Radioresistance in Glioblastoma and the Development of Radiosensitizers

Md Yousuf Ali 1,2,3, Claudia R. Oliva 2,3, Abu Shadat M. Noman 4,5, Bryan G. Allen 2,3, Prabhat C. Goswami 2,3, Yousef Zakharia 6, Varun Monga 6, Douglas R. Spitz 2,3, John M. Buatti 3 and Corinne E. Griguer 2,3,*

1 Interdisciplinary Graduate Program in Human Toxicology, University of Iowa, Iowa City, IA 52242, USA; ali-mdyousuf@uiowa.edu
2 Free Radical & Radiation Biology Program, Department of Radiation Oncology, Holden Comprehensive Cancer Center, University of Iowa, Iowa City, IA 52242, USA; claudia-oliva@uiowa.edu (C.R.O.); bryan-allen@uiowa.edu (B.G.A.); prabhat-goswami@uiowa.edu (P.C.G.); douglas-spitz@uiowa.edu (D.R.S.)
3 Department of Radiation Oncology, University of Iowa Hospitals and Clinics, Iowa City, IA 52242, USA; john-buatti@uiowa.edu
4 Department of Biochemistry and Molecular Biology, The University of Chittagong, Chittagong 4331, Bangladesh; asmnoman.bmb@cu.ac.bd
5 Department of Pathology, McGill University, Montreal, QC H3A 2B4, Canada
6 Department of Internal Medicine, University of Iowa, Iowa City, IA 52242, USA; yousef-zakharia@uiowa.edu (Y.Z.); varun-monga@uiowa.edu (V.M.)
* Correspondence: corinne-griguer@uiowa.edu

Received: 21 July 2020; Accepted: 28 August 2020; Published: 3 September 2020

Simple Summary: Numerous mechanisms of glioblastoma (GBM) radioresistance have been identified but have not yet resulted in development of effective radiosensitizer that can increase the efficacy of radiotherapy. In this review, the authors review the mechanisms of GBM radioresistance along with current status of radiation treatment and imaging techniques used in GBM diagnosis and radiotherapy. In addition, they summarize the current GBM radiosensitizers that are being investigated or enrolled in clinical trials. This review emphasizes on the importance of developing an effective radiosensitizer to increase the outcome of GBM radiotherapy. The authors highlight the importance of discovering of novel mechanism(s) of GBM radioresistance that will lead in developing an effective radiosensitizer.

Abstract: Ionizing radiation is a common and effective therapeutic option for the treatment of glioblastoma (GBM). Unfortunately, some GBMs are relatively radioresistant and patients have worse outcomes after radiation treatment. The mechanisms underlying intrinsic radioresistance in GBM has been rigorously investigated over the past several years, but the complex interaction of the cellular molecules and signaling pathways involved in radioresistance remains incompletely defined. A clinically effective radiosensitizer that overcomes radioresistance has yet to be identified. In this review, we discuss the current status of radiation treatment in GBM, including advances in imaging techniques that have facilitated more accurate diagnosis, and the identified mechanisms of GBM radioresistance. In addition, we provide a summary of the candidate GBM radiosensitizers being investigated, including an update of subjects enrolled in clinical trials. Overall, this review highlights the importance of understanding the mechanisms of GBM radioresistance to facilitate the development of effective radiosensitizers.

Keywords: glioblastoma; radioresistance; radiosensitizer
1. Introduction

Glioblastoma (GBM) is the most common adult primary malignant brain tumor and is also the most lethal [1,2]. Median progression-free and overall survival after initial diagnosis are 6.2–7.5 and 14.6–20.5 months, respectively, even with a highly aggressive standard-of-care treatment consisting of maximum safe surgical resection, radiation therapy, and chemotherapy [3–8]. In light of this grim prognosis, substantial effort has been invested to improve the overall survival of patients with GBM. However, over the last decade, all preclinical strategies that have shown promise for improving the outcome of GBM treatments have failed to provide an overall survival benefit in large randomized clinical trials [4–6,9,10]. The main reason for these failures is attributed to the development of resistance to standard therapeutic options for GBM, which include radiotherapy with concomitant chemotherapy. In particular, the development of adaptive radioresistance has been a major challenge. In the hope of identifying a method to overcome this urgent clinical problem, significant research has focused on defining the molecular mechanisms of adaptive radioresistance in GBM. This review presents a brief synopsis of historic advances in GBM diagnosis and treatment, along with reported findings from pre-clinical studies and the clinical trials of candidate radiosensitizers in GBM.

2. History and Current Status of GBM Detection and Imaging Techniques

Standard-of-care treatment for GBM includes surgical resection of the tumor, followed by radiotherapy with concomitant daily temozolomide (TMZ) chemotherapy. Successful surgical resection of the GBM tumor and radiotherapy largely depend on proper tumor imaging and diagnosis. The appearance and location of the tumor suggest both the diagnosis and surgical approach, as well as the safety of resection. Diagnostic imaging of GBM and other brain tumors has significantly improved throughout the last century (Figure 1). The first successful image-based diagnosis of any brain tumor was achieved using X-rays in 1904, roughly a decade after the discovery of X-rays by Wilhelm Röentgen in 1895 [11]. In the next few decades, several other techniques for diagnosing brain tumors were developed and used, such as skull radiographs, pneumoencephalography, ventriculography, myelography, and cerebral angiography [11–16]. Among these techniques, pneumoencephalography was the first technique that allowed visualization, although indirect, of the living brain. The first pediatric brain tumor was diagnosed and reported in 1952 using this technique [17]. In 1954, the utility of nuclear scanning using radioisotopes for localizing brain tumors was reported [18]. Nuclear scanning was the first noninvasive method available to localize brain tumors and has been used consistently ever since. A new era of neuroimaging-based diagnosis began in 1971, after the invention of the computed tomography (CT) scanner by Sir Godfrey N. Hounsfield. For the next decade, CT was widely used and described as the most accurate technique for diagnosing brain tumors [19–22]. Beginning in the 1980s, however, the popularity of CT for brain tumor diagnosis began to decline as studies started to report better diagnosis clarity with magnetic resonance imaging (MRI). MRI provides vastly improved soft-tissue contrast, high spatial resolution, and rapid widespread availability [23]. The later introduction of spiral or helical CT technology, which allows the array detector to spin continuously around the patient, afforded even greater improvements, including the ability to obtain many more images with far greater speed. The greatest advantage of spiral CT for brain tumor imaging has been the consequent ability to create CT angiograms and conduct time-dependent blood perfusion measurements [24].

Despite the improvements to CT, MRI is the preferred imaging technique to characterize gliomas, with approximately 80% of primary malignant brain tumors characterized by MRI [25]. MRI is more sensitive than CT, as indicated by the better correlation of gross and microscopic autopsy findings with MRI than with CT. Compared with MRI, CT usually provides poorer resolution and underrepresents the size of brain tumors [26]. In 1984, when MRI was first used for brain tumor imaging [23], it was still considered an anatomic imaging method. This characterization changed after the deployment of diffusion-weighted imaging (DWI) and diffusion tensor imaging (DTI) [27]. These advancements were based on the concept that tumors have a higher cellular density than normal tissue, so fluid in the
Comparison of apparent diffusion coefficients calculated with DWI can distinguish tumor from non-tumor tissue [28,29] and even GBM from central nervous system lymphoma [30,31]. Moreover, dynamic contrast-enhanced (DCE) MRI and dynamic susceptibility (DSC) MRI are two other advanced MRI techniques that can help in monitoring physiological and biological processes in GBM [32,33]. DSC and DCE-MRI are based on modulation and modification of T1 and T2 relaxation time. Although T1 and T2 relaxation are naturally present as signal contrast mechanisms, intrinsic changes in these mechanisms due to disease processes can be quite subtle [34]. Therefore, exogenous contrast agents (CAs), such as gadolinium-based gadopentetate dimeglumine, are sometimes used in clinical oncology MRI studies. DCE-MRI is performed by modulating T1 relaxation time using exogenous CAs. The method is based on the exchange of exogenous CAs between the intravascular compartment and the interstitial tissue. The time course of the diffusion of the contrast agent from the blood pool into tissues through leaky blood vessels is measured to accomplish DCE-MRI. DSC-MRI is based on the drop in the T2 signal after the injection of gadolinium-based CAs and the magnetic susceptibility of the particular tissue [35]. Localized MR spectroscopy imaging (MRSI) is another advanced modality of MRI that depends on the metabolic characteristics of tissue for evaluation of brain tumors. MRSI uses unique spectra originating from nuclei such as proton (1H), phosphorus (31P), and carbon (13C) spectra to measure brain metabolites [36,37]. Conventional MRI provides anatomical information and differences in the morphological structure of the brain tumor. However, anatomical images based on MRI do not provide all the information about molecular changes in response to therapy. MRSI can provide that complimentary information, as it non-invasively maps metabolic profiles and dynamics of the GBM tumor [36,38]. MRSI can perform both steady-state and kinetic analysis of cancer metabolism in vivo and can detect a variety of metabolites [39,40]. Therefore, combining the molecular information provided by MRSI with anatomical information from conventional MRI would provide a better strategy for GBM patient management. MRSI has been reported for use in GBM treatment planning and follow-up of patients after radio- and chemotherapy [41–43]. All these advancements in MRI are crucial for detecting the response of patients to radiotherapy and managing GBM radioresistance. Lately, positron emission tomography (PET) has also been used along with MRI to provide additional insight into the biology of gliomas, which can improve planning for surgery and radiotherapy [44,45]. Additional potential imaging information that may impact GBM treatment includes radiomics, the conversion of biomedical images into quantitative data.

**Figure 1.** Timeline of important discoveries and events that led to current imaging techniques for detecting glioblastoma (GBM) and other brain tumors. CT, computed tomography; MRI, magnetic resonance imaging; PET, positron emission tomography.
Radiomics allows advanced non-invasive assessment of complex imaging features obtained by MRI that may serve as biomarkers \cite{46,47} of disease aggressivity or response. Although these major advances in imaging techniques have substantially improved our ability to diagnose brain tumors, including GBM, overall survival and prognosis for patients with GBM continues to be poor, mostly due to inherent and developed resistance against standard-of-care therapy.

3. Treatment Options for GBM/History of GBM Treatment

Despite the growing number of preclinical studies and clinical trials for GBM, current treatment options have not made significant gains in improving patient survival. GBM treatment is particularly challenging because of the primary location, intrinsic heterogeneity, and infiltrating growth pattern of these tumors. Standard-of-care treatment for GBM includes surgical resection of the tumor, followed by radiotherapy with concomitant daily temozolomide (TMZ) chemotherapy followed by additional TMZ therapy. Surgical treatment in GBM aims for maximal surgical resection, thereby improving conditions for complimentary treatments with chemo- and radiotherapy. Extent of resection (EOR) by surgery is an important treatment-related predictor, as more extensive surgical removal is associated with longer life expectancy \cite{48–50}. Surgical resection is followed by concurrent TMZ and radiotherapy. TMZ is given in a dose of 75 mg/m$^2$/day for six weeks and radiotherapy is given in 30 fractions, totaling 60 Gy, followed by six maintenance cycles of TMZ (150–200 mg/m$^2$/day for the first five days of a 28-day cycle) \cite{5,51}. GBM tumors with epigenetic silencing of the MGMT (O$^6$-methylguanine-DNA methyltransferase) DNA-repair gene by promoter methylation seem to benefit the most from the addition of TMZ \cite{52}. A recent study \cite{53} suggests that dual alkylator therapy with temozolomide and lomustine might improve survival compared with standard temozolomide therapy in patients with newly diagnosed glioblastoma with a MGMT-methylated promoter. Most patients eventually experience tumor recurrence or progression. Recurrent GBM tumors are often resistant to temozolomide. Moreover, standard-of-care treatment for recurrent GBM is not well defined. Recurrent patients can benefit from repeat surgery if a total gross resection is possible \cite{54}. Lomustine is used as a second course of alkylating agent to treat most recurrent GBM patients who are eligible for salvage therapy. Bevacizumab is also given as a single agent in some countries including the USA, but not in the European Union. A combination of bevacizumab and lomustine is considered for treatment of recurrent GBM patients with rapidly progressing disease \cite{55}. However, in most cases, these chemotherapies either in combination or alone have failed to show prolongation of overall survival in recurrent GBM patients\cite{56,57}. Therefore, irrespective of the treatment method, most patients diagnosed with primary GBM die within two years.

Until the 1970s, surgery was the only option to treat glioma, with radiation used only as a palliative treatment. However, the scenario changed when several studies in the late 1970s reported the successful use of radiation in treating malignant gliomas in clinical trials \cite{58–60}. Over the next few decades, surgical removal of the tumor, followed by radiotherapy, became the standard-of-care therapy for patients with GBM. In 2005, Stupp and colleagues introduced temozolomide (TMZ), a DNA alkylator, in combination with radiation therapy \cite{3}. Surgical removal of the tumor, followed by concomitant radiotherapy and chemotherapy with TMZ, has remained the standard-of-care treatment for GBM since 2005 \cite{61}. Recently, the application of tumor-treating fields (TTFs), which involves the continuous delivery of low-intensity electric fields alternating at an intermediate frequency, has been viewed by some as a promising cancer treatment. TTF therapy has been shown to improve both progression-free and overall survival in GBM \cite{8}. Despite some initial skepticism, application of TTFs to the shaved head through a transducer connected to a portable device has been reported to be effective in patients with GBM in a randomized clinical trial \cite{62}. Moreover, a variety of molecular targeted therapies have been tried both clinically and pre-clinically, such as leflunomide targeting platelet derived growth factor receptor (PDGFR) \cite{63}, erlotinib targeting epidermal growth factor receptor \cite{64}, tipifarnib targeting Ras \cite{65}, temsirolimus targeting mTOR \cite{66}, and enzasturin targeting PKC-β \cite{67}. However, nothing is known of the impact of immunotherapy regimens on mechanisms of radioresistance in GBM.
Not all patients respond to these therapies in a similar way. High genetic and molecular variation in GBM tumors makes it difficult to predict individual responses to specific therapeutics. Thus, it is not surprising that despite all these treatment options, the median survival for GBM patients has not dramatically improved. Tumor recurrence, which is almost inevitable after a median survival of 32–36 weeks, further complicates treatment efforts [68–70]. Treatment of these recurrent tumors is exceptionally challenging. Reirradiation and stereotactic radiotherapy have been used to treat recurrent GBM tumors [71,72]; however, these and other salvage options are limited by cumulative toxicity [73]. So far, many clinical trials with different chemotherapeutic and recently immunotherapeutic agents administered as single agents or in combinational therapy have been conducted for recurrent GBM [57,74–78], yet none of these combinations has reliably improved survival, highlighting the urgency to find new GBM treatment options.

4. Current Status of Radiation Treatment in GBM and Emergence of Radioresistance

Although most patients with GBM have been treated following the same general protocols over the last decade, radiation therapy has changed substantially over this period as a result of better instrumentation and improvements in imaging technology [79]. These changes include an upgrade in radiotherapy technology from 2-dimensional whole-brain radiotherapy to 3-dimensional conformal radiotherapy, and more recently to intensity-modulated radiation therapy (IMRT) and volumetric arc radiation therapy (VMAT) [80,81]. IMRT techniques limit the exposure of normal tissues to radiation by delivering non-uniform, computationally optimized radiation to the tumor. In addition, fractionated stereotactic radiation has been suggested and used in several clinical trials for patients with recurrent GBM. Stereotactic guidance further improves the accuracy of treatment delivery to a radiographically identified target. With fractionation, the total dose of radiation is split into many smaller fractions and administered over a span of several weeks, which improves the radiobiological impact on tumors versus normal tissues, which repair damage more quickly. An approach known as hypofractionated radiotherapy allows the total dose of radiation to be split into larger doses, thus fewer fractions, and administered over a shorter period, which may improve convenience, although it does not appear to improve overall survival of patients [73,82–84]. A limited number of studies combining advanced image-based targeting of GBM with dose escalation suggest a small benefit in outcomes [85–87]. Thus, despite major advances in radiation technology, the overall outcome of radiotherapy in GBM remains far from optimal, as tumors are inherently resistant and develop increased resistance to radiation, especially upon recurrence.

5. Mechanisms of GBM Radioresistance

Over the years, many studies have been devoted to elucidating the mechanism of GBM radioresistance. So far, a variety of mechanisms have been implicated to explain GBM radioresistance. All these previously reported mechanisms have identified several key factors, including tumor microenvironment, hypoxia, metabolic alteration, glioma stem cells, tumor heterogeneity, microRNAs, cell cycle, and DNA damage and repair (Figure 2), that contribute to the development of GBM radioresistance. These underlying mechanisms of GBM radioresistance have been discussed in detail in the following subsections.
5.1. Tumor Microenvironment

GBM tumor microenvironment plays a major role in tumorigenesis and progression of GBM. Cellular composition of the GBM tumor microenvironment includes non-neoplastic stromal cells, normal and reactive astrocytes, fibroblasts, extracellular and vascular pericytes, glioma stem cells, and immune cells [88,89]. The tumor microenvironment also includes different biomolecules produced by all cell types within the tumor to support its growth and progression. All these complex networks of various cells and biomolecules in the tumor microenvironment contribute to the radiation response [90]. Several studies have reported that glioma cells irradiated in vivo in xenograft models show resistance to radiation, whereas same cells grown in vitro exhibit susceptibility to radiation, thus indicating the role of the tumor microenvironment in vivo in developing radioresistance [91,92]. The GBM microenvironment is anatomically compartmentalized in what is referred to as tumor niches, where the signaling arising in stromal and tumor cells converges and regulates tumor progression and proliferation [93–95]. Glioma stem cells also reside in these microenvironment niches. Recent evidence suggests that microenvironment niches provide glioma stem cells with a variety of mechanisms to obstruct chemotherapies and radiotherapies, thus developing resistance [96].

