Adult precision medicine: learning from the past to enhance the future

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Abstract

Despite therapeutic advances for other malignancies, gliomas remain challenging solid tumors to treat. Complete surgical resection is nearly impossible due to gliomas' diffuse infiltrative nature, and treatment is hampered by restricted access to the tumors due to limited transport across the blood–brain barrier. Recent advances in genomic studies and next-generation sequencing techniques have led to a better understanding of gliomas and identification of potential aberrant signaling pathways. Targeting the specific genomic abnormalities via novel molecular therapies has opened a new avenue in the management of gliomas, with encouraging results in preclinical studies and early clinical trials. However, molecular characterization of gliomas revealed significant heterogeneity, which poses a challenge for targeted therapeutic approaches. In this context, leading neuro-oncology researchers and clinicians, industry innovators, and patient advocates convened at the inaugural annual Remission Summit held in Orlando, FL in February 2019 to discuss the latest advances in immunotherapy and precision medicine approaches for the treatment of adult and pediatric brain tumors and outline the unanswered questions, challenges, and opportunities that lay ahead for advancing the duration and quality of life for patients with brain tumors. Here, we provide historical context for precision medicine in other cancers, present emerging approaches for gliomas, discuss their limitations, and outline the steps necessary for future success. We focus on the advances in small molecule targeted therapy, as the use of immunotherapy as an emerging precision medicine modality for glioma treatment has recently been reviewed by our colleagues.

Key Points

- For precision medicine to be successful in gliomas, targets should be 'drivers' and drugs must reach targets in therapeutic concentrations.

- Future clinical trial design should include molecular classification, highlight certain biomarkers and promote Bayesian adaptive randomization designs.

Definition

Precision medicine for adult brain tumors is an innovative treatment approach tailored to the genetic profile of both the patient and cancer.[1] Precision medicine approaches for adult brain tumors have yielded some success, specifically in the targeting of BRAF, H3K27 demethylation, and *NTRK* fusions.[2,3] However, spatial and temporal heterogeneity is an important challenge that the brain cancer field must address before precision medicine can be considered a viable option for brain

tumor patients. Spatial heterogeneity has elegantly been described in a published transcriptional atlas,[4] where genomic alterations and gene expression patterns are shown to differ between from the leading edge, infiltrating tumor, cellular tumor, pseudopalisading cells around necrosis, and microvascular proliferation regions of glioblastoma (GBM) resected tumors.[4] Temporal heterogeneity, which describes the changes in gene expression patterns occurring between the time of diagnosis, treatment, and ultimately tumor recurrence, was highlighted when a review of phase III clinical trial (ACT-IV) investigating rindopepimut with temozolomide for patients with newly diagnosed, epidermal growth factor receptor variant III (*EGFRvIII*)-expressing GBM revealed a loss of *EGFRvIII* expression in approximately 60% of recurrent tumor tissues over time, independently of the treatment arm.[5] Other commonly used molecular targets displaying temporal heterogeneity between primary and recurrent GBM include *FGFR-2, FGFR-3, ALK, PDGFRA, PDGFRB*, VEGFR2/KDR, *EGFR*, and *MET*.[6] In GBM isocitrate dehydrogenase (IDH) wild-type tumors, molecular events appear stable in nearly 80% of occurrences, whereas changes to mutational status do frequently occur (nearly 90% of *TERT* and 60% of *EGFR* mutations).[7] It is important that the research and data generated from using precision medicine in the treatment of other cancers be leveraged in the application of precision medicine for adult brain tumors.

Successes in Other Cancers

The success of precision medicine is evident in many cancer types, with therapies targeting *ERBB2* (*HER2*) amplification in breast cancer,[8,9] breakpoint cluster region *(BCR)-ABL* fusion gene in chronic myelogenous leukemia,[10,11] epidermal growth factor receptor (*EGFR*) mutations in non-small-cell lung cancer (NSCLC),[12,13] anaplastic lymphoma kinase (*ALK*) fusions in non-small-cell lung carcinoma,[14,15] and proto-oncogene B-Raf (*BRAF*) mutations in melanoma.[16–18] Identification of these specific genetic alterations have led to the development of standardized treatment plans focused on targeting the protein products of these alterations.[19]

