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Vaccine strategies for glioblastoma: progress and future directions

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Abstract

Recent advances in glioblastoma therapy have led to optimism that more effective therapies will improve outcomes. Immunotherapy is a promising approach that has demonstrated the potential to eradicate cancer cells with cellular-level accuracy while minimizing damage to surrounding healthy tissue. Several vaccination strategies have been evaluated for activity against glioblastoma in clinical trials. These include peptide vaccines, polyvalent dendritic cell vaccines, heat shock protein vaccines and adoptive immunotherapy. In this review, we highlight clinical trials representative of each of these approaches and discuss strategies for integrating these therapies into routine patient care.

Keywords

chemotherapy; glioblastoma; immunotherapy; radiation therapy; tumor-associated antigen; vaccine

Recent advances in surgical techniques and innovations in the development and delivery of adjuvant therapies have improved the prognosis for patients with glioblastoma multiforme (GBM). Nonetheless, the outlook remains poor with a median life expectancy of 20 months [1,2] and a 3-year survival rate of only 10% [3]. Conventional therapies have succeeded in reliably delaying disease progression; however, the potential of these therapies to eradicate GBM is constrained by the tumor's invasiveness, location and resistance to radiation and chemotherapy. Although metastasis outside the CNS is rare [4], GBM infiltrates normal brain tissue well beyond the radiographic borders of the tumor, precluding cure with surgical resection alone [5–7]. Radiation and chemotherapy are valuable adjuvant therapies; however, specific cell populations are chemo- and radio-resistant, resulting in inevitable tumor recurrence [8]. Specifically, failure to eliminate cancer stem cells likely plays a

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critical role in recurrence as this population has the capacity to repopulate the tumor bulk as well as differentiate into critical supporting structures [9,10].

The observation that infection sometimes precedes tumor regression predates modern medicine; however, the first attempts at purposeful manipulation of the immune system to fight cancer is credited to William Coley, who noted regression of a sarcoma following a case of erysipelas (streptococcal infection). Based on this observation, Coley administered killed bacteria vaccines (Coley's Toxins) to a series of cancer patients and reported occasionally impressive results [11].

It was well into the next century, however, before rigorous scientific study of the immune system provided the tools necessary to understand and improve upon Coley's approach. These efforts were introduced into widespread clinical consciousness in 2006 with the US FDA approval of Gardasil® (Merck, NJ, USA), the first preventative anticancer vaccine for cervical cancer. The approval of sipuleucel-T (Provenge®; Dendreon, WA, USA), a dendritic cell vaccine for hormone-resistant metastatic prostate cancer, in 2010 [12] and ipilimumab (Yervoy™; Bristol-Myers Squibb, NY, USA), a monoclonal antibody directed against the immune checkpoint CTLA-4, for metastatic melanoma in 2011 [13]; however, ushered in a new era in cancer immunotherapy, as these are the first active immunologic agents with proven activity against solid tumors.

Bolstered by these recent advances, immunotherapy appears to have arrived as a weapon in the oncology armamentarium. The potential of immunotherapy to target tumors with cellular-level accuracy makes this approach uniquely appealing for eliminating GBM cells that have infiltrated healthy brain tissue. However, there are a number of challenges still facing successful use of immunotherapy against CNS tumors.

Patients with high-grade gliomas have long been known to exhibit alterations in local and systemic immune responses [14,15]. Although an in-depth discussion of the immunosuppressive microenvironment of GBM is beyond the scope of this review, we have reviewed this topic in detail elsewhere [16]. The addition of steroids, radiation therapy and lymphodepleting cytotoxic agents further suppresses immune function [17]. In addition, immunotherapeutic agents may produce delayed and variable radiographic responses, even in patients who exhibit significant clinical benefit [18], complicating evaluation of the effectiveness of an immunologically active agent, and potentially delaying critical clinical decision-making.

Despite these challenges, given the current limitations of GBM therapies and the emergence of immune-based therapies in other cancers, at this juncture it is prudent to take stock of the current state of immunotherapy and envision how this approach might be employed in routine clinical practice. Two distinct sets of challenges must be confronted in accomplishing this goal. First, preclinical research must elucidate and exploit immune targets that reliably and safely generate antitumor responses in the CNS. These therapies must then be implemented against a rapidly progressive disease in an immunologically fragile patient population. This review has two purposes. First, we will highlight select vaccines in clinical trials in order to evaluate the current status of GBM immunotherapy. We

will then draw from preclinical studies and experiences in other tumors to consider how this burgeoning treatment modality might be integrated into the care of GBM patients.

