Introduction

Glioblastoma (GBM) is the most frequent and aggressive malignant primary brain tumor with only about 12% of patients surviving beyond 36 months (long-term survivors).[1,2] According to the latest Central Brain Tumor Registry of the USA statistical report, the age-adjusted incidence rate for GBM is 3.19/100,000. The incidence of GBM increases with age and peaks at 75–84 years (14.93/100,000), being more common in males (3.97/100,000).[3]

The current treatment strategy for GBM patients combines maximal surgical resection, followed by radiotherapy (RT) with concomitant and adjuvant temozolomide (TMZ).[4,5] Complete surgical resection is virtually impossible due to the infiltrative nature of these tumors, yet gross total resection is still a positive prognostic marker. Concurrent adjuvant RT in combination with TMZ represents the standard of care for patients with newly diagnosed GBM, but still <5% of patients survive for longer than 5 years after diagnosis.[6-8]

Decades of molecular studies have identified key genetic abnormalities in human GBMs, including the following: (1) dysregulation of growth factor signaling pathways via amplification and mutational activation of receptor tyrosine kinase (RTK) genes; (2) activation of the phosphatidylinositol-3-OH kinase (PI3K) pathway; and (3) inactivation of the p53 and retinoblastoma tumor suppressor.
The former corresponds to de novo epigenetic alterations in both DNA and histones, altering phosphate-dependent manner. These homologous enzymes decarboxylate KG into O(6)-ketoglutarate (KG), and this “neomorphic” mutation renders the IDH enzyme to reduce αKG into 2-hydroxyglutarate in the nicotinamide adenine dinucleotide phosphate-dependent manner. Mutant IDH1 or IDH2 are correlated with increased histone methylation, causing epigenetic alterations in both DNA and histones, altering gene expression and promoting oncogenic transformation.

Nowadays, mutations in IDH1 are commonly established as a hallmark molecular feature of secondary GBM (~70% of secondary GBM, compared with 5–20% in primary GBM) who have predominant localization of GBM in the frontal and temporal lobes. Since primary GBM is a clinically defined entity and the presence of IDH1/2 mutations has been shown to be inversely related to or even mutually exclusive of epidermal growth factor receptor (EGFR) and phosphatase and tensin homolog (PTEN) abnormalities, which are hallmarks of primary GBM, IDH-mutated GBM lesions may represent genetically “secondary” GBM tumors. Moreover, the IDH mutation status is stable during the progression of lower-grade gliomas to secondary GBMs. Mutations in the IDH genes are thought to cause glioma-CpG island methylator phenotype (G-CIMP) within the proneural GBM subgroup. IDH mutations seem to require cooperating mutations in TP53 and ATRX, and they are less frequently detected in primary GBMs.

**Morphological Diagnosis**

Malignant gliomas are histologically heterogeneous and invasive tumors that are derived from glia. The World Health Organization (WHO) classification system groups gliomas into 4 histological grades defined by increasing degrees of undifferentiation, anaplasia, and aggressiveness. Malignant gliomas, the most common form of gliomas, consist of WHO grade IV tumors (GBM) and grade III tumors (anaplastic astrocytoma, oligodendroglioma, and oligoastrocytoma). GBMs account for approximately 60–70% of malignant gliomas and is characterized histologically by considerable cellularity and mitotic activity, microvascular proliferation, necrosis and they are also recalcitrant to radio/chemotherapy. Primary (de novo, approximately 95% of cases) GBMs manifest rapidly, without evident of less malignant precursor lesions, after a short clinical history. Secondary GBMs (approximately 5% of cases) develop more slowly by progression from low-grade diffuse astrocytoma and anaplastic astrocytoma. GBM and other malignant gliomas are highly invasive, infiltrating surrounding brain parenchyma, yet they are typically confined to the central nervous system (CNS) and do not metastasize. Unfortunately, WHO morphological classification is based on subjective criteria, lacks reproducibility, and remains imperfect in its ability to predict individual outcomes.