5.2. Hypoxia

Hypoxia is common in solid tumors because the rapid tumor growth outpaces the growth of blood vessels, preventing the homogenous diffusion of oxygen to all tumor regions [97]. GBM tumors contain hypoxic regions detected by MRI and microscopic analysis [98], and hypoxia-inducible factors (HIFs) have been shown to contribute critically to GBM tumorigenesis by regulating the tumorigenic capacity of glioma stem cells [99]. In addition, it was reported in the middle of the last century that oxygen concentration influences the response of mammalian cells to radiation [100]. The majority of DNA damage caused by conventional radiotherapy in normoxic conditions is mediated...
by reactive oxygen species (ROS) such as $O_2\cdot^−$, $H_2O_2$, and OH. However, the free radical-generated oxidative stress-inducing capacity of radiotherapy decreases in hypoxic conditions, so it is not surprising that hypoxia leads to the development of radioresistance. Marampon and colleagues reported that regulation of the functional interplay among extracellular signal-related kinases (ERKs), DNA-dependent protein kinase catalytic subunit (DNA-PKcs), and HIF1-α mediated by hypoxia causes radioresistance in GBM [101]. Upon activation by hypoxia, HIF2-α was shown to activate OCT4, a stem cell transcription factor. Upon activation, OCT4 regulates the self-renewal and differentiation of stem cells. Thus, hypoxia can induce radioresistance by increasing stemness in glioma cell populations [102–105]. Poorly structured blood vessel networks can also result in irregular and fluctuating tumor tissue perfusion. These fluctuations lead to periods of poor and better oxygenation, exposing cells to periods of hypoxia followed by periods of reoxygenation in a cyclic manner [106]. This phenomenon, known as cycling hypoxia, has been reported to induce GBM radioresistance by triggering a substantial increase in HIF1-α activity [107]. Investigators are exploring a variety of mechanisms to minimize hypoxia and reduce radioresistance in GBM. For example, improving intratumoral oxygenation has been reported to increase glioma radiosensitivity in vitro and in vivo. Tracing and applying increasing doses of radiation in hypoxic regions is also being investigated [108,109]. However, more studies are needed to target GBM hypoxia to improve the response to radiotherapy in patients.

5.3. Metabolic Alteration

Reprogramming of cellular energetics, or metabolic alteration, is a hallmark of cancer [110] and has an important role in the progression of GBM and other brain tumors. The modification of metabolism and mitochondrial bioenergetics detected in GBM cells fuels survival, proliferation, and invasion. Emerging reports suggest that metabolic alteration also mediates resistance to standard-of-care therapies in GBM [111–119]. In particular, high rates of glycolysis have been correlated with GBM radioresistance, and inhibition of the glycolytic pathway has been shown to reduce this resistance in vitro and in vivo [120,121]. The reductant NADPH is a major source of electrons for most cellular antioxidant systems mediated by glutathione and thioredoxin, playing a critical role in redox metabolism and facilitating survival against numerous pro-oxidants, such as radiation. In IDH1wtGBM, the wild-type IDH1 mediates the production of NADPH in response to radiation, facilitating radioresistance. Conversely, knockdown of wild-type IDH1 has been reported to reduce the level of NADPH, making GBM cells radiosensitive in vitro and in vivo [122,123]. High tumor expression of the ATPase family, AAA domain-containing 3A (ATAD3A), a nuclear DNA-encoded mitochondrial protein involved in maintaining mitochondrial functions, and communication between the endoplasmic reticulum (ER) and mitochondria, have been shown to correlate with the development of GBM radioresistance [124]. Moreover, it has been reported that mitochondrial ATP-sensitive potassium channels are overexpressed in glioma and control glioma radioresistance by regulating ROS-induced ERK activation [124]. Knockdown of TP53-induced glycolysis and apoptosis regulator (TIGAR) has been shown to radiosensitize glioma cells to radiation [125]. TIGAR, an early target of p53, can increase the level of NADPH, redirecting glucose into the pentose phosphate pathway. Increased NADPH helps cells to deal with redox stress. Therefore, it is possible that TIGAR induces radioresistance by helping GBM cells to handle radiation-induced redox stress. Altogether, numerous studies show a correlation between metabolic alterations and GBM radioresistance, although the direct mechanistic link between metabolic reprogramming and GBM radioresistance remains to be elucidated.

5.4. Glioma Stem Cells

In recent years, cancer stem cells (CSCs), also known as tumor initiating cells, have been extensively reported in different cancer types. CSCs are a subpopulation of cells within a tumor mass that have the ability to self-renew and differentiate into diverse types of tumor cells [126,127]. Several studies have demonstrated the existence of self-renewing tumorigenic cells in GBM and other gliomas that show
multilineage differentiation potential and stem cell marker expression, and which are thus referred to as glioma stem cells [128–132]. In addition, glioma stem cells have been shown to propagate as therapy-resistant cells [133–135], as shown in Figure 3. Glioma initiating cells (GICs) are resistant to radiation and are directly correlated with patients’ outcomes [136]. GICs can be characterized by the expression of a group of markers such as SOX2, OCT4, NANOG, OLIG2, NESTIN, ID1, CD133, CD15, CD44, and A2B5 [137–145]. The fraction of glioma cells expressing CD133, a marker for both neural and GICs [130], increases after irradiation. CD133-positive GICs preferentially activate DNA damage checkpoint proteins such as Chk1 and Chk2 in response to radiation, carrying out the repair of radiation-induced DNA damage more effectively than CD133–negative cells. Thus, an enhanced DNA damage repair capacity likely underlies, at least in part, the radioreistance of CD133-positive GICs [134]. It was also shown that GICs become radioreistant through the overexpression of proliferating cell nuclear antigen (PCNA)-associated factor (PAF). [146]. PAF is a DNA damage-regulated factor that controls the accessibility of DNA translesion synthesis (TLS) enzymes to PCNA, thereby facilitating DNA damage bypass [147]. After irradiation of GICs, PAF associates with PCNA to release TLS Pol η, resulting in restoration of error-free DNA synthesis and, in turn, glioma stem cell proliferation and radioreistance [146]. Moreover, high expression of cathepsin L, a lysosomal endopeptidase enzyme, mediates radioreistance in GICs. Interestingly, knockdown of cathepsin L in patient-derived GICs led to decreased expression of CD133 and reduced phosphorylation of DNA damage checkpoint proteins, restoring radiosensitivity [148]. However, further studies are needed to determine how cathepsin L promotes these effects. Overall, these studies show that the presence of GICs in GBM might play a critical role in promoting radioreistance.

Figure 3. GSCs can self-renew, initiate tumors, and survive radiotherapy. The cells that survive radiotherapy can give rise to a population of cells that are resistant to radiation. Created with BioRender (Science Suite Inc., Toronto, Ontario, Canada).

5.5. GBM Tumor Heterogeneity

Tumor heterogeneity is characterized by the presence of different cell populations or clones having distinct genetic or molecular profiles within a tumor or among different individual tumors originating from the same tumor. Intertumoral heterogeneity is defined by distinct genetic alterations present in individual tumors originating in the same organ, whereas intratumoral heterogeneity is characterized by distinct genetic alterations within the same tumor [149,150]. Intratumoral heterogeneity is further complicated by the presence of different cell types [151]. GICs residing in the GBM microenvironment niche play a major role in tumor heterogeneity. GSCs are characterized by their ability to regenerate, whereas GBM initiating cells are a subpopulation of GSCs that are CD133+ and are capable of tumor
initiation in orthotopic mouse models [152]. GSCs and GBM initiating cells have been shown to contribute to GBM radioresistance through increased activation of DNA damage checkpoint pathways and intrinsic hyperactivation of PI3/Akt and PTEN pathways [134,153]. The differences in molecular and genetic signatures of these different cells within a single tumor cause differential responses to radiotherapy among specific cell populations. Upon treatment with radiation, the radioresistant populations eventually become dominant, leading to an overall increase in tumor resistance [154].

Intratumoral heterogeneity creates a major challenge in the treatment of GBM. Heterogeneity has been detected among tumors from different patients, yet molecular analysis of patient-derived GBM tissue has shown genetic diversity within regions of individual tumors as well [155–157]. Single-cell RNA sequencing and integrated genomic analysis of GBM tissues have shown unique transcriptional programs within individual tumors and clinically relevant subtypes [150,158]. Moreover, molecular analysis has revealed multiple cellular subclones within a single GBM tumor. Genomic analysis of GBM tumors has identified four major subtypes based on gene expression patterns, namely classical, pro-neural, neural, and mesenchymal. Alterations in the expression of EGFR, NF1, and PDGFRα/IDH1 genes identify classical, mesenchymal, and pro-neural subtypes, respectively, whereas the neural subtype is defined by the expression of several neural markers such as NEFL, GABRA1, SYT1, and SLC12A5 [159]. However, subsequent studies have redefined the transcriptional subtypes of GBM into three clinically relevant classes, designated as proneural, mesenchymal, and classical [160]. GBM tumors also vary in the status of several other genes, with such variety including differences in isocitrate dehydrogenase (IDH) mutation and O6-methylguanine-DNA methyl transferase (MGMT) promoter methylation [161,162]. IDH is an enzyme that catalyzes the decarboxylation of isocitrate to α-ketoglutarate. IDHs have three isoforms, namely IDH1, 2, and 3. The majority of IDH mutations in GBM involve R132 of IDH1 [163,164]. The R132 IDH1 mutation is more common in secondary GBM than in primary GBM [161,165]. IDH1 mutation has a better prognosis, although exceptions have also been reported [166,167]. IDH1 mutations have been reported to radiosensitize glioma cells by epigenetic downregulation of TIGAR. Moreover, IDH1 silencing can improve the response of GBM cells to radiation by reducing the level of NADPH [123,168]. MGMT encodes for a DNA repair enzyme that repairs and detoxifies TMZ-induced DNA damage [169]. A combination of IDH1 mutation and MGMT methylation has been reported to better predict the outcome of TMZ and radiotherapy than either IDH1 or MGMT alone [170,171]. As the combination of MGMT methylation and IDH1 show a correlation with better patient outcomes following radiotherapy, it remains to be investigated if these two mechanisms can be targeted in radioresistant GBM cells.

5.6. MicroRNAs

MicroRNAs are small non-coding RNAs that usually inhibit gene expression at the posttranscriptional level. Altered expression of several microRNAs has been reported in different cancers [172–175], and the role of microRNAs in GBM has been studied extensively [176,177]. A literature survey conducted in 2013 reported that around 235 microRNAs are overexpressed and 95 are downregulated in GBM, compared with normal brain tissue [178]. Notably, microRNAs have been shown to effectively regulate radiation-related signal transduction pathways in GBM, and many studies have reported that the radiosensitivity of GBM can be altered by targeting these microRNAs. For example, miR-124 was found to increase the radiosensitivity of glioma cells by targeting and inhibiting CDK4 [179,180]. In addition, Patryk and colleagues have shown that overexpression of miR-1 and miR-221/222 confer radioresistance in GBM cells by regulating AKT, independently of PTEN status. Upon activation by miR-221/222 after irradiation, AKT modulates DNA-PKcs expression to enhance DNA damage repair (DDR) activity and thereby promote radioresistance [181]. Another study reported that miR-1, miR-125a, miR-150, and miR-425 induce radioresistance in GBM through upregulation of the cell cycle checkpoint response [182]. Thus, these studies show that different microRNAs can regulate GBM radioresistance by modulating Akt signaling, cell cycle checkpoint responses, and DDR activity.
5.7. Cell Cycle, DNA Repair and Other Signaling Pathways

Several studies have reported the role of the DNA repair pathways in GBM radioresistance. Marampon and colleagues reported that histone deacetylase (HDAC)-4 and -6 promote radioresistance in GBM by inducing double strand break (DSB) repair [183]. Overexpression of α-6 integrin also causes radioresistance in GBM by increasing the efficiency of DDR [184]. Furthermore, overexpression of EGFR and EGFRvIII cause radioresistance in GBM by activating both homologous recombination and nonhomologous end joining. EGFRvIII has been shown to activate a key enzyme, DNA-PKcs, involved in DSB repair [185,186]. BMI1, a component of the polycomb repressive complex 1 (PRC1), is associated with the proliferation of high-grade gliomas and other cancer types [187–190]. BMI1 was also reported to confer radioresistance to GBM by recruiting DDR machinery [191].

GBM is a heterogeneous tumor that often harbors anomalies in a variety of signaling pathways. Alterations in several molecular and signaling pathways have been shown to be involved in inducing radioresistance in GBM [192]. One of the pathways reported to be intricately involved in this resistance is the Notch signaling pathway. This signaling pathway is important in the maintenance of a variety of cells, including neural stem cells, and is known to play an important role in cancer stem cells [193–196]. Inhibition of Notch 1 and 2 restores radiosensitivity in glioma stem cells, and Notch has been reported to induce radioresistance in GBM through regulation of the PI3-kinase/Akt pathway [197].

In general, the PI3-kinase/Akt signaling pathway is involved in numerous important cellular functions, including cell proliferation, migration, differentiation, metabolism, and apoptosis [198]. Moreover, abnormal activation of the PI3-kinase/Akt pathway is detected in multiple cancer types, including GBM, and is associated with poor prognosis and survival in patients [199]. In GBM, the increase in expression and activity of AKT contributes to tumor progression, recurrence, and radioresistance. Radiation activates Akt in GBM and thereby contributes to the development of radioresistance [200]. Akt has also been shown to be correlated with poor progression-free and overall survival of GBM patients [201–203]. Activation of AKT can enhance DNA damage repair (DDR) by promoting γ-H2AX foci resolution in irradiated glioma cells [186], whereas downregulation of AKT facilitates unreparable DNA double strand breaks (DSB) in irradiated U251 glioma cells [204,205]. In addition, the transmembrane protein leucine-rich repeats and immunoglobin-like domains protein 1 (LRIG1) has been reported to alter GBM radioresistance by modulating the Akt pathway [206]. LRIG1 is expressed in several human tissues and organs and is described as a tumor suppressor [207]. Irradiation causes downregulation of LRIG1 in radioresistant U251R cells [206]. Overexpression of LRIG1 in U251R cells significantly reduced EGF signaling and AKT phosphorylation, increasing DNA damage and susceptibility to radiation [206], indicating that downregulation of LRIG1 contributes to radioresistance. Expression of PTEN, an important gene in the PI3-kinase/Akt pathway, is also frequently altered in GBM [208]. Loss or mutation of PTEN leads to activation of Akt, resulting in resistance to radiotherapy. Depletion of PTEN has also been shown to sensitize tumor cells to therapies that induce DNA damage, such as radiation [209]. A recent study reported that pharmaceutical inhibition of PTEN phosphorylation at tyrosine 240 sensitizes GBM cells to radiation by attenuating DNA damage repair mediated by nuclear PTEN [210]. Together, these studies indicate that overactivation of Akt signaling promotes GBM radioresistance by modulating DDR and reducing radiation-induced DNA DSBs.

The tumor suppressor p53 is one of most frequently deregulated genes in human cancer and is positioned in the center of the regulatory network controlling cell proliferation, survival, and genome integrity [211]. Around 40%–50% of GBMs have p53 mutations [212,213], and a lack of p53-mediated apoptosis could be a factor in therapy resistance in GBM. Indeed, it has been reported that the failure of p53 to induce p21BAX expression causes radioresistance in GBM-derived cells [214].

Constitutive activation of the JAK/STAT pathway is also common in many cancers. STAT3 is a redox-sensitive transcription factor that is required for the maintenance of stemness in GBM cells [215,216]. In GBM cells, irradiation promotes the nuclear translocation and activation of STAT3, promoting malignancy, and STAT3 activation is high in CD133-positive radioresistant GBM cells and recurrent tumors [217,218]. However, inhibition of STAT3 activity triggers the activation of ERK1/2,
which allows GBM cells to survive radiotherapy. Therefore, dual inhibition of STAT3 and ERK1/2 is necessary to sensitize glioma cells to radiation [219]. Another transcription factor, forkhead box protein M1 (FOXM1), which is vital for cell proliferation, cell cycle progression, tissue homeostasis, and DNA damage repair, has been shown to regulate metastasis in different cancers [220,221]. In GBM, high tumor expression of FOXM1 is associated with poor prognosis [222]. Furthermore, in irradiated GBM cells, FOXM1 was shown to mediate radioresistance in a manner that involves direct interaction with STAT3 and is dependent on STAT3 activation [223]. Finally, inhibition of suppressors of cytokine signaling (SOCS), which can regulate JAK/STAT signaling transduction, has been shown to increase radioresistance in glioma cells [224]. In particular, SOCS3 has been implicated in GBM radioresistance, and methylation of the SOCS3 promoter may be associated with poor prognosis in patients with GBM [225]. Thus, radiation-induced inhibition of SOCS proteins in glioma cells may activate the JAK/STAT pathway, promoting radioresistance [224]. Overall, these studies show that the JAK/STAT pathway has a major role to play in GBM radioresistance.

The Wnt signaling pathway, best known for critically controlling neural patterning and organ development, has long been described as an important contributor to CSC maintenance in various cancers, including GBM [226,227]. Overexpression of Wnt/β-catenin has been correlated with GBM aggressiveness and poor prognosis of patients [228,229]. Activation of Wnt signaling has also been shown to confer resistance to radiation. For example, multiple Wnt signaling-related genes, including APC, FZD1, LEFT1, TCF4, and WISP1, are overexpressed in radiosensitive GBM cells [230]. Moreover, inhibition of the Wnt/β-catenin pathway restored radiosensitivity in GBM cells displaying adaptive radioresistance [231]. Upon activation, β-catenin translocates to and accumulates in the nucleus, resulting in activation of β-catenin target genes such MMP-2 and MMP-9. Irradiation was shown to mediate these effects in glioma cells, and the activation of MMP-2 and MMP-9 after irradiation induced tumor spreading and invasion [232,233]. Although the above studies show correlation of Wnt/β-catenin pathway activation with GBM radioresistance, more research is needed to elucidate the specific mechanisms by which the Wnt pathway promotes this radioresistance.