Current approaches

Peptide vaccines

Peptide vaccines involve administration of tumor-associated antigens (TAAs) in a pro-inflammatory context (usually via coadministration of an adjuvant) as a means of priming the immune system against cancer cells. This strategy requires antigen cross-presentation, but is otherwise similar to vaccination strategies commonly implemented in infectious disease [19].

The first step in selectively targeting tumor cells is identifying appropriate TAAs. Ideally, the expression of a targeted TAA is restricted to malignant cells (or malignant cells and nonvital tissues) and plays a critical role in tumor progression. Several TAAs common in other tumors have also been targeted in GBM, including HER-2, TRP-2, gp100, MAGE-1, IL-13Rα2 and AIM-2 [20]. However, owing to many of these antigens also being expressed in normal tissues, T cells directed against these antigens are subject to negative selection in the thymus. Conversely, neoantigens are not subject to negative selection and may represent more ideal immunologic targets.

EGF receptors (EGFRs) have been shown to drive tumor progression in a variety of cancers by regulating cell proliferation, differentiation and survival, as well as modulating downstream signaling pathways involved in invasion and angiogenesis [21,22]. Furthermore, aberrant EGFR activity has been observed in the majority of solid tumors, including GBM [23–29]. In the late 1980s, Burt Vogelstein, along with his postdoctoral fellow Albert Wong, and Darell Bigner codiscovered a variant of EGFR commonly expressed in GBMs [30,31]. This EGFR variant (EGFRvIII) is rarely expressed in normal tissues but is the most common variant of the EGFR in GBM, being expressed in 27–67% of tumors [23,32].

The first trial to demonstrate that targeting EGFRvIII with a peptide vaccine was safe and potentially efficacious was ACTIVATE [33]. This trial utilized a 14-amino acid peptide from the EGFRvIII protein (PEPvIII) conjugated to keyhole limpet hemocyanin. In this study, 18 subjects with immunohistochemical confirmation of EGFRvIII positivity were enrolled and received the vaccine in combination with radiation therapy and temozolomide. Three vaccines were administered in 2-week intervals with the first administered 2 weeks following surgery. After initial dosing, the vaccine was administered once per month until radiographic progression was noted. Median time to progression (14.2 months) and median survival (26 months) in this study compared favorably with historical controls. In addition, immune-based assays performed on sera from patients who received the vaccine showed increased titers of anti-EGFRvIII antibodies, as well as an increase in $CD8^+$, IFNγ-producing T cells. Notably, 82% of tumors that recurred did not express EGFRvIII. The authors attributed this to immunoediting under immunologic pressure and interpreted this finding as support for the immunologic activity of the vaccine. Other authors, however, have

cautioned against this interpretation and noted that independent confirmation of this finding is warranted [34].

While the ACTIVATE trial was ongoing, Stupp *et al.* demonstrated a survival benefit with surgery, radiation and continuous daily temozolomide followed by six cycles of adjuvant temozolomide [3]. The Stupp protocol subsequently became standard of care and was implemented in the ACTIVATE II trial [35]. Twenty-one subjects with confirmation of EGFRvIII expression were enrolled and received the PEPvIII vaccine, CDX-110 (Celldex Therapeutics, MA, USA). The first dose was administered within 6 weeks following completion of chemotherapy and radiation with an additional two doses administered at 2-week intervals. Vaccination was continued at 1-month intervals thereafter until the time of tumor progression. The results of this study corroborated the findings of the ACTIVATE trial as median time to progression was 15.2 months and median overall survival was 23.6 months [35].

A larger Phase II study of CDX-110 is currently in progress [36]. Termed ACT III, this study is a randomized, multicenter clinical trial with 81 patients enrolled. The treatment group received the Stupp protocol in addition to CDX-110, while the control group received the Stupp protocol alone. Based on interim data from the first 40 patients, the authors reported that 70% of patients were progression-free at 5.5 months [36].

