REVIEW

For reprint orders, please contact: reprints@futuremedicine.com

Emerging biomarkers in anaplastic oligodendroglioma: implications for clinical investigation and patient management

Solmaz Sahebjam¹, Mairéad G McNamara¹ & Warren P Mason^{*1}

Practice Points

- In anaplastic oligodendroglial tumors, chromosome 1p19q codeletion has been established as a predictive marker of response to treatment with procarbazine, lomustine and vincristine chemotherapy in addition to radiation therapy. Status of 1p19q codeletion should be identified in these patients prior to clinical decision-making.
- *MGMT* promoter hypermethylation confers improved prognosis in anaplastic oligodendroglial tumors.
- *IDH1* and *IDH2* have been shown to frequently be mutated in oligodendroglial tumors and are associated with better prognosis.
- Mutations of *CIC* and *FUBP1* have been closely associated with 1p19q codeletion and *IDH* mutations, but their clinical significance remains unknown.
- *CDKN2A* deletion has been associated with a poor response to procarbazine, lomustine and vincristine chemotherapy. However, this marker needs to be validated prospectively in clinical trials prior to its use in clinical decision-making.

SUMMARY Oligodendrogliomas are heterogeneous tumors with a variable response to treatment. This clinical variability underlines the urgent need for markers that can reliably aid diagnosis and guide clinical decision-making. Long-term follow-up data from the EORTC 26951 and RTOG 9402 clinical trials in newly diagnosed anaplastic oligodendroglioma have established chromosome 1p19q codeletion as a predictive marker of response to procarbazine, lomustine and vincristine chemotherapy in anaplastic oligodendrogliomas. In addition, *MGMT* promoter hypermethylation has been strongly associated with glioma CpG island hypermethylation phenotype (G-CIMP⁺) status, this has been suggested as an epiphenomenon of genome-wide methylation, conferring a more favorable prognosis. Molecular profiling of these tumors has identified several other markers with potential clinical significance: mutations of *IDH*, *CIC*, *FUBP1* and *CDKN2A* require further validation before they can be implemented as clinical decision-making tools. Additionally, recent data on the clinical significance of intrinsic glioma subtyping appears promising. Indeed, existing evidence suggests that comprehensive analyses such as intrinsic glioma subtyping or G-CIMP status are superior to single molecular markers. Clearly, with evolving treatment strategies and in the era of individualized therapy, broader omics-based molecular evaluations are required to improve outcome prediction and to identify patients who will benefit from specific treatment strategies.

¹Pencer Brain Tumor Centre, Princess Margaret Cancer Center, 610 University Avenue, Toronto, ON, M5G 2M9, Canada

^{*}Author for correspondence: Tel.: +1 416 946 2277; Fax: +1 416 946 2284; warren.mason@uhn.ca

Oligodendrogliomas are the second most common primary brain tumors in adults, accounting for approximately 2% of all primary brain neoplasms in adults [101]. These tumors are quite heterogeneous with a highly variable clinical course and response to treatment. Results of two pivotal Phase III trials, EORTC 26951 and RTOG 9402, have underlined the importance of comprehensive characterization of these tumors with the goal of effective patient selection for specific molecularly targeted therapies (Table 1) [1,2]. In this review, we focus on oligodendroglial biomarkers that are established and in development, and summarize the relevant clinical experience incorporating these molecular genetic features.

Codeletion of 1p19q

Codeletion of 1p19q resulting in loss of heterozygosity is a frequent finding in oligodendrogliomas occurring in 50–76% of these tumors [2,3]. Recently published results of the long-term follow-up of the patients enrolled into the RTOG 9402 and EORTC 26951 studies demonstrated that the addition of procarbazine, lomustine and vincristine (PCV) chemotherapy to radiation therapy (RT) dramatically improved overall survival (OS) in patients with codeleted 1p19q anaplastic oligodendrogliomas and anaplastic oligoastrocytomas, when compared with radiation alone [1,2].

The randomized EORTC 26951 investigated the addition of six cycles of adjuvant-standard PCV chemotherapy subsequent to RT of 59.4 Gy in 33 fractions [1,4] in patients with anaplastic oligodendrogliomas or anaplastic oligoastrocytomas [1,4]. This study reported a significant difference in OS, the primary end point, of 11.7 months with the addition of PCV

chemotherapy to RT (hazard ratio [HR]: 0.75; 95% CI: 0.60–0.95) [1]. While patients with 1p19q codeleted tumors derived more benefit from the addition of PCV chemotherapy to RT (OS not reached in the RT/PCV group vs 112 months in the RT group; HR: 0.56; 95% CI: 0.31–1.03; p = 0.0594), in patients with non-codeleted tumors, the risk reduction was lower (OS: 25 vs 21 months for the RT/PCV and the RT alone groups, respectively; HR: 0.83; 95% CI: 0.62–1.10) [1].