6. Radiosensitizers in GBM and Other Cancer Treatment

Radiotherapy is still the most common treatment option across many tumor types. Around 50% of all cancer patients receive radiation during the course of their treatment, which constitutes 40% of all curative treatments for cancer [234–236]. Improved technologies and knowledge about radiation treatment methods have increased the use of radiation therapy. However, there is still a wide range of obstacles and challenges, which have already been discussed in this paper. These challenges, such as the presence of CSCs, tumor heterogeneity, and metabolic alterations, make it difficult to use radiotherapy alone to cure tumors, not only in GBM but in other cancers as well [237–239]. In this regard, the use of radiosensitizers has been described as an excellent option for making radiotherapy more effective without increasing the dose of radiation, which may then be detrimental to normal tissues [240–242]. Radiosensitizers increase cell sensitivity to radiotherapy by altering the activity of cell factors that modulate the deleterious effects of radiation. Mechanisms of radiosensitization involve inhibiting intracellular thiols [243,244], creating cytotoxic substances [245], inhibiting repair biomolecules [246], and mimicking the electrophilic activity of oxygen [247,248]. The radiosensitizing effects of these mechanisms were related mainly to effects on the DDR pathway induced by radiotherapy. However, over time, the use of radiosensitizers has become a multifaceted approach [249,250]. Some established chemotherapeutic agents have been used as radiosensitizers and have been reported to successfully enhance the efficacy of radiotherapy in clinical trials for different cancers [251,252]. For example, the chemotherapeutic agent gemcitabine has been shown to be an effective radiosensitizer in the treatment of many cancers, such as breast, ovarian, non-small cell lung, pancreatic, and bladder cancers [253–255]. In addition, the small molecule inhibitor of c-MET, crizotinib, has been shown to enhance the radiosensitivity of KRAS-mutant colorectal cancers that are resistant to cetuximab [256].
Pretreatment of breast and lung cancer cells with a novel estrone analog also increases sensitivity to radiation [257].

As discussed earlier, one of the main reasons for radioresistance in GBM and other solid tumors is the presence of hypoxic regions within the tumor. Therefore, oxygen-mimicking compounds have been investigated as potential radiosensitizers in different cancers [258–260]. In particular, compounds containing a nitro group that has the same electron affinity as oxygen have been described to have radiosensitizing effects [242,261–264]. Furthermore, oxygen carriers and agents that can produce oxygen, such as hydrogen peroxide, have also been described as potential radiosensitizers [265,266]. However, insufficient and poorly formed blood vessels in the tumor microenvironment make it difficult to increase tumor oxygenation therapeutically [267]. Therefore, the alternative approach of reducing mitochondrial respiration has been investigated as a method for increasing oxygenation in hypoxic tumor regions [268]. This approach, termed metabolic radiosensitization, reduces the cellular metabolic demand for oxygen by reducing mitochondrial oxidative metabolism. In this regard, Benaj and colleagues have shown that papaverine, an inhibitor of mitochondrial complex I, increases tumor oxygenation and thus sensitizes the cells of hypoxic lung and breast tumors—but not healthy, normoxic tissues—to radiation in mouse models [269].

As radiotherapy is an integral component of the standard-of-care therapy for GBM, the use of radiosensitizers has been promoted as a potential treatment option for GBM as well [270,271], and many chemotherapeutic agents have been investigated [272–275]. However, most potential GBM radiosensitizers have not progressed to clinical trials due to a lack of promising preclinical data (Table 1).

| Name of the Radiosensitizers | Effect                                                                 | References       |
|------------------------------|----------------------------------------------------------------------|------------------|
| Gemcitabine                  | Initiates DNA damage by incorporating gemcitabine triphosphate, an active metabolite of gemcitabine, instead of nucleotide deoxycytidine triphosphate (dCTP) | [272,276]       |
| Gö6976                       | Protein kinase inhibitor                                             | [277]           |
| Talazoparib                  | PARP inhibitor                                                        | [278]           |
| MEK162                      | MAPK inhibitor                                                        | [279]           |
| Erlotinib                   | EGFR inhibitor                                                        | [280]           |
| Everolimus                   | mTOR inhibitor                                                        | [281]           |
| Valproate                    | HDAC                                                                 | [282]           |
| Vorinostat                   | HDAC inhibitor                                                        | [283]           |
| Vandetanib                   | VEGFR2 inhibitor                                                      | [284]           |
| Enzastaurin                  | Protein Kinase C (PKC) inhibitor                                      | [285]           |
| Talampanel                   | alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor antagonist | [286]           |
| TMZ                          | Alkylates/methylates DNA at N-7 or O-6 positions of guanine residue    | [287]           |
| Bortezomib                   | Proteasome inhibitor                                                  | [288]           |
| Resveratrol                  | STAT3 inhibitor                                                       | [218]           |
| Veliparib                    | PARP inhibitor                                                        | [289]           |
| Adavosertib                  | WEE1 inhibitor                                                        | [290]           |
| Chloroquine                  | Inhibits autophagy and induces apoptosis                              | [291,292]       |
| Ascorbate                    | Pro-oxidant                                                           | [293]           |
| RRx-001                      | Macrophage-stimulating agent                                          | [294]           |
Of the proposed radiosensitizers that were effective in preclinical studies and thus evaluated to phase I/II clinical trials (Table 2), most failed to improve progression-free and overall survival and did not progress to phase III. However, we are still awaiting the results of phase II clinical trials for some agents, as shown in Table 2. Therefore, continued research into the mechanisms of radioresistance is needed to identify novel candidate radiosensitizers.

Table 2. List of current and previous clinical trials of radiosensitizers for GBM treatment.

| Study ID      | Phase | Diagnosis                                      | Treatment                                         | Outcomes                                                                                                                                 |
|---------------|-------|-----------------------------------------------|--------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------|
| NCT01752491   | I     | GBM                                           | Ascorbate, TMZ, and radiotherapy                 | No dose-limiting toxicities [293]                                                                                                     |
| NCT01465347   | I & II| GBM                                           | Trans sodium crocetinate (TSC), TMZ, and radiotherapy | No adverse effects. Suggests radiotherapy and TSC combination is beneficial for GBM treatment. No significant difference in overall survival [297]. |
| NCT00185861   | I     | Recurrent malignant glioma                    | Arsenic trioxide (ATO) and stereotactic radiotherapy | ATO and fractionated stereotactic radiotherapy is well-tolerated [298].                                                                 |
| NCT04205357   | I     | Recurrent GBM                                 | Sulfasalazine and stereotactic radiotherapy       | Study ongoing, recruiting patients                                                                                                     |
| NCT02871843   | I     | GBM, oligodendroglioma, anaplastic oligodendroglioma | RRx-001, TMZ, and radiotherapy                    | Study ongoing                                                                                                                          |
| NCT00302159   | II    | High-grade gliomas                            | Valproic acid (VPA), TMZ, and radiotherapy        | No adverse effects; VPA in combination with TMZ and radiotherapy can improve outcome [282]                                               |
| NCT00305864   | I & II| GBM                                           | Motexafin gadolinium, TMZ, and radiotherapy       | No adverse effects; no significant improvement in overall survival [295]                                                                |
| NCT03562430   | II    | GBM                                           | NVX-108, TMZ, and radiotherapy                    | Study ongoing                                                                                                                          |
| NCT03672721   | I & II| GBM                                           | Carboplatin and radiotherapy                     | Study ongoing                                                                                                                          |
| NCT02376532   | I     | GBM                                           | Chloroquine, TMZ, and radiotherapy                | No adverse effects reported [305]                                                                                                     |
| NCT02432417   | II    | GBM                                           | Chloroquine, TMZ, and radiotherapy                | Study ongoing                                                                                                                          |
| NCT01849146   | I     | Newly diagnosed and recurrent GBM             | Adavosertib, TMZ, and radiotherapy                | Study ongoing                                                                                                                          |
| NCT03423628   | I     | GBM                                           | AZD1390 and Radiotherapy                         | Study ongoing                                                                                                                          |
7. Conclusions

It is clear from the literature that GBM remains a very deadly cancer, despite the myriad research efforts and clinical trials with agents designed to improve the treatment outcome. Moreover, in patients with newly diagnosed or recurrent GBM, outcomes with radiotherapy have not improved for years. Radiosensitizers have been considered and remain a viable option for improving the outcome of therapy in GBM but have not yet achieved this potential. Overall, more research is necessary to fully understand the mechanisms of GBM radioresistance and improve the outcomes of patients with this deadly disease.

Author Contributions: Conceptualization, M.Y.A., C.R.O., A.S.M.N., B.G.A., P.C.G., Y.Z., V.M., D.R.S., J.M.B., and C.E.G.; methodology, M.Y.A., and C.E.G.; software, N/A; validation, N/A; formal analysis, M.Y.A., C.R.O., and C.E.G.; investigation, M.Y.A., C.R.O., and C.E.G.; resources, M.Y.A., C.R.O., D.R.S., J.M.B., and C.E.G.; data curation, N/A; writing—original draft preparation, M.Y.A., C.R.O., and C.E.G.; writing—review and editing, M.Y.A., C.R.O., A.S.M.N., B.G.A., P.C.G., Y.Z., V.M., D.R.S., J.M.B., and C.E.G.; visualization, M.Y.A., C.R.O., and C.E.G.; supervision, C.E.G.; project administration, C.R.O., and C.E.G.; funding acquisition, C.R.O., J.M.B., and C.E.G. All authors have read and agreed to the published version of the manuscript.

Funding: This study was supported by National Institute of Neurological Disorders and Stroke: U01NS093663, National Cancer Institute: R01CA160821.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Kohler, B.A.; Ward, E.; McCarthy, B.J.; Schymura, M.J.; Ries, L.A.G.; Eheman, C.; Jemal, A.; Anderson, R.N.; Ajani, U.A.; Edwards, B.K. Annual Report to the Nation on the Status of Cancer, 1975-2007, Featuring Tumors of the Brain and Other Nervous System. J. Natl. Cancer Inst. 2011, 103, 714–736. [CrossRef] [PubMed]

2. Davis, M.E. Glioblastoma: Overview of Disease and Treatment. Clin. J. Oncol. Nurs. 2016, 20, S2–S8. [CrossRef] [PubMed]

3. Stupp, R.; Mason, W.P.; van den Bent, M.J.; Weller, M.; Fisher, B.; Taphoorn, M.J.; Belanger, K.; Brandes, A.A.; Marosi, C.; Bogdahn, U.; et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N. Engl. J. Med. 2005, 352, 987–996. [CrossRef] [PubMed]

4. Gilbert, M.R.; Dignam, J.J.; Armstrong, T.S.; Wefel, J.S.; Blumenthal, D.T.; Vogelbaum, M.A.; Colman, H.; Chakravarti, A.; Pugh, S.; Won, M.; et al. A randomized trial of bevacizumab for newly diagnosed glioblastoma. N. Engl. J. Med. 2014, 370, 699–708. [CrossRef]

5. Gilbert, M.; Wang, M.; Aldape, K.D.; Stupp, R.; Hegi, M.; Jaekle, K.A.; Armstrong, T.S.; Wefel, J.S.; Won, M.; Blumenthal, D.T.; et al. Dose-Dense Temozolomide for Newly Diagnosed Glioblastoma: A Randomized Phase III Clinical Trial. J. Clin. Oncol. 2013, 31, 4085–4091. [CrossRef]

6. Chiotis, O.; Wick, W.; Mason, W.; Henriksson, R.; Saran, F.; Nishikawa, R.; Carpentier, A.F.; Hoang-Xuan, K.; Kavan, P.; Cernea, D.; et al. Bevacizumab plus Radiotherapy–Temozolomide for Newly Diagnosed Glioblastoma. N. Engl. J. Med. 2014, 370, 709–722. [CrossRef]

7. Mandel, J.; Yust-Katz, S.; Patel, A.J.; Cachia, D.; Liu, D.; Park, M.; Yuan, Y.; A Kent, T.; De Groot, J.F. Inability of positive phase II clinical trials of investigational treatments to subsequently predict positive phase III clinical trials in glioblastoma. Neuro Oncol. 2017, 20, 113–122. [CrossRef]

8. Stupp, R.; Taillibert, S.; Kanner, A.A.; Kesari, S.; Steinberg, D.M.; Toms, S.A.; Taylor, L.P.; Lieberman, F.; Silvani, A.; Fink, K.L.; et al. Maintenance Therapy With Tumor-Treating Fields Plus Temozolomide vs Temozolomide Alone for Glioblastoma. JAMA 2015, 314, 2535–2543. [CrossRef]

9. Westphal, M.; Heese, O.; Steinbach, J.P.; Schnell, O.; Schackert, G.; Mehdorn, H.M.; Schulz, D.; Simon, M.; Schlegel, U.; Senft, C.; et al. A randomised, open label phase III trial with nimotuzumab, an anti-epidermal growth factor receptor monoclonal antibody in the treatment of newly diagnosed adult glioblastoma. Eur. J. Cancer 2015, 51, 522–532. [CrossRef]

10. Stupp, R.; Hegi, M.; Gorlia, T.; Erridge, S.C.; Perry, J.; Hong, Y.-K.; Aldape, K.D.; Lhermitte, B.; Pietsch, T.; Gruijcic, D.; et al. Cilengitide combined with standard treatment for patients with newly diagnosed glioblastoma with methylated MGMT promoter (CENTRIC EORTC 26071-22072 study): a multicentre, randomised, open-label, phase 3 trial. Lancet Oncol. 2014, 15, 1100–1108. [CrossRef]
11. Pfahler, G. Cerebral skiagraphy: transactions of the American Roentgen Ray Society—5th Annual Meeting. *Am. J. Roentgenol. Radium. Ther. Nucl. Med.* 1904, 4, 174–186.

12. Adson, A.W.; Ott, W.O.; Crawford, A.S. A Study of Ventriculography. *Radiology* 1924, 2, 65–73. [CrossRef]

13. Schwartz, C.W. The Giomas Roentgenologically Considered. *Radiology* 1936, 27, 419–432. [CrossRef]

14. Forestier, J. Actual Technic of Examination of the Spinal Cavities with Lipiodol 1. *Radiol.* 1928, 11, 481–489. [CrossRef]

15. Scott, W.G.; Seaman, W.B. Developments in cerebral angiography with rapid serialized X-ray exposures on roll film 9 1/2 inches wide. *Radiology* 1951, 56, 15–30. [CrossRef] [PubMed]

16. Hoeffner, E.G.; Mukherji, S.K.; Srinivasan, A.; Quint, D.J. Neuoradiology Back to the Future: Brain Imaging. *Am. J. Neuroradiol.* 2011, 33, 5–11. [CrossRef] [PubMed]

17. Smith, A.B. Brain Tumors in Childern. *Radiology* 1952, 58, 688–695. [CrossRef]

18. Seaman, W.B.; Ter-Pogossian, M.M.; Schwartz, H.G. Localization of Intracranial Neoplasms with Radioactive Isotopes. *Radiology* 1954, 62, 30–36. [CrossRef]

19. New, P.F.; Scott, W.R.; Schnur, J.A.; Davis, K.R.; Taveras, J.M. Computerized Axial Tomography with the EMI Scanner. *Radiology* 1974, 110, 109–123. [CrossRef]

20. Baker, H.L.; Houser, O.W.; Campbell, J.K. National Cancer Institute study: evaluation of computed tomography in the diagnosis of intracranial neoplasms. I. Overall results. *Radiology* 1980, 136, 91–96. [CrossRef]

21. Potts, D.G.; Abbott, G.F.; Von Sneidern, J.V. National Cancer Institute study: evaluation of computed tomography in the diagnosis of intracranial neoplasms. III. Metastatic tumors. *Radiology* 1980, 136, 657–664. [CrossRef] [PubMed]

22. New, P.F.; Aronow, S.; Hesselink, J.R. National Cancer Institute study: evaluation of computed tomography in the diagnosis of intracranial neoplasms. IV. Meningiomas. *Radiology* 1980, 136, 665–675. [CrossRef] [PubMed]

23. Araki, T.; Inouye, T.; Suzuki, H.; Machida, T.; Iio, M. Magnetic resonance imaging of brain tumors: measurement of T1. Work in progress. *Radiology* 1984, 150, 95–98. [CrossRef] [PubMed]

24. Castillo, M. History and Evolution of Brain Tumor Imaging: Insights through Radiology. *Radiology* 2014, 273, 111–125. [CrossRef] [PubMed]

25. Ostrom, Q.T.; Gittleman, H.; Fulop, J.; Liu, M.; Blanda, R.; Kromer, C.; Wolinsky, Y.; Kruchko, C.; Barnholtz-Sloan, J.S. CBTRUS Statistical Report: Primary Brain and Central Nervous System Tumors Diagnosed in the United States in 2008–2012. *Neuro Oncol.* 2015, 17, iv1–iv62. [CrossRef] [PubMed]

26. Whelan, H.T.; Clanton, J.A.; Wilson, R.E.; Tulipan, N.B. Comparison of CT and MRI brain tumor imaging in treatment evaluation of glioblastomas: Clinical relevance of current and future techniques. *J. Magn. Reson. Imaging* 2018, 49, 11–22. [CrossRef] [PubMed]

27. Alexander, A.L.; Lee, J.E.; Lazar, M.; Field, A.S. Diffusion tensor imaging of the brain. *Neurotherapeutics* 2007, 4, 316–329. [CrossRef]

28. Yanagihara, T.K.; Wang, T. Diffusion-weighted imaging of the brain for glioblastoma: Implications for radiation oncology. *Appl. Radiat. Oncol.* 2014, 5–13.