There are two clinical trials of CDX-110 that are currently enrolling [201]. ACT IV is a PhaseIII clinical trial of CDX-110 in patients with newly diagnosed GBM. This is a two-arm, double-blind, randomized study in which half of the patients will receive CDX-110, a control cohort will receive a KLH vaccine and both arms will receive standard treatment based on the Stupp protocol. All patients will be followed until death. ReACT is a clinical trial of CDX-110 in patients with recurrent GBM. ReACT is a two-arm, double-blind, Phase II study that will compare the CDX-110 vaccine in combination with bevacizumab with bevacizumab plus keyhole limpet hemocyanin. Patients will be treated until radiographic evidence of disease progression is noted. At the time of progression, treatment with CDX-110 will be terminated and patients will be permitted to receive other therapies. A second group of patients previously refractory to bevacizumab will also be enrolled and will receive bevacizumab plus CDX-110. Additional information on the ACT IV and ReACT trials is available on the Celldex website [201]. Rindopepimut (CDX-110) is being studied in combination with temozolomide in newly diagnosed patients in a European trial currently underway [202]. In this study, 140 total patients will be enrolled. A summary of completed EGFRvIII peptide vaccine trials is provided in Table 1 and ongoing EGFRvIII peptide vaccine trials are summarized in Table 2.

While considerable effort has been invested in peptide vaccines targeting EGFRvIII, it should be noted that many other antigens are also being targeted using peptide vaccines. One particularly interesting vaccine, IMA950, consists of 11 synthetic TAAs and is in early Phase I testing in Europe. This vaccine is unique because nine of the TAAs bind to the HLA class I allele A*02, while two of them bind to HLA class II alleles. The theoretical advantage of this strategy is that both cytotoxic T lymphocytes (CTLs) and T-helper cells are expected to have antitumor activity [37].

Dendritic cell vaccines

Dendritic cells (DCs) are a subset of leukocytes that are derived from $CD34^+$ bone marrow progenitor cells and function primarily in immune surveillance and antigen presentation [38]. DCs are professional APCs, circulating throughout the body and surveying the local environment using receptors for pathogen-associated molecular patterns. Once the immature DC recognizes a protein structure that is unique to a pathogen and becomes activated, the foreign protein is endocytosed and the DC migrates to T-cell rich areas of lymph nodes, where the foreign peptide is presented on MHC class I or II [39]. Once activated, DCs express proinflammatory cytokines and are potent stimulators of both CD4 and CD8 T cells [40]. Clinical trials of DCs pulsed with TAAs have shown some promise, despite heterogeneity in dose, route, location of administration and adjuvant [20,41–50].

The first study to use DCs loaded with the EGFRvIII peptide was the VICTOR I study [51]. In this trial, DCs were preloaded with the EGFRvIII peptide conjugated to KLH. Fifteen patients were initially enrolled, although only 12 received the vaccine as three patients experienced progression during radiotherapy. Three doses were administered within the first 2 weeks following surgery and two additional doses were administered at 2-week intervals thereafter. The results of this study demonstrated that the vaccine was safe and suggested a potential survival benefit as the median time to progression of 18.7 months and median survival of 22.8 months compared favorably with historical controls [51].

The ICT-107 vaccine (Immunocellular Therapeutics, CA, USA) and DC-Vax Brain (Northwest Biotherapeutics, MD, USA) contain DCs loaded with multiple TAAs. The ICT-107 vaccine uses DCs loaded with TRP-2, GP100, HER2, MAGE-1, IL13Rα2 and AIM-2. A Phase I study of this vaccine in newly diagnosed GBM patients reported a progression-free survival of 16.9 months and a median overall survival of 38.4 months [20]. Vaccination with DC-Vax Brain (Northwest Biotherapeutics) included administration of a Toll-like receptor agonist and reported similar results in a Phase I study of DCs pulsed with autologous tumor lysate [44]. Of note, only minor toxicities have been reported in these DC vaccine trials. Despite these studies being underpowered to demonstrate a survival benefit, the results further support the safety and potential efficacy of DC tumor vaccines. A summary of select DC vaccine studies is provided in Table 3. Ongoing and actively recruiting clinical trials, as obtained through the NIH clinical trials database [203], is provided in Table 4.

Heat shock protein vaccines

Heat shock proteins (HSPs) aid in protein folding, regulate apoptosis and modulate immune responses [52–54]. HSPs are overexpressed in the cytoplasm and on the cell surface of GBM cells [55]. This overexpression is likely due to a combination of the hypoxic GBM microenvironment and the high metabolic demands of rapidly proliferating tumor cells [56]. HSPs have been shown to interact with many proteins known to drive tumorigenesis, including EGFR, PDGF receptor, FAK, AKT, p53 and PI3K [57–59].