The Phase III randomized RTOG 9402 trial investigated the addition of intensive PCV chemotherapy (four cycles every 6 weeks before RT) to involved-field RT (59.4 Gy in 33 fractions) in an unselected population with anaplastic oligodendrogliomas and anaplastic oligoastrocytomas [2]. Similar to EORTC 26951, patients with codeleted tumors had improved OS compared with their counterparts with non-codeleted tumors (PCV plus RT arm: 14.7 vs 2.6 years; p < 0.001; RT alone arm: 7.3 vs 2.7 years; p < 0.001) and derived survival benefit from treatment with PCV plus RT (14.7 vs 7.3 years; HR: 0.59; 95% CI: 0.37–0.95; p = 0.03). However, median survival of patients with non-codeleted tumors were similar in both arms (2.6 vs 2.7 years; HR: 0.85; 95% CI: 0.58–1.23; p = 0.39) [2].

It is noteworthy that, in both trials, patients with 1p19q codeleted tumors who were initially treated in the RT-alone arm, had inferior survival despite more intense salvage chemotherapy at progression [1,2].

Given the results of these two studies, it is imperative to determine the 1p19q status of anaplastic oligodendroglial tumors for treatment planning. In cases with 1p19q allelic loss, RT alone is no longer considered an adequate

Table 1. Molecular alterations and their impact on overall survival in the RTOG 9402 and EORTC 26951 trials.							
Trial	Median OS (months)						Ref.
	1p19q: codeleted vs retained	MGMT promoter: methylated vs unmethylated	CIMP: CIMP+ vs CIMP-	IDH: mutated vs wild-type	CDKN2A: deleted vs retained	10q: loss vs intact	
RTOG 9402							
All patients	RR: 0.121	–	–	–	RR: 4.9	RR: 1.82	[2,29]
RT arm	87.6 vs 32.4	–	–	–	–	–	
PCV + RT	176.4 vs 31.2	–	–	–	–	–	
EORTC 26951							
All patients	123 vs 23	–	67.44 vs 14.88	–	–	–	[1,17]
RT	111.8 vs 21.1	43.3 vs 15.6	–	64.8 vs 14.7	–	–	
PCV + RT	NR vs 25	70.9 vs 16.6	–	NR vs 19	–	–	
CIMP: CpG island hypermethylation phenotype; NR: Not reached; OS: Overall survival; PCV: Procarbazine, lomustine and vincristine; RR: Relative risk; RT: Radiation therapy.							

treatment and these patients should receive chemotherapy in addition to RT.

The remarkable chemosensitivity of tumors with 1p19q codeletion has raised the possibility of using chemotherapy alone as an initial treatment strategy. **Figure 1** demonstrates the imaging response of a patient with an anaplastic oligoastrocytoma after nine cycles of treatment with temozolomide. However, the long-term clinical benefit of a strategy that uses temozolomide chemotherapy as a primary therapy for anaplastic gliomas with 1p19q codeletion has yet to be studied in randomized clinical trials, and is one of the questions that the CODEL study is designed to answer [102].

The optimal treatment of anaplastic gliomas without 1p19q codeletion remains unclear. In both of the aforementioned anaplastic oligodendroglial trials, patient selection was only based on histology, and some enrolled patients with non-codeleted oligodendroglial tumors still responded to chemotherapy [1,2,5]. The analysis of the RTOG 9402 study showed that survival curves of patients with non-codeleted tumors diverged in favor of PCV plus RT [2]. The ongoing, international, randomized Phase III CATNON study is addressing the value of the addition of temozolomide chemotherapy to irradiation for newly diagnosed 1p19q intact anaplastic gliomas. In this trial, following diagnosis, patients are randomly assigned to one of four treatment arms: RT only; RT with concomitant daily temozolomide; RT followed by adjuvant temozolomide; and RT with concomitant and adjuvant temozolomide [103].

Although RTOG 9402 and EORTC 26951 provided convincing evidence that, in the setting of PCV for anaplastic oligodendrogliomas and anaplastic oligoastrocytomas, codeletion of 1p19q is both a predictive and prognostic biomarker, our knowledge of the identity and function of the affected genes remains limited.