29. Schminda, K.M. Diffusion-weighted MRI as a biomarker for treatment response in glioma. *CNS Oncol.* 2012, 1, 169–180. [CrossRef]

30. Guo, A.C.; Cummings, T.J.; Dash, R.C.; Provenzale, J.M. Lymphomas and High-Grade Astrocytomas: Comparison of Water Diffusibility and Histologic Characteristics. *Radiology* 2002, 224, 177–183. [CrossRef]

31. Kono, K.; Inoue, Y.; Nakayama, K.; Shakudo, M.; Morino, M.; Ohata, K.; Wakasa, K.; Yamada, R. The role of diffusion-weighted imaging in patients with brain tumors. *Am. J. Neuroradiol.* 2001, 22, 1081–1088. [PubMed]

32. Kalpathy-Cramer, J.; Gerstner, E.R.; Emblem, K.; Andronesi, O.C.; Rosen, B. Advanced Magnetic Resonance Imaging of the Physical Processes in Human Glioblastoma. *Cancer Res.* 2014, 74, 4622–4637. [CrossRef]

33. Van Dijken, B.R.; Van Laar, P.J.; Smits, M.; Dankbaar, J.W.; Enting, R.H.; Van Der Hoorn, A. Perfusion MRI in treatment evaluation of glioblastomas: Clinical relevance of current and future techniques. *J. Magn. Reson. Imaging* 2018, 49, 11–22. [CrossRef] [PubMed]

34. Stevenson, V.; Parker, G.J.; Barker, G.J.; Birnie, K.; Tofts, P.S.; Miller, D.; Thompson, A. Variations in T1 and T2 relaxation times of normal appearing white matter and lesions in multiple sclerosis. *J. Neurol. Sci.* 2000, 178, 81–87. [CrossRef]
35. Østergaard, L. Principles of cerebral perfusion imaging by bolus tracking. *J. Magn. Reson. Imaging* 2005, 22, 710–717. [CrossRef]
36. Nelson, S.J. Multivoxel magnetic resonance spectroscopy of brain tumors. *Mol. Cancer Ther.* 2003, 2, 497–507.
37. Blueml, S.; Moreno-Torres, A.; Shic, F.; Nguyen, C.H.; Ross, B.D. Tricarboxylic acid cycle of glia in the in vivo human brain. *NMR Biomed.* 2002, 15, 1–5. [CrossRef]
38. Howe, F.A.; Opstad, K.S. 1H MR spectroscopy of brain tumours and masses. *NMR Biomed.* 2003, 16, 123–131. [CrossRef]
39. Kurhanewicz, J.; Vigneron, D.B.; Brindle, K.; Chekmenev, E.Y.; Comment, A.; Cunningham, C.H.; DeBerardinis, R.J.; Green, G.G.; Leach, M.O.; Rajan, S.S.; et al. Analysis of Cancer Metabolism by Imaging Hyperpolarized Nuclei: Prospects for Translation to Clinical Research. *Neoplasia* 2011, 13, 81–97. [CrossRef]
40. Pinker, K.; Stadlbauer, A.; Bogner, W.; Gruber, S.; Helbich, T.; Pinker, K. Molecular imaging of cancer: MR spectroscopy and beyond. *Eur. J. Radiol.* 2012, 81, 566–577. [CrossRef]
41. Andronesi, O.C.; Esmaeili, M.; Borra, R.J.H.; Emblem, K.; Gerstner, E.R.; Plotkin, S.R.; Chi, A.S.; Eichler, A.F.; Dietrich, J.; et al. Early changes in glioblastoma metabolism measured by MR spectroscopic imaging during combination of anti-angiogenic cediranib and chemoradiation therapy are associated with survival. *NPJ Precis. Oncol.* 2017, 1, 20. [CrossRef] [PubMed]
42. Arias-Ramos, N.; Ferrer-Font, L.; Lope-Piedrafita, S.; Mocioiu, V.; Julia-Sapé, M.; Pumarola, M.; Arús, C.; Candiota, A.P. Metabolomics of Therapy Response in Preclinical Glioblastoma: A Multi-Slice MRSI-Based Volumetric Analysis for Noninvasive Assessment of Temozolomide Treatment. *Metabolites* 2017, 7, 20. [CrossRef] [PubMed]
43. Laprie, A.; Ken, S.; Fillerton, L.; Lubrano, V.; Vieillevigne, L.; Tensaouti, F.; Catalaa, I.; Boetto, S.; Khalifa, J.; Attal, J.; et al. Dose-painting multicenter phase III trial in newly diagnosed glioblastoma: the SPECTRO-GLIO trial comparing arm A standard radiochemotherapy to arm B radiochemotherapy with simultaneous boost guided by MR spectroscopic imaging. *BMC Cancer* 2019, 19, 167. [CrossRef] [PubMed]
44. Schlemmer, H.-P.W.; Pichler, B.J.; Schmand, M.; Burbur, Z.; Michel, C.; Ladebeck, R.; Jattke, K.; Townsend, D.; Nagm, A.; Jacob, P.K.; et al. Simultaneous MR/PET Imaging of the Human Brain: Feasibility Study. *Radiology* 2008, 248, 1028–1035. [CrossRef]
45. Pichler, B.J.; Kolb, A.; Nägele, T.; Schlemmer, H.P. PET/MRI: Paving the Way for the Next Generation of Clinical Multimodality Imaging Applications. *J. Nucl. Med.* 2010, 51, 333–336. [CrossRef]
46. Chaddad, A.; Kucharczyk, M.J.; Daniel, P.; Sabri, S.; Jean-Claude, B.J.; Niazi, T.; Abdulkarim, B. Radiomics in Glioblastoma: Current Status and Challenges Facing Clinical Implementation. *Front. Oncol.* 2019, 9, 374. [CrossRef]
47. Chen, C.; Ou, X.; Wang, J.; Guo, W.; Ma, X. Radiomics-Based Machine Learning in Differentiation Between Glioblastoma and Metastatic Brain Tumors. *Front. Oncol.* 2019, 9, 806. [CrossRef]
48. Wolbers, J.G. Novel strategies in glioblastoma surgery aim at safe, supra-maximum resection in conjunction with local therapies. *Chin. J. Cancer* 2014, 33, 8–15. [CrossRef]
49. Li, Y.M.; Suki, D.; Hess, K.; Sawaya, R. The influence of maximum safe resection of glioblastoma on survival in 1229 patients: Can we do better than gross-total resection? *J. Neurosurg.* 2016, 124, 977–988. [CrossRef]
50. Sanai, N.; Polley, M.Y.; McDermott, M.W.; Parsa, A.T.; Berger, M.S. An extent of resection threshold for newly diagnosed glioblastomas. *J. Neurosurg.* 2011, 115, 3–8. [CrossRef]
51. Fernandes, C.; Costa, A.; Osório, L.; Lago, R.C.; Linhares, P.; Carvalho, B.; Caeiro, C.; De Vleeschouwer, S. Current Standards of Care in Glioblastoma Therapy. In *Glioblastoma*; De Vleeschouwer, S., Ed.; Codon Publications: Brisbane, Australia, 2017; pp. 197–241.
52. Hegi, M.E.; Diserens, A.C.; Gorlia, T.; Hamou, M.F.; de Tribolet, N.; Weller, M.; Kros, J.M.; Hainfellner, J.A.; Mason, W.; Mariani, L.; et al. MGMT gene silencing and benefit from temozolomide in glioblastoma. *N. Engl. J. Med.* 2005, 352, 997–1003. [CrossRef] [PubMed]
53. Herrlinger, U.; Tzaridis, T.; Mack, F.; Steinbach, J.P.; Schlegel, U.; Sabel, M.; Hau, P.; Kortmann, R.-D.; Krex, D.; Grauer, O.; et al. Lomustine-temozolomide combination therapy versus standard temozolomide therapy in patients with newly diagnosed glioblastoma with methylated MGMT promoter (CeTeG/NOA09): A randomised, open-label, phase 3 trial. *Lancet* 2019, 393, 678–688. [CrossRef]
54. Suchorska, B.; Weller, M.; Tabatabai, G.; Senft, C.; Hau, P.; Sabel, M.C.; Herrlinger, U.; Ketter, R.; Schlegel, U.; Marosi, C.; et al. Complete resection of contrast-enhancing tumor volume is associated with improved survival in recurrent glioblastoma—results from the DIRECTOR trial. *Neuro Oncol.* 2016, 18, 549–556. [CrossRef] [PubMed]

55. Weller, M.; Le Rhun, E.; Preusser, M.; Tonni, J.-C.; Roth, P. How we treat glioblastoma. *ESMO Open* 2019, 4, e000520. [CrossRef] [PubMed]

56. Gramatzki, D.; Roth, P.; Rushing, E.; Weller, J.; Andratschke, N.; Hofer, S.; Korol, D.; Regli, L.; Pangalu, A.; Pless, M.; et al. Bevacizumab may improve quality of life, but not overall survival in glioblastoma: An epidemiological study. *Ann. Oncol.* 2018, 29, 1431–1436. [CrossRef] [PubMed]

57. Wick, W.; Gorlia, T.; Bendszus, M.; Taphoorn, M.; Sahm, F.; Harting, I.; Brandes, A.A.; Taal, W.; Domont, J.; Idbaih, A.; et al. Lomustine and Bevacizumab in Progressive Glioblastoma. *N. Engl. J. Med.* 2017, 377, 1954–1963. [CrossRef]

58. Walker, M.D.; Strike, T.A.; Sheline, G.E. An analysis of dose-effect relationship in the radiotherapy of malignant gliomas. *Int. J. Radiat. Oncol.* 1979, 5, 1725–1731. [CrossRef]

59. Frankel, S.A.; German, W.J. Glioblastoma multiforme; review of 219 cases with regard to natural history, pathology, diagnostic methods, and treatment. *J. Neurosurg.* 1958, 15, 489–503. [CrossRef]

60. Sheline, G.E. Radiation therapy of brain tumors. *Cancer* 1977, 39, 873–881. [CrossRef]

61. Yabroff, K.R.; Harlan, L.; Zeruto, C.; Abrams, J.; Mann, B. Patterns of care and survival for patients with glioblastoma multiforme diagnosed during 2006. *Neuro Oncol.* 2012, 14, 351–359. [CrossRef]

62. Stupp, R.; Taillibert, S.; Kanner, A.; Read, W.; Steinberg, D.M.; Lhermitte, B.; Toms, S.; Idbaih, A.; Ahluwalia, M.S.; Fink, K.; et al. Effect of Tumor-Treating Fields Plus Maintenance Temozolomide vs Maintenance Temozolomide Alone on Survival in Patients With Glioblastoma. *JAMA* 2017, 318, 2306–2316. [CrossRef] [PubMed]

63. Vlassenbroeck, A.G.; Thiessen, B.; Beattie, B.J.; Malkin, M.G.; Blasberg, R.G. Evaluation of early response to SU101 target-based therapy in patients with recurrent supratentorial malignant gliomas using FDG PET and Gd-DTPA MRI. *J. Neurooncol.* 2000, 46, 249–259. [CrossRef] [PubMed]

64. Prados, M.D.; Lamborn, K.R.; Chang, S.; Burton, E.; Butowski, N.; Malec, M.; Kapadia, A.; Rabbitt, J.; Page, M.S.; Fedoroff, A.; et al. Phase I study of erlotinib HCl alone and combined with temozolomide in patients with stable or recurrent malignant gliomas. *Neuro Oncol.* 2006, 8, 67–78. [CrossRef] [PubMed]

65. Cloughesy, T.F.; Kuhn, J.; Robbins, H.I.; Abrey, L.; Wen, P.; Fink, K.; Lieberman, F.S.; Mehta, M.; Chang, S.; Yung, A.; et al. Phase I Trial of Tipifarnib in Patients With Recurrent Malignant Glioma Taking Enzyme-Inducing Antiepileptic Drugs: A North American Brain Tumor Consortium Study. *J. Clin. Oncol.* 2005, 23, 6647–6656. [CrossRef]

66. Chang, S.M.; Kuhn, J.; Wen, P.; Greenberg, H.; Schiff, D.; Conrad, C.; Fink, K.; Robbins, H.I.; Cloughesy, T.; DeAngelis, L.M.; et al. Phase I/pharmacokinetic study of CCI-779 in patients with recurrent malignant glioma on enzyme-inducing antiepileptic drugs. *Investig. New Drugs* 2004, 22, 427–435. [CrossRef] [PubMed]

67. Kreisl, T.N.; Fidler, J.; Kotliarova, S.; Abutman, J.; Albert, P.S.; Kim, L.; Musib, L.; Thornton, D.; Fine, H.A. A phase I/II trial of enzastaurin in patients with recurrent high-grade gliomas. *Neuro Oncol.* 2010, 12, 181–189. [CrossRef] [PubMed]

68. Choucair, A.K.; Levin, V.A.; Gutin, P.H.; Davis, R.L.; Silver, P.; Edwards, M.S.B.; Wilson, C.B. Development of multiple lesions during radiation therapy and chemotherapy in patients with gliomas. *J. Neurosurg.* 1986, 65, 654–658. [CrossRef]

69. Ammialati, M.; Galicich, J.H.; Arbit, E.; Liao, Y. Reoperation in the treatment of recurrent intracranial malignant gliomas. *Neurosurgery* 1987, 21, 607–614. [CrossRef]

70. Loeffler, J.; Alexander, E.; Hochberg, F.H.; Wen, P.Y.; Morris, J.H.; Schoene, W.C.; Siddon, R.L.; Morse, R.H.; Black, P.M. Clinical patterns of failure following stereotactic interstitial irradiation for malignant gliomas. *Int. J. Radiat. Oncol.* 1990, 19, 1455–1462. [CrossRef]

71. Dhermain, F.; De Crevoisier, R.; Parker, F.; Ciocca, C.; Kaliski, A.; Beaudre, A.; Lefkopoulos, D.; Armand, J.-P.; Haie-Meder, C. Récidives dans les tumeurs gliales: place de la radiothérapie [Role of radiotherapy in recurrent gliomas]. *Bull. Cancer* 2004, 91, 883–889.

72. Combs, S.E.; Debus, J.; Schulz-Ertner, D. Radiotherapeutic alternatives for previously irradiated recurrent gliomas. *BMC Cancer* 2007, 7, 167. [CrossRef] [PubMed]
73. Fogh, S.E.; Andrews, D.W.; Glass, J.; Curran, W.; Glass, C.; Champ, C.E.; Evans, J.J.; Hyslop, T.; Pequignot, E.; Downes, B.; et al. Hypofractionated Stereotactic Radiation Therapy: An Effective Therapy for Recurrent High-Grade Gliomas. *J. Clin. Oncol.* 2010, 28, 3048–3053. [CrossRef] [PubMed]

74. Batchelor, T.; Mulholland, P.; Neyns, B.; Nabors, B.; Campron, M.; Wick, A.; Mason, W.; Mikkelsen, T.; Phuphanich, S.; Ashby, L.S.; et al. Phase III Randomized Trial Comparing the Efficacy of Cediranib As Monotherapy, and in Combination With Lomustine, Versus Lomustine Alone in Patients With Recurrent Glioblastoma. *J. Clin. Oncol.* 2013, 31, 3212–3218. [CrossRef] [PubMed]

75. Kong, D.-S.; Lee, J.-I.; Kim, W.S.; Son, M.J.; Lim, D.H.; Lim, S.T.; Park, K.; Kim, J.H.; Eoh, W.; Nam, D.-H. A pilot study of metronomic temozolomide treatment in patients with recurrent temozolomide-refractory glioblastoma. *Oncol. Rep.* 2006, 16, 1117–1121. [CrossRef] [PubMed]

76. Friedman, H.S.; Prados, M.D.; Wen, P.Y.; Mikkelsen, T.; Schiff, D.; Abrey, L.E.; Yung, W.K.; Paleologos, N.; Nicholas, M.K.; Jensen, R.; et al. Bevacizumab Alone and in Combination With Irinotecan in Recurrent Glioblastoma. *J. Clin. Oncol.* 2009, 27, 4733–4740. [CrossRef] [PubMed]

77. Zustovich, F.; Lombardi, G.; Della Puppa, A.; Rotilio, A.; Scienza, R.; Pastorelli, D. A phase II study of cisplatin and temozolomide in heavily pre-treated patients with temozolomide-refractory high-grade malignant glioma. *Anticancer. Res.* 2009, 29, 4275–4279.

