors that lack either of these molecular markers
- The independent prognostic importance of *O*⁶-methylguanine-DNA methyltransferase (*MGMT*) promoter methylation remains to be determined
- No molecular marker has yet been validated as predicting a favorable response to chemotherapy or radiation therapy

nuclei, variably prominent perinuclear clearing, and a relatively even cellular distribution. Finally, low-grade oligoastrocytomas exhibit a mixed phenotype in which some tumor cells show astrocytic differentiation while others show oligodendroglial differentiation.

Molecular markers**Tumor protein p53 pathway abnormalities**

Molecular abnormalities involving the p53 pathway are early events in the molecular pathogenesis of diffuse astrocytomas.² Population-based studies have shown that the incidence of *TP53* mutations is ≈53% in diffuse astrocytoma, 44% in oligoastrocytoma, and 13% in oligodendroglioma (Table 1).⁶

The *TP53* gene itself is the most commonly mutated p53 pathway gene in low-grade gliomas; however, molecular abnormalities involving other genes in the pathway—such as *p14^{ARF}* (p14 alternative reading frame product of cyclin-dependent kinase inhibitor 2A) or *MDM2* or *MDM4* p53 binding protein homolog—have also been described. Hypermethylation of *p14^{ARF}* has been detected in 33% of diffuse astrocytomas,⁷ and methylation of the *p14^{ARF}* promoter is thought to be an early event in tumor development.^{7,8} PCR analysis of 27 recurrent or progressed astrocytomas showed that hypermethylation of *p14^{ARF}* and *MGMT* was relatively frequent, occurring in 26% and 63% of cases, respectively.⁹ Except in one case, hypermethylation involved either *p14^{ARF}* or *MGMT*, and homozygous deletion of *p14^{ARF}* was not found in any of the cases. The nature of abnormalities involving the *MDM2* gene is less clear. One study found DNA amplifications in a majority of a small number of diffuse astrocytomas, with overexpression of *MDM2* in the amplified region.¹⁰ However, a subsequent study, which included mostly glioblastomas, found no evidence of *MDM2* amplification among diffuse astrocytomas.¹¹

Deletions involving chromosomes 1p and 19q

Deletion of chromosomes 1p and/or 19q represents a well-recognized molecular abnormality most strongly associated with oligodendrogliomas that exhibit the classic microscopic features of uniformly round nuclei and small nucleoli, an even cellular distribution, and prominent perinuclear clearing ('haloes').¹² Several

reports have documented the frequencies of 1p deletion, 19q deletion and 1p19q codeletion status among diffuse astrocytomas, oligodendrogliomas and oligoastrocytomas (Table 1).^{6,13–15} Reported frequencies of codeletion range from 0–10% for astrocytomas, 21–59% for oligoastrocytomas, and 39–70% for oligodendrogliomas.^{13–15} A report published in 2009 documented the relative homogeneity of 1p19q chromosomal loss within a given tumor.¹⁶ In most codeleted cases, an unbalanced translocation involving chromosomes 1 and 19 is thought to mediate the combined deletions of 1p and 19q.^{17,18} Loss of 1p and/or 19q and mutations involving *TP53* are virtually always mutually exclusive events.

The two methods most commonly used in clinical neuropathology to detect 1p and 19q loss are PCR-based loss of heterozygosity (LOH) studies and fluorescence *in situ* hybridization (FISH) analysis. Both techniques have relatively high sensitivity and specificity, although FISH analysis does not require normal DNA for comparison and also allows chromosomal copy number assessment.¹⁹ The ability to detect polysomy among codeleted anaplastic oligodendrogliomas could predict early recurrence,²⁰ but this advantage afforded by FISH analysis has not been verified among low-grade gliomas. The loci of interest for FISH analysis must be carefully selected, since both astrocytic and oligodendroglial tumors commonly show 1p deletions, and 1p36 deletions are relatively common among astrocytic tumors.²¹ Other methods, such as multiplex ligation-dependent probe amplification, have yet to be widely adopted.²²