The BRAF kinase inhibitor vemurafenib was shown to improve survival in patients with untreated *BRAF*^{V600E}-mutated metastatic melanoma, with over a 50% response rate.[20] After implementing the *BRAF*^{V600E} inhibitors dabrafenib and vemurafenib into standard practice for the treatment of *BRAF*-mutated melanomas, further studies demonstrated improved efficacy when combining these drugs with inhibition of the downstream target mitogen-activated protein kinase (MAPK). The combinations of dabrafenib plus trametinib, vemurafenib plus cobimetinib, and encorafenib plus binimetanib have shown improved response and delay in acquired resistance in *BRAF*-mutated melanoma patients.[16,21]

Isocitrate dehydrogenase is a particularly attractive target for therapeutic intervention since *IDH* mutations appear to be an early event in carcinogenesis.[22] IDH inhibitors have received positive attention following research in leukemia, in which approximately 10% of patients with acute myeloid leukemia (AML) harbor *IDH1* mutations.

Ivosidenib, an IDH1 oral inhibitor, has been shown to be well tolerated and induce durable remissions in AML patients with poor prognosis.[23] Recently, both ivosidenib and the IDH2 inhibitor enasidenib have received approval from the Food and Drug Administration (FDA) for treatment of refractory *IDH*-mutated AML patients.[24] Ivosidenib and vorasidenib, a brain-penetrant IDH1/2 inhibitor, have both been evaluated in recurrent IDH-mutated gliomas with a significant subset of grade II non-enhancing tumors showing prolonged stable disease.[25] A phase III trial evaluating vorasidenib in newly diagnosed *IDH*-mutated grade II gliomas is currently underway (INDIGO trial).

The approval of EGFR tyrosine kinase inhibitors (TKIs) has allowed a dramatic improvement in clinical outcomes for patients with *EGFR*-mutated NSCLC. EGFR is a cell surface membrane receptor that controls intracellular signal transduction pathways regulating cell proliferation, apoptosis, angiogenesis, adhesion, and motility. Although first-generation (erlotinib, gefitinib) and second-generation (afatinib) TKIs have been standard of care for initial management of *EGFR*-mutant NSCLC, newer data suggest improved outcomes with front-line treatment with the third-generation agent, osimertinib.[26] Osimertinib was approved by the FDA in 2018 as a first-line treatment for people with advanced NSCLC with specific *EGFR* mutations, based on earlier findings from the FLAURA trial showing a median overall survival of 38.6 months among participants in the osimertinib treatment group, compared with 31.8 months in the erlotinib/gefitinib group.[27] In addition, osimertinib has been shown to cross the blood–brain barrier (BBB) with activity demonstrated against brain and leptomeningeal metastases.[28–30]

BRAF, IDH1, and *EGFR* mutations are also frequently found in gliomas. Thus, the benefits of targeting products of these genes in other cancers may provide useful insights for furthering the care of brain tumor patients. However, several limitations, specific to the biology and location of gliomas, have hampered the progress of targeted therapies in these tumors.

Inherent Limitations for Precision Medicine for Gliomas

The success of precision oncology is largely dependent on the identification of targetable biological features in the tumor. This poses a great challenge, especially for high-grade gliomas, as these tumors are inherently heterogeneous. Cells within gliomas differ in their morphology, the underlying gene expression, and genetic mutational landscape.[31–34] Thus, any selected therapeutic target might be expressed by most, but unlikely by all cells within a tumor, leading to incomplete elimination of cancer cells. Genetic, epigenetic, and microenvironmental factors can all contribute to the intratumoral heterogeneity.[35] In glioma, 4 major cellular subtypes (pro-neural, neural, mesenchymal, and classical) have been recently identified.[32] These subpopulations are present at various proportions in different patients' tumors; however, the relevance of this complex biology in the development of therapy resistance is not yet understood. Furthermore, no therapeutic benefit

has been connected to a specific subtype. Glioma cells' ability to shift between these different phenotypes is of particular concern, as it decreases the likelihood of therapeutic success with a single-agent targeted treatment, unless it very efficiently targets an overarching mechanism. In addition, recent studies have highlighted the temporal molecular heterogeneity leading to differential mutational profiles of the tumors at recurrence following treatment, which might be driving universal therapeutic resistance in adult patients with diffuse glioma.[8,36] Therefore, understanding the fundamental biology of glioma at the molecular level is paramount and the relevance of any molecular data has to be discussed with any subsequent treatment inflicting (resistance) pressure.