78. Reardon, D.A.; Brandes, A.A.; Omuro, A.; Mulholland, P.; Lim, M.; Wick, A.; Baehring, J.; Abluwalia, M.S.; Roth, P.; Bähr, O.; et al. Effect of Nivolumab vs Bevacizumab in Patients With Recurrent Glioblastoma. *JAMA Oncol.* 2020, 6, 1–8. [CrossRef]

79. Halasz, L.M.; Soltys, S.G.; Breneman, J.C.; Chan, M.D.; Laack, N.N.; Minniti, G.; Kirkpatrick, J. Treatment of Gliomas: A Changing Landscape. *Int. J. Radiat. Oncol.* 2017, 98, 255–258. [CrossRef]

80. Briere, T.M.; McAleer, M.F.; Levy, L.B.; Yang, J.N. Sparing of normal tissues with volumetric arc radiation therapy for glioblastoma: single institution clinical experience. *Radiat. Oncol.* 2017, 12, 79. [CrossRef]

81. Shafer, R.; Nichol, A.; Vollans, E.; Fong, M.; Nakano, S.; Moiseenko, V.; Schmuland, M.; Ma, R.; McKenzie, M.; Otto, K. A Comparison of Volumetric Modulated Arc Therapy and Conventional Intensity-Modulated Radiotherapy for Frontal and Temporal High-Grade Gliomas. *Int. J. Radiat. Oncol.* 2010, 76, 1177–1184. [CrossRef]

82. Combs, S.E.; Thilmann, C.; Edler, L.; Debus, J.; Schulz-Ertner, D. Efficacy of Fractionated Stereotactic Reirradiation in Recurrent Gliomas: Long-Term Results in 172 Patients Treated in a Single Institution. *J. Clin. Oncol.* 2005, 23, 8863–8869. [CrossRef] [PubMed]

83. Cho, K.H.; Hall, W.A.; Gerbi, B.J.; Higgins, P.D.; McGuire, W.A.; Clark, H.B. Single dose versus fractionated stereotactic radiotherapy for recurrent high-grade gliomas. *Int. J. Radiat. Oncol.* 1999, 45, 1133–1141. [CrossRef]

84. Wang, T.J.C.; Wu, C.-C.; Jani, A.; Estrada, J.; Ung, T.; Chow, D.; Soun, J.E.; Saad, S.; Qureshi, Y.H.; Gartrell, R.; et al. Hypofractionated radiation therapy versus standard fractionated radiation therapy with concurrent temozolomide in elderly patients with newly diagnosed glioblastoma. *Pr. Radiat. Oncol.* 2016, 6, 306–314. [CrossRef]

85. Tsien, C.; Moughan, J.; Michalski, J.M.; Gilbert, M.R.; Purdy, J.; Simpson, J.; Kresel, J.J.; Curran, W.J.; Diaz, A.; Mehta, M.; et al. Phase I Three-Dimension Conformal Radiation Dose Escalation Study in Newly Diagnosed Glioblastoma: Radiation Therapy Oncology Group Trial 98-03. *Int. J. Radiat. Oncol.* 2009, 73, 699–708. [CrossRef] [PubMed]

86. Cordova, J.S.; Shu, H.-K.G.; Liang, Z.; Gurbani, S.S.; Cooper, L.A.D.; Holder, C.A.; Olson, J.J.; Kairdolf, B.; Schreibmann, E.; Neill, S.G.; et al. Whole-brain spectroscopic MRI biomarkers identify infiltrating margins in glioblastoma patients. *Neuro Oncol.* 2016, 18, 1180–1189. [CrossRef]

87. Cao, Y.; Tseng, C.-L.; Balter, J.M.; Teng, F.; Parmar, H.A.; Sahgal, A. MR-guided radiation therapy: transformative technology and its role in the central nervous system. *Neuro Oncol.* 2017, 19, ii16–ii29. [CrossRef]

88. Schiffer, D.; Annovazzi, L.; Mazzucco, M.; Mellai, M. The Microenvironment in Gliomas: Phenotypic Expressions. *Cancers* 2015, 7, 2352–2359. [CrossRef]

89. Schiffer, D.; Annovazzi, L.; Casalone, C.; Corona, C.; Mellai, M. Glioblastoma: Microenvironment and Niche Concept. *Cancers* 2018, 11, 5. [CrossRef]
90. Mannino, M.; Chalmers, A. Radioresistance of glioma stem cells: Intrinsic characteristic or property of the ‘microenvironment-stem cell unit’? *Mol. Oncol.* 2011, *5*, 374–386. [CrossRef]

91. Jamal, M.; Rath, B.H.; Tsang, P.S.; Camphausen, K.; Tofilon, P.J. The Brain Microenvironment Preferentially Enhances the Radioresistance of CD133+ Glioblastoma Stem-like Cells. *Neoplasia* 2012, *14*, 150–158. [CrossRef]

92. Farace, C.; Oliver, J.A.; Melguizo, C.; Alvarez, P.; Bandiera, P.; Rama, A.R.; Malaguarnera, G.; Ortiz, R.; Madeddu, R; Prados, J. Microenvironmental Modulation of Decorin and Lumican in Temozolomide-Resistant Glioblastoma and Neuroblastoma Stem-Like Cells. *PLoS ONE* 2015, *10*, e0134111. [CrossRef] [PubMed]

93. Charles, N.A.; Holland, E.C.; Gilbertson, R.; Glass, R.; Kettenmann, H. The brain tumor microenvironment. *Glia* 2012, *60*, 502–514. [CrossRef] [PubMed]

94. Lorger, M. Tumor Microenvironment in the Brain. *Cancers* 2012, *4*, 218–243. [CrossRef] [PubMed]

95. Calabrese, C.; Poppleton, H.; Kocak, M.; Hogg, T.L.; Fuller, C.; Hamner, B.; Oh, E.Y.; Gaber, M.W.; Finklestein, D.; Allen, M.; et al. A Perivascular Niche for Brain Tumor Stem Cells. *Cancer Cell* 2007, *11*, 69–82. [CrossRef] [PubMed]

96. Fidoamore, A.; Cristiano, L.; Antonosante, A.; D’Angelo, M.; Di Giacomo, E.; Astarita, C.; Giordano, A.; Ippoliti, R.; Benedetti, E.; Cimini, A. Glioblastoma Stem Cells Microenvironment: The Paracrine Roles of the Niche in Drug and Radioresistance. *Stem Cells Int.* 2016, *2016*, 1–17. [CrossRef] [PubMed]

97. Muz, B.; De La Puente, P.; Azab, F.; Azab, A.K. The role of hypoxia in cancer progression, angiogenesis, metastasis, and resistance to therapy. *Hypoxia* 2015, *3*, 83–92. [CrossRef]

98. Kaur, B.; Khwaja, F.W.; Severson, E.A.; Matheny, S.L.; Brat, D.J.; Van Meir, E.G. Hypoxia and the hypoxia-inducible-factor pathway in glioma growth and angiogenesis1. *Neuro-oncology* 2005, *7*, 134–153. [CrossRef]

99. Li, Z.; Bao, S.; Wu, Q.; Wang, H.; Eyler, C.; Cao, Y.; Lathia, J.; McLendon, R.E.; et al. Hypoxia-Inducible Factors Regulate Tumorigenic Capacity of Glioma Stem Cells. *Cancer Cell* 2009, *15*, 501–513. [CrossRef]

100. Gray, L.H.; Conger, A.D.; Ebert, M.; Hornsey, S.; Scott, O.C.A. The Concentration of Oxygen Dissolved in Tissues at the Time of Irradiation as a Factor in Radiotherapy. *Br. J. Radiol.* 1953, *26*, 638–648. [CrossRef]

101. Marampon, F.; Gravina, G.L.; Zani, B.; Popov, V.M.; Fratticci, A.; Cerasani, M.; Di Genova, D.; Mancini, M.; Ciccarelli, C.; Ficorella, C.; et al. Hypoxia sustains glioblastoma radioresistance through ERKs/DNA-PKcs/HIF-1α functional interplay. *Int. J. Oncol.* 2014, *44*, 2121–2131. [CrossRef]

102. Gustafsson, M.V.; Zheng, X.; Pereira, T.; Gradin, K.; Jin, S.; Lundkvist, J.; Ruas, J.L.; Poellinger, L.; Lendahl, U.; Bondesson, M. Hypoxia Requires Notch Signaling to Maintain the Undifferentiated Cell State. *Dev. Cell* 2005, *9*, 617–628. [CrossRef]

103. Covello, K.L.; Kehler, J.; Yu, H.; Gordan, J.D.; Arsham, A.M.; Hu, C.-J.; Labosky, P.A.; Simon, M.C.; Keith, B. HIF-2 regulates Oct-4: effects of hypoxia on stem cell function, embryonic development, and tumor growth. *Genes Dev.* 2006, *20*, 557–570. [CrossRef] [PubMed]

104. Li, P.; Zhou, C.; Xu, L.; Xiao, H. Hypoxia Enhances Stemness of Cancer Stem Cells in Glioblastoma: An In Vitro Study. *Int. J. Med. Sci.* 2013, *10*, 399–407. [CrossRef] [PubMed]

105. Heddeleton, J.M.; Li, Z.; E McLendon, R.; Hjelmeland, A.B.; Rich, J.N. The hypoxic microenvironment maintains glioblastoma stem cells and promotes reprogramming towards a cancer stem cell phenotype. *Cell Cycle* 2009, *8*, 3274–3284. [CrossRef] [PubMed]

106. Michiels, C.; Tellier, C.; Feron, O. Cycling hypoxia: A key feature of the tumor microenvironment. *Biochim. Biophys. Acta* 2016, *1866*, 76–86. [CrossRef] [PubMed]

107. Hsieh, C.-H.; Lee, C.-H.; Liang, J.-A.; Yu, C.-Y.; Shyu, W.-C. Cycling hypoxia increases U87 glioma cell radioresistance via ROS induced higher and long-term HIF-1 signal transduction activity. *Oncol. Rep.* 2010, *24*, 1629–1636. [CrossRef] [PubMed]

108. Gérard, M.; Corrozier-Dulmont, A.; Lesueur, P.; Collet, S.; Chérel, M.; Bourgeois, M.; Stefan, D.; Limkin, E.J.; Perrio, C.; Guillamo, J.-S.; et al. Hypoxia Imaging and Adaptive Radiotherapy: A State-of-the-Art Approach in the Management of Glioma. *Front. Mol. Biol.* 2019, *6*, 117. [CrossRef]

109. McGee, M.C.; Hamner, J.B.; Williams, R.F.; Rosati, S.F.; Sims, T.L.; Ng, C.Y.; Gaber, M.W.; Calabrese, C.; Wu, J.; Nathwani, A.C.; et al. Improved Intratumoral Oxygenation Through Vascular Normalization Increases Glioma Sensitivity to Ionizing Radiation. *Int. J. Radiat. Oncol.* 2010, *76*, 1537–1545. [CrossRef]

110. Hanahan, D.; Weinberg, R.A. Hallmarks of Cancer: The Next Generation. *Cell* 2011, *144*, 646–674. [CrossRef]
111. Venneti, S.; Thompson, C.B. Metabolic Reprogramming in Brain Tumors. Annu. Rev. Pathol. Mech. Dis. 2017, 12, 515–545. [CrossRef]

112. Zhou, W.; Wahl, D.R. Metabolic Abnormalities in Glioblastoma and Metabolic Strategies to Overcome Treatment Resistance. Cancers 2019, 11, 1231. [CrossRef] [PubMed]

113. Libby, C.J.; Tran, A.N.; Scott, S.E.; Griguer, C.; Hjelmeland, A.B. The pro-tumorigenic effects of metabolic alterations in glioblastoma including brain tumor initiating cells. Biochim Biophys Acta Rev. Cancer 2018, 1869, 175–188. [CrossRef] [PubMed]

114. Duman, C.; Yaqubi, K.; Hoffmann, A.; Acikgöz, A.A.; Korshunov, A.; Bendszus, M.; Herold-Mende, C.; Liu, H.-K.; Alfonso, J. Acyl-CoA-Binding Protein Drives Glioblastoma Tumorigenesis by Sustaining Fatty Acid Oxidation. Cell Metab. 2019, 30, 274–289.e5. [CrossRef]

115. Oliva, C.R.; Halloran, B.; Hjelmeland, A.B.; Vazquez, A.; Bailey, S.M.; Sarkaria, J.N.; Griguer, C.E. Griguer, IGFBP6 controls the expansion of chemoresistant glioblastoma through paracrine IGF2/IGF1R signaling. Cell Commun. Signal 2018, 16, 61. [CrossRef] [PubMed]

116. Oliva, C.R.; Markert, T.; Ross, L.J.; White, E.L.; Rasmussen, L.; Zhang, W.; Everts, M.; Moellering, D.; Bailey, S.M.; Suto, M.J.; et al. Identification of Small Molecule Inhibitors of Human Cytochrome Oxidase That Target Chemoresistant Glioma Cells. J. Biol. Chem. 2016, 291, 24188–24199. [CrossRef] [PubMed]

117. Oliva, C.R.; Zhang, W.; Langford, C.; Suto, M.J.; Griguer, C.E. Repositioning chlorpromazine for treating chemoresistant glioma through the inhibition of cytochrome c oxidase bearing the COX4-1 regulatory subunit. Oncotarget 2017, 8, 37568–37583. [CrossRef]

118. Oliva, C.R.; Moellering, D.; Gillespie, G.Y.; Griguer, C.E. Acquisition of Chemoresistance in Gliomas Is Associated with Increased Mitochondrial Coupling and Decreased ROS Production. PLoS ONE 2011, 6, e24665. [CrossRef]

119. Oliva, C.R.; Nozell, S.E.; Diers, A.; McClugage, S.G.; Sarkaria, J.N.; Markert, J.M.; Darley-Usmar, V.M.; Bailey, S.M.; Gillespie, G.Y.; Landar, A.; et al. Acquisition of Temozolomide Chemoresistance in Gliomas Leads to Remodeling of Mitochondrial Electron Transport Chain. J. Biol. Chem. 2010, 285, 39759–39767. [CrossRef]

120. Wolf, A.; Agnihotri, S.; Micallef, J.; Mukherjee, J.; Sabha, N.; Cairns, R.; Hawkins, C.; Guha, A. Hexokinase 2 is a key mediator of aerobic glycolysis and promotes tumor growth in human glioblastoma multiforme. J. Cell Biol. 2011, 192, 313–326. [CrossRef]

121. Vartanian, A.; Agnihotri, S.; Wilson, M.R.; Burrell, K.E.; Tonge, P.D.; Alamsahebpour, A.; Jalali, S.; Taccone, M.S.; Mansouri, S.; Golbourn, B.; et al. Targeting hexokinase 2 enhances response to radio-chemotherapy in glioblastoma. Oncotarget 2016, 7, 69518–69535. [CrossRef]

122. Calvert, A.E.; Chalastanis, A.; Wu, Y.; Hurley, L.A.; Kouri, F.M.; Bi, Y.; Kachman, M.; May, J.L.; Bartom, E.T.; Hua, Y.; et al. Cancer-Associated IDH1 Promotes Growth and Resistance to Targeted Therapies in the Absence of Mutation. Cell Rep. 2017, 19, 1858–1873. [CrossRef] [PubMed]

123. Wahl, D.R.; Dresser, J.; Wilder-Romans, K.; Parsels, J.D.; Zhao, S.G.; Davis, M.; Zhao, L.; Kachman, M.; Wernisch, S.; Burant, C.F.; et al. Glioblastoma Therapy Can Be Augmented by Targeting IDH1-Mediated NADPH Biosynthesis. Cancer Res. 2016, 77, 960–970. [CrossRef] [PubMed]

124. You, W.-C.; Chiuo, S.-H.; Huang, C.-Y.; Chiang, S.-F.; Yang, C.-L.; Sudhakar, J.N.; Lin, T.-Y.; Chiang, I.-P.; Shen, C.-C.; Cheng, W.-Y.; et al. Mitochondrial protein ATPase family, AAA domain containing 3A correlates with radioresistance in glioblastoma. Neuro Oncol. 2013, 15, 1342–1352. [CrossRef] [PubMed]

125. Peña-Rico, M.A.; Calvo-Vidal, M.N.; Villalonga-Planells, R.; Martinez-Soler, F.; Gimenez-Bonafe, P.; Navarro-Sabaté, À.; Tortosa, A.; Bartrons, R.; Manzano, A. TP53 induced glycolysis and apoptosis regulator (TIGAR) knockdown results in radiosensitization of glioma cells. Radiother. Oncol. 2011, 101, 132–139. [CrossRef]

126. Visvader, J.E.; Lindeman, G.J. Cancer stem cells in solid tumours: accumulating evidence and unresolved questions. Nat. Rev. Cancer 2008, 8, 755–768. [CrossRef]

127. Rosen, J.M.; Jordan, C.T. The Increasing Complexity of the Cancer Stem Cell Paradigm. Science 2009, 324, 1670–1673. [CrossRef]

128. Ignatova, T.N.; Kukekov, V.G.; Laywell, E.D.; Suslov, O.N.; Vrionis, F.D.; Steindler, D.A. Human cortical glial tumors contain neural stem-like cells expressing astroglial and neuronal markers in vitro. Glia 2002, 39, 193–206. [CrossRef]
Cancers 2020, 12, 2511

129. Hemmati, H.D.; Nakano, I.; Lazareff, J.A.; Masterman-Smith, M.; Geschwind, D.H.; Bronner, E.M.; Kornblum, I.H. Cancerous stem cells can arise from pediatric brain tumors. *Proc. Natl. Acad. Sci. 2003*, *100*, 15178–15183. [CrossRef]

130. Singh, S.K.; Clarke, I.D.; Terasaki, M.; Bonn, E.V.; Hawkins, C.; Squire, J.; Dirks, P.B. Identification of a cancer stem cell in human brain tumors. *Cancer Res. 2003*, *63*, 5821–5828.

131. Lathia, J.D.; Gallagher, J.; Heedleston, J.M.; Wang, J.; Eyler, C.E.; MacSwords, J.; Wu, Q.; Vasanji, A.; McLendon, R.E.; Hjelmeland, A.B.; Dewhirst, M.W.; Bigner, D.D.; Rich, J.N. Glial stem cells promote radioresistance by preferential activation of the DNA damage response. *Nature 2006*, *444*, 756–760. [CrossRef]

132. Chen, J.; Li, Y.; Yu, T.-S.; McKay, R.M.; Kernie, S.G.; Parada, L.F. A restricted cell population propagates glioblastoma growth after chemotherapy. *Nature 2012*, *488*, 522–526. [CrossRef] [PubMed]

133. Kim, Y.; Joo, K.M.; Jin, J.; Nam, D.H. Cancer Stem Cells and Their Mechanism of Chemo-Radiation Resistance. *Int. J. Stem Cells 2009*, *2*, 109–114. [CrossRef] [PubMed]

134. Ligon, K.L.; Huillard, E.; Mehta, S.; Kesari, S.; Liu, H.; Alberta, J.A.; Bachoo, R.M.; Kane, M.; Louis, D.N.; Depinho, R.A.; et al. Olig2-Regulated Lineage-Restricted Pathway Controls Replication Competence in Neural Stem Cells and Malignant Glioma. *Neuron 2007*, *53*, 503–517. [CrossRef]

135. Veselská, R.; Kuglik, P.; Cejpek, P.; Svachova, H.; Neradil, J.; Relichova, J. Nestin expression in the cell lines derived from glioblastoma multiforme. *BMC Cancer 2006*, *6*, 32. [CrossRef] [PubMed]

136. Anido, J.; Sáez-Borderías, A.; González-Juncà, A.; Rodón, L.; Folch, G.; Carmona, M.A.; Prieto-Sánchez, R.M.; Barba, I.; Martínez-Saez, E.; Prudkin, L.; et al. TGF-β Receptor Inhibitors Target the CD44high/Id1high Glioma-Initiating Cell Population in Human Glioblastoma. *Cancer Cell 2010*, *18*, 655–668. [CrossRef]

137. Liu, G.; Yuan, X.; Zeng, Z.; Tunici, P.; Ng, H.; Abdulkadir, I.R.; Lu, L.; Irvin, D.; Black, K.L.; Yu, J.S. Analysis of gene expression and chemoresistance of CD133+ cancer stem cells in glioblastoma. *Mol. Cancer 2006*, *5*, 67. [CrossRef] [PubMed]

138. Smits, G.; Stamenovic, I.; Yu, Q. CD44 attenuates activation of the hippo signaling pathway and is a prime therapeutic target for glioblastoma. *Cancer Res. 2010*, *70*, 2455–2464. [CrossRef]