In a recent sequencing analysis of oligodendrogliomas, 1p19q codeletion and mutations of *CIC*, *FUBP1* and *IDH* frequently clustered as genetic molecular derangements [6–8].

CIC is a large gene of 20 exons located on chromosome 19q13.2 and encodes the mammalian homolog of the *Drosophila* transcriptional repressor Capicua. Capicua represses genes downstream of the RAS/MAPK signaling pathway and has been shown to be involved in the ErbB and Wnt oncogenic signaling cascades [7,9]. Yip *et al.* performed exome sequencing

on a set of 16 oligodendrogliomas with 1p19q codeletion and validated their results by deep sequencing 13 additional tumors. Somatic mutations and insertion/deletions in the *CIC* gene were detected in 20 out of 29 tumors (69%) [7]. These mutations were mainly clustered in exon 5 and 20 of the gene and were highly associated with 1p19q codeletion and *IDH1/2* mutations [7]. In addition, the authors studied the prognostic significance of these mutations but could not detect any significant correlation with recurrence, progression-free survival (PFS) or OS [7]. Bettegowda *et al.* also reported similar findings in oligodendrogliomas. In this series, *CIC* mutations were only found in cases with 19q loss (18 out of 27 cases; 67%) [6].

The *FUBP1* gene, located on the chromosome region 1p31.1, encodes FUBP1 [10,11]. FUBP1 binds specifically to single-stranded sequences of the human oncogene *c-MYC*, particularly the FUSE component of the gene and, by forming a complex with PUF60, represses the expression of *MYC* [6,10]. Therefore, loss-of-function mutations in *FUBP1* remove the negative effect of the *FUBP1*–*PUF60*–*FUSE* complex and, ultimately, lead to *MYC* activation [6]. Mutations of *FUBP1* have been reported in 15% of cases with oligodendrogliomas and are mainly frameshift and nonsense mutations clustering in the *FUBP1* DNA binding site spanning exons 5–14 [6,8]. At present, the clinical significance of these mutations is unclear.

Furthermore, 1p19q codeletion appears to be a component of a cluster of molecular genetic alterations that portends improved prognosis. Two studies have shown that the proneural gene expression signature, as defined by The Cancer Genome Atlas Project, is enriched in oligodendrogliomas, particularly those with the 1p19q codeletion [12,13]. This signature, which accounts for 74% of tumors, has also been associated with longer survival [13].

In addition to displaying the proneural signature, 1p19q codeleted oligodendrogliomas are associated with a constellation of positive prognostic markers, including methylation of the *MGMT* promoter, *IDH1* mutations and CIMP*.

In summary, the chromosome 1p19q codeletion has been established as a predictive marker of response to treatment with PCV chemotherapy in addition to RT, and patients with anaplastic oligodendrogliomas harboring 1p19q allelic loss should receive both treatment modalities.

Figure 1. MRI scan in a patient with 1p19q codeleted anaplastic oligodendroglioma. Images from (A) before and (B) after nine cycles of treatment with temozolomide, demonstrating improvement of contrast enhancement. The patient received 12 cycles of temozolomide and the response was durable for 13 months.

***MGMT* promoter hypermethylation**

Epigenetic silencing of the *MGMT* promoter by hypermethylation has been associated with a more favorable prognosis and response to alkylating agents in glioblastoma [14]. Subsequent studies in oligodendroglioma have indicated that *MGMT* promoter hypermethylation, occurring in up to 71% of cases, may also have a prognostic impact in this population [15,16].

A *post hoc* analysis of the EORTC 26951 trial revealed that *MGMT* promoter hypermethylation was correlated with a significantly superior PFS (PCV plus RT arm: 55.6 vs 9.8 months; RT alone arm: 15.2 vs 7.1 months) and OS (PCV plus RT arm: 70.9 vs 16.3 months; RT alone arm: 43.3 vs 15.6 months) in both treatment arms [1,15]. The prognostic relevance of *MGMT* promoter methylation in anaplastic gliomas was also shown in data from the NOA-04 Phase III trial. In this trial, patients with WHO grade III gliomas (anaplastic oligodendrogliomas, anaplastic oligoastrocytomas and anaplastic astrocytomas) were randomly

assigned to receive RT (total dose of 60 Gy in 1.8–2.0-Gy fractions), PCV chemotherapy or temozolomide. Similarly, *MGMT* promoter methylation was associated with significantly better time to treatment failure and PFS in all treatment arms [16].