***MGMT* promoter methylation**

MGMT is a DNA repair enzyme that removes alkyl groups from the *O*⁶ position of guanine—a function that underlies the development of resistance to alkylating agent therapy in some patients.²³ Methylation of the *MGMT* promoter confers increased sensitivity of glioblastoma to the effects of alkylating agents, presumably by decreasing *MGMT* activity.^{24,25}

MGMT expression has been documented in both low-grade and high-grade gliomas. *MGMT* activity studies and anti-*MGMT* immunohistochemical staining have shown that ≈15% of diffuse astrocytomas and glioblastomas lack *MGMT* activity,²⁶ and *MGMT* expression is detectable by immunohistochemical analysis in ≈68% of cases (Table 1).²⁷

The important issues of testing methodology and heterogeneity of protein expression have been addressed in the literature. In contrast with previous studies that used formalin-fixed, paraffin-embedded (FFPE) tissue, *MGMT* methylation status determination on frozen brain tumor samples by means of methylation-specific PCR analysis showed that methylated *MGMT* was present in ≈93% of low-grade gliomas.²⁸ The enhanced detection rate was attributed to the use of frozen rather than FFPE tissue.

The issue of protein heterogeneity was addressed in a study of glioblastoma surgical biopsy and subtotal resection specimens, and multiple cell lines.²⁹ PCR analysis demonstrated notable intratumoral heterogeneity of *MGMT* methylation, while western blot studies

Table 1 | Frequencies of selected molecular abnormalities among low-grade gliomas

Molecular abnormality	Astrocytoma (%) (grade II)	Oligoastrocytoma (%) (grade II)	Oligodendroglioma (%) (grade II)
<i>TP53</i> mutations ⁶	53	44	13
1p/19q codeletion ^{13–15}	0–10	21–59	39–70
<i>MGMT</i> hypermethylation ²⁸	11	27	62
<i>PTEN</i> mutations ^{34,36}	0	0	0
<i>PTEN</i> promoter methylation ³⁸	43	67	50
<i>IDH1</i> mutations ^{50,54–57,59}	59–88	50–100	68–82
<i>IDH2</i> mutations ^{54,56}	1–7	1	4–5

Abbreviations: *IDH*, isocitrate dehydrogenase; *MGMT*, O⁶-methylguanine-DNA methyltransferase; *PTEN*, phosphatase and tensin homolog; *TP53*, tumor protein p53.

and immunohistochemical analysis revealed marked heterogeneity of protein expression. Low-grade gliomas were not analyzed in this study, but would be expected to produce similar results. A study of the equally important issue of concordance between *MGMT* methylation status and protein expression found only poor to moderate correlation between *MGMT* protein expression, as measured by immunohistochemical analysis, and PCR-based *MGMT* methylation status in glioblastoma samples.²⁴ To our knowledge, similar comparison studies have not been performed on large samples of low-grade gliomas.

Of note, changes in *MGMT* methylation after tumor progression have also been observed. A study of *MGMT* promoter methylation status among paired grade II oligodendrogliomas and oligoastrocytomas that progressed to anaplastic (grade III) tumors found that 31% of tumors with initial 1p chromosomal deletion showed a gain in methylation at progression, compared with 87.5% for tumors with chromosome 1p intact.³⁰

Phosphatase and tensin homolog

PTEN exerts its putative tumor suppressor function via inhibition of the phosphatidylinositol 3-kinase–Akt signaling pathway.³¹ Immunohistochemical analysis has failed to detect *PTEN* expression in normal human brain tissue,³² although some studies using immunoblot analysis have shown marked *PTEN* expression in such tissue.³³

A study published in 1999 identified *PTEN* mutations in 28% of glioblastomas, 7% of anaplastic astrocytomas, and 0% of low-grade gliomas through the use of denaturing gel electrophoresis followed by DNA sequencing.³⁴ Subsequent reports that employed various mutation detection methods, including LOH analysis³⁵ and direct sequencing,³⁶ have confirmed the presence of *PTEN* mutations in ≈30% of glioblastoma cases and the near-complete absence of *PTEN* mutations in low-grade gliomas (Table 1). Analyses of *PTEN* protein expression carried out by western blot, immunoblot assays and immunohistochemistry have consistently demonstrated an inverse relationship between tumor grade and *PTEN* expression in adult gliomas.^{32,33} These observations have led most investigators to conclude that *PTEN* mutations are relatively late genetic events that occur in a subset of gliomas during evolution from low-grade to high-grade tumors. A study published in 2007 reported that,

in contrast to adult gliomas, pediatric gliomas that had undergone malignant transformation had an unexpectedly high frequency of *PTEN* deletion, as measured by FISH analysis.³⁷ This study underscores the important genetic differences between pediatric and adult glioma biology.