HSP vaccines are a subclass of protein vaccines that are composed of HSPs bound to tumor peptides. In this setting, the HSP is believed to be a proinflammatory signal while the

peptide provides a specific immunologic target. Evidence for this comes from preclinical studies that have demonstrated that antitumor immunity is specific to HSPs derived from tumor cells rather than normal tissue [60–62]. In addition, neither tumor-derived HSPs nor TAAs alone induced antitumor immunity. However, when TAAs were complexed to HSPs, antitumor immunity was restored. However, when carried by albumin in human serum, the same peptides did not result in antitumor activity [63]. These data suggest that the HSP–peptide complex is required to induce antitumor immunity.

APCs mediate the antitumor immune response in HSP vaccines via interaction with the cell surface receptors CD91, TLR-4 and CD14 [63–68]. Following recognition of the HSP–peptide complex by APCs, TAAs are endocytosed and presented in the context of MHC class I to prime CD8 T cells [69]. In this process, proinflammatory cytokines, including TNF-α, IL-1β, IL-12, IL-6 and GM-CSF, are released from the activated APC, supporting immune cell activation.

The only HSP vaccine with results reported in GBM is the ProPhage (Agenus, MA, USA) vaccine (formerly HSPPC-96), which is composed of the 96 kDa HSP, gp96, complexed to endogenous tumor peptides [70]. This vaccine is manufactured using multistep affinity and nonaffinity chromatography and is currently under clinical investigation for GBM along with a variety of other solid and blood-borne cancers [71–73]. Results of the first Phase I study in patients with recurrent GBM showed promising results (Table 5). In patients receiving four vaccines, 11/12 patients responded with a median survival of 47 weeks. Brain biopsy revealed $CD4^+$, $CD8^+$, $CD56^+$ and IFN-γ-producing cells [74]. A summary of actively recruiting trials for HSP vaccines is also provided in Table 5.

Adoptive immunotherapy

Adoptive immunotherapy involves *ex vivo* activation of autologous immune cells and subsequent infusion back into the patient. This strategy has primarily used lymphocyte-activated killer cells and CTLs. Lymphocyte-activated killer cells are generated by culturing autologous peripheral lymphocytes with IL-2, generating T cells and NK cells. These cells are then injected intra-tumorally or -venously, where they can become activated by host APCs. Clinical trials using adoptive immunotherapy with LAK cells have reported variable toxicity and efficacy (Table 6).

Other studies have used tumor-infiltrating T lymphocytes or T cells from draining lymph nodes (Table 7). In comparison with studies with LAK cells, peripheral transfer of CTLs was associated with only minor toxicities. Conversely, more serious toxicities, including transient cerebral edema and hemorrhage, were reported in studies where CTLs were injected directly into the tumor cavity [75,76].

Implementing immunotherapy for GBM

Experience with conventional cancer therapies suggests that combination therapy is generally superior to monotherapy [3,2]. This principle likely holds true for immunotherapy as well. For example, data from a Phase I trial of combination immunotherapy with ipilimumab and bevacizumab in advanced melanoma suggest that combination therapy can

be administered safely and may be superior to either therapy alone [77]. In addition, tumors may more readily develop resistance against single-agent immunotherapy. As previously mentioned, GBM has been reported to develop resistance in clinical trials of the EGFRvIII peptide vaccine by downregulating expression of the targeted antigen [33]. If this is the case, one strategy for combating immunoediting may be combining EGFRvIII vaccination with antineoplastic therapies with activity against non-EGFRvIII expressing cells. Finally, given the pace of GBM progression and the potential consequences of delaying treatment, upfront, multimodal combination therapy has obvious appeal.

Combination with chemotherapy

Lymphodepletion is a common side effect of many chemotherapeutic agents and is generally considered counterproductive to immunotherapy. However, other effects of chemotherapy include spilling of TAAs [78] and upregulation of pro-inflammatory molecules [79]. In addition, some agents, such as cyclophosphamide, may selectively deplete immunosuppressive T regs [80]. Locally harnessing these proinflammatory effects while avoiding systemic immune suppression has generally proven challenging. For example, combining sipuleucel-T with chemotherapy in patients with metastatic prostate cancer has not demonstrated a reliable benefit in survival or disease control [81].

