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Current strategies and novel immunotherapeutic approaches for overcoming immune resistance in glioblastoma

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Abstract

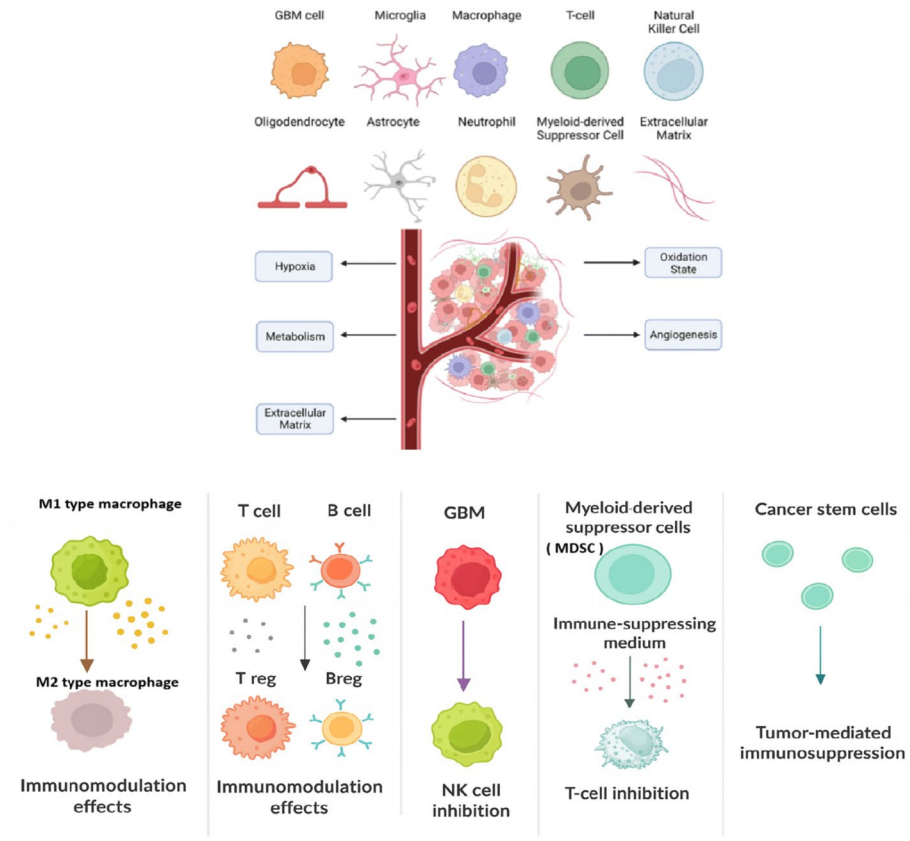
Glioblastoma (GBM) is the most aggressive primary malignant brain tumor, characterized by rapid proliferation, extensive invasion, and significant genetic heterogeneity. Despite the availability of standard treatments such as surgical resection, radiotherapy, and chemotherapy, the prognosis for GBM patients remains poor, with a median survival of approximately 15 months. Recent advances in immunotherapy have introduced innovative approaches aimed at leveraging the immune system to specifically target and eliminate GBM cells. These strategies include cytokine-based therapies, immune checkpoint inhibitors, chimeric antigen receptor (CAR) T-cell and natural killer (NK) cell therapies, RNA-based immunotherapies, and nanoparticle-mediated drug delivery systems. Furthermore, emerging technologies such as CRISPR/Cas9 gene editing, exosome-based delivery, STING pathway activation, and AI-guided personalized treatment have shown promise in overcoming the immunosuppressive tumor microenvironment and enhancing therapeutic efficacy. This review provides a comprehensive overview of these cutting-edge approaches, discussing their mechanisms, clinical potential, current limitations, and future directions for the development of more effective immunotherapies for GBM.

Keywords Glioblastoma, Immunotherapy, Gene editing, Nanoparticles, CAR-T, Exosomes, Tumor microenvironment

1 Introduction

Glioblastoma (GBM) is the most common and aggressive primary malignant brain tumor in adults, classified as a grade IV astrocytoma by the World Health Organization (WHO). Despite extensive research efforts, the prognosis for GBM patients remains dismal, with a median overall survival of approximately 15 months and a 5-year survival rate of less than 5% [1, 2]. This poor prognosis is primarily due to the highly infiltrative

Graphical Abstract



nature of GBM, which makes complete surgical resection challenging, and its intrinsic resistance to conventional therapies such as radiotherapy and chemotherapy. Additionally, the presence of the blood–brain barrier (BBB) significantly limits the delivery of therapeutic agents to the tumor site, further complicating treatment [3].