However, none of these studies supported the notion that *MGMT* promoter methylation is predictive for the response to alkylating chemotherapy [15,16]. Of note, *MGMT* promoter hypermethylation was associated with 1p19q loss in anaplastic oligodendrogliomas [16].

A subsequent report using samples from EORTC 26951 and the Erasmus Medical Centre (Rotterdam, The Netherlands) revealed that *MGMT* promoter hypermethylation is part of more general genome-wide hypermethylation profile that confers a favorable prognosis [17]. In this study, two prognostically different molecular subgroups of anaplastic oligodendrogliomas were identified, as defined by either predominantly hypermethylated or unmethylated CpG islands. Survival of the

subgroup with CpG island hypermethylation phenotype (CIMP⁺) tumors was significantly longer than the unmethylated (CIMP⁻) subgroup (5.62 vs 1.24 years). There was also a strong association between CIMP status and *MGMT* promoter hypermethylation ($p = 0.61$; $p < 0.001$), which led the authors to hypothesize that *MGMT* promoter hypermethylation may well be an epiphenomenon of genome-wide hypermethylation that signifies favorable prognosis [17].

Based on current evidence, *MGMT* promoter hypermethylation should not guide the treatment strategy for patients with anaplastic oligodendroglial tumors, although it may provide valuable prognostic information.

IDH1 & IDH2 mutations

IDH1 and IDH2 are the cytoplasmic and mitochondrial NADP-dependent isocitrate dehydrogenases, respectively [18]. Mutations of *IDH1* and *IDH2* occur in more than 70% of oligodendrogliomas, and mainly affect amino acid 132 of *IDH1* (*IDH1*^{R132MUT}) and the analogous amino acid of *IDH2* (*IDH2*^{R172MUT}) [16,19–21].

IDH1 appears to function as a tumor suppressor, and mutational inactivation leads to tumorigenesis, in part through the induction of the HIF-1 pathway [21,22]. Mutations of *IDH1* are considered to be very early genetic events that occur before *TP53* mutations or loss of 1p19q [21]. These mutations frequently appear in oligodendroglial tumors, occurring in up to 80% of low-grade oligodendrogliomas and 86% of anaplastic oligodendrogliomas [16,19,21]. Mutations in the *IDH2* gene are of lower frequency and are generally nonoverlapping with tumors containing *IDH1* mutations [16,19].

Exactly how mutant *IDH* promotes tumorigenesis is unknown. Mutations of *IDH* have been shown to increase the intracellular concentration of 2-hydroxyglutarate, a potential oncometabolite [20]. Recently, *IDH1* somatic mutations have been found to be very tightly associated with the glioma CpG island hypermethylation phenotype (G-CIMP⁺) across all glioma tumors [23–25]. In a study by Turcan *et al.*, 98% of the G-CIMP⁺ WHO grade II and III gliomas possessed either an *IDH1* or *IDH2* mutation, and were associated with markedly better clinical end points [25]. These authors demonstrated a causal relationship between *IDH* mutations and the G-CIMP⁺ phenotype and postulated that *IDH* mutations

lead to G-CIMP⁺ by modulating patterns of methylation on a genome-wide scale [25]. One of the suggested mechanisms of DNA hypermethylation in *IDH1* or *IDH2* tumors occurs by disrupting the function of the TET family of 5-methylcytosine hydroxylases [26,27]. TET hydroxylases are α -ketoglutarate (α -KG)-dependent enzymes with a proposed role in DNA demethylation [26]. Mutations of *IDH1* or *IDH2* cause both a reduction in α -KG and an accumulation of 2-hydroxyglutarate, the latter of which acts as an antagonist of α -KG *in vitro* [26,27]. These changes impair TET function, and lead to aberrant DNA hypermethylation and generation of the CIMP⁺ phenotype [26,27].

The favorable prognostic impact of *IDH* mutations has been reported in several clinical trials and case series [1,16,19]. In the NOA-04 trial, *IDH1* mutations were positively associated with a longer PFS irrespective of treatment arm, histology, 1p19q status or *MGMT* promoter hypermethylation. In fact, in the multivariate model, *IDH1* mutation reduced the risk of progression (HR: 0.48; 95% CI: 0.29–0.77; $p = 0.0021$) more than 1p19q codeletion (HR: 0.47; 95% CI: 0.3–0.83; $p = 0.0092$), *MGMT* promoter hypermethylation (HR: 0.59; 95% CI: 0.37–1.1; $p = 0.0216$) or histology (HR: 0.63; 95% CI: 0.38–1.0; $p = 0.0425$) [16]. In the multivariate model of EORTC 26951, *IDH* mutation was an independent factor with a significant OS HR reduction (HR: 0.356) [1].