**Genetics Variation of Glioblastoma**

**Isocitrate dehydrogenase mutations**

The first genome-wide exon sequencing effort for glioma identified heterozygous hotspot mutations at codon 132 (most commonly R132H) in isocitrate dehydrogenase 1 (IDH1) in 12% of GBM. These mutations change the enzymatic activity of the cytoplasmic and peroxisomal IDH1. The same holds true for codon 172 mutations in the mitochondrial IDH2 gene. These homologous enzymes decarboxylate isocitrate to α-ketoglutarate (αKG), and this “neomorphic” mutation renders the IDH enzyme to reduce αKG into 2-hydroxyglutarate in the nicotinamide adenine dinucleotide phosphate-dependent manner. Mutant IDH1 or IDH2 are correlated with increased histone methylation, causing epigenetic alterations in both DNA and histones, altering gene expression and promoting oncogenic transformation.

**Telomerase reverse transcriptase promoter mutations**

Recently, novel somatic mutations in the promoter region of telomerase reverse transcriptase (TERT) have been identified in malignant melanomas, as well as being associated with increased telomerase expression and activity. The tumors derived from cell populations with low self-renewal capacity generally depend on alterations that keep telomerase activity, while epigenetic alteration maintains telomerase activity in tumor types arisen from self-renewing stem cells. The two most common mutations are located at C228T and C250T, with identical hotspots also found in gliomas. The highest incidence was identified among most tumors harboring 1p/19q co-deletion and IDH mutations (98%), as well as IDH wild-type (IDH wt) tumors with EGFR amplification (92%). The former corresponds to oligodendroglioma, while the latter corresponds to primary GBMs. The frequency of TERT mutations is relatively
low in diffuse and anaplastic astrocytomias (19% and 25%, respectively). In the study by Killela et al., patients with TERT promoter mutations alone (i.e., no IDH mutation) had the poorest overall survival (OS) (median 11.3 months), patients with tumors without TERT or IDH1/2 mutations had a slightly better survival (median 16.6 months), while patients with only IDH1 mutant GBM had the best survival (median 42.3 months). Although another study with 358 patients found no significant difference in OS between TERT mutant and TERT wild-type (IDH wt) GBM, the role of TERT promoter mutations may provide a tool to identify non-IDH mutant GBMs.

**Epidermal growth factor receptor aberrations**
The range of high-amplitude focal copy-number aberrations in adult GBM highlights a key role of EGFR amplifications (43% of cases) which co-occurred with EGFR intragenic deletions and/or point mutations. EGFR mutations were accompanied by regional DNA amplification in the majority of cases, leading to a wide range of mutation allelic frequencies. The prominent intragenic deletions in GBM target parts of the gene encoding either the extracellular domain of EGFR (exons 2–7, the deletion of which forms EGFR variant III) or the carboxyl terminus, and these deletions are always correlated with amplification and co-expression of the wild-type EGFR. EGFR was recently shown to be activated by recurrent translocations in 7% of GBM samples: It was most frequently fused in-frame to septin 14 or phosphoserine phosphatase as the 3’ gene segment. Overall, 57% of GBM showed evidence of mutation, rearrangement, altered splicing, and/or focal amplification of EGFR.

**PTEN alterations**
Loss of heterozygosity (LOH) at chromosome 10q23 occurs at high frequency in a variety of human tumors. LOH at 10q23 occurs in ~70% of GBMs. Mutations of PTEN were detected in 31–44% of GBM. PTEN is a negative regulator of the phosphoinositide 3-kinase pathway, a major signaling pathway that stimulates cellular proliferation in response to growth factor stimulation. PTEN deletions were more common in GBM, except classical grade II/III gliomas. PTEN deletions were fairly common across all gene expressions subtypes, but absent in IDH1 mutant tumors. PTEN loss and deletion were associated with incremental increases in AKT pathway activity. Several studies demonstrated that patients with loss of function mutations of PTEN generally had shorter survival than patients with PTEN retention. However, PTEN loss was not associated with worse OS in newly diagnosed GBM patients of the TMZ era.