Although *PTEN* mutation is exceedingly rare in low-grade gliomas and relatively common in glioblastoma, *PTEN* promoter methylation is common in grade II gliomas and quite frequent in secondary glioblastomas.³⁸ These observations suggest that *PTEN* promoter methylation represents an epigenetic mechanism that results in *PTEN* inactivation and tumor development in low-grade gliomas. Some studies based on non-CNS tumors have shown a positive correlation between *PTEN* methylation status and protein expression by western blot and/or immunohistochemical analysis,^{39,40} although other studies failed to corroborate this finding.^{41,42}

BRAF mutations

Mutations in the chromosome 7 (7q34) gene *BRAF*, the protein product of which activates the mitogen-activated protein kinase (MAPK) pathway, are thought to contribute to the tumorigenesis of a variety of human malignancies.⁴³ In 2008, *BRAF* abnormalities were described among low-grade gliomas—namely pilocytic astrocytomas⁴⁴—and overexpression of the gene product, B-Raf, was thought to drive MAPK pathway activation.⁴⁵

Array-comparative genomic hybridization and FISH analysis of the *BRAF* locus revealed copy number gains of *BRAF* in 23–38% of adult low-grade gliomas, including diffuse astrocytomas, depending on the presence or absence of associated chromosome 7 gains.⁴⁵ A later study described a tandem duplication at 7q34 that produced a novel oncogenic *BRAF* fusion gene (*KIAA1549:BRAF*).⁴⁶ This duplication occurred in 66% of pilocytic astrocytomas, but was not seen in any of 50 diffusely infiltrating astrocytomas. Thus, while both diffuse astrocytomas and pilocytic astrocytomas may show gains involving 7q34, the *KIAA1549:BRAF* fusion seems to be specific for pilocytic astrocytomas.⁴⁷ The oncogenic fusion *SRGAP3:RAF1*, which is rarely observed, as well as the Val600Glu point mutation and a trinucleotide insertion at codon 598 in *BRAF*, are additional mechanisms that result in MAPK activation in pilocytic astrocytoma.⁴⁸

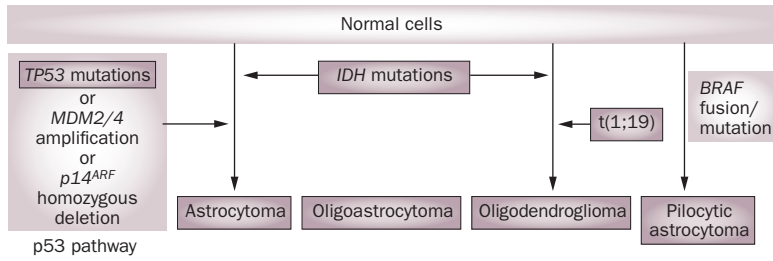


Figure 1 | A model for the development and progression of astrocytic and oligodendroglial tumors. Abbreviations: *BRAF*, v-raf murine sarcoma viral oncogene homolog B1; *IDH*, isocitrate dehydrogenase; *MDM2/4*, MDM2 and MDM4 p53 binding protein homologs; *p14^{ARF}*, p14 alternative reading frame product of cyclin-dependent kinase inhibitor 2A; *t(1;19)*, translocation involving chromosomes 1 and 19 resulting in 1p and 19q loss; *TP53*, tumor protein p53. Permission obtained from Oxford University Press Ltd © Ichimura, K. *et al. Neuro. Oncol.* **11**, 341–347 (2009).