Given the available evidence, patients with tumors harboring *IDH1* or *IDH2* mutations have a more favorable prognosis. However, the treatment strategy should not be changed based on *IDH1* and *IDH2* status.

Chromosome 10q loss

Allelic loss of 10q occurs in 10–20% of oligodendrogliomas with a higher frequency reported in anaplastic oligodendrogliomas [28,29].

In two case series, loss of the 10q chromosome has been correlated with a significantly shorter PFS and OS [28,30]. In a report of 130 patients with oligodendroglial tumors, 10q loss conferred a much shorter PFS (7.9 vs 40.5 months; $p < 0.0001$) and lower 3-year survival rate (3 vs 86.5%; $p < 0.0001$) irrespective of 1p19q status [28]. Similar results were reported in a study by Horbinski *et al.* where the PFS and OS of patients with anaplastic oligodendrogliomas were significantly shorter if 10q was deleted [30]. However, 10q status was not associated with

survival of patients enrolled in the RTOG 9402 trial [29].

Therefore, the prognostic value of 10q loss and its impact on clinical decisions in the management of patients with oligodendroglial tumors remains unclear.

Deletion of the CDKN2A gene

Although chromosome 9p loss and homozygous deletion of the CDKN2A locus are rare findings in low-grade oligodendrogliomas, 42% of the anaplastic oligodendrogliomas have 9p loss, homozygous deletion of the CDKN2A gene or both [31]. Indeed, these alterations have been implicated in the progression of WHO grade II gliomas to anaplastic oligodendrogliomas [31].

Furthermore, single-nucleotide polymorphism array analysis of anaplastic oligodendrogliomas has shown that chromosome 9p and the CDKN2A locus are the most frequently altered genomic regions in the 1p19q codeleted tumors, either through genomic loss (32.4%) or copy-neutral losses of heterozygosity (13.2%). Recurring copy-neutral losses of heterozygosity in the CDKN2A locus has been suggested as one of the mechanisms of CDKN2A silencing and expression regulation [32].

The clinical significance of chromosome 9p loss and homozygous deletion of the CDKN2A locus is a focus of ongoing investigation. Cairncross et al., by analyzing the tumors from RTOG 9402, demonstrated that anaplastic oligodendrogliomas with CDKN2A deletion respond poorly to PCV chemotherapy, with a median survival time of less than 2 years. Moreover, anaplastic oligodendrogliomas that progress via the CDKN2A gene deletion pathway appear to represent a more resistant and aggressive subset of tumors [29].

In summary, CDKN2A deletion has been associated with a poor response to PCV chemotherapy.

However, further validation is required prior to its use in clinical decision-making.

Intrinsic glioma subtypes

Expression profiling has provided an objective method to classify tumors based on unique molecular signatures. Intrinsic (unsupervised) glioma subtypes (IGSs) are molecularly similar tumors that can be identified based on gene expression analysis [33–35]. Six IGSs have been established: IGS-9, -16, -17, -18, -22 and -23. Distinct genetic alterations have been associated with different IGSs. For example, IDH1 mutations are significantly more prevalent in IGS-9 and -17 subtypes [34]. Gravendeel et al. have demonstrated that intrinsic subtyping of gliomas is a better predictor of survival than histologic characterization [34].

Erdem-Eraslan et al. have examined the prognostic relevance of the IGS, in a subset of patients enrolled in EORTC 26951 [33]. One hundred and forty samples were assigned to one of the six IGSs previously defined by Gravendeel et al. [33,34]. Interestingly, IGSs were highly prognostic for both OS and PFS. The median OS times for IGS-9, -17, -18 and -23 were 8.5, 2.8, 1.2 and 1.0 years, respectively, and the median PFS times were 5.7, 1.8, 0.5 and 0.5 years, respectively. Additionally, this study demonstrated that the IGS-9 subtype derived significant PFS and OS benefit from the addition of PCV chemotherapy to RT, while IGS-18 or -23 subtypes had no appreciable OS benefit [33]. In this analysis, the IGS-9 subtype was associated with more frequent 1p19q codeletion, IDH1 mutations and MGMT hypermethylation. However, 1p19q codeletion was only found in 63% of IGS-9 tumors and only 70% had IDH1 mutations. These findings suggest that IGSs may provide stronger predictive information than that is provided by 1p19q codeletion and MGMT hypermethylation status [5,33].