**Other novel genetic aberrations**
In a smaller fraction of primary GBMs (about 3%), a fusion of the tyrosine kinase coding region of fibroblast growth factor receptor 1 (FGFR1) to the transforming acidic coiled-coil (TACC) coding domain of TACC1 (or fusion of FGFR3 to TACC3) results in constitutive kinase activity. In transcriptome profiling of 272 gliomas from CGGA, 67 in-frame fusion transcripts were identified, including three recurrent fusion transcripts: FGFR3-TACC3, RNF213-SLC26A11, and PTPRZ1-MET (fusion transcript involving the protein tyrosine phosphatase, receptor-type, Z polypeptide 1 gene and the MET proto-oncogene, ZM). ZM fusion was found in three of 20 (15%) specimens. Exogenous expression of the ZM fusion in the U87MG GBM line enhanced cell migration and invasion. Clinically, patients afflicted with ZM fusion harboring GBMs survived poorly relative to those afflicted with non-ZM-harboring. Therefore, recurrent fusion events that involve RTK-encoding genes might be a promising therapeutic target and provide a strong rationale for the inclusion of these patients in future stratified clinical trials using different RTK inhibitors. Table 1 summarizes all of the above described and other genetic alterations and related altered signaling pathways in primary versus secondary GBM.

**Molecular Classification**
The phenotype of a tumor is the result of the genotype and the influence of the tumor’s environment on the tumor. One would expect that molecular diagnostics will contribute to a better classification of brain tumors [Tables 2–4]. Phillips described three subclases of high-grade gliomas (termed proneural, mesenchymal, and proliferative) associated with different outcomes; specifically, prolonged survival of the proneural subclass. Similar classification of GBMs

| Genetic abnormalities | Frequency (%) | Major altered signaling pathways |
|-----------------------|---------------|----------------------------------|
| Secondary GBM         |               |                                  |
| IDH mutation          | 60–80         | Metabolism                       |
| ATRX mutation or loss | 57            | Genome integrity                 |
| TP53 mutation         | 65            | p53 pathway                      |
| RB1 loss              | 43            | Rb pathway                       |
| CDKN2A loss           | 19            | Rb pathway                       |
| PTEN loss             | 41            | PI3K signaling                   |
| PTPRZ1-MET fusion     | 15            | RTK signaling                    |
| Primary GBM           |               |                                  |
| TERT promoter mutation| 60–80         | Telomere maintenance             |
| NF1 loss              | 10–18         | MAPK signaling                   |
| PTEN loss             | 36–41         | PI3K signaling                   |
| PI3K mutation         | 15–25         | PI3K signaling                   |
| TP53 mutation         | 28–35         | p53 pathway                      |
| EGFR VIII             | 25–50         | RTK signaling                    |
| EGFR ampl.            | 36–60         | RTK signaling                    |
| PDGFRα ampl.          | 10–13         | RTK signaling                    |
| RB1 loss              | 14            | Rb pathway                       |
| CDKN2A loss           | 31–78         | Rb pathway                       |
| FGFR3-TACC3 fusion    | 23–60         | RTK signaling                    |
was also detected in a larger cohort of mixed gliomas.\[66\] In 2010, unsupervised clustering of gene expression data from adult GBM samples from the TCGA identified four different molecular subtypes: Proneural, neural, classical, and mesenchymal.\[41\] Proneural GBMs were subdivided into G-CIMP-positive and G-CIMP-negative GBM subsets on the basis of characteristic DNA methylation patterns that strongly correspond with IDH1 mutation status.\[27,67\] Another later study, which compared DNA methylation patterns across both pediatric and adult patients with GBM, found a similar clustering in tumors from adult patients and further identified three more distinct clusters that predominantly consisted of children and adolescents.\[68\] Recently, Liu et al. profiled the genetic features of multifocal GBM and found that M-GBMs had no IDH1, ATRX, or PDGFRA mutations, significantly associated with the mesenchymal subtype. They also identified the CYB5R2 gene to be hypomethylated and overexpressed in M-GBMs.\[69\]