Isocitrate dehydrogenase mutations

IDH1 and *IDH2* are NADP-dependent enzymes that normally catalyze the production of α-ketoglutarate from isocitrate.⁴⁹ Since the initial publication by Parsons *et al.*⁵⁰ that recognized the link between *IDH1* mutations and glioblastoma, additional reports have provided further insight into potential mechanisms of *IDH*-related glioma pathogenesis, the types and frequencies of *IDH* mutations, and the diagnostic utility of *IDH* mutation and mutant protein detection among low-grade gliomas.

Mutations involving codon 132 of *IDH1* result in reduced enzymatic activity of mutant *IDH1* proteins compared with wild-type *IDH1*, as a result of substantially impaired isocitrate binding.⁵¹ One study suggested that inactivation of *IDH1* through mutation results in induction of the hypoxia-inducible factor-1α (*HIF-1α*) pathway.⁵¹ A subsequent study, however, demonstrated that *IDH1* mutations result in the production and accumulation of D-2-hydroxyglutarate (D-2HG), which may act as an oncogenic metabolite.⁵² Glioma development has been associated with elevated 2HG levels in the rare autosomal recessive condition hydroxyglutaric aciduria, but the L-2HG enantiomer, rather than the D-2HG form that Dang *et al.*⁵² described, is the accumulated metabolite in this condition.⁵³ Thus, the role of 2HG in glioma pathogenesis remains unclear.

Several studies have reported the surprising frequency of *IDH* mutations among gliomas. Various detection methods, including direct sequencing analysis,⁵⁴ have identified *IDH1* mutations in 59–88% of diffuse astrocytomas, 68–82% of oligodendrogliomas, 50–100% of oligoastrocytomas, 50–78% of anaplastic astrocytomas, 49–75% of anaplastic oligodendrogliomas, 63–100% of anaplastic oligoastrocytomas, 3–7% of primary glioblastomas, and 50–88% of secondary glioblastomas (Table 1).^{50,54–59} Among the different types of *IDH1* mutations, the vast majority result in an amino acid substitution at residue 132 (Arg132His).⁵⁴

The data obtained to date indicate that *IDH2* mutations are present in 1–7% of diffuse astrocytomas, 4–5% of oligodendrogliomas, ≤1% of oligoastrocytomas,

1–4% of anaplastic astrocytomas, 5–8% of anaplastic oligodendrogliomas, and ≤6% of anaplastic oligoastrocytomas, but are not seen in either primary or secondary glioblastomas (Table 1).^{54,56}

Some—but not all—reports have detected *IDH* mutations among pilocytic astrocytomas.^{55,57,58} Importantly, *IDH* mutations have not been detected among ependymomas,^{55–58} medulloblastomas,^{55,56,58} schwannomas,^{55,58} meningiomas,^{55,58} peripheral neuroectodermal tumors, or dysembryoplastic neuroepithelial tumors.⁵⁸ Early studies showed no evidence of *IDH* mutations among non-CNS tumors,^{56,60} but later reports documented the presence of *IDH1* codon 132 mutations in small numbers of prostate carcinomas and B-acute lymphoblastic leukemias.⁶¹ Other studies have reported *IDH1*^{62–64} and *IDH2*^{63,64} mutations in a small percentage of patients with acute myeloid leukemia.

Unlike *TP53* mutations and 1p19q codeletion, which are mutually exclusive events among most low-grade gliomas, the vast majority of *IDH1* mutations are seen in *TP53*-mutated or 1p19q-codeleted tumors.⁵⁸ This observation, combined with the finding that *IDH* mutations have not been shown to occur later than either *TP53* mutations or 1p19q loss,⁶⁵ supports the contention that *IDH* mutations are very early events in the development of infiltrating gliomas (Figure 1).

In addition to mutation frequency analysis, the diagnostic utility of *IDH* mutation or mutant protein detection among low-grade gliomas has been explored. One group performed mutational analysis on a series of glial tumors and so-called ‘reactive’ lesions, and found *IDH1* or *IDH2* mutations in 49% of the gliomas, but not in any of the non-neoplastic samples.⁶⁶ A mouse monoclonal antibody targeting the *IDH1* Arg132His substitution seems to show high specificity for this mutation and can label individual infiltrating tumor cells.⁶⁷ A study published in 2009 confirmed the utility of immunohistochemistry in distinguishing infiltrating glioma from reactive gliosis.⁶⁸ Finally, combined *IDH* and *BRAF* analysis could enable a confident distinction to be made between pilocytic astrocytoma and diffuse astrocytoma.⁴⁷ This differentiation would be especially helpful in the setting of small biopsy samples that show overlapping morphological features.