Table 2. Molecular alterations in anaplastic oligodendroglioma, and their prognostic and predictive value.

Molecular alteration	Prognostic value	Predictive value	Ref.
Chromosome 1p19q codeletion	Association with favorable prognosis	Association with response to PCV chemotherapy	[1,2]
MGMT promoter methylation	Association with favorable prognosis	–	[1,15]
IDH1 and IDH2 mutations	Association with favorable prognosis	–	[1,16,19]
Chromosome 10q loss	Its prognostic value remains unclear. Association with poor prognosis has been reported	–	[28–30]
Deletion of the CDKN2A gene	–	Association with a poor response to PCV chemotherapy	[29]

PCV: Procarbazine, lomustine and vincristine.

Conclusion & future perspective

At present, 1p19q codeletion status is critical for the management of patients with oligodendroglioma, and data on *IDH* mutations, *MGMT* promoter hypermethylation and chromosome 10q loss may provide treating physicians with additional information on prognosis (Table 2). Furthermore, next-generation sequencing technologies have significantly expanded our knowledge of oligodendroglial tumors and led to the identification of new genetic alterations, such as *CIC* and *FUBP1* mutations.

However, the utility of these markers in planning treatment strategies and designing clinical trials remains to be fully established. Emerging evidence suggests that more comprehensive analyses, such as intrinsic glioma subtyping or G-CIMP status, are superior to single molecular markers. However, this highly specialized technology requires further validation in clinical trials, and the complexity of data

analysis, cost of the procedure and tissue availability remain limiting factors that need to be addressed before intrinsic glioma subtyping can become a widespread test. In summary, emerging data provide hope that comprehensive characterization of oligodendroglial tumors through broader omics-based molecular evaluations will improve outcome prediction and identification of patients who will benefit from specific treatment strategies.

Financial & competing interests disclosure

The authors have no relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending, or royalties.

No writing assistance was utilized in the production of this manuscript.

References

Papers of special note have been highlighted as:

- of interest
  - of considerable interest
- 1 van den Bent MJ, Brandes AA, Taphoorn MJ *et al.* Adjuvant procarbazine, lomustine, and vincristine chemotherapy in newly diagnosed anaplastic oligodendroglioma: long-term follow-up of EORTC brain tumor group study 26951. *J. Clin. Oncol.* 31(3), 344–350 (2013).
  - Provides long-term follow-up data on EORTC 26951 and concludes that the addition of six cycles of procarbazine, lomustine and vincristine (PCV) after 59.4 Gy of radiation therapy (RT) increases both overall survival (OS) and progression-free survival in anaplastic oligodendroglial tumors, and that 1p19q codeleted tumors derive more benefit than non-codeleted tumors.
  - 2 Cairncross G, Wang M, Shaw E *et al.* Phase III trial of chemoradiotherapy for anaplastic oligodendroglioma: long-term results of RTOG 9402. *J. Clin. Oncol.* 31(3), 337–343 (2013).
  - Provides long-term follow-up data on RTOG 9402 and concludes that PCV plus RT significantly increases OS in the subset of patients with codeleted 1p19q anaplastic oligodendroglial tumors.
  - 3 Okamoto Y, Di Patre PL, Burkhard C *et al.* Population-based study on incidence, survival rates, and genetic alterations of low-grade diffuse astrocytomas and oligodendrogliomas. *Acta Neuropathol.* 108(1), 49–56 (2004).
  - 4 van den Bent MJ, Carpentier AF *et al.* Adjuvant procarbazine, lomustine, and vincristine improves progression-free survival but not overall survival in newly diagnosed anaplastic oligodendrogliomas and oligoastrocytomas: a randomized European Organisation for Research and Treatment of Cancer Phase III trial. *J. Clin. Oncol.* 24(18), 2715–2722 (2006).
  - 5 de Groot JF. Prediction of response to chemotherapy in anaplastic glioma: how many markers does it take? *J. Clin. Oncol.* 31(3), 297–298 (2013).
  - 6 Bettgowda C, Agrawal N, Jiao Y *et al.* Mutations in *CIC* and *FUBP1* contribute to human oligodendroglioma. *Science* 333(6048), 1453–1455 (2011).
  - 7 Yip S, Butterfield YS, Morozova O *et al.* Concurrent *CIC* mutations, *IDH* mutations, and 1p19q loss distinguish oligodendrogliomas from other cancers. *J. Pathol.* 226(1), 7–16 (2012).
  - *CIC* mutations were shown to be highly associated with 1p19q codeletion and *IDH1/2* mutations, suggesting a functional interaction between *CIC* mutations, *IDH1/2* mutations and 1p19q codeletion.
  - 8 Sahm F, Koelsche C, Meyer J *et al.* *CIC* and *FUBP1* mutations in oligodendrogliomas, oligoastrocytomas and astrocytomas. *Acta Neuropathol.* 123(6), 853–860 (2012).
  - 9 Lee CJ, Chan WI, Scotting PJ. *CIC*, a gene involved in cerebellar development and ErbB signaling, is significantly expressed in medulloblastomas. *J. Neurooncol.* 73(2), 101–108 (2005).
  - 10 Duncan R, Bazar L, Michelotti G *et al.* A sequence-specific, single-strand binding protein activates the far upstream element of c-myc and defines a new DNA-binding motif. *Genes Dev.* 8(4), 465–480 (1994).
  - 11 Hsiao HH, Nath A, Lin CY, Foltz-Stogniew EJ, Rhoades E, Braddock DT. Quantitative characterization of the interactions among c-myc transcriptional regulators FUSE, FBP, and FIR. *Biochemistry* 49(22), 4620–4634 (2010).
  - 12 Ducray F, Idhah A, de Reynies A *et al.* Anaplastic oligodendrogliomas with 1p19q codeletion have a proneural gene expression profile. *Mol. Cancer* 7, 41 (2008).
  - 13 Cooper LA, Gutman DA, Long Q *et al.* The proneural molecular signature is enriched in oligodendrogliomas and predicts improved survival among diffuse gliomas. *PLoS ONE* 5(9), e12548 (2010).
  - 14 Hegi ME, Diserens AC, Gorlia T *et al.* *MGMT* gene silencing and benefit from temozolomide in glioblastoma. *N. Engl. J. Med.* 352(10), 997–1003 (2005).
  - 15 van den Bent MJ, Dubbink HJ, Sanson M *et al.* *MGMT* promoter methylation is