The recent reports published on the Nature Genetics and NEJM were comprehensively analyzed by whole-exome sequencing and/or targeted deep sequencing as well as array comparative genomic hybridization. In the Nature Genetics article,\[70\] grade II and III gliomas were divided into and exhausted by the genetically well-defined type I–III subtypes. Type III tumors represented the IDH wild-type grade II and III tumors in the current cohort, showing an OS rate more similar to that of GBM. Similarly, the report\[71\] from TCGA research network independently identified similar groups, using unsupervised clustering analyses of DNA mutation, RNA expression, DNA copy number, and DNA methylation data. The integration of genome-wide data from multiple platforms delineated three molecular classes of lower-grade gliomas (grade II/III gliomas) that were more concordant with IDH, 1p/19q, and TP53 status than with histologic class. This multi-platform approach yielded three groups similar to those initially described by Jiao’s model.\[58\] The large majority of lower-grade gliomas without an IDH mutation had genomic aberrations and clinical behavior strikingly similar to those found in primary GBM.

The report\[72\] from Mayo Clinic and UCSF defined a priori groups that were based on the presence or absence of TERT promoter mutations, IDH mutations, and 1p/19q codeletion and found consistent associations between the molecular groups and age at diagnosis, survival, patterns of acquired alterations, and germline variants across the three data sets. The group with only TERT mutations has a high prevalence

### Table 2: Phillips classifications of GBM based on transcription profiling

| Classifications                  | Subgroups          | Mesenchymal |
|----------------------------------|--------------------|-------------|
| Patient age (years)              | Older (~50)        | Older (~50) |
| Biological process               | Proliferation      | Angiogenesis|
| Chromosome alterations            | Gain of 7 and loss of 10 or 10q | PTEN loss |
| EGFR/PTEN                        | EGFR normal/PTEN intact | PTEN loss |

EGFR: Epidermal growth factor receptor; PTEN: Phosphatase and tensin homolog; GBM: Glioblastoma.

### Table 3: TCGA classifications of GBM based on transcription and methylation profiling

| Classifications                  | Subgroups          |
|----------------------------------|--------------------|
| G-CIMP+                          | G-CIMP−            |
| Genetic alteration               | IDH/TP53/ATRX      | 4q ampl.    |
| Phenotype                        | Oligodendrogliotic | Neuron      |
| Prognosis                        | Best                | 7p ampl.    |
| Chemotherapy                     | Resistant           | Astrocytic  |

TCGA: The cancer genome atlas; GBM: Glioblastoma; G-CIMP: Glioma-CpG island methylator phenotype; ampl.: Amplification; IDH: Isocitrate dehydrogenase; NF1: Neurofibromatosis 1; RB1: Retinoblastoma 1.

### Table 4: DKFZ classifications of GBM based on methylation profiling

| Classifications | Subgroups |
|-----------------|-----------|
| IDH             | RTK I “PDGFRA” | RTK II “classic” |
| Median age (years) | 40       | 36         | 58         |
| Genetic alteration | IDH      | PDGFRA ampl. | EGFR ampl. |
| Tumor location   | Frontal and temporal | Hemispheric | Hemispheric |
| Prognosis        | Favorable | Poor       | Poor       |

DKFZ: Deutsches Krebsforschungszentrum (German Cancer Research Center); GBM: Glioblastoma; RTK: Receptor tyrosine kinase; PDGFRA: Platelet-derived growth factor receptor alpha; EGFR: Epidermal growth factor receptor; IDH: Isocitrate dehydrogenase; Ampl.: Amplification.
of loss of chromosome 4 and acquired PIK3CA or PIK3R1 mutations. Gliomas with only TERT mutations are primarily grade IV gliomas. These tests (for IDH mutations, 1p/19q codeletion, and TERT promoter alterations) can be used to define five principal groups of gliomas with characteristic distributions of age at diagnosis, clinical behavior, acquired genetic alterations, and associated germline variants.

Application of Genetics Study in Clinical Practice

Over the past decade, insights into the molecular pathology of gliomas have significantly improved both our biological understanding of neoplasms as well as our abilities to diagnose tumors and estimate their prognosis and likelihood of response to specific therapies. To discuss the inclusion of molecular information into the next WHO classification of CNS tumors, a meeting under the sponsorship of the International Society of Neuropathology (ISN) has been held in Haarlem, the Netherlands. According to the ISN-Haarlem consensus, “integrated” diagnosis was established through the usage of ATRX, IDH1-R132H IHC, 1p/19q analyses, and IDH sequencing in the diagnosis of diffuse gliomas.