The central nervous system (CNS) has historically been considered an immune-privileged site due to the absence of conventional lymphatic vessels and the protective role of the BBB [4]. However, recent evidence has shown that the CNS maintains a unique and tightly regulated immune environment that allows for limited surveillance and immune activity [5, 6]. Some of these mechanisms, such as restricted antigen presentation [7], local immunosuppressive signaling [8], and specialized interactions between glial cells and immune cells [9], remain active in GBM. These features contribute to the tumor's ability to evade immune surveillance and create a profoundly immunosuppressive microenvironment that hinders effective anti-tumor immunity [10, 11].

In recent years, immunotherapy has emerged as a promising strategy to address these challenges by activating the patient's immune system to specifically target and eliminate GBM cells. Unlike traditional treatments that primarily target rapidly dividing tumor cells, immunotherapy aims to overcome the immune-suppressive microenvironment of GBM and harness the body's natural defense mechanisms to achieve durable tumor control [12]. Early immunotherapeutic approaches, such as cytokine-based therapies and immune checkpoint inhibitors, have demonstrated potential in preclinical studies but

have faced significant challenges in clinical translation due to the complex immunosuppressive networks within the GBM tumor microenvironment [13].

To overcome these barriers, researchers are now exploring a range of innovative strategies, including exosome-based delivery, CRISPR/Cas9 gene editing, STING pathway activation, microbiome modulation, and AI-guided personalized treatment. These approaches aim to not only enhance immune activation but also precisely target the diverse cellular and molecular mechanisms that drive GBM progression and immune escape. For instance, exosomes and nanoparticles are being developed as efficient drug carriers capable of crossing the BBB, while gene editing technologies such as CRISPR/Cas9 offer the potential to reprogram immune cells for enhanced anti-tumor activity.

This review provides a comprehensive overview of these emerging immunotherapeutic strategies, highlighting their mechanisms, advantages, limitations, and potential for clinical translation. By integrating recent advances in biotechnology, immunology, and computational biology, these approaches hold the potential to significantly improve the treatment outcomes for GBM patients and transform the landscape of neuro-oncology.

2 Standard treatment approaches for GBM

The WHO classifies grade III and IV astrocytic tumors, including oligodendroglioma (grade III) and oligoastrocytoma (grade III), as part of a broader category of malignant central nervous system (CNS) tumors known as gliomas [14]. Diagnosis of malignant astrocytoma is based on histopathological examination. The WHO recommends using the St. Anne-Mayo grading system for confirming the diagnosis of malignant astrocytoma (grade III), which requires the presence of at least two of the following four criteria: (1) nuclear atypia, (2) mitosis, (3) endothelial proliferation, and (4) necrosis [1, 2].

GBM (grade IV) is identified by meeting at least three of these four criteria. Since the likelihood of systemic dissemination is low, this classification relies solely on pathological findings. According to the Central Brain Tumor Registry of the United States (CBTRUS), from 2017 to 2021, a total of 445,792 primary CNS tumors were recorded, of which 6.3% were GBM, making it the most prevalent type of CNS tumor [15].

GBM is more commonly diagnosed in men than women, with a male-to-female ratio of approximately 1.5:1, and it is more frequent in Caucasians compared to African Americans, with a ratio of 2:1. Studies have shown that the incidence of GBM increases with age, with the highest prevalence observed among individuals aged 75–84 years. The average age at diagnosis is around 64 years [16].

Importantly, advanced age is associated with immunosenescence and immune dysregulation, which significantly affect therapeutic outcomes [17, 18]. Elderly patients, particularly those in their 70s and 80s, exhibit reduced T-cell proliferative capacity, diminished diversity in the T-cell repertoire, and a shift toward immunosuppressive populations such as myeloid-derived suppressor cells (MDSCs) and regulatory T cells (Tregs) [19–21]. In addition, they often experience chronic "inflammaging," a state of persistent low-grade inflammation that paradoxically coexists with impaired immune responses [22, 23]. These age-related immune alterations can blunt the effectiveness of immunotherapies such as checkpoint inhibitors, cancer vaccines, or adoptive T-cell therapies [24, 25]. Clinical analyses consistently demonstrate that GBM patients over 65 years of age show worse survival and weaker responses to immunotherapy, in part due to these immunosenescent changes [26, 27]. Consequently, ongoing and future immunotherapy trials are

stratifying patients by age and investigating interventions such as senolytics or immune rejuvenation strategies to improve therapeutic efficacy in older adults [28].