Trials of some chemoimmunotherapy combinations, however, support the potential efficacy of this approach. For example, a recent trial of ipilimumab plus dacarbazine in 502 patients with metastatic melanoma found that the addition of ipilimumab improved overall survival compared with dacarabzine alone (11.2 vs 9.1 months) and carried a risk of adverse events comparable with ipilimumab alone [82]. A smaller Phase II study compared this combination with ipilimumab and reported a trend toward increased overall survival with combination therapy (11.4 vs 14.3 months) [83].

Preclinical data suggest that local drug delivery may be superior to systemic administration for the treatment of intracranial tumors in combinatorial chemoimmunotherapy regimens. Local delivery of chemotherapy has several theoretical advantages, including circumventing the blood–brain barrier and harnessing local proinflammatory effects while minimizing systemic immune toxicity. For example, a preclinical study found that local delivery of carmustine using biodegradable wafers in combination with local delivery of IL-2 demonstrated a synergistic survival benefit in a rat glioma model [84]. This effect does not seem to be limited to a specific chemotherapy, as biodegradable polymers loaded with a variety of chemotherapeutic agents have been shown to not only improve survival, but also augment the local inflammatory response in the setting of local IL-2 administration [85].

Local delivery of immunotherapy may also be advantageous for some agents. Immune responses in the brain require coordination of local and systemic immune activation [86]; thus, some therapies may be more immunologically active when delivered directly to the CNS than when administered systemically [87,88]. IL-2 provides the most striking example. Preclinical data suggest that intracranial implantation of nonreplicating tumor cells transduced with IL-2 affords protection against intracranial tumor challenge in a murine glioma model; however, this protective effect is not observed when the cells are implanted subcutaneously [89].

There have been no clinical trials of combinatorial chemoimmunotherapy for malignant gliomas to date. Trials of chemotherapy and systemically delivered IL-2 in other tumors, however, have been generally disappointing [90–92]. This is perhaps not surprising in light of the preclinical studies discussed above and the well-established role of autocrine IL-2 signaling in CD8 T-cell function [93]. Of course, in meta-static cancer, local drug delivery is difficult to achieve given the inherent difficulty of targeting occult metastases and circulating tumor cells. In brain tumors, however, a variety of technologies ranging from biodegradable polymers to micro-chips are capable of delivering agents directly to the tumor site [94–96]. Furthermore, retrospective data suggest that local delivery of bis-chloroethylnitrosourea combined with systemic temozolomide is safe and may produce favorable median survival in patients with GBM [1].

Combination with radiation therapy

Radiation therapy has long been a mainstay of GBM treatment for its direct cytotoxic effects, but the immunologic effects of ionizing radiation are only beginning to be understood [97]. The hypothesis that localized radiation therapy can produce systemic antitumor activity (the abscopal effect) dates back to the early 1950s [98]. Clinical documentation of this phenomenon, however, has been scarce. The most convincing evidence for an immune-mediated abscopal effect in humans comes from a recent case report of a patient with metastatic melanoma receiving ipilimumab [99]. This patient received focused radiation therapy to a paraspinal mass and experienced regression of several lesions outside the irradiated field. Interestingly, this response corresponded with serologic evidence of an immune response against the TAA NY-ESO-1.

The abscopal effect has not been documented in primary or metastatic brain tumors. Recently, however, the case of a patient with a previously observed abscopal effect who developed a brain metastasis and underwent focal radiation therapy to this lesion and treatment with ipilimumab was reported [100]. This patient exhibited regression of his extracranial metastases following intracranial radiation therapy with a corresponding increase in anti-MAGEA3 titer and evidence of a new serologic response to PASD1. In this prime-boost scenario, it is difficult to determine if the second response resulted from irradiation of the patient’s intracranial lesion, ipilimumab administration or both. Nevertheless, taken together, these reports suggest that the combination of radiation therapy and ipilimumab may promote offsite tumor regression and, furthermore, provide evidence that radiation therapy has immunologic activity in the CNS. It should be noted that radiation therapy has been associated with a fall in CD4 count in glioma patients [101]. More detailed immunologic evaluation may provide a putative mechanism for this phenomenon, allowing for identification of therapeutic targets.

Combination with surgery

Surgery is the cornerstone of therapy for high-grade gliomas and must be evaluated as an integral part of any prospective immunotherapy regimen. The use of perioperative corticosteroids is an obvious consideration. Further study is needed to delineate the deleterious effects of corticosteroids on specific immunotherapy regimens. This information may then be used to design treatment regimens that minimize these effects. In addition, the

consequences of delaying adjuvant immunotherapy while a patient recovers from surgery must be evaluated. Neo-adjuvant immunotherapy may potentially obviate some of these adverse effects; however, in CNS tumors, the theoretical advantages of neo-adjuvant immunotherapy should be tempered with the potential for iatrogenic edema prior to surgical decompression.