Management and outcome

To date, molecular markers have generally not had a major role in treatment decisions. Standard management of low-grade glioma begins with tissue confirmation of the diagnosis, and most specialists would favor maximal safe tumor resection over biopsy despite absence of level 1 supporting evidence.^{69,70} Fractionated radiotherapy has well-established benefits in terms of prolonging time to tumor progression and radiographic response.⁷¹ Radiotherapy at diagnosis does not improve overall survival rates compared with radiotherapy at progression⁷¹ and, in addition, radiotherapy conveys a risk of cognitive decline that is unrelated to tumor progression.⁷² Radiotherapy is, therefore, often deferred in ‘low-risk’ low-grade glioma patients (those <40 years

of age with well-controlled symptoms and no evidence of radiographic progression). Nonetheless, almost all patients with low-grade glioma receive radiotherapy at some point during their treatment, usually as the first non-surgical therapy. Chemotherapy with lipid-soluble alkylating agents, particularly temozolomide, also has documented activity in low-grade glioma. Chemotherapy is most commonly employed to treat post-radiation tumor progression. To date, combined radiochemotherapy in low-grade glioma, consisting of radiotherapy plus procarbazine, lomustine and vincristine (PCV), has not demonstrated a favorable effect on overall survival rates compared with radiotherapy alone (with chemotherapy at progression), although a favorable influence on progression-free survival was observed.⁷³

Ongoing phase III trials in low-grade glioma seek to determine the role of temozolomide in management of these tumors. The first trial, European Organization for Research and Treatment of Cancer protocol 22033-26033 (launched in 2005), randomly assigned patients to either temozolomide or fractionated radiotherapy, and completed accrual at the beginning of 2010. The second trial, a US study involving all major US cooperative groups (ECOG E3F05), opened to accrual in late 2009 and randomly assigned patients to standard radiotherapy alone or with daily temozolomide, followed by up to 12 cycles of standard temozolomide. The former trial used 1p status while the latter used the presence or absence of 1p19q codeletion as a stratification factor. Both trials plan to incorporate analyses of *MGMT* promoter methylation status. *IDH* mutational status was not considered within the design of either trial, although this parameter will, undoubtedly, be analyzed.

Clinical trials have not focused on the effects of *PTEN* or *BRAF* abnormalities in low-grade gliomas, owing to the rarity of these abnormalities. *TP53* mutational status, which is cumbersome to determine and is outweighed in importance by 1p19q status, has not been systematically analyzed. Instead, efforts have been directed at trying to understand the prognostic (in terms of overall survival) and predictive (in terms of treatment response) importance of the molecular abnormalities involving 1p19q and the *MGMT* and *IDH* genes.

Several studies have examined 1p19q or 1p status as a prognostic factor for overall survival. Most^{17,74–82} but not all^{6,13,83–85} of these studies found that codeletion (or deletion of 1p) was prognostic. Whether 1p19q codeletion predicts favorable therapeutic response is also unclear. Some studies have suggested that codeleted 1p19q status predicts response to radiation^{17,86} or chemotherapy;^{78,87,88} however, most studies have been unable to confirm codeletion as a predictor of chemotherapy response.^{80,81,89–91}

Few studies have explored *MGMT* status in low-grade glioma. Two studies relying on univariate analysis have suggested that *MGMT* promoter methylation is associated with prolonged overall survival,^{81,92} but three multivariate analyses, as well as other studies examining *MGMT* expression by immunohistochemistry^{27,76} or methylation⁸⁴ analysis, found no association between promoter methylation and overall survival.^{59,80,92} Furthermore, one of the

two studies associating promoter methylation with overall survival noted a very strong correlation between *MGMT* status and 1p19q codeletion—a probable prognostic factor for survival.⁸¹ The literature on *MGMT* as a predictor of response to chemotherapy is similarly mixed. Two studies, reported in 2009⁸¹ and 2010 (W. Taal, personal communication), did not find promoter methylation to be predictive of response to temozolomide, in contrast to another study reported in 2006.²⁸ In the earlier study, a remarkable 93% of low-grade gliomas demonstrated *MGMT* promoter methylation.