As mentioned in the previous section, several potential therapeutic targets for gliomas have been successfully pursued in other tumor types. Apart from the cellular heterogeneity issue, some of these drugs have failed trials in gliomas due to insufficient drug penetration into the tumor tissue.[37,38] Thus, overcoming the BBB and ensuring sufficient distribution of the drug will be required for advances in precision neuro-oncology.

Another challenge in effective targeted therapies for glioma is the difficulty of biomarker assessment. Since repeat on-treatment biopsy is not possible, it is hard to evaluate treatment effects at early timepoints, which in other tumor types is a valuable guide for tailoring the treatment to a particular tumor and patient. Thus, for gliomas, longitudinal follow-up after treatment is solely based on radiological imaging, which is not suitable for measuring the effects of treatment every few days. Many of the abovementioned limitations could be overcome, and we discuss in detail the most significant approaches that could drive the field forward below.

Requirements for Success

Identify Targets That Are "Drivers"

CNS tumors were among the first tumor types undergoing molecular characterization by the Cancer Genome Atlas Network.[33,34] Advances in next-generation sequencing (NGS) provided an extensive view of genomic alterations frequently found in these tumors.[39,40] Most brain tumor-related genes play crucial roles in mitogen-activated signal transduction, cellular immortalization, or DNA repair. Highly expressed receptor tyrosine kinases (RTKs), including EGFR and platelet-derived growth factor receptor A (PDGFRA), are products of frequent amplifications in GBM.[40] Despite extensive efforts to use drugs that target these molecules, most have failed to produce durable responses in clinical trials. Even if the drugs can cross the BBB in adequate concentrations, they may not be sufficiently potent to inhibit the targeted molecular pathway. Many of these genes can activate redundant signaling pathways, making drugs directed at single targets insufficient.[41] Moreover, the RTKs are often amplified only in a subset of cells within a tumor, thus generating a heterogeneous mosaic of gene expression.[42]

The identification of mutations that are uniformly present in all cancer cells could have a higher clinical impact when considering targeted therapies for gliomas. One such case is the mutation of *IDH1*, which is present in up to 70% of lower-grade gliomas and most secondary GBMs.[43,44] Upcoming classifications will increasingly consider molecular characteristics in the nomenclature of glioma. Round 2 of the cIMPACT classification updates has recommended re-classifying *IDH*-mutated GBM as astrocytoma, *IDH* mutant, WHO grade IV. The WHO grade IV designation for *IDH*-mutant astrocytoma would involve the presence of necrosis or microvascular proliferation on histology or genetic alteration with *CDKN2A/B* homozygous deletion.[45] Mutations in the *IDH1* gene generate a new catalytic activity of the resulting IDH enzyme, production of 2-hydroxyglutarate, and alterations in the metabolism and epigenetic regulation of these tumor cells.[46] Moreover, these mutations are clonal, suggesting an early event in tumor evolution[47] and providing an attractive opportunity for drug design. The value of targeting mutant *IDH1* is controversial as the early occurrence of *IDH1* mutations may be a "hit-and-run" event[48] that facilitates malignant transformation but may not be essential for the survival of fully malignant cells later in tumor evolution. Studies of IDH inhibitors are ongoing in the glioma population, with the oral IDH1 inhibitor AG120 (ivosidenib) and the IDH1/2 inhibitor AG-881 (vorasidenib) showing favorable safety profiles in phase I clinical trials and a subset of patients with prolonged stable disease.[25,49,50] Early clinical trials of IDH1 inhibitors in glioma have shown some effects,[51] with a high percentage of patients with prolonged stable disease. These promising initial results have led to an ongoing phase III placebo-controlled trial evaluating vorasidenib in *IDH*-mutated grade II gliomas following initial surgery (INDIGO trial, NCT04164901). Another example of a ubiquitous mutation in cancers is a mutation of the *hTERT* promoter,[52] which results in the upregulation of telomere lengthening protein hTERT and epigenetic reprogramming of cancer cells. As it is overexpressed in 90% of all human tumors, and increases cellular aging defense mechanisms, hTERT protein has been extensively studied as a therapeutic target in glioma.[53] However, as in the case of RTKs, the presence of alternative mechanisms for telomere lengthening[54] has rendered the hTERT-targeting agents tested in these trials ineffective. Nonetheless, hTERT remains a potentially promising target and the focus of significant research.