139. Ogden, A.T.; Waziri, A.E.; Lochhead, R.A.; Fusco, D.; Lopez, K.; Ellis, J.A.; Kang, J.; Assanah, M.; McKhann, G.M.; Sisti, M.B.; et al. Identification of A2B5+/CD133− tumor-initiating cells in adult human gliomas. *Neurosurgery 2008*, *62*, 505–515. [CrossRef] [PubMed]

140. Ong, D.S.T.; Hu, B.; Ho, Y.W.; Sauvé, C.-E.G.; Bristow, C.A.; Wang, Q.; Multani, A.S.; Chen, P.; Nezi, L.; Jiang, S.; et al. PAF promotes stemness and radioresistance of glioma stem cells. *Proc. Natl. Acad. Sci. USA 2017*, *114*, E9086–E9095. [CrossRef] [PubMed]

141. Tawada, M.; Hashimoto, Y.; Sato, K.; Ito, K.; Nakamura, T. Identification of a neural crest cell as a neural stem cell by the expression of CD44. *Cell Stem Cell 2009*, *5*, 67. [CrossRef] [PubMed]

142. Bao, S.; Wu, Q.; McLendon, R.E.; Hao, Y.; Shi, Q.; Hjelmeland, A.B.; Dewhirst, M.W.; Bigner, D.D.; Rich, J.N. Glial stem cells promote radioresistance by preferential activation of the DNA damage response. *Nature 2006*, *444*, 756–760. [CrossRef]

143. Son, M.J.; Woolard, K.; Nam, D.H.; Lee, J.; Fine, H.A. SSEA-1 Is an Enrichment Marker for Tumor-Initiating Cells in Human Glioblastoma. *Cell Stem Cell 2009*, *4*, 444–453. [CrossRef] [PubMed]

144. Xu, Y.; Stamenovic, I.; Yu, Q. CD44 attenuates activation of the hippo signaling pathway and is a prime therapeutic target for glioblastoma. *Cancer Res. 2010*, *70*, 2455–2464. [CrossRef]

145. Ogden, A.T.; Waziri, A.E.; Lochhead, R.A.; Fusco, D.; Lopez, K.; Ellis, J.A.; Kang, J.; Assanah, M.; McKhann, G.M.; Sisti, M.B.; et al. Identification of A2B5+/CD133− tumor-initiating cells in adult human gliomas. *Neurosurgery 2008*, *62*, 505–515. [CrossRef] [PubMed]

146. Ong, D.S.T.; Hu, B.; Ho, Y.W.; Sauvé, C.-E.G.; Bristow, C.A.; Wang, Q.; Multani, A.S.; Chen, P.; Nezi, L.; Jiang, S.; et al. PAF promotes stemness and radioresistance of glioma stem cells. *Proc. Natl. Acad. Sci. USA 2017*, *114*, E9086–E9095. [CrossRef] [PubMed]
148. Wang, W.; Long, L.; Wang, L.; Tan, C.; Fei, X.; Chen, L.; Huang, Q.; Liang, Z.-Q. Knockdown of Cathepsin L promotes radiosensitivity of glioma stem cells both in vivo and in vitro. *Cancer Lett.* 2016, 371, 274–284. [CrossRef]

149. Friedmann-Morvinski, D. Glioblastoma Heterogeneity and Cancer Cell Plasticity. *Crit. Rev. Oncog.* 2014, 19, 327–336. [CrossRef]

150. Patel, A.P.; Tirosch, I.; Trombetta, J.J.; Shalek, A.K.; Gillespie, S.M.; Wakimoto, H.; Cahill, D.; Nahed, B.V.; Curry, W.T.; Martuza, R.L.; et al. Single-cell RNA-seq highlights intratumoral heterogeneity in primary glioblastoma. *Science* 2014, 344, 1396–1401. [CrossRef]

151. Soeda, A.; Hara, A.; Kunisada, T.; Yoshimura, S.-I.; Iwama, T.; Park, D.M. The Evidence of Glioblastoma Heterogeneity. *Sci. Rep.* 2015, 5, 7979. [CrossRef]

152. Singh, S.K.; Hawkins, C.; Clarke, I.D.; Squire, J.A.; Bayani, J.; Hide, T.; Henkelman, R.M.; Cusimano, M.D.; Dirks, P.B. Identification of human brain tumour initiating cells. *Nature* 2004, 432, 396–401. [CrossRef]
[PubMed]

153. Castellino, R.C.; Durden, D.L. Mechanisms of Disease: the PI3K–Akt–PTEN signaling node—an intercept point for the control of angiogenesis in brain tumors. *Nat. Clin. Pr. Neurol.* 2007, 3, 682–693. [CrossRef]
[PubMed]

154. Meyer, M.; Reimand, J.; Lan, X.; Head, R.; Zhu, X.; Kushida, M.; Bayani, J.; Pressey, J.C.; Lionel, A.C.; Dirks, P.B. Identification of human brain tumour initiating cells. *Cancer Cell* 2008, 32, 1807–1812. [CrossRef]
[PubMed]

155. Oba-Shinjo, S.M.; Marie, S.K.N.; et al. Frequent ATRX, CIC, FUBP1 and IDH1 mutations refine the classification of malignant gliomas. *Oncotarget* 2015, 6, 274–284. [CrossRef]
[PubMed]

156. Shapiro, J.R.; Yung, W.K.; Shapiro, W.R. Isolation, karyotype, and clonal growth of heterogeneous glioblastoma cell lines. *Acta Neuropathol.* 1980, 49, 23–30. [CrossRef]
[PubMed]

157. Zalcberg, J.R.; Golgher, D. Molecular Genetics of Glioblastomas. *Cancer Lett.* 2016, 371, 274–284. [CrossRef]

158. Wang, W.; Long, L.; Wang, L.; Tan, C.; Fei, X.; Chen, L.; Huang, Q.; Liang, Z.-Q. Knockdown of Cathepsin L promotes radiosensitivity of glioma stem cells both in vivo and in vitro. *Cancer Lett.* 2016, 371, 274–284. [CrossRef]

159. Tavaré, S. Intratumor heterogeneity in human glioblastoma reflects cancer evolutionary dynamics. *Proc. Natl. Acad. Sci. USA* 2013, 110, 4009–4014. [CrossRef]

160. Wang, Q.; Hu, B.; Hu, X.; Kim, H.; Squatrito, M.; Scarpace, L.; Decarvalho, A.C.; Lyu, S.; Li, P.; Li, Y.; et al. Integrated genomic analysis identifies clinically relevant subtypes of glioblastoma characterized by abnormalities in PDGFRα, IDH1, EGFR, and NF1. *Cancer Cell* 2010, 17, 98–110. [CrossRef]

161. Parsons, D.W.; Jones, S.; Zhang, X.; Lin, J.C.-H.; Leary, R.J.; Angenendt, P.; Mankoo, P.; Carter, H.; Siu, I.-M.; Gallia, G.L.; et al. An Integrated Genomic Analysis of Human Glioblastoma Multiforme. *Science* 2008, 321, 1807–1812. [CrossRef]
[PubMed]

162. Verhaak, R.G.; Hoadley, K.A.; Purdom, E.; Wang, V.; Qi, Y.; Wilkerson, M.D.; Miller, C.R.; Ding, L.; Golub, T.; Mesirov, J.P.; et al. Integrated Genomic Analysis Identifies Clinically Relevant Subtypes of Glioblastoma Characterized by Abnormalities in PDGFRA, IDH1, EGFR, and NF1. *Cancer Cell* 2010, 17, 98–110. [CrossRef]
[PubMed]

163. Sottoriva, A.; Spiteri, I.; Piccirillo, S.G.M.; Touloumis, A.; Collins, V.P.; Marioni, J.C.; Curtis, C.; Watts, C.; Tavaré, S. Intratumor heterogeneity in human glioblastoma reflects cancer evolutionary dynamics. *Proc. Natl. Acad. Sci. USA* 2013, 110, 4009–4014. [CrossRef]

164. Wang, W.; Long, L.; Wang, L.; Tan, C.; Fei, X.; Chen, L.; Huang, Q.; Liang, Z.-Q. Knockdown of Cathepsin L promotes radiosensitivity of glioma stem cells both in vivo and in vitro. *Cancer Lett.* 2016, 371, 274–284. [CrossRef]

165. Frueh, L.; Mohammadi, M.; Grotzer, M.A.; Ewen, M.; Breneman, D.K.; Batist, G.; Burger, P.G.; Chen, X.; Choi, Y.H.; De Smedt, C.; et al. Glioblastoma Stem Cells Deregulate Cellular Metabolism in Glioma. *Neuro Oncol.* 2014, 16, 1363–1372. [CrossRef] [PubMed]

166. Hartmann, C.; Hentschel, B.; Wick, W.; Capper, D.; Felsberg, J.; Simon, M.; Westphal, M.; Schackert, G.; Meyermann, R.; Pietsch, T.; et al. Patients with IDH1 wild type anaplastic astrocytomas exhibit worse prognosis than IDH1-mutated glioblastomas, and IDH1 mutation status accounts for the unfavorable prognostic effect of higher age: implications for classification of gliomas. *Acta Neuropathol.* 2010, 120, 707–718. [CrossRef]

167. Jiao, Y.; Killela, P.J.; Reitman, Z.J.; Rasheed, B.A.; Heaphy, C.M.; De Wilde, R.F.; Rodriguez, F.J.; Rosenberg, S.; Oba-Shinjo, S.M.; Marie, S.K.N.; et al. Frequent ATRX, CIC, FUBP1 and IDH1 mutations refine the classification of malignant gliomas. *Oncotarget* 2012, 3, 709–722. [CrossRef]
168. Yin, N.; Xie, T.; Zhang, H.; Chen, J.; Yu, J.; Liu, F. IDH1-R132H mutation radiosensitizes U87MG glioma cells via epigenetic downregulation of TIGAR. *Onco. Lett.* 2019, 19, 1322–1330. [CrossRef]

169. Pegg, E.A. Mammalian O6-alkylguanine-DNA alkyltransferase: regulation and importance in response to alkylating carcinogenic and therapeutic agents. *Cancer Res.* 1990, 50, 6119–6129.

170. Yang, P.; Zhang, W.; Wang, Y.; Peng, X.; Chen, B.; Qiu, X.; Li, G.; Li, S.; Wu, C.; Yao, K.; et al. IDH mutation and MGMT promoter methylation in glioblastoma: results of a prospective registry. *Oncotarget* 2015, 6, 40896–40906. [CrossRef]

171. Molenaar, R.J.; Verbaan, D.; Lamba, S.; Zanon, C.; Jeuken, J.W.; Boots-Sprenger, S.H.; Wesseling, P.; Hulsebos, T.J.; Troost, D.; Van Tilborg, A.A.; et al. The combination of IDH1 mutations and MGMT methylation status predicts survival in glioblastoma better than either IDH1 or MGMT alone. *Neuro Oncol.* 2014, 16, 1263–1273. [CrossRef]

172. Bartel, B. MicroRNAs: Target Recognition and Regulatory Functions. *Cell* 2009, 136, 215–233. [CrossRef] [PubMed]

173. Fabian, M.R.; Sonenberg, N.; Filipowicz, W. Regulation of mRNA Translation and Stability by microRNAs. *Annu. Rev. Biochem.* 2010, 79, 351–379. [CrossRef] [PubMed]

174. Spizzo, R.; Nicoloso, M.S.; Croce, C.M.; Calin, G.A. SnapShot: MicroRNAs in Cancer. *Cell* 2009, 137, 586–586.e1. [PubMed]

175. Ryan, B.; Robles, A.I.; Harris, C.C. Genetic variation in microRNA networks: the implications for cancer research. *Nat. Rev. Cancer* 2010, 10, 389–402. [CrossRef]

176. Banelli, B.; Forlani, A.; Alleman, G.; Morabito, A.; Pistillo, M.P.; Romani, M. MicroRNA in Glioblastoma: An Overview. *Int. J. Genom.* 2017, 2017, 1–16. [CrossRef]

177. Sana, J.; Busek, P.; Fadrus, P.; Besse, A.; Radova, L.; Vecera, M.; Reguli, S.; Sromova, L.S.; Hilser, M.; Lipina, R.; et al. Identification of microRNAs differentially expressed in glioblastoma stem-like cells and their association with patient survival. *Sci. Rep.* 2018, 8, 2836. [CrossRef]

178. Møller, H.G.; Rasmussen, A.P.; Andersen, H.H.; Johnsen, K.B.; Henriksen, M.; Duroux, M. A Systematic Review of MicroRNA in Glioblastoma Multiforme: Micro-modulators in the Mesenchymal Mode of Migration and Invasion. *Mol. Neurobiol.* 2012, 47, 131–144. [CrossRef]

179. Deng, X.; Ma, L.; Wu, M.; Zhang, G.; Jin, C.; Guo, Y.; Liu, R. miR-124 radiosensitizes human glioma cells by targeting CDK4. *J. Neurooncol.* 2013, 114, 263–274. [CrossRef]

180. Toraih, E.A.; El-Wazir, A.; Abdallah, H.Y.; Tantawy, M.A.; Fawzy, M.S. Deregulated MicroRNA Signature Following Glioblastoma Irradiation. *Cancer Control.* 2019, 26, 1–8. [CrossRef]

181. Li, W.; Guo, F.; Wang, P.; Hong, S.; Zhang, C. miR-221-222 confers radioresistance in glioblastoma cells through activating Akt independent of PTEN status. *Curr. Mol. Med.* 2014, 14, 185–195. [CrossRef]

182. Moskwa, P.; Zinn, P.O.; Choi, Y.E.; Shukla, S.A.; Fendler, W.; Chen, C.C.; Lu, J.; Golub, T.R.; Hjelmeland, A.; Chowdhury, D. A functional screen identifies miRs that induce radioresistance in glioblastomas. *Mol. Cancer Res.* 2014, 12, 1767–1778. [CrossRef] [PubMed]

183. Marampon, F.; Megiorni, F.; Camero, S.; Crescioli, C.; McDowell, H.P.; Sferra, R.; Vetuschi, A.; Pompili, S.; Ventura, L.; De Felice, F.; et al. HDAC4 and HDAC6 sustain DNA double strand break repair and stem-like phenotype by promoting radioreistance in glioblastoma cells. *Cancer Lett.* 2017, 397, 1–11. [CrossRef] [PubMed]

184. Kovalski-Chauvel, A.; Modesto, A.; Gouazé-Andersson, V.; Baricault, L.; Gilhodes, J.; Delmas, C.; Lemarié, A.; Toulas, C.; Cohen-Jonathan-Moyal, E.; Seva, C. Alpha-6 integrin promotes radioresistance of glioblastoma by modulating DNA damage response and the transcription factor Zeb1. *Cell Death Dis.* 2018, 9, 872. [CrossRef] [PubMed]

185. Mukherjee, B.; McEllin, B.; Camacho, C.V.; Tomimatsu, N.; Sirasanagandala, S.; Nanpapag, S.; Hatanpaa, K.J.; Mickey, B.; Madden, C.; Maher, E.; et al. EGFRvIII and DNA double-strand break repair: a molecular mechanism for radioresistance in glioblastoma. *Cancer Res.* 2009, 69, 4252–4259. [CrossRef] [PubMed]

186. Golding, S.E.; Morgan, R.N.; Adams, B.R.; Hawkins, A.J.; Povirk, L.F.; Valerie, K. Pro-survival AKT and ERK signaling from EGFR and mutant EGFRvIII enhances DNA double-strand break repair in human glioma cells. *Cancer Biol. Ther.* 2009, 8, 730–738. [CrossRef] [PubMed]

187. Oliva, C.R.; Markert, T.; Gillespie, G.Y.; Griguer, C.E. Nuclear-encoded cytochrome c oxidase subunit 4 regulates BMI1 expression and determines proliferative capacity of high-grade gliomas. *Oncotarget* 2015, 6, 4330–4344. [CrossRef]
188. Godlewski, J.; Nowicki, M.O.; Bronisz, A.; Williams, S.; Otsuki, A.; Nuovo, G.; Raychaudhury, A.; Newton, H.B.; Chiocca, E.A.; Lawler, S. Targeting of the Bmi-1 Oncogene/Stem Cell Renewal Factor by MicroRNA-128 Inhibits Glioma Proliferation and Self-Renewal. Cancer Res. 2008, 68, 9125–9130. [CrossRef]

189. Lessard, J.; Sauvageau, G. Bmi-1 determines the proliferative capacity of normal and leukaemic stem cells. Nature 2003, 423, 255–260. [CrossRef]

190. Jacobs, J.L.; Kieboom, K.; Marino, S.; A DePinho, R.; Van Lohuizen, M. The oncogene and Polycomb-group gene bmi-1 regulates cell proliferation and senescence through the ink4a locus. Nature 1999, 397, 164–168. [CrossRef]

191. Facchino, S.; Abdouh, M.; Chatoo, W.; Bernier, G. BMI1 Confers Radioresistance to Normal and Cancerous Neural Stem Cells through Recruitment of the DNA Damage Response Machinery. J. Neurosci. 2010, 30, 10096–10111. [CrossRef]

192. Han, X.; Xue, X.; Zhou, H.; Zhang, G. A molecular view of the radioresistance of gliomas. Oncotarget 2017, 8, 100931–100941. [CrossRef] [PubMed]

193. Bolos, V.; Blanco, M.; Medina, V.; Aparicio, G.; Díaz-Prado, S.; Grande, E.; Blanco, M. Notch signalling in cancer stem cells. Clin. Transl. Oncol. 2009, 11, 11–19. [CrossRef]

194. Dontu, G.; Jackson, K.W.; McNicholas, E.; Kawamura, M.J.; Abdallah, W.M.; Wicha, M.S. Role of Notch signaling in cell-fate determination of human mammary stem/progenitor cells. Breast Cancer Res. 2004, 6, 1–11. [CrossRef] [PubMed]

195. Hitoshi, S.; Alexson, T.; Tropepe, V.; Donoviel, D.; Elia, A.J.; Conlon, R.A.; Mak, T.W.; Bernstein, A.; Sabaawy, H.E. Notch pathway molecules are essential for the maintenance, but not the generation, of mammalian neural stem cells. Genes Dev. 2002, 16, 846–858. [CrossRef] [PubMed]

196. Hitoshi, S.; Seaberg, R.M.; Koscik, C.; Alexson, T.; Kusunoki, S.; Kazamawa, T.; Tsuji, S.; Van Der Kooy, D. Primitive neural stem cells from the mammalian epiblast differentiate to definitive neural stem cells under the control of Notch signaling. Genes Dev. 2004, 18, 1806–1811. [CrossRef]

197. Wang, J.; Wakeman, T.P.; Latha, J.D.; Hjelmeland, A.B.; Wang, X.-F.; White, R.R.; Rich, J.N.; Sullenger, B.A. Notch Promotes Radioresistance of Glioma Stem Cells. Stem Cells 2009, 28, 17–28. [CrossRef] [PubMed]

198. Manning, B.D.; Cantley, L.C. AKT/PKB Signaling: Navigating Downstream. Cell 2007, 129, 1261–1274. [CrossRef] [PubMed]

199. Kwiatkowska, A.; Symons, M. Signaling determinants of glioma cell invasion. Adv. Exp. Med. Biol. 2013, 786, 121–141.