Mutations in *IDH1* and *IDH2* are strongly linked to improved overall survival in low-grade gliomas. Low-grade glioma patients with *IDH1* mutation had a median survival of 151 months, compared with 60 months in patients with nonmutated tumors. When gliomas of all grades were considered in a multivariate model, including 1p19q and *MGMT* methylation status, *IDH1* mutations and, to a lesser extent, 1p19q codeletion, independently conveyed improved overall survival, while *MGMT* methylation was not associated with improved survival.⁵⁹ These results followed an identical pattern to that seen in a 2010 EORTC report on *IDH1* and *IDH2* mutations in anaplastic oligodendroglial tumors.⁹³ Recognizing the strong association between *IDH* mutations and 1p19q codeletion is important;^{56,59,94} virtually all codeleted tumors also carry an *IDH* mutation. *IDH* mutation also conveys improved overall survival in low-grade gliomas that lack 1p19q codeletion.^{94,95} The only study assessing *IDH* status as a predictor of response to temozolomide in low-grade gliomas did not find this marker to be predictive.⁹⁵

Some tentative conclusions can be drawn from the above data. *IDH* mutations and 1p19q codeletion seem to be independent markers of overall survival. At present, none of these markers is a convincing or prospectively validated predictor of response to therapy; the data are strongest for 1p19q. Thus, use of these markers to assign patients to chemotherapy seems premature. Ongoing randomized clinical trials, already stratified according to 1p or 1p19q status, will also need to analyze results on the basis of *IDH* mutational status. Conceivably, future trials will need to stratify patients into prognostically distinct subgroups harboring 1p19q codeletion (almost all of which will also have *IDH* mutation), *IDH* mutation without 1p19q codeletion, and neither *IDH* mutation nor 1p19q codeletion.⁹⁴

Finally, a distinct group of gliomas exhibiting characteristic patterns of promoter DNA methylation—termed the CpG island methylator phenotype (CIMP)—was reported in 2010.⁹⁶ The CIMP seems to be relatively common among low-grade gliomas, especially oligodendroglomas, and tends to be seen in younger patients who show improved survival. Interestingly, presence of the CIMP is strongly associated with *IDH1* mutations.

Conclusions

The current gold standard for the diagnosis of low-grade gliomas relies on histological examination of tumor tissue, but the application of various molecular testing

methods has uncovered important genetic abnormalities, some of which provide useful prognostic information about patient outcome and treatment. The abnormalities include *TP53* mutations, chromosome 1p and 19q deletion status, *MGMT* promoter methylation status, *PTEN* and *BRAF* mutations, and *IDH* gene mutations. Various testing methods, from protein expression analysis by immunohistochemistry to mutation detection by PCR-based assays, have provided insights into the relative frequencies of these abnormalities among low-grade gliomas. Incorporation of these molecular findings into strategies for patient management has been the aim of various clinical trials, which have focused on 1p19q deletion status, *MGMT* methylation status, and *IDH* mutational status.

Review criteria

Articles for this review were selected from searches of full-text, English language papers in the PubMed database. Search terms included: "glioma", "astrocytoma", "oligodendroglioma", "oligoastrocytoma", "pilocytic astrocytoma", "glioblastoma", "incidence", "prevalence", "molecular markers", "methylation", "mutation", "p53", "p14ARF", "1p", "19q", "deletion", "BRAF", "MGMT", "PTEN", "IDH", "mutations", "antibody", "immunohistochemistry", "fluorescence in situ hybridization", "amplification", "polymerase chain reaction", "clinical trial", "management", "phase III", "prognosis" and "temozolomide". For search result retrieval, no limits were placed on the year of publication. The reference lists of some original articles were consulted to ensure the completeness of citations.

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Author contributions

T. D. Bourne and D. Schiff both contributed equally to this work in terms of research, writing, editing and reviewing.