An attractive strategy for more effective treatment of brain tumors has been to test targets for which tailored therapies have been developed against other tumor types. In particular, melanoma and hematologic malignancies have been early adopters of precision medicine, with targets that are relevant to brain tumors. Case reports have suggested a treatment response after targeting *BRAF*-mutated gliomas with BRAF kinase inhibitors that have demonstrated efficacy in the treatment of melanoma. However, the evidence is limited due to a lack of large randomized studies as well as a limited sample size, and it is estimated that less than 3% of high-grade gliomas harbor *BRAF*^{V600E} mutations.[55] A recent phase II study of dabrafenib and trametinib in patients with recurrent

gliomas harboring *BRAF^V600E* mutations has shown durable responses in 29% of patients and 62% in patients with lower-grade gliomas.[56]

How could we identify the potential drug targets that are crucial for the survival of glioma or other CNS tumor cells? While many of the tumor-initiating genetic events and pathways leading to tumor cell proliferation/death have been identified, more research is needed to understand the networks of pathways and the interplay between genetic and epigenetic alterations that drive CNS tumor growth. Inhibiting common downstream signaling pathways, such as proteasomes using the proteasome inhibitor marizomib[57] (NCT03345095) or nuclear export with Selinexor,[58] could be crucial in order to overcome redundant signaling pathways and heterogeneity. In gliomas, communicating networks that render tumor cells resistant against cytotoxic therapies were identified.[59] Synapses on the connections originating from the glioma cells allow tumor cells to retrieve neuronal input that fosters growth.[60] Single-cell sequencing further revealed that primary brain tumors recapitulate many steps of neurodevelopment.[61] Thus, exploring the intersection of cancer biology and neuroscience will open new opportunities for repurposing drugs used in neurological conditions.[62] These include glutamate inhibitors, such as perampanel, which can cross the BBB, and could inhibit glioma cell proliferation. A list of selected published case reports and clinical trial results highlighting targets that are currently being tested in precision medicine for brain tumors is given in Table 1.

In addition, our current insight into new targeted therapies will be significantly broadened by the use of preclinical tumor models that better recapitulate the complexity of brain tumors. Currently employed preclinical glioma models include carcinogen (ethyl-nitrosourea)-induced gliomas in animals, in vitro glioma cell cultures derived from human or animal gliomas, patient-derived and murine glioma xenograft models (subcutaneous, orthotopic), and transgenic mouse models (eg, with conditional expression of oncogenes or loss of tumor-suppressor genes).[63] However, the intratumoral heterogeneity of diffuse gliomas cannot be recapitulated to the fullest in those in vitro or in vivo models. Genetically engineered animal models lack the diversity of the tumor-driving mutations. While patient-derived xenografts retain high levels of heterogeneity, they need to be established in immunocompromised animals, which lack key subsets of immune cells that shape the tumor landscape in human gliomas. In addition, for most therapies, the BBB, which is relatively intact in large parts of diffuse glioma, is a true barrier, and modeling this barrier is a challenging task. Increased efforts in improving patient tumor-derived xenografts models[64] and in vitro cerebral organoid systems,[65,66] as well as progress in microfluidic[67,68] and single-cell technologies[33,34,69,70] that can assist in reproducing crucial aspects of brain tumors such as the BBB and molecular heterogeneity, will allow us to learn more about the interactions between distinct subtypes of glioma cells and their microenvironment. Systems biology and mathematical modeling[71,72] will also help us in finding new, more effective ways to target the dependencies of brain tumors. Although preclinical models do not perfectly recapitulate human gliomas, these models may be very useful if carefully selected based on the targeted therapy to be tested.