Exposure to ionizing radiation is a recognized risk factor for developing malignant gliomas, including GBM. Despite advancements in surgical techniques, radiotherapy, and chemotherapeutic regimens, the prognosis for GBM remains poor, characterized by high recurrence rates and limited long-term survival. Recent studies indicate that the median overall survival for GBM patients is approximately 15 months, with one-year survival rates around 43% and five-year survival rates ranging between 5 and 10%. These statistics underscore the aggressive nature of GBM and highlight the urgent need for more effective therapeutic strategies and early diagnostic approaches to improve patient outcomes [29, 30]. Patients with anaplastic astrocytoma have a slightly better prognosis, with a one-year survival rate of 61% and a five-year survival rate of 27% [31]. Currently, the median survival for patients diagnosed with GBM is approximately 12–15 months, whereas for those with anaplastic astrocytoma, the median survival ranges between 36 and 60 months [32]. Typically, patients with GBM present symptoms such as headaches, neurological disturbances, altered mental status, personality changes, seizures, nausea, vomiting, and increased intracranial pressure. At diagnosis, poor prognostic indicators include advanced age, low Karnofsky Performance Status (KPS), inoperable tumors, and histological features consistent with GBM [33].

The standard treatment protocol for suspected GBM patients involves surgical resection followed by focal radiotherapy and concurrent chemotherapy. High-dose corticosteroids, particularly dexamethasone, are frequently administered to reduce tumor-associated edema and alleviate neurological symptoms [32]. Additionally, anticonvulsant therapy is initiated when seizures are present. The first step in managing GBM patients is surgical tumor resection. A common limitation of surgical intervention is the tumor's location in critical brain regions responsible for speech and other vital functions. Recent findings suggest that resecting at least 78% of the tumor volume significantly enhances patient survival. Surgical success is influenced by factors such as patient age, KPS, tumor volume, and the extent of resection. After surgery, patients who undergo 78% tumor resection have a median survival of 12.5 months, whereas those who receive temozolomide chemotherapy experience an extension of survival to approximately 16 months. Since 2005, temozolomide has been widely accepted as a standard therapeutic agent for GBM [34]. Researchers assessed the combination of temozolomide and radiotherapy in GBM patients. They reported an average survival of 12.1 months for patients who received only radiotherapy, compared to 14.6 months for those treated with a combination of temozolomide and radiotherapy. Additionally, the two-year survival rate for patients receiving combination therapy was 26.5%, compared to only 10.4% for those who underwent radiotherapy alone [35].

3 Tumor immunosuppressive landscape

3.1 Tumor microenvironment (TME)

The GBM microenvironment is profoundly immunosuppressive, enabling tumor progression and immune evasion. It comprises both cellular and non-cellular elements, including tumor-associated macrophages (TAMs), microglia, Tregs, myeloid-derived suppressor cells (MDSCs), immunosuppressive cytokines, and an extracellular matrix (ECM) that together create a "hostile soil" for immune attack [36]. TAMs (microglia