While these are critical practical considerations, it is perhaps more interesting to consider the immunologic effects of surgery itself. For example, in preclinical cancer models, cytoreductive surgery has been demonstrated to restore anti-tumor immune activity to deactivated immune cells [102]. The mechanisms of this response are not well characterized, but may be secondary to increased T-cell trafficking or sudden reduction of antigen load. Surgery also affords direct access to the tumor, facilitating local delivery of immunologically active agents and affording an opportunity to tailor therapies to the immunologic microenvironment of the patient's tumor. One such example is the use of autologous whole-cell lysate to load DCs or HSP vaccines [103]. Another example is the strategy of harvesting tumor-infiltrating lymphocytes, expanding and activating these cells *ex vivo* and infusing them for adoptive T-cell therapy. The latter approach has been evaluated in clinical trails for melanoma, with encouraging results [104].

Future perspective

Immunotherapy is a promising approach in GBM due to its potential to eradicate neoplastic cells while sparing normal tissue and generating durable antitumor activity. In addition, immunotherapy may effectively target specific cell populations [105], an approach that could be expanded to eliminate cells that are resistant to chemotherapy and ionizing radiation. Significant efforts are underway to develop immunologically active agents against GBM, including a number of completed and ongoing clinical trials [16]. The results of these trials will provide further insight into mechanisms of escape and guide the development of future therapies.

We believe that in addition to identifying new immunologic targets and developing therapies to exploit these targets, the next 5–10 years will see a significant effort in understanding the immunologic effects of conventional GBM therapies and developing safe, and potentially powerful combination immunotherapy regimens. Several classes of immunotherapeutic agents are currently in development. In addition to active immunotherapy, adoptive cell transfer and antibodies against immune checkpoints, such as CTLA-4 and PD-1, and other targets with immunologic activity such as VEGF, hold the potential to generate robust antitumor immune responses.

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Executive summary

Current approaches

- The most extensively studied vaccine strategies have been peptide, dendritic cell and heat shock protein vaccines.
- Passive immunotherapies, including adoptive immunotherapy have also been evaluated with variable results.
- EGF receptor vIII is the most frequently targeted glioblastoma multiforme (GBM) neoantigen.
- Other tumor-associated antigens, including TRP-2, GP100, HER2, MAGE-1, IL13Rα2 and AIM-2 have also been targeted in polyvalent vaccines.
- Dendritic cell vaccines for GBM have generally been well tolerated with minimal side effects.
- Although some of these agents have demonstrated some activity against GBM, to-date none have been shown to be superior to current standard of care.

Implementing immunotherapy for GBM

- Combination immunotherapy is likely superior to monotherapy and is currently being tested in clinical trials for CNS and non-CNS malignancies.
- Understanding the immunologic effects of traditional therapies for GBM, such as chemotherapy, ionizing radiation and surgery, will prove critical for integrating immunotherapy into routine clinical practice.
- Local delivery of chemotherapy may be more immunologically advantageous than systemic delivery.
- Radiation has been shown to have both immuno-suppressive and -stimulatory effects. A better understanding of these effects will allow for development of combination regimens in which ionizing radiation is used to augment antitumor immune responses.
- Surgery will likely remain the mainstay of therapy for GBM; however, the immunologic consequences of surgery are poorly understood. Immunotherapies must be integrated into patient care in a manner that does not delay or complicate surgery and ideally takes advantage of postoperative inflammation and access to tumor tissue, including tumor-infiltrating immune cells.

Table 1

Summary of completed trials using EGF receptor vIII peptide vaccines.

Study	Phase	Patients (n)	Antigen	Trial results	Ref.
Sampson *et al*. (2010)	II	18	EGFRvIII	TTP 14.2 months Median survival 26 months	[33]
Del Vecchio *et al.* (2012) (preliminary results only)	II	21	EGFRvIII	Median survival 23.6 months	[35]

EGFR: EGF receptor; TTP: Time to progression.

Table 2

Ongoing clinical trials using peptide vaccines in patients with high-grade glioma.