Find Currently Available Drugs to Hit the Targets

Novel treatments for gliomas are urgently needed, but the process of developing new agents that reach clinical application is highly time-consuming and expensive. Therefore, drug repurposing has been employed to facilitate and accelerate the discovery of new cancer therapies.[73,74] Several compounds that have been successfully used for non-oncology indications have been repositioned for the treatment of gliomas, based on findings of tumor cell characteristics that might render them sensitive to those therapeutic agents. Those repurposed drugs have key advantages, such as known drug CNS activity, BBB penetration, pharmacokinetics/pharmacodynamics, and clinical safety information.[75] Furthermore, they are less costly than newly marketed drugs and have already been approved for clinical use by regulatory authorities. Antiepileptic drugs, such as valproic acid, levetiracetam, and talampanel, have been investigated, but no favorable survival outcome has been associated with their use in clinical studies for GBM.[76–79] The antidiabetic drug metformin has also been investigated as an adjunct to GBM treatment due to its modulatory effects on metabolism; however, pooled retrospective analyses of clinical trial data have not produced conclusive evidence supporting its use in the treatment of GBM.[80] Disulfiram, a drug used to manage chronic alcoholism, has recently been found to have preclinical activity in glioma[81–84] and have been evaluated in clinical trials (NCT01777919). Mebendazole, an FDA-approved anthelmintic drug, has been shown to exhibit antitumor effects by inhibiting protein kinases and potentially induce microtubule destabilization in preclinical glioma models.[85] Due to mebendazole's efficacy in tumor suppression, and evidence from laboratory studies suggesting that it can help improve the effectiveness of radiotherapy and temozolomide,[86,87] numerous clinical trials have been underway to investigate this possibility (NCT01729260, NCT01837862, and NCT02644291). In addition, as gliomas are a molecularly complex disease with high dynamic heterogeneity,

Table 1. Summary of Targeted Therapies for Gliomas in Completed or Currently Enrolling Clinical Trials

Target	Drug	Clinical Trial
BRAF	Vemurafenib	NCT01524978
RAS/RAF/ MEK/ERK	TAK-580	NCT03429803
	Dabrafenib/Trametinib	NCT02034110, NCT02684058
	Encorafenib/Binimetinib	NCT03973918
EGFR	ABT-414	NCT01800695
H3 K27M	Panobinostat	NCT02717455
IDH1/2	*Vorasidenib* (AG-881)	NCT04164901
VEGF(R)	Bevacizumab	NCT00345163
	Regorafenib	NCT03970447

multi-targeted approaches have been explored, such as in the Coordinated Undermining of Survival Paths by 9 repurposed drugs (CUSP9) trial[88] by the International Initiative for Accelerated Improvement of Glioblastoma Care.[89] The initially proposed regimen consisted of 9 already-marketed drugs that had not been previously used for oncological indications but showed some inhibitory effects on signaling pathways that promote GBM growth. Preliminary results confirming the safety of CUSP9 (version 3) combined with metronomic temozolomide were presented at the Society for Neuro-Oncology meeting in 2018. Whether these regimens have any activity remains to be determined.

Furthermore, the integration of modern OMICS technology, systems biology, and high-throughput drug screens would allow for a more systematic approach and individually tailored, patient-specific therapeutic strategy. This type of approach was explored by Byron et al.[90] in a single-arm clinical trial (NCT02060890). While this study did not show therapeutic efficacy, it demonstrated that a molecularly driven, integrative approach is technically feasible. A more collaborative and transparent setting with respect to industry-generated drugs, granting access to deprioritized drugs and related information, would greatly facilitate and accelerate successful drug repurposing for the treatment of malignant gliomas. An example of such an initiative is the Discovering New Therapeutic Uses for Existing Molecules program, which represents a collaboration between the NIH National Center for Advancing Translational Sciences and pharmaceutical companies such as Astra Zeneca, Janssen Research and Development, LLC, and Pfizer Inc.[91] Moreover, widely accessible comprehensive data libraries integrating results from OMICS analyses as well as molecular and clinical data on available drugs, such as the drug repurposing hub or the ReFRAME library, need to be developed.[92,93] Additional integrative platforms and funding opportunities would further promote drug repurposing and the discovery of novel treatments for malignant gliomas.