and macrophages): TAMs can constitute up to ~ 30% of the GBM mass. They include yolk-sac–derived brain-resident microglia and infiltrating monocyte-derived macrophages. Notably, these populations localize differently: microglia are enriched in peritumoral regions, whereas infiltrating macrophages cluster in perivascular niches. Both subtypes typically adopt an M2-like phenotype, releasing IL-10, TGF-β, and VEGF, which support angiogenesis, promote tumor growth, and inhibit cytotoxic T-cell activity [37]. TAMs also downregulate MHC and co-stimulatory molecules, impair antigen presentation, and contribute to therapeutic resistance, including reduced responsiveness to anti-VEGF therapy [38]. High densities of $CD163^+$ M2-polarized TAMs strongly correlate with poor prognosis in GBM patients [39]. Tregs: Tregs are enriched in GBM and often outnumber effector T cells within tumors. Their expansion is driven by TGF-β, which induces $FOXP3^+$ Treg differentiation, and IL-10, which sustains their pool. Tregs potently suppress anti-tumor immunity by inhibiting proliferation and cytokine production of effector T cells. Elevated Treg signatures in GBM tumors correlate with shorter overall survival, underscoring their clinical significance [40]. Myeloid-derived suppressor cells (MDSCs): MDSCs accumulate in both the tumor and circulation of GBM patients. They suppress T and NK cell activity through arginase-1, inducible nitric oxide synthase (iNOS), reactive oxygen species, and cytokine secretion [36]. MDSCs also enhance Treg development, further reinforcing a pro-tumor environment [41]. Elevated MDSC levels in patient blood and tumors are associated with worse prognosis, while preclinical depletion or reprogramming of MDSCs restores T-cell activity and improves immunotherapy efficacy [42]. Extracellular matrix (ECM) and soluble factors: The ECM, enriched in glycoproteins and proteoglycans, acts as both a physical and biochemical barrier. Molecules like tenascin-C restrict T-cell infiltration into glioma tissue and sequester cytokines, perpetuating immunosuppression [39]. Soluble mediators such as prostaglandin E_2 (PGE_2), TGF-β, and IL-10, abundantly produced by GBM cells and TAMs, further reinforce immune exclusion [37]. The net result is an immunologically "cold" TME that presents a major obstacle to successful immunotherapy [36].

3.2 Mechanisms of immunosuppression

GBM suppresses both local and systemic immune responses through multiple molecular pathways.

Immunosuppressive cytokines: TGF-β and IL-10 are central mediators. TGF-β drives $CD4^+$ T cells toward $FOXP3^+$ Treg differentiation, inhibits effector $CD8^+$ T-cell cytolytic molecules (perforin, granzymes, IFN-γ), and downregulates activating receptors such as NKG2D on NK cells [43]. TGF-β2 also reduces HLA-DR expression on GBM cells, impairing neoantigen presentation [44]. IL-10, secreted by both GBM and TAMs, blocks antigen presentation, inhibits dendritic cell maturation, and expands Treg populations [45]. PGE_2 synergizes by promoting MDSC expansion [46].

Immune checkpoint receptor–ligand interactions: PD-L1 expression on GBM and infiltrating myeloid cells engages PD-1 on T cells, recruiting SHP2 phosphatases that inhibit TCR/CD28 signaling cascades (PI3K–Akt, NF-κB, NFAT), driving exhaustion and anergy [43]. CTLA-4, expressed on Tregs and activated T cells, competes with CD28 for CD80/CD86 on APCs, blocking co-stimulation and suppressing IL-2 production [47]. FasL (CD95L), expressed on GBM endothelium or tumor cells, triggers apoptosis of Fas^+ tumor-infiltrating lymphocytes, eliminating tumor-specific T cells at the invasive

margin [48]. Together, these checkpoint-mediated interactions form a "molecular shield" against effector immunity.

IDO1 pathway: Indoleamine 2,3-dioxygenase 1 (IDO1) is often upregulated in GBM. By catabolizing tryptophan into kynurenine, IDO1 induces immunosuppression via two mechanisms: (i) tryptophan depletion activates the GCN2 stress pathway in effector T cells, halting proliferation and inducing apoptosis; (ii) kynurenine engages the aryl hydrocarbon receptor (AhR) on T and NK cells, downregulating activating receptors (e.g., NKG2D, NKp46) and impairing cytotoxicity [48]. IDO1 activity skews the immune balance toward Tregs and MDSCs, while suppressing effector T/NK cell responses [49].

Extracellular vesicles (EVs): GBM-derived EVs (exosomes and microvesicles) act as potent immunosuppressive agents. They carry PD-L1, FasL, IL-10, TGF-β, and regulatory microRNAs [44]. Functionally, EVs dose-dependently inhibit $CD4^+$ and $CD8^+$ T-cell receptor–mediated activation, proliferation, and cytokine secretion. In vitro studies show that this suppression can be partially reversed by PD-1 blockade, confirming EV PD-L1's role [50]. EV uptake by monocytes reprograms them into tumor-supportive phenotypes, extending local suppression into systemic compartments [51]. These findings highlight EVs as critical drivers of GBM immune escape. The immunosuppressive microenvironment caused by GBM is summarized in Fig. 1.

3.3 Metabolic checkpoints (Lactate & adenosine)

A hallmark of GBM's immunosuppressive environment is metabolic reprogramming, which creates additional barriers to effective immunity.