200. Li, H.-F.; Kim, J.-S.; Waldman, T. Radiation-induced Akt activation modulates radioresistance in human glioblastoma cells. Radiat. Oncol. 2009, 4, 43. [CrossRef]

201. Mehta, M.; Khan, A.J.; Danish, S.; Haffty, B.G.; Sabaawy, H.E. Radiosensitization of Primary Human Glioblastoma Stem-like Cells with Low-Dose AKT Inhibition. Mol. Cancer Ther. 2015, 14, 1171–1180. [CrossRef] [PubMed]

202. Matsutani, T.; Nagai, Y.; Mine, S.; Murai, H.; Saeki, N.; Iwadate, Y. Akt/protein kinase B overexpression as an accurate prognostic marker in adult diffuse astrocytoma. Acta Neurochir. (Wien) 2009, 151, 263–268. [CrossRef] [PubMed]

203. Chakravarti, A.; Zhai, G.; Suzuki, Y.; Sarkesh, S.; Black, P.M.; Muzikansky, A.; Loeffler, J.S. The prognostic significance of phosphatidylinositol 3-kinase pathway activation in human gliomas. J. Clin. Oncol. 2004, 22, 1926–1933. [CrossRef] [PubMed]

204. Chautard, E.; Loubau, G.; Tchirkov, A.; Chassagne, J.; Vermet-Desroches, C.; Morel, L.; Verrelle, P. Akt signaling pathway: a target for radiosensitizing human malignant glioma. Neuro Oncol. 2010, 12, 434–443. [PubMed]

205. Kao, G.D.; Jiang, Z.; Fernandes, A.M.; Gupta, A.K.; Maity, A. Inhibition of Phosphatidylinositol-3-OH Kinase/Akt Signaling Impairs DNA Repair in Glioblastoma Cells following Ionizing Radiation. J. Biol. Chem. 2007, 282, 21206–21212. [CrossRef] [PubMed]

206. Yang, J.-A.; Liu, B.-H.; Shao, L.-M.; Guo, Z.-T.; Yang, Q.; Wu, L.-Q.; Ji, B.-W.; Zhu, X.-N.; Zhang, S.-Q.; Li, C.-J.; et al. LRIG1 enhances the radiosensitivity of radioresistant human glioblastoma U251 cells via attenuation of the EGFR/Akt signaling pathway. Int. J. Clin. Exp. Pathol. 2015, 8, 3850–3950. [PubMed]

207. Malik, U.; Javed, A. LRIGs: A Prognostically Significant Family with Emerging Therapeutic Competence against Cancers. Curr. Cancer Drug Targets 2016, 17, 3–16. [CrossRef] [PubMed]
208. The Cancer Genome Atlas Research Network. Comprehensive genomic characterization defines human glioblastoma genes and core pathways. *Nature* 2008, 455, 1061–1068. [CrossRef]
209. McCabe, N.; Hanna, C.; Walker, S.M.; Gonda, D.; Li, J.; Wikstrom, K.; Savage, K.I.; Butterworth, K.T.; Chen, C.; Harkin, D.P.; et al. Mechanistic Rationale to Target PTEN-Deficient Tumor Cells with Inhibitors of the DNA Damage Response Kinase ATM. *Cancer Res.* 2015, 75, 2159–2165. [CrossRef]
210. Ma, J.; Benitez, J.A.; Li, J.; Miki, S.; De Albuquerque, C.P.; Galatro, T.; Orellana, L.; Zanca, C.; Reed, R.; Boyer, A.; et al. Inhibition of Nuclear PTEN Tyrosine Phosphorylation Enhances Glioma Radiation Sensitivity through Attenuated DNA Repair. *Cancer Cell* 2019, 33, 816. [CrossRef]
211. Kastenhuber, E.R.; Lowe, S.W. Putting p53 in Context. *Cell* 2005, 123, 783–792. [CrossRef]
212. Wu, J.K.; Ye, Z.; Darras, B.T. Frequency of p53 tumor suppressor gene mutations in human primary brain tumors. *Neurosurgery* 1993, 33, 824–830. [PubMed]
213. Chen, P.; Iavarone, A.; Fick, J.; Edwards, M.; Prados, M.; Israel, M.A. Constitutional p53 mutations associated with brain tumors in young adults. *Cancer Genet. Cytogenet.* 1995, 82, 106–115. [CrossRef]
214. Sherry, M.M.; Reeves, A.; Wu, J.K.; Cochran, B.H. STAT3 is required for proliferation and maintenance of multipotency in glioblastoma stem cells. *Stem Cells* 2009, 27, 2383–2392. [CrossRef]
215. Guryanova, O.A.; Wu, Q.; Cheng, L.; Lathia, J.D.; Huang, Z.; Yang, J.; MacSwords, J.; Eyler, C.E.; McLendon, R.E.; Heddleston, J.M.; et al. Nonreceptor Tyrosine Kinase BMX Maintains Self-Renewal and Tumorigenic Potential of Glioblastoma Stem Cells by Activating STAT3. *Cancer Cell* 2011, 19, 498–511. [CrossRef] [PubMed]
216. Kesanakurti, D.; Chetty, C.; Maddirela, D.R.; Gujrati, M.; Rao, J.S. Essential role of cooperative NF-κB and Stat3 recruitment to ICAM-1 intronic consensus elements in the regulation of radiation-induced invasion and migration in glioma. *Oncogene* 2012, 32, 5144–5155. [CrossRef]
217. Yang, Y.-P.; Chang, Y.-L.; Huang, P.-I.; Chiou, G.-Y.; Tseng, L.-M.; Chiou, S.-H.; Chen, M.-H.; Chen, M.-T.; Shih, Y.-H.; Chang, C.-H.; et al. Resveratrol suppresses tumorigenicity and enhances radiosensitivity in primary glioblastoma tumor initiating cells by inhibiting the STAT3 axis. *J. Cell. Physiol.* 2011, 227, 976–993. [CrossRef]
218. Xie, B.; Zhang, L.; Hu, W.; Fan, M.; Jiang, N.; Duan, Y.; Jiang, D.; Xiao, W.; Fragoso, R.C.; Lam, K.S.; et al. Dual blockage of STAT3 and ERK1/2 eliminates radioresistant GBM cells. *Redox Biol.* 2019, 24, 101189. [CrossRef]
219. Raychaudhuri, P.; Park, H.J. FoxM1: a master regulator of tumor metastasis. *Cancer Res.* 2011, 71, 4329–4333. [CrossRef]
220. Alvarez-Fernández, M.; Medema, R. Novel functions of FoxM1: from molecular mechanisms to cancer therapy. *Front. Oncol.* 2013, 3, 30. [CrossRef]
221. Lee, Y.; Kim, K.H.; Kim, D.G.; Cho, H.J.; Kim, Y.; Rheey, J.; Shin, K.; Seo, Y.J.; Choi, Y.-S.; Lee, J.-I.; et al. FoxM1 Promotes Stemness and Radio-Resistance of Glioblastoma by Regulating the Master Stem Cell Regulator Sox2. *PLoS ONE* 2015, 10, e0137703. [CrossRef] [PubMed]
222. Maachani, U.B.; Shankavaram, U.; Kramp, T.; Tofilon, P.J.; Camphausen, K.; Tandle, A.T. FOXM1 and STAT3 interaction confers radioresistance in glioblastoma cells. *Oncotarget* 2016, 7, 77365–77377. [CrossRef] [PubMed]
223. Ventero, M.P.; Fuentes-Baile, M.; Quereda, C.; Perez-Valeciano, E.; Alenda, C.; Garcia-Morales, P.; Esposito, D.; Dorado, P.; Manuel Barbera, V.; Saceda, M. Radiotherapy resistance acquisition in Glioblastoma. Role of SOCS1 and SOCS3. *PLoS ONE* 2019, 14, e0212581. [CrossRef] [PubMed]
224. Martini, M.; Pallini, R.; Luongo, G.; Cenci, T.; Lucantoni, C.; LaRocca, I.M. Prognostic relevance of SOCS3 hypermethylation in patients with glioblastoma multiforme. *Int. J. Cancer* 2008, 123, 2955–2960. [CrossRef]
225. Reya, T.; Clevers, H. Wnt signalling in stem cells and cancer. *Nature* 2005, 434, 843–850. [CrossRef]
226. Jin, X.; Jeon, H.-Y.; Joo, K.M.; Kim, J.K.; Jin, J.; Kim, S.H.; Kang, B.G.; Beck, S.; Lee, S.J.; Park, A.-K.; et al. Frizzled 4 Regulates Stemness and Invasiveness of Migrating Glioma Cells Established by Serial Intracranial Transplantation. *Cancer Res.* 2011, 71, 3066–3075. [CrossRef]
228. Rossi, M.; Magnoni, L.; Miracco, C.; Mori, E.; Tosi, P.; Pirtoli, L.; Tini, P.; Oliveri, G.; Cosci, E.; Bakker, A. β-catenin and Gli1 are prognostic markers in glioblastoma. *Cancer Biol. Ther.* 2011, 11, 753–761. [CrossRef]

229. Tompa, M.; Kalovits, F.; Nagy, A.; Kalman, B. Contribution of the Wnt Pathway to Defining Biology of Glioblastoma. *Neuro Molecular Med.* 2018, 20, 437–451. [CrossRef]

230. Lee, Y.; Lee, J.-K.; Ahn, S.H.; Lee, J.; Nam, D.-H. WNT signaling in glioblastoma and therapeutic opportunities. *Lab. Investig.* 2015, 96, 137–150. [CrossRef]

231. Mccord, M.; Mukouyama, Y.-S.; Gilbert, M.R.; Jackson, S. Targeting WNT Signaling for Multifaceted Glioblastoma Therapy. *Front. Cell. Neurosci.* 2017, 11, 318. [CrossRef]

232. Kim, Y.; Kim, K.H.; Lee, H.; Yang, H.; Kim, D.; Kang, W.; Jin, J.; Joo, K.M.; Lee, J.; Nam, D.-H. Wnt activation is implicated in glioblastoma radioresistance. In Proceedings of the 103rd Annual Meeting of the American Association for Cancer Research, Chicago, IL, USA, 31 March–4 April 2012. Abstract nr 3458.

233. Dong, Z.; Zhou, L.; Han, N.; Zhang, M.; Lyu, X. Wnt/β-catenin pathway involvement in ionizing radiation-induced invasion of U87 glioblastoma cells. *Strahlenther Onkol.* 2015, 191, 672–680. [CrossRef] [PubMed]

234. Barnett, G.C.; West, C.M.L.; Dunning, A.M.; Elliott, R.M.; Coles, C.E.; Pharoah, P.D.P.; Burnet, N.G. Normal tissue reactions to radiotherapy: towards tailoring treatment dose by genotype. *Nat. Rev. Cancer* 2009, 9, 134–142. [CrossRef] [PubMed]

235. Delaney, G.; Jacob, S.; Featherstone, C.; Barton, M. The role of radiotherapy in cancer treatment. *Cancer* 2005, 104, 1129–1137. [CrossRef] [PubMed]

236. Baskar, R.; Lee, K.A.; Yeo, R.; Yeoh, K.-W. Cancer and Radiation Therapy: Current Advances and Future Directions. *Int. J. Med Sci.* 2012, 9, 193–199. [CrossRef]

237. Marie-Egyptienne, D.T.; Lohse, I.; Hill, R. Cancer stem cells, the epithelial to mesenchymal transition (EMT) and radioresistance: Potential role of hypoxia. *Cancer Lett.* 2013, 341, 63–72. [CrossRef]

238. Krause, M.; Dubrovskova, A.; Linge, A.; Baumann, M. Cancer stem cells: Radioresistance, prediction of radiotherapy outcome and specific targets for combined treatments. *Adv. Drug Deliv. Rev.* 2017, 109, 63–73. [CrossRef]

239. Lalla, R.V.; Treister, N.; Sollecito, T.; Schmidt, B.; Patton, L.L.; Mohammadi, K.; Hodges, J.S.; Brennan, M.T. OraRad Study Group Oral complications at 6 months after radiation therapy for head and neck cancer. *Oral Dis.* 2017, 23, 1134–1143. [CrossRef]

240. Shenoy, M.A.; Singh, B.B. Chemical Radiosensitizers in Cancer Therapy. *Cancer Investig.* 1992, 10, 533–551. [CrossRef]

241. Wang, H.; Mu, X.; He, H.; Zhang, X.-D. Cancer Radiosensitizers. *Trends Pharmacol. Sci.* 2018, 39, 24–48. [CrossRef]

242. Wardman, P. Chemical radiosensitizers for use in radiotherapy. *Clin. Oncol. (R. Coll. Radiol.)* 2007, 196, 397–417. [CrossRef]

243. Hodgkiss, R.; Middleton, R. Enhancement of Misonidazole Radiosensitization by an Inhibitor of Glutathione Biosynthesis. *Int. J. Radiat. Biol. Relat. Stud. Phys. Chem. Med.* 1983, 43, 179–183. [CrossRef] [PubMed]

244. Zhao, Y.; Seefeldt, T.; Chen, W.; Carlson, L.; Stoebner, A.; Hanson, S.; Foll, R.; Matthees, D.P.; Palakurthi, S.; Guan, X. Increase in thiol oxidative stress via glutathione reductase inhibition as a novel approach to enhance cancer sensitivity to X-ray irradiation. *Free. Radic. Biol. Med.* 2009, 47, 176–183. [CrossRef] [PubMed]

245. Bache, M.; Zschornak, M.P.; Passin, S.; Kessler, J.; Wichmann, H.; Kappler, M.; Paschke, R.; Kaluderovic, G.N.; Kommera, H.; Taubert, H.; et al. Increased betulinic acid induced cytotoxicity and radiosensitivity in glioma cells under hypoxic conditions. *Radiat. Oncol.* 2011, 6, 111. [CrossRef]

246. Raleigh, D.R.; Haas-Kogan, D.A. Molecular targets and mechanisms of radiosensitization using DNA damage response pathways. *Futur. Oncol.* 2013, 9, 219–233. [CrossRef]

247. Adams, G.E. Chemical radiosensitization of hypoxic cells. *Br. Med Bull.* 1973, 29, 48–53. [CrossRef] [PubMed]

248. Fowler, J.F.; Adams, E.G.; Denekamp, J. Radiosensitizers of hypoxic cells in solid tumors. *Cancer Treat. Rev.* 1976, 3, 227–256. [CrossRef]

249. Goel, S.; Ni, D.; Cai, W. Harnessing the Power of Nanotechnology for Enhanced Radiation Therapy. *ACS Nano* 2017, 11, 5233–5237. [CrossRef]
250. Kunz-Schughart, L.A.; Dubrovska, A.; Peitzsch, C.; Ewe, A.; Aigner, A.; Schellenburg, S.; Muders, M.H.; Hampel, S.; Cirillo, G.; Lemma, F.; et al. Nanoparticles for radiooncology: Mission, vision, challenges. *Biomater.* 2017, 12, 155–184. [CrossRef]

251. Lawrence, T.S.; Blackstock, A.; McGinn, C. The mechanism of action of radiosensitization of conventional chemotherapeutic agents. *Semin. Radiat. Oncol.* 2003, 13, 13–21. [CrossRef]

252. Wilson, G.D.; Bentzen, S.M.; Harari, P. Biologic Basis for Combining Drugs With Radiation. *Semin. Radiat. Oncol.* 2006, 16, 2–9. [CrossRef]

253. Shewach, D.S.; Lawrence, T.S. Radiosensitization of human solid tumor cell lines with gemcitabine. *Semin. Oncol.* 1996, 23, 65–71. [PubMed]

254. Lawrence, T.S.; Eisbruch, A.; McGinn, C.J.; Fields, M.T.; Shewach, D.S. Radiosensitization by gemcitabine. *Oncol. (Williston Park)* 1999, 13, 55–60. [PubMed]

255. Azria, D.; Jacot, W.; Prost, P.; Culine, S.; Ychou, M.; Lemanski, C.; Dubois, J.-B. Gemcitabine et radiations ionisantes: radiosensibilisation ou association radiochimiothérapie [Gemcitabine and ionizing radiations: radiosensitization or radio-chemotherapy combination]. *Bull. Cancer* 2002, 89, 369–379. [PubMed]

256. Cuneo, K.C.; Mehta, R.K.; Kurapati, H.; Thomas, D.G.; Lawrence, T.S.; Nyati, M.K. Enhancing the Radiation Response in KRAS Mutant Colorectal Cancers using the c-Met Inhibitor Crizotinib. *Transl. Oncol.* 2018, 12, 209–216. [CrossRef]

257. Nolte, E.M.; Joubert, A.; Lakier, R.; Van Rensburg, A.; Mercier, A.E. Exposure of Breast and Lung Cancer Cells to a Novel Estrone Analog Prior to Radiation Enhances Bcl-2-Mediated Cell Death. *Int. J. Mol. Sci.* 2018, 19, 2887. [CrossRef]