Demonstrate That These Drugs Reach the Target in Therapeutic Concentrations

Achieving effective concentrations of anticancer drugs within solid tumors is difficult as these are challenging to measure. In brain tumors, exclusion of therapeutic agents due to the BBB coupled with the limited and likely non-uniform distribution of compounds within the brain and tumor region makes adequate drug delivery a critical challenge.[75] Published reports of in vivo animal and human clinical studies evaluating drug levels and target inhibition in the brain provide an initial resource for evaluating the potential CNS activity of a compound. In addition, in silico predictive models have been developed to aid in predicting BBB penetration.[94] Clinical trials that attempt to assess the extent of drug delivery to targeted GBM tissue typically involve treatment of patients prior to surgery; drug concentration is then measured in a small sample of the contrast-enhancing resected tumor. However, these analyses need to take into account that most of the tumor remaining after surgery is non-enhancing and infiltrative,

likely with different genomic changes and a more intact BBB when compared with the resected tumor.[95] Therefore, understanding the differences in drug distribution between the enhancing and non-enhancing tumor tissue, as well as the potential differences in the biological response to the drug in these distinct regions, is critically important. Future therapeutic planning will thus need to consider BBB penetration of the selected therapy and/or potential methods to circumvent this barrier to effective drug delivery. Another challenge is the fact that the drug concentrations measured are typically total drug concentration and what is likely more relevant is the free-drug concentration. This is hard to measure and requires labor-intensive approaches such as microdialysis or modeling approaches with uncertain reliability.[96]

Modify or Circumvent the BBB to Increase Drug Delivery to the Brain

One method for optimizing the delivery of drugs to gliomas would be to modify or circumvent the BBB. Various techniques have been researched in an effort to increase BBB permeability and optimize treatment success. Magnetic resonance-guided focused ultrasound (MRgFUS), coupled with injected microbubbles, has been used to disrupt the BBB in a way that is both temporary and customizable. Animal studies have shown enhanced drug delivery following MRgFUS, with no axonal or neuronal injury, and a recent phase I study has shown early safety and feasibility, though further investigation is warranted.[97] Another phase I study conducted in recurrent GBM patients recently showed that pulsed ultrasound was well tolerated and may increase the effectiveness of systemic drug therapies, such as carboplatin, in the brain without inducing neurotoxicity.[98] Laser Interstitial Thermal Therapy (LITT) has also been shown to temporarily disrupt the BBB, and research is ongoing to analyze the effects on drug delivery. In 2016, one of the pivotal clinical trials testing these effects reported that BBB permeability increased within 1–2 weeks after LITT, with resolution over 4–6 weeks, suggesting a window of opportunity for drug delivery.[99] Recently, preclinical data evaluating Tumor Treating Fields' effects on the BBB have indicated their potential for increasing BBB permeability, and further studies are being conducted.[100] In addition to physical methods of BBB disruption, several pharmacological approaches have also been tested. Cereport (RMP-7), a bradykinin B2 receptor agonist, transiently increases the permeability of the BBB.[101,102] It has been shown to effectively enhance chemotherapeutic effect when combined with carboplatin,[103] as well as improve the delivery of other drugs into the CNS.[104] More recently, the A2A adenosine receptor signaling pathway has also been targeted to achieve BBB disruption.[105–107] Co-administration of regadenoson, an FDA-approved adenosine receptor agonist used for cardiac stress testing, has been shown to increase temozolomide levels in rat brain by 60%, without affecting plasma concentrations,[108] but had no effect in a pilot study on BBB in normal human CNS.[109] An initial study in GBM patients did not show improvement in temozolomide concentrations in the brain.[110] Whether these approaches disrupt the BBB sufficiently to

significantly enhance the delivery of therapeutic agents remains to be seen. Alternative dosing strategies may be required to maximize the benefit of transient BBB disruption.