Lactate: Due to the Warburg effect, GBM cells produce large amounts of lactate, leading to acidification of the TME. Lactate skews macrophages toward the M2 phenotype, impairs T-cell proliferation and cytotoxicity, and suppresses NK cell degranulation. Acidic conditions also reduce TCR signaling, reinforcing immune dysfunction [52].

Adenosine: GBM and stromal cells upregulate CD39 and CD73 ecto-enzymes, which hydrolyze ATP into adenosine, particularly under hypoxic conditions [53]. Adenosine binds to A2A receptors on T and NK cells, suppressing MAPK, NF-κB, and NFAT signaling, reducing IL-2/IFN-γ/TNF-α production, and upregulating inhibitory checkpoints (PD-1, CTLA-4, LAG-3) [54]. Adenosine also promotes $FOXP3^+$ Treg and M2

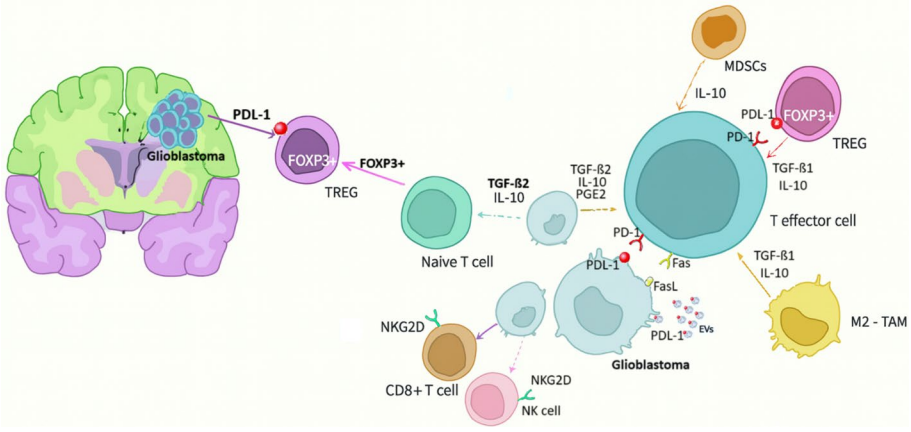


Fig. 1 Illustration of the immunosuppressive microenvironment created by glioblastomas. MDSCs refer to MDSCs, while NK stands for natural killer cells

macrophage expansion, while impairing NK and dendritic cell activity [55]. This positions adenosine as a key "metabolic immune checkpoint [56]."

Therapeutic implications: Targeting these metabolic barriers is a growing strategy. A2A antagonists (e.g., ciforadenant/CPI-444) and anti-CD73 antibodies restore T-cell activity in preclinical models, with early-phase trials including GBM cohorts [57]. Blocking lactate production/export via LDHA or MCT4 inhibitors reduces M2 polarization and relieves T-cell dysfunction [52]. Combination therapies—such as PD-1 blockade plus A2A inhibition plus stereotactic radiation—are under investigation [58]. Challenges remain, as tumor metabolism is redundant and systemic inhibition may affect normal physiology, but correlative studies suggest GBM patients with lower lactate/adenosine activity have improved immune responses and outcomes [59] (Table 1).

4 Conventional immunotherapy

4.1 Immune checkpoint inhibitors

Immune checkpoint inhibitors (ICIs) are monoclonal antibodies designed to block inhibitory pathways that tumors exploit to evade immune detection and destruction. The most extensively studied immune checkpoints in GBM are PD-1/PD-L1 and CTLA-4. These checkpoints normally function to maintain immune homeostasis and prevent autoimmunity, but tumors often overexpress their ligands to inhibit effective anti-tumor immune responses [60].

The PD-1/PD-L1 pathway involves the binding of PD-L1, expressed on tumor cells, to the Programmed Death-1 (PD-1) receptor on T cells, resulting in the suppression of T cell activation and proliferation. Blocking this pathway with antibodies such as nivolumab or pembrolizumab has demonstrated clinical efficacy in other cancers like melanoma and non-small cell lung cancer (NSCLC) [61, 62]. However, clinical trials in GBM have yielded mixed results due to the highly immunosuppressive tumor microenvironment and low mutational burden of GBM (Khasraw et al., 2020) [63]. For example, in the CheckMate-143 phase III trial, nivolumab failed to improve overall survival compared to bevacizumab in recurrent GBM patients, highlighting the limitations of single-agent PD-1 blockade [64] (Fig. 2).