- prognostic but not predictive for outcome to adjuvant PCV chemotherapy in anaplastic oligodendroglial tumors: a report from EORTC Brain Tumor Group Study 26951. *J. Clin. Oncol.* 27(35), 5881–5886 (2009).
- Provides information on the clinical significance of *MGMT* promoter hypermethylation. Analysis of the data from the EORTC 26951 trial showed that *MGMT* promoter hypermethylation was correlated with a significantly superior progression-free survival and OS in both PCV/RT and RT-only arms.
- 16 Wick W, Hartmann C, Engel C *et al.* NOA-04 randomized Phase III trial of sequential radiochemotherapy of anaplastic glioma with procarbazine, lomustine, and vincristine or temozolomide. *J. Clin. Oncol.* 27(35), 5874–5880 (2009).
  - 17 van den Bent MJ, Gravendeel LA, Gorlia T *et al.* A hypermethylated phenotype is a better predictor of survival than *MGMT* methylation in anaplastic oligodendroglial brain tumors: a report from EORTC 26951. *Clin Cancer Res.* 17(22), 7148–7155 (2011).
  - Demonstrates that there is a strong association between glioma CpG island hypermethylation phenotype status and *MGMT* promoter methylation, suggesting that *MGMT* promoter methylation may be part of a genome-wide methylation profile that confers favorable prognosis.
  - 18 Reitman ZJ, Jin G, Karoly ED *et al.* Profiling the effects of isocitrate dehydrogenase 1 and 2 mutations on the cellular metabolome. *Proc. Natl Acad. Sci. USA* 108(8), 3270–3275 (2011).
  - 19 Yan H, Parsons DW, Jin G *et al.* *IDH1* and *IDH2* mutations in gliomas. *N. Engl. J. Med.* 360(8), 765–773 (2009).
  - Demonstrates that mutations of *IDH1* and *IDH2* occur in more than 70% of oligodendrogliomas and confer favorable prognosis. They mainly affect the amino acid 132 of *IDH1* (*IDH1*^{R132MUT}) and analogous amino acid of the *IDH2* gene (*IDH2*^{R172MUT}).
  - 20 Lu C, Ward PS, Kapoor GS *et al.* *IDH* mutation impairs histone demethylation and results in a block to cell differentiation. *Nature* 483(7390), 474–478 (2012).
  - 21 Watanabe T, Nobusawa S, Kleihues P, Ohgaki H. *IDH1* mutations are early events in the development of astrocytomas and oligodendrogliomas. *Am. J. Pathol.* 174(4), 1149–1153 (2009).
  - 22 Zhao S, Lin Y, Xu W *et al.* Glioma-derived mutations in *IDH1* dominantly inhibit *IDH1* catalytic activity and induce HIF-1 α . *Science* 324(5924), 261–265 (2009).
  - 23 Nounshmehr H, Weisenberger DJ, Diefes K *et al.* Identification of a CpG island methylator phenotype that defines a distinct subgroup of glioma. *Cancer Cell* 17(5), 510–522 (2010).
  - 24 Laffaire J, Everhard S, Idbaih A *et al.* Methylation profiling identifies 2 groups of gliomas according to their tumorigenesis. *Neuro Oncol.* 13(1), 84–98 (2011).
  - 25 Turcan S, Rohle D, Goenka A *et al.* *IDH1* mutation is sufficient to establish the glioma hypermethylator phenotype. *Nature* 483(7390), 479–483 (2012).