Study	Phase	Estimated enrollment (n)	Status	Primary outcome	Ref.
Phase II study of CDX-110 in patients with glioblastoma multiforme (ACT III)	II	82	Ongoing	Progression-free survival	[202]
Phase I rindopepimut after conventional radiation in children with diffuse intrinsic pontine gliomas	I	12	Recruiting	Safety	[202]
A study of rindopepimut/GM-CSF in patients with relapsed EGFRvIII-positive glioblastoma (ReACT)	II	95	Recruiting	Progression-free survival	[202]
Phase III study of rindopepimut/GM-CSF in patients with newly diagnosed glioblastoma (ACT IV)	III	440	Recruiting	Overall survival	[202]
A trial investigating the IMA950 vaccine and GM-CSF for glioblastoma	I	45	–	Safety	[202]
An international, randomized, double-blind, controlled Phase II study of rindopepimut/GM-CSF with adjuvant temozolomide in patients with newly diagnosed, surgically resected, EGFRvIII-positive glioblastoma (IRRADIANCE)	II	140	–	Progression-free survival	[203]

EGFR: EGF receptor.

Table 3

Summary of trials using dendritic cell vaccines in patients with glioblastoma multiforme.

Study	Phase	Patients (n)	Number of injected DCs	DC loading	Location of injection	Trial result(s)	Ref.
Phuphanich *et al.* (2012)	I	17	1×10^7	Peptide epitopes: HER2, TRP-2, gp100, MAGE-1, IL13Rα2 and AIM-2. HLA-A1 and/or HLA-A2	ID	PFS: 16.9 months Median survival: 38.4 months	[20]
Iwami *et al.* (2012)	I	8	1×10^7	IL13Rα2	ID	SD: 1 Regression: 1	[106]
Prins *et al.* (2011)	I	23	–	Tumor cell lysate	ID	Median survival: 31.4 months	[44]
Okada *et al.* (2011)	I/II	13	–	Peptide epitopes EphA2, IL-13Rα2, YKL-40 and gp100	ID	SD: 6 CR: 1 PR: 2 Median survival for recurrent GBM: 12 months	[107]
Sampson *et al.* (2009)	I	12	1×10^8	EGFRvIII-specific peptide conjugated to keyhole limpet hemocyanin	ID	TTP: 10.2 months Median survival: 18.7 months	[45]
Wheeler *et al.* (2008)	II	32	1×10^7 to 4×10^7	Tumor cell lysate	ID	TTP: 208 vs 167 days for controls	[47]
De Vleeschouwer *et al.* (2008)	I/II	56	0.7×10^6 to 1×10^6	Tumor cell lysate	ID	12-month OS: 37.4% 24-month OS: 14.8% 36-month OS: 11.1%	[41]
Walker *et al.* (2008)	I	13	1×10^6	Tumor cell lysate	ID	1-year survival: 38%	[46]
Yamanaka *et al.* (2005)	I/II	24	1×10^7	Tumor cell lysate	ID or ID and IT	PR: 1 MR: 3 SD: 10	[49]
Kikuchi *et al.* (2004)	I	15	3.6×10^6 to 32.3×10^6	Tumor cell lysate	ID	PR: 4 NC: 1 MR: 1	[42]
Yamanaka *et al.* (2003)	I/II	10	10×10^6 to 32×10^6	Tumor cell lysate	ID and IT	MR: 2 NC: 4	[48]
Kikuchi *et al.* (2001)	I	8	2.4×10^6 to 8.7×10^6	Tumor cell lysate	ID	SD: 6	[43]
Yu *et al.* (2001)	I	9	1×10^6	MHC class I peptides	ID	Median survival: 455 vs 257 days for controls	[50]

CR: Complete response; DC: Dendritic cell; EGFR: EGF receptor; GBM: Glioblastoma multiforme; ID: Intradermal; IT: Intratumoral; MR: Minor response; NC: No change; OS: Overall survival; PFS: Progression-free survival; PR: Partial response; SD: Stable disease; TTP: Time to progression.

Table 4

Ongoing clinical trials using dendritic cell vaccines in patients with high-grade glioma.