258. Rey, S.; Schito, L.; Koritzinsky, M.; Wouters, B. Molecular targeting of hypoxia in radiotherapy. *Adv. Drug Deliv. Rev.* 2017, 109, 45–62. [CrossRef]

259. Horsman, M.R.; Overgaard, J. The impact of hypoxia and its modification of the outcome of radiotherapy. *J. Radiat. Res.* 2016, 57, i90–i98. [CrossRef]

260. Bousquet, P.A.; Meltzer, S.; Sønstevold, L.; Esbensen, Y.; Dueland, S.; Flatmark, K.; Sitter, B.; Bathen, T.F.; Seierstad, T.; Redalen, K.R.; et al. Markers of Mitochondrial Metabolism in Tumor Hypoxia, Systemic Inflammation, and Adverse Outcome of Rectal Cancer. *Transl. Oncol.* 2019, 12, 76–83. [CrossRef]

261. Chapman, J.D.; Webb, R.G.; Borsa, J. Radiosensitization of mammalian cells by p-nitroacetophenone. I. Characterization in asynchronous and synchronous populations. *Int. J. Radiat. Biol. Relat. Stud. Phys. Chem. Med.* 1971, 19, 561–573. [CrossRef]

262. Barker, H.E.; Paget, J.T.E.; Khan, A.A.; Harrington, K.J. The tumour microenvironment after radiotherapy: mechanisms of resistance and recurrence. *Nat. Rev. Cancer* 2015, 15, 409–425. [CrossRef]

263. Overgaard, J.; Hansen, H.S.; Andersen, A.; Højlem-Hansen, M.; Jørgensen, K.; Sandberg, E.; Berthelsen, A.; Hammer, R.; Pedersen, M. Misonidazole combined with split-course radiotherapy in the treatment of invasive carcinoma of the larynx and pharynx: report from the DAHANCA 2 study. *Int. J. Radiat. Oncol.* 1989, 16, 1065–1068. [CrossRef]

264. Urtasun, R.C.; Chapman, J.D.; Feldstein, M.L.; Band, R.P.; Rabin, H.R.; Wilson, A.F.; Marynowski, B.; Starreveld, E.; Shnитka, T. Peripheral neuropathy related to misonidazole: Incidence and pathology. *Br. J. Cancer Suppl.* 1978, 3, 271–275. [PubMed]

265. Takaoka, T.; Shibamoto, Y.; Matsuo, M.; Sugie, C.; Murai, T.; Ogawa, Y.; Miyakawa, A.; Manabe, Y.; Kondo, T.; Nakajima, K.; et al. Biological effects of hydrogen peroxide administered intratumorally with or without irradiation in murine tumors. *Cancer Sci.* 2017, 108, 1787–1792. [CrossRef] [PubMed]

266. Koch, C.J.; Oprysko, P.R.; Shuman, A.L.; Jenkins, W.T.; Brandt, G.; Evans, S. Radiosensitization of hypoxic tumor cells by dodecafluoropentane: a gas-phase perfluorochemical emulsion. *Cancer Res.* 2002, 62, 3626–3629.

267. Harrison, D.K.; Vaupel, P. Heterogeneity in Tissue Oxygenation: From Physiological Variability in Normal Tissues to Pathophysiological Chaos in Malignant Tumours. *Adv. Exp. Med. Biol.* 2014, 812, 25–31.

268. Gallez, B.; Neveu, M.-A.; Danhier, P.; Jordan, B.F. Manipulation of tumor oxygenation and radiosensitivity through modification of cell respiration. A critical review of approaches and imaging biomarkers for therapeutic guidance. *Biochim. Biophys. Acta Bioenerg.* 2017, 1858, 700–711. [CrossRef]
Benej, M.; Hong, X.; Vibhute, S.; Scott, S.; Wu, J.; Graves, E.; Le, Q.-T.; Koong, A.C.; Giaccia, A.J.; Yu, B.; et al. Papaverine and its derivatives radiosensitize solid tumors by inhibiting mitochondrial metabolism. *Proc. Natl. Acad. Sci. USA* **2018**, *115*, 10756–10761. [CrossRef]

Chang, J.E.; Khuntia, D.; Robins, H.I.; Mehta, M.P. Radiotherapy and radiosensitizers in the treatment of glioblastoma multiforme. *Clin. Adv. Hematol. Oncol.* **2007**, *5*, 894–915.

Bayin, N.S.; Ma, L.; Placantonakis, D.G.; Barcellos-Hoff, M.H. Evaluation of Radioresponse and Radiosensitizers in Glioblastoma Organotypic Cultures. In *Glioblastoma*. *Methods in Molecular Biology*; Placantonakis, D., Ed.; Humana Press: New York, NY, USA, 2018; Volume 1741, pp. 171–182.

Sigmond, J.; Honeywell, R.J.; Postma, T.J.; Dirven, C.M.F.; De Lange, S.M.; Van Der Born, K.; Laan, A.C.; Baayen, J.C.A.; Van Groeningen, C.J.; Bergman, A.M.; et al. Gemcitabine uptake in glioblastoma multiforme: potential as a radiosensitizer. *Ann. Oncol.* **2009**, *20*, 182–187. [CrossRef]

Van Nijterik, K.A.; Berg, J.V.; Stalpers, L.J.; LaFleur, M.V.M.; Leenstra, S.; Slotman, B.J.; Hulsebos, T.J.; Sminia, P. Differential Radiosensitizing Potential of Temozolomide in MGMT Promoter Methylated Glioblastoma Multiforme Cell Lines. *Int. J. Radiat. Oncol.* **2007**, *69*, 1246–1253. [CrossRef]

Metro, G.; Fabi, A.; Mirri, M.A.; Vidiri, A.; Pace, A.; Carosi, M.; Russillo, M.; Maschio, M.; Giannarelli, D.; Pellegrini, D.; et al. Phase II study of fixed dose rate gemcitabine as radiosensitizer for newly diagnosed glioblastoma multiforme. *Cancer Chemother. Pharmacol.* **2009**, *65*, 391–397. [CrossRef] [PubMed]

Setua, S.; Oubera, M.; Piccirillo, S.G.M.; Watts, C.; Welland, M. Cisplatin-tethered gold nanospheres for multimodal chemo-radiotherapy of glioblastoma. *Nanoscale* **2014**, *6*, 10865–10873. [CrossRef] [PubMed]

Ruiz van Haperen, V.W.; Veerman, G.; Vermorken, J.B.; Peters, G.J. 2′,2′-Difluoro-deoxycytidine (gemcitabine) incorporation into RNA and DNA of tumour cell lines. *Biochem. Pharmacol.* **1993**, *46*, 762–766. [CrossRef]

Ouedraogo, Z.G.; Müller-Barthélémy, M.; Kemeny, J.-L.; Dedieu, V.; Biau, J.; Khalil, T.; Raœelfils, L.I.; Granzotto, A.; Pereira, B.; Beaudoin, C.; et al. STAT3 Serine 727 Phosphorylation: A Relevant Target to Radiosensitize Human Glioblastoma. *Brain Pathol.* **2015**, *25*, 18–30. [CrossRef]

Lesueur, P.; Chevalier, F.; El-Habr, E.; Junier, M.-P.; Chneiweiss, H.; Castéra, L.; Muller, E.; Stefan, D.; Saintigny, Y. Radiosensitization Effect of Talazoparib, a Parp Inhibitor, on Glioblastoma Stem Cells Exposed to Low and High Linear Energy Transfer Radiation. *Sci. Rep.* **2018**, *8*, 1–12. [CrossRef]

Narayan, R.S.; Gasol, A.; Slangen, P.L.; Cornelissen, F.M.; Lagerweij, T.; Veldman, H.Y.; Dik, R.; Berg, J.V.; Slotman, B.J.; Wurdinger, T.; et al. Identification of MEK162 as a Radiosensitizer for the Treatment of Glioblastoma. *Mol. Cancer Ther.* **2017**, *17*, 347–354. [CrossRef]

Prados, M.D.; Chang, S.M.; Butowski, N.; DeBoer, R.; Parvataneni, R.; Carliner, H.; Kabuubi, P.; Ayers-Ringler, J.; Rabitt, J.; Page, M.; et al. Phase II Study of Erlo tinib Plus Temozolomide During and After Radiation Therapy in Patients With Newly Diagnosed Glioblastoma Multiforme or Gliosarcoma. *J. Clin. Oncol.* **2009**, *27*, 579–584. [CrossRef]

Chinnaiyan, P.; Won, M.; Wen, P.Y.; Rojiani, A.M.; Werner-Wasik, M.; Shih, H.A.; Ashby, L.S.; Yu, H.-H.M.; Stieber, V.; Malone, S.C.; et al. A randomized phase II study of everolimus in combination with chemoradiation in newly diagnosed glioblastoma: results of NRG Oncology RTOG 0913. *Neuro Oncol.* **2017**, *20*, 666–673. [CrossRef]

Krauze, A.V.; Myrehaug, S.D.; Chang, M.G.; Holdford, D.J.; Smith, S.; Shih, J.; Tofilon, P.J.; Fine, H.A.; Camphausen, K. A Phase 2 Study of Concurrent Radiation Therapy, Temozolomide, and the Histone Deacetylase Inhibitor Valproic Acid for Patients With Glioblastoma. *Int. J. Radiat. Oncol.* **2015**, *92*, 986–992. [CrossRef]

Galanis, E.; Anderson, S.K.; Miller, C.R.; Sarkaria, J.N.; Jaeckle, K.; Buckner, J.C.; Ligon, K.L.; Ballman, K.V.; Moore, D.F.; Jr; Nebozyn, M.; et al. Phase I/II trial of vorinostat combined with temozolomide and radiation therapy for newly diagnosed glioblastoma: results of Alliance N0874/ABTC 02. *Neuro Oncol.* **2018**, *20*, 546–556. [CrossRef]

Lee, E.Q.; Kaley, T.J.; Duda, D.G.; Schiff, D.; Lassman, A.B.; Wong, E.T.; Mikkelsen, T.; Purow, B.W.; Muzikansky, A.; Ancukiewicz, M.; et al. A Multicenter, Phase II, Randomized, Noncomparative Clinical Trial of Radiation and Temozolomide with or without Vandetanib in Newly Diagnosed Glioblastoma Patients. *Clin. Cancer Res.* **2015**, *21*, 3610–3618. [CrossRef] [PubMed]
285. Butowski, N.; Chang, S.M.; Lamborn, K.R.; Polley, M.; Pieper, R.; Costello, J.F.; Vandenberg, S.; Parvataneni, R.; Nicole, A.; Sneed, P.K.; et al. Phase II and pharmacogenomics study of enzastaurin plus temozolomide during and following radiation therapy in patients with newly diagnosed glioblastoma multiforme and gliosarcoma. *Neuro Oncol.* **2011**, *13*, 1331–1338. [CrossRef] [PubMed]

286. Grossman, S.A.; Ye, X.; Chamberlain, M.; Mikkelsen, T.; Batchelor, T.; Desideri, S.; Piantadosi, S.; Fisher, J.; Fine, H.A. Talapampanel With Standard Radiation and Temozolomide in Patients With Newly Diagnosed Glioblastoma: A Multicenter Phase II Trial. *J. Clin. Oncol.* **2009**, *27*, 4155–4161. [CrossRef] [PubMed]

287. Carlson, B.L.; Grogan, P.T.; Mladek, A.C.; Schroeder, M.A.; Kitange, G.J.; Decker, P.A.; Giannini, C.; Wu, W.; Ballman, K.V.; James, C.D.; et al. Radiosensitizing Effects of Temozolomide Observed in vivo only in a Subset of O6-Methylguanine-DNA Methyltransferase Methylated Glioblastoma Multiforme Xenografts. *Int. J. Radiat. Oncol.* **2009**, *75*, 212–219. [CrossRef]

288. Kong, X.-T.; Nguyen, N.T.; Choi, Y.J.; Zhang, G.; Nguyen, H.N.; Filka, E.; Green, S.; Yong, W.H.; Liu, L.M.; Green, R.M.; et al. Phase II Study of Bortezomib Combined With Temozolomide and Regional Radiation Therapy for Upfront Treatment of Patients With Newly Diagnosed Glioblastoma Multiforme: Safety and Efficacy Assessment. *Int. J. Radiat. Oncol.* **2018**, *100*, 1195–1203. [CrossRef]

289. Jue, T.R.; Nozue, K.; Lester, A.; Joshi, S.; Schroder, L.B.W.; Whittaker, S.P.; Nixdorf, S.; Rapkins, R.W.; Khosraw, M.; McDonald, K.L. Veliparib in combination with radiotherapy for the treatment of MGMT unmethylated glioblastoma. *J. Transl. Med.* **2017**, *15*, 61. [CrossRef]

290. Sarcar, B.; Kahali, S.; Prabhu, A.H.; Shumway, S.D.; Xu, Y.; DeMuth, T.; Chinnaiyan, P. Targeting radiation-induced G2 checkpoint activation with the Wee-1 inhibitor MK-1775 in glioblastoma cell lines. *Mol. Cancer Ther.* **2011**, *10*, 2405–2414. [CrossRef]

291. Ye, H.; Chen, M.; Cao, F.; Huang, H.; Zhan, R.; Zheng, X. Chloroquine, an autophagy inhibitor, potentiates radiosensitivity of glioma initiating cells by inhibiting autophagy and activating apoptosis. *BMC Neurol.* **2016**, *16*, 178. [CrossRef]

292. Allen, B.G.; Bodeker, K.L.; Smith, M.C.; Monga, V.; Sandhu, S.; Hohl, R.J.; Carlisle, T.L.; Brown, H.; Hollenbeck, N.J.; Vollstedt, S.; et al. First-in-Human Phase I Clinical Trial of Pharmacologic Ascorbate Combined with Radiation and Temozolomide for Newly Diagnosed Glioblastoma. *Clin. Cancer Res.* **2019**, *25*, 6590–6597. [CrossRef]

293. Oronsky, B.; Seicinski, J.; Ning, S.; Peehl, D.; Oronsky, A.; Cabrales, P.; Bednarski, M.; Knox, S. RRX-001, a novel dinitroazetidine radiosensitizer. *Investig. New Drugs* **2016**, *34*, 371–377. [CrossRef] [PubMed]

294. Brachman, D.G.; Pugh, S.L.; Ashby, L.S.; Thomas, T.A.; Dunbar, E.M.; Narayan, S.; Robins, H.I.; Muro, K.; Helenowski, I.; Grimm, S.A.; Marymont, M.; Chandler, J.P.; Muro, K.; Newman, S.B.; Levy, R.M.; Jovanovic, B.; McCarthy, K.; Rockhill, J.K.; Won, M.; et al. Phase I/II trials of Temozolomide, Motexafin Gadolinium, and 60-Gy fractionated radiation for newly diagnosed supratentorial glioblastoma multiforme: final results of RTOG 0513. *Int. J. Radiat. Oncol.* **2015**, *89*, 961–967. [CrossRef] [PubMed]

295. Graham, K.; Unger, E.C. Overcoming tumor hypoxia as a barrier to radiotherapy, chemotherapy and immunotherapy in cancer treatment. *Int. J. Nanomed.* **2018**, *13*, 6049–6058. [CrossRef] [PubMed]

296. Gainer, J.L.; Sheehan, J.P.; Larner, J.M.; Jones, D.R. Trans sodium crocetinate with temozolomide and radiation therapy for glioblastoma multiforme. *J. Neurosurg.* **2017**, *126*, 460–466. [CrossRef]

297. Grimm, S.A.; Marymont, M.; Chandler, J.P.; Muro, K.; Newman, S.B.; Levy, R.M.; Jovanovic, B.; McCarthy, K.; Raizer, J.J. Phase I Study of arsenic trioxide and temozolomide in combination with radiation therapy in patients with malignant gliomas. *J. Neuro-Oncology* **2012**, *110*, 237–243. [CrossRef]

298. Kunthekar, P.; Grimm, S.; Chandler, J.; Mehta, M.; Marymont, M.; Levy, R.; Muro, K.; Helenowski, I.; McCarthy, K.; Fountas, L.; et al. A phase II trial of arsenic trioxide and temozolomide in combination with radiation therapy for patients with malignant gliomas. *J. Neurooncol.* **2017**, *133*, 589–594. [CrossRef]

299. Bell, J.B.; Eckerdt, F.; Dhruv, H.D.; Finlay, D.; Peng, S.; Kim, S.; Kroczyńska, B.; Beauchamp, E.M.; Alley, K.; Clymer, J.; et al. Differential Response of Glioma Stem Cells to Arsenic Trioxide Therapy Is Regulated by MNK1 and mRNA Translation. *Mol. Cancer Res.* **2017**, *15*, 460–466. [CrossRef]

300. Takeuchi, S.; Wada, K.; Nagatani, K.; Otani, N.; Osada, H.; Navashiro, H. Sulfasalazine and temozolomide with radiation therapy for newly diagnosed glioblastoma. *Neurol. India* **2014**, *62*, 42. [CrossRef]
303. Carruthers, R.; Ahmed, S.U.; Strathdee, K.; Gomez-Roman, N.; Amoah-Buahin, E.; Watts, C.; Chalmers, A. Abrogation of radioresistance in glioblastoma stem-like cells by inhibition of ATM kinase. *Mol. Oncol.* **2014**, 9, 192–203. [CrossRef]

304. Durant, S.T.; Zheng, L.; Wang, Y.; Chen, K.; Zhang, L.; Zhang, T.; Yang, Z.; Riches, L.; Trinidad, A.G.; Fok, J.H.L.; et al. The brain-penetrant clinical ATM inhibitor AZD1390 radiosensitizes and improves survival of preclinical brain tumor models. *Sci. Adv.* **2018**, 4, eaat1719. [CrossRef] [PubMed]

305. Compter, I.; Eekers, D.; Hoeben, A.; Rouschop, K.; Reymen, B.; Ackermans, L.; Beckervordersantforth, J.; Bauer, N.; Anten, M.; Wesseling, P.; et al. CHLOROBRAIN phase IB trial: The addition of chloroquine, an autophagy inhibitor, to concurrent radiation and temozolomide for newly diagnosed glioblastoma. *Ann. Oncol.* **2019**, 30, v154. [CrossRef]

© 2020 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).