RT plus concomitant and adjuvant TMZ chemotherapy is the current standard of care for patients with GBM. Several clinical trials have displayed that MGMT promoter methylation correlated with prolonged progression-free and OS in patients with GBM receiving alkylating drug chemotherapy. In 2012, two independent randomized trials in elderly patients with GBM assessed RT alone versus TMZ chemotherapy alone as an initial treatment. Subgroup analyses of both trials showed better outcome for chemotherapy in patients with MGMT promoter methylated tumors but reduced survival in patients with unmethylated tumors. Recently, the CGGA project delineated that patients with IDH wild-type GBM who underwent RT + TMZ exhibited significantly longer survival times compared to the patients who were assigned to the RT alone treatment. However, among patients with IDH mutation tumors, the survival pattern of patients undergoing RT + TMZ or RT was comparable. These results strongly suggest that treatment strategies for elderly patients with GBM should be individualized dependent on IDH and MGMT.

In addition, due to the high heterogeneity of GBM, each of which may respond differently to one targeted therapy, there has been considerable interest in identifying molecular markers that predict response to a specific molecular targeted therapy. Bevacizumab, a monoclonal antibody against vascular endothelial growth factor, being granted approval by the US Food and Drug Administration for treating recurrent GBM in 2009. However, it does not benefit OS in either recurrent GBM or newly diagnosed GBM. The presence of EGFR overexpression and EGFR mutations in GBM could predict the activity of EGFR-targeted drugs in patients with these aberrations. However, these potential treatment approaches still have not been clear with contradictory findings in previous clinical trials.

It was demonstrated that a point mutation in IDH1R132H, expressed in gliomas and other tumors, is presented on human major histocompatibility complex (MHC) class II and induces a mutation-specific CD4+ antitumor T-cell response in patients and a syngeneic tumor model in MHC-humanized mice. Conceptually, patients with low-grade and anaplastic gliomas, secondary GBM with a high prevalence of the IDH1 (R132H) mutation represent a patient population that may particularly benefit from an IDH1R132H specific tumor vaccine.

Conclusions

These recurrent genetic aberrations occur in a specific context of cellular origin, co-oncogenic hits and are present in distinct patient populations. Primary and secondary GBMs are distinct disease entities that affect different age groups of patients and develop through distinct genetic aberrations. These differences are important, especially because they may affect sensitivity to radio- and chemo-therapy and should thus be considered in the identification of targets for novel therapeutic approaches. The biological distinction of GBM subgroups should therefore guide the design of future clinical trials.

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Conflicts of interest

There are no conflicts of interest.

References

1. Sturm D, Bender S, Jones DT, Lichter P, Grill J, Becher O, et al. Paediatric and adult glioblastoma: Multiform (epi) genomic culprits emerge. Nat Rev Cancer 2014;14:92-107. doi: 10.1038/nrc3655.
2. Seyfried TN, Flores R, Poff AM, D'Agostino DP, Mukherjee P. Metabolic therapy: A new paradigm for managing malignant brain cancer. Cancer Lett 2015;356(2 Pt A):289-300. doi: 10.1016/j.canlet.2014.07.015.
3. Ostrom QT, Gittleman H, Farah P, Ondracek A, Chen Y, Wolinsky Y, et al. CBTRUS statistical report: Primary brain and central nervous system tumors diagnosed in the United States in 2006-2010. Neuro Oncol 2013;15 Suppl 2:ii1-56. doi: 10.1093/neuonc/not151.
4. Thomas AA, Brennan CW, DeAngelis LM, Omuro AM. Emerging therapies for glioblastoma. JAMA Neurol 2014;71:1437-44. doi: 10.1001/jamaneurol.2014.1701.
5. Omuro A, DeAngelis LM. Glioblastoma and other malignant gliomas: A clinical review. JAMA 2013;310:1842-50. doi: 10.1001/jama.2013.280319.
6. Stupp R, Mason WP, van den Bent MJ, Weller M, Fisher B, Taphoorn MJ, et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N Engl J Med 2005;352:987-96. doi: 10.1056/NEJMoa043330.
7. Stupp R, Hegi ME, Mason WP, van den Bent MJ, Taphoorn MJ, Janzer RC, et al. 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;10:459-66. doi: 10.1016/S1470-2045(09)70025-7.
Specific chromosomal abnormalities in Malignant astrocytic glioma: Genetics, biology, and paths to treatment. Genes Dev 2007;21:2683‑710. doi: 10.1101/gad.1596707.