Pharmacologic approaches are an important method to circumvent the BBB, especially when intervention with a device is contraindicated due to tumor location and/or size. Transmembrane diffusion and transporters are 2 important pathways for substances to cross the BBB. Transmembrane diffusion is a non-saturable process that is enhanced when a drug is hydrophobic and small in terms of molecular weight. However, one important limitation of a drug's hydrophobicity/high lipid solubility is that its uptake by peripheral tissues is increased, thus decreasing the drug's blood concentration and amount reaching the BBB, in the case of glioma-targeting drugs. Transporter-targeted drug delivery systems have emerged as promising platforms for efficient drug delivery. Transporters use a saturable transport system that allows the uptake of large, hydrophilic substances. Furthermore, transporters for peptides and regulatory proteins can be selective of the areas they target within the central nervous system.[111]

Osmotic BBB opening is a method used to increase drug concentration within the CNS. Hypertonic mannitol is infused into the vessel, with barrier disruption within the tumor evaluated by either contrast-enhanced computed tomography or radionucleotide scanning. Early clinical studies using this method were able to identify increased methotrexate levels in the brain that corresponded with contrast enhancement on imaging.[112] However, several issues with this approach have limited its widespread use. First, the procedure remains labor intensive and invasive, with complications related to catheter use including thrombosis as well as groin hematoma post catheterization. Other issues that arise are related to BBB disruption and include seizures and other neurological symptoms.[113] The costs associated with osmotic BBB opening procedures also remain an issue. In addition, infusion in one vessel is a major limitation, since tumors are not contained within the distribution of only one vessel, and thus multiple vessels must be infused. Lastly, this approach has been tested for several decades, with the initial phase I study performed in 1979, but there have not been significant clinical improvements.[114]

Convection-enhanced delivery (CED) is another mechanism that has been used in GBM treatment that utilizes a pressure gradient to drive fluid flow throughout the tumor. In comparison to diffusion-based mechanisms, CED delivers drugs evenly, at increased quantities, and throughout larger tissue volumes. Limitations of CED include the challenging selection of agents that specifically target the intra- and peritumoral regions while minimizing toxicity to normal cells within the peritumoral region. Challenges also include trajectory planning in the use of more than one cannula to target the intra- and peritumoral regions. Furthermore, cannula design for CED must take into consideration several key principles, which include minimizing injury to normal cells, combining flexibility with increased speed of drug delivery, and optimizing the durability of the device. Improvements in cannula design have allowed for increased flexibility (once the rigid stylet has been removed following insertion) and longer duration for infusion of therapy, as demonstrated by cannulas used in recent clinical trials,[115,116] they are still a limiting factor when considering CED.[117]

Biodegradable polymers have been used in GBM and implanted after surgical tumor debulking to allow local drug delivery, while minimizing systemic side effects.[118] Furthermore, polymeric delivery allows prolonged and controlled exposure to the drug. Further studies are being explored to expand polymeric delivery to various chemotherapies and biologics focusing on novel targets.[119] There remain questions regarding whether this approach allows sufficient diffusion of therapeutic agents into the tumor against bulk flow.

Design Clinical Trials With Goals That Progress Logically

Design and implementation of a clinical trial typically require the full support of a major institution, given the resources required to successfully achieve its goals, which are mainly to determine whether new treatments are both safe and effective. These goals should be reasonably achievable based on existing literature and preclinical data. Progression through the phases of any trial will determine the safety profile of any interventional arm and also allow for the monitoring of toxicity. These data will allow for the quantification of the intervention and increase its broad applicability. Patients should typically be monitored during the trial for biomarkers that are specific for the intervention in question. Finally, in addition to overall survival, logical endpoints should include 6-month progression-free survival as well as durable response rates. As life expectancy increases, greater emphasis on quality of life endpoints will be paramount to ensure meaningful response is achieved.