  - 26 Figueroa ME, Abdel-Wahab O, Lu C *et al.* Leukemic *IDH1* and *IDH2* mutations result in a hypermethylation phenotype, disrupt TET2 function, and impair hematopoietic differentiation. *Cancer Cell* 18(6), 553–567 (2010).
  - 27 Xu W, Yang H, Liu Y *et al.* Oncometabolite 2-hydroxyglutarate is a competitive inhibitor of α -ketoglutarate-dependent dioxygenases. *Cancer Cell* 19(1), 17–30 (2011).
  - 28 Ramirez C, Bowman C, Maurage CA *et al.* Loss of 1p, 19q, and 10q heterozygosity prospectively predicts prognosis of oligodendroglial tumors – towards individualized tumor treatment? *Neuro Oncol.* 12(5), 490–499 (2010).
  - 29 Cairncross JG, Ueki K, Zlatescu MC *et al.* Specific genetic predictors of chemotherapeutic response and survival in patients with anaplastic oligodendrogliomas. *J. Natl Cancer Inst.* 90(19), 1473–1479 (1998).
  - 30 Horbinski C, Nikiforova MN, Hobbs J *et al.* The importance of 10q status in an outcomes-based comparison between 1p/19q fluorescence *in situ* hybridization and polymerase chain reaction-based microsatellite loss of heterozygosity analysis of oligodendrogliomas. *J. Neuropathol. Exp. Neurol.* 71(1), 73–82 (2012).
  - 31 Bigner SH, Matthews MR, Rasheed BK *et al.* Molecular genetic aspects of oligodendrogliomas including analysis by comparative genomic hybridization. *Am. J. Pathol.* 155(2), 375–386 (1999).
  - 32 Idbaih A, Ducray F, Dehais C *et al.* SNP array analysis reveals novel genomic abnormalities including copy neutral loss of heterozygosity in anaplastic oligodendrogliomas. *PLoS ONE* 7(10), e45950 (2012).
  - 33 Erdem-Eraslan L, Gravendeel LA, de Rooij J *et al.* Intrinsic molecular subtypes of glioma are prognostic and predict benefit from adjuvant procarbazine, lomustine, and vincristine chemotherapy in combination with other prognostic factors in anaplastic oligodendroglial brain tumors: a report from EORTC 26951. *J. Clin. Oncol.* 31(3), 328–336 (2013).
  - Provides information on the prognostic relevance of intrinsic (unsupervised) glioma subtyping in anaplastic oligodendroglial tumors and shows that the intrinsic (unsupervised) glioma subtype is highly prognostic for OS in patients who were treated with radiation and PCV.
  - 34 Gravendeel LA, Kouwenhoven MC, Gevaert O *et al.* Intrinsic gene expression profiles of gliomas are a better predictor of survival than histology. *Cancer Res.* 69(23), 9065–9072 (2009).
  - 35 French PJ, Swagemakers SM, Nagel JH *et al.* Gene expression profiles associated with treatment response in oligodendrogliomas. *Cancer Res.* 65(24), 11335–11344 (2005).
  - Websites
  - 101 Central Brain Tumor Registry of the United States. www.cbtrus.org
  - 102 Radiation therapy or radiation therapy and temozolomide or temozolomide alone in treating patients with newly diagnosed anaplastic glioma. www.clinicaltrials.gov/ct2/show/NCT00887146?term=NCT00887146&rank=1
  - 103 Radiation therapy with or without temozolomide in treating patients with anaplastic glioma. www.clinicaltrials.gov/ct2/show/NCT00626990?term=NCT00626990&rank=1