Ohiaki H, Dessen P, Jourde P, Horstmann S, Nishikawa T, Di Patre PL, et al. Genetic pathways to glioblastoma: A population‑based study. Cancer Res 2004;64:6892‑9. doi: 10.1158/0008‑5472.CAN‑04‑1337.

Nonoguchi N, Ohta T, Oh JE, Kim YH, Kleihues P, Ohgaki H. TERT promoter mutations in primary and secondary glioblastomas. Acta Neuropathol 2013;126:931‑7. doi: 10.1007/s00401‑013‑1163‑0.

Louis DN. Molecular pathology of malignant gliomas. Annu Rev Pathol 2006;1:97‑117. doi: 10.1146/annurev.pathol.1.110304.100403.

Ricard D, Idiahi A, Ducray F, Lahutte M, Hoang‑Xuan K, Delattre JY. Primary brain tumours in adults. Lancet 2012;379:1984‑96. doi: 10.1016/s0140‑6736(11)61346‑9.

van den Bent MJ. Interobserver variation of the histopathological diagnosis in clinical trials on glioma: A clinician’s perspective. Acta Neuropathol 2010;120:297‑304. doi: 10.1007/s00401‑010‑0725‑7.

Parsons DW, Jones S, Zhang X, Lin JC, Leary RJ, Angenendt P, et al. Tumor‑speciﬁc mutations identiﬁed by genome‑wide analysis of DNA copy number and DNA methylation. Science 2010;321:1637‑40. doi: 10.1126/science.1164328.

Cairns RA, Harris IS, Mak TW. Regulation of cancer cell metabolism. Nat Rev Cancer 2011;11:85‑95. doi: 10.1038/nrc2981.

Horn S, Figl A, Rachakonda PS, Fischer C, Sucker A, Gast A, et al. TERT promoter mutations in familial and sporadic melanoma. Science 2013;339:959‑61. doi: 10.1126/science.1230062.

van den Bent MJ. Tumor heterogeneity is an active process maintained by a putative protein tyrosine phosphatase gene mutated in human brain, breast, and prostate cancer. Science 1997;275:1943‑7. doi: 10.1126/science.275.5308.1943.
et al.

Identification of a CpG island methylator

Loss of tumor suppressor PTEN function increases B7-H1

Gene expression profiling reveals molecularly and clinically

ATRX and IDH1-R132H immunohistochemistry with

Molecular subtypes of glioblastoma are relevant to lower
grade glioma. PLoS One 2014;9:e91216. doi: 10.1371/journal.

p0091216.

Smith JS, Tachibana I, Passe SM, Huntley BK, Borell TJ, Iurria N,
et al. PTEN mutation, EGFR amplification, and outcome in patients
with anaplastic astrocytoma and glioblastoma multiforme. J Natl
Cancer Inst 2001;93:1246-56. doi: 10.1093/jnci/93.16.1246.

Sano T, Lin H, Chen X, Langford LA, Koul D, Bonds ML,
et al. Differential expression of MMAC1/PTEN in glioblastoma
multiforme: Relationship to localization and prognosis. Cancer Res
1999;59:1820-4.

Choe G, Horvath S, Cloughesy TF, Crosby K, Seligson D, Palotie A,
et al. Analysis of the phosphatidylinositol 3'-kinase signaling pathway
in glioblastoma patients in vivo. Cancer Res 2003;63:2742-6.

Carico C, Núñez M, Mukherjee D, Elramsisy A, Dantis J, Hu J,
et al. Loss of PTEN is not associated with poor survival in newly
diagnosed glioblastoma patients of the temozolomide era. PLoS One
2012;7:e33684. doi: 10.1371/journal.pone.0033684.