For clinical trials for gliomas, radiologic endpoints to assess tumor response have been continuously updated by the Response Assessment in Neuro-Oncology (RANO) working groups,[120] which discuss gliomas, brain metastases, and leptomeningeal disease. The earliest RANO working group, RANO-high-grade glioma (RANO-HGG), aimed to address issues surrounding pseudo-progression following completion of fractionated radiation treatment and concomitant chemotherapy, as well as pseudo-response following the use of antiangiogenic treatments affecting vascular permeability.[121] The immunotherapy RANO (iRANO) working group has provided guidance on radiographic assessment for patients receiving immunotherapy to allow for close monitoring with imaging over the first 3 months of starting immunotherapy, when patients typically have radiographic worsening in the setting of clinical stability. This recommendation was developed from observations in other solid tumors that show transient radiographic worsening prior to response, which is similar to the phenomenon of pseudo-progression post-radiation and is often interpreted as immune-mediated inflammatory changes.[121,122] Future directions for RANO include assessment of PET imaging in the management of glioma patients, involving both prognostic information and response to treatment. As a supplement to MRI-based

assessment, PET imaging through amino acid PET tracers *O*-(2-[18F]-fluoroethyl)-ʟ-tyrosine and 3,4-dihydroxy-6-[18F]-fluoro-ʟ-phenylalanine has been shown to accurately determine changes related to progressive tumor, versus treatment effects related to pseudo-progression or delayed radiation-related changes.[121,123] Advances in MR imaging techniques and utilization of PET imaging will play a significant role in identifying radiologic endpoints in future glioma studies.

Next-generation sequencing and molecular testing of gliomas have become more widespread due to a greater emphasis on molecular characterization to be integrated with histological diagnosis, as described in the updated 2016 World Health Organization Classification of Tumors of the Central Nervous System[124] and further elaborated in the next update as already discussed by the cIMPACT approach. Molecular classification may reduce concerns of histological ambiguity in biopsied tissues, while leveraging our understanding of certain biomarkers to be predictive, prognostic, and/or diagnostic.[125]

In addition, a number of recent platform trials with Bayesian adaptive randomization designs, such as the INSIGhT[126] and GBM AGILE[127] trials, are attempting to accelerate the development of novel targeted agents and match biomarkers to specific agents. For the INSIGhT trial, experimental arms are compared with a common control arm of fractionated radiation therapy with concomitant temozolomide, followed by adjuvant temozolomide. The initial stage uses experimental arms with defined genomic biomarkers that are tested simultaneously. The trial evolves as the acquired results alter the randomization probabilities of treatment effects on progression-free survival. The model incorporates biomarker-specific probability and methods incorporate Bayesian estimation. The results that accumulate during the second stage may lead to several treatment arms dropping off the study due to low probability affecting overall survival with potential for new arms being added. GBM AGILE similarly uses 2 stages and employs Bayesian estimation in their statistical method. The initial stage employs adaptive randomization among various treatment arms (considering clinical and biomarker endpoints) to evaluate multiple treatment options simultaneously and move treatments that hold promise into the second stage which utilizes fixed randomization versus control to accelerate the clinical trial process. The same principles are used in platform basket trials, such as the N2M2 trial,[128] that is assigning patients into multiple arms base on tumor genotype.

Summary

Precision medicine offers an individualized treatment plan based on the patient's tumor molecular markers and genetic signature. Tumor heterogeneity must be addressed by identifying targets that are "drivers." Modeling systems using advanced mathematics and computational biology can aid in target identification while inhibition of downstream signaling pathways may address their redundancy. Once drugs that reach the desired target are identified, we must demonstrate effective drug delivery that maintains therapeutic concentration in non-contrast-enhancing tumors. Clinical trials with novel adaptive designs will allow targeted drug testing but maintain flexibility to allow pivoting to different treatment pathways at the earliest time a drug is suspected to be ineffective. Clinical trials remain the preferred recommendation for both newly diagnosed and recurrent GBM patients, as standard-of-care treatment is simply not good enough for the majority of patients due to poor prognosis. Given the lack of competing therapies, NGS panels that can provide information on potentially actionable targets remain a very attractive tool when tailoring treatment for individual patients. However, if the drug cannot reach the desired target in therapeutic concentrations, then the patient may sustain systemic toxicities without a meaningful therapeutic response. Tailored treatments must consider ease of drug delivery, effective therapeutic concentrations, and the adverse effects when assessing efficacy.

Keywords

Funding

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