Study	Phase	Estimated enrollment (n)	Status	Primary outcome(s)	Ref.
Phase II feasibility study of DC vaccination for newly diagnosed GBM	II	11	Ongoing	Tumor-specific, cytotoxic T-cell response	[202]
Vaccine therapy in treating patients with malignant glioma	I	8	Ongoing	Dose-limiting toxicity, survival, tumor progression and cellular immune response	[202]
Vaccine therapy in treating patients with newly diagnosed GBM	I	16	Ongoing	Safety and feasibilty	[202]
DC cancer vaccine for high-grade glioma (GBM-Vax)	II	56	Recruiting	PFS	[202]
Safe study of DC-based therapy targeting tumor stem cells in glioblastoma	I/II	20	Recruiting	Adverse events	[202]
Efficacy and safety of autologous DC vaccination in GBM after complete surgical resection	II	37	Recruiting	PFS	[202]
Vaccine therapy in treating patients with malignant glioma	I	18	Ongoing	Dose-limiting toxicity	[202]
DC vaccination for patients with solid tumors	I/II	10	Recruiting	Immunogenicity	[202]
Study of a drug [DCVax®-L] to treat newly diagnosed GBM brain cancer	III	300	Recruiting	PFS	[202]
A study of ICT-107 immunotherapy in GBM	II	200	Recruiting	OS	[202]
Vaccine therapy in treating patients undergoing surgery for recurrent GBM	I	50	Recruiting	Safety and feasibility	[202]

DC: Dendritic cell; GBM: Glioblastoma multiforme; OS: Overall survival; PFS: Progression-free survival.

Table 5

Clinical trials using heat shock proteins in patients with high-grade glioma.

Study	Phase	Estimated enrollment (n)	Trial results	Primary outcome(s)	Ref.
Completed					
Crane *et al.* (2012)	I	12	Median survival: 47 weeks in immunologic responders	–	[74]
Ongoing					
HSPPC-96 vaccine with temozolomide in patients with newly diagnosed GBM (HeatShock)	II	55	Ongoing	Safety, survival	[202]
GP96 HSP–peptide complex vaccine in treating patients with recurrent or progressive glioma	I/II	50	Ongoing	Safety, maximum tolerated dose, toxicity and PFS	[202]

GBM: Glioblastoma multiforme; HSP: Heat shock protein; PFS: Progression-free survival.

Table 6

Clinical trials using lymphokine-activated killer cells.

Study	Phase	Patients (n)	Results	Ref.
Dillman *et al.* (2009)	I/II	33	Median survival: 20.5 months	[108]
Dillman *et al.* (2004)	I/II	40	Median survival: 17.5 months	[109]
Sankhla *et al.* (1996)	I	10	PR: 2	[110]
Hayes *et al.* (1995)	I	15	Median survival following reoperation: 53 weeks	[111]
Boiardi *et al.* (1994)	I	9	CR: 1 PR: 2 SD: 4	[112]
Jeffes *et al.* (1993)	I	19	Median survival: 37 weeks	[113]
Blancher *et al.* (1993)	I	13	Tumor progression noted after 4–12 weeks	[114]
Lillehei *et al.* (1991)	I	20	Median survival: 63 weeks	[115]
Barba *et al.* (1989)	I	10	PR: 1	[116]
Jacobs *et al.* (1986)	I	9	No PR or SD	[117]

CR: Complete response; PR: Partial response; SD: Stable disease.

Table 7

Clinical trials using cytotoxic T lymphocytes.

Study	Phase	Patients (n)	Results	Ref.
Tsuboi *et al.* (2003)	I	10	CR: 1 PR: 4 Median survival: 5 months Overall response rate: 50%	[76]
Wood *et al.* (2000)	I	9	PR: 3 Survival >4 years: 2	[118]
Plautz *et al.* (2000)	I	9	PR: 3 (1 GBM and 2 grade III survival >4 years)	[119]
Sloan *et al.* (2000)	I	19	CR: 1 PR: 7 Median survival: 12 months	[120]
Quattrocchi *et al.* (1999)	I	6	CR: 1 PR: 2	[75]
Tsurushima *et al.* (1999)	I	4	PR: 3 SD: 1	[121]
Plautz *et al.* (1998)	I	10	SD: 1 4 patients alive after 1 year	[122]
Kruse *et al.* (1997)	I	5	Transient toxicities: Survival (AO) >30 months Survival (AA) >28 months	[123]
Holladay *et al.* (1996)	I	15	No PR or SD Disease-free survival >8 months: 7	[124]
Kitahara *et al.* (1987)	I	5	PR: 2 1 patient alive >104 weeks	[125]

AA: Anaplastic astrocytoma; AO: Anaplastic oligodendroglioma; CR: Complete response; GBM: Glioblastoma multiforme; PR: Partial response; SD: Stable disease.