Singh D, Chan JM, Zoppoli P, Niola F, Sullivan R, Castano A,
et al. Transforming fusions of FGFR and TACC genes in human
glioblastoma. Science 2012;337:1231-5. doi: 10.1126/science.

220834.

Parker BC, Annala MJ, Cordell DE, Granberg KJ, Sun Y, Ji P,
et al. The tumorgenic FGFR3-TACC3 gene fusion escapes miR-99a
regulation in glioblastoma. J Clin Invest 2013;123:855-65. doi:
10.1172/JCI67144.

Jiao Y, Killela PJ, Reitman ZJ, Rasheed AB, Hapchyn C, de Wilde RF,
et al. Frequent ATRX, CIC, FUBP1 and IDH1 mutations refine the
classification of malignant gliomas. Oncotarget 2012;3:709-22.
doi: 10.18632/oncotarget.588.

Crespo I, Vital AL, Gonzalez-Tablas M, Patino Medel C, Otero A,
Lopes MC, et al. Molecular and genomic alterations in glioblastoma
multiforme. Am J Pathol 2015;185:1820-33. doi: 10.1016/j.

ajpath.2015.02.023.

Bao ZS, Chen HM, Yang MY, Zhang CB, Yu K, Ye WL,
et al. RNA-seq of 272 gliomas revealed a novel, recurrent PTPRZ1-MET
effusion transcript in secondary glioblastomas. Genome Res
2014;24:1765-73. doi: 10.1101/gr.151266.113.

Weller M, Pfister SM, Wick W, Hegi ME, Reifenberger G, Stupp R,
Molecular neuro-oncology in clinical practice: A new horizon. Lancet
2015;386:1264-75. doi: 10.1016/S0140-6736(15)00556-7.

De Stefano AL, Fucetti A, Frattini V, Labussiere M, Mohkatur K,
Zoppoli P, et al. Detection, characterization, and inhibition of
FGFR-TACC fusions in IDH wild-type glioma. Clin Cancer Res
2015;21:3307-17. doi: 10.1158/1078-0432.CCR-14-2199.

Li X, Diehn M, Watson N, Bollen AW, Alapde KD, Nicholas KM,
et al. Gene expression profiling reveals molecularly and clinically
distinct subtypes of glioblastoma multiforme. Proc Natl Acad Sci U S
A 2005;102:5814-9. doi: 10.1073/pnas.0408270102.

Shi R, Shi T, Kim CJ, Horvath S, Liu Y, Cloughesy TF, et al.
Gene expression profiling identifies molecular subtypes of gliomas.
Oncogene 2003;22:4918-23. doi: 10.1038/sj.onc.1206753.

Phillips HS, Kharbanda S, Chen R, Forrest WF, Soriano RH,
Wu TD, et al. Molecular subclasses of high-grade glioma predict
prognosis, delineate a pattern of disease progression, and resemble
stages in neurogenesis. Cancer Cell 2006;9:157-73. doi: 10.1016/j.
cell.2006.02.019.

Greenwald LA, Kouwenhoven MC, Gevaert O, de Rooi JJ,
Stubbs AP, Duijm JE, et al. Intrinsic gene expression profiles of
gliomas are a better predictor of survival than histology. Cancer Res
2009;69:9065-72. doi: 10.1158/0008-5472.CAN-09-2307.

Noushmehr H, Weisenberger DJ, Diefes K, Phillips HS, Pujara K,
Berman BP, et al. Identification of a CpG island methylation
phenotype that defines a distinct subgroup of glioma. Cancer Cell
2010;17:510-22. doi: 10.1016/j.ccc.2010.03.017.

Sturm D, Witt H, Hovestadt V, Khuong-Quang DA, Jones DT,
Konermann C, et al. Hotspot mutations in H3F3A and IDH1 define
distinct epigenetic and biological subgroups of glioblastoma. Cancer
Cell 2012;22:425-37. doi: 10.1016/j.ccc.2012.08.024.

Liu Q, Liu Y, Li W, Wang X, Sawaya R, Lang FF, et al. Genetic,
epigenetic, and molecular landscapes of multifocal and multicentric
glioblastoma. Acta Neuropathol 2015;130:587-97. doi: 10.1007/
s00401-015-1470-8.

Suzuki H, Aoki K, Chiba K, Sato Y, Shiozawa Y, Shiraiishi Y, et al.
Mutational landscape and clonal architecture in grade II and III
gliomas. Nat Genet 2015;47:458-68. doi: 10.1038/ng.3273.

Weller M, Pfister SM, Wick W, Hegi ME, Reifenberger G, Stupp R,
Molecular predictors of progression-free and overall survival in patients
with newly diagnosed glioblastoma: A prospective translational study of
the German Glioma Network. J Clin Oncol 2009;27:5743-50. doi: 10.1200/JCO.2009.23.0805.

Reuss DE, Sahm F, Schirrmeister F, Wiencke J, Schildkraut C,
et al. Temozolomide chemotherapy alone versus radiotherapy alone
for malignant astrocytoma in the elderly: The NOA-08 randomised,
phase 3 trial. Lancet Oncol 2010;11:1501-8. doi: 10.1016/s1470-
2045(10)70165-6.

Cohen MH, Shen YL, Keegan P, Pazdur R. FDA drug approval
summary: Bevacizumab (Avastin) as treatment of recurrent
glioblastoma multiforme. Oncologist 2009;14:1311-8. doi: 10.1634/
theoncologist.2009-0121.
84. Kreisl TN, Kim L, Moore K, Duic P, Royce C, Stroud I, et al. Phase II trial of single-agent bevacizumab followed by bevacizumab plus irinotecan at tumor progression in recurrent glioblastoma. J Clin Oncol 2009;27:740-5. doi: 10.1200/JCO.2008.16.3055.

85. Friedman HS, Prados MD, Wen PY, Mikkelson T, Schiﬀ D, Abrey LE, et al. Bevacizumab alone and in combination with irinotecan in recurrent glioblastoma. J Clin Oncol 2009;27:4733-40. doi: 10.1200/JCO.2008.19.8721.

86. Gilbert MR, Dignam JJ, Armstrong TS, Wefel JS, Blumenthal DT, Vogelbaum MA, et al. A randomized trial of bevacizumab for newly diagnosed glioblastoma. N Engl J Med 2014;370:699-708. doi: 10.1056/NEJMoa1308573.

87. Chinot OL, Wick W, Mason W, Henriksson R, Saran F, Nishikawa R, et al. Bevacizumab plus radiotherapy-temozolomide for newly diagnosed glioblastoma. N Engl J Med 2014;370:709-22. doi: 10.1056/NEJMoa1308345.

88. Neyns B, Sadones J, Joosens F, Bouttens F, Verbeke L, Baurain JF, et al. Stratified phase II trial of cetuximab in patients with recurrent high-grade glioma. Ann Oncol 2009;20:1596-603. doi: 10.1093/annonc/mdp032.

89. Yung WK, Vredenburgh JJ, Cloughesy TF, Nghiemphu P, Klencke B, Gilbert MR, et al. Safety and efficacy of erlotinib in first-relapse glioblastoma: A phase II open-label study. Neuro Oncol 2010;12:1061-70. doi: 10.1093/neuonc/noq072.

90. Schumacher T, Bunse L, Wick W, Platten M. Mutant IDH1: An immunotherapeutic target in tumors. Oncoimmunology 2015;3:e974392. doi: 10.4161/2162402X.2014.974392.

91. Schumacher T, Bunse L, Pusch S, Sahm F, Wiestler B, Quandt J, et al. A vaccine targeting mutant IDH1 induces antitumour immunity. Nature 2014;512:324-7. doi: 10.1038/nature13387.

92. Ledford H. Metabolic quirks yield tumour hope. Nature 2014;508:158-9. doi: 10.1038/508158a.

93. Yaqub F. Inhibition of mutant IDH1 in acute myeloid leukaemia. Lancet Oncol 2015;16:e9. doi: 10.1016/s1470-2045(14)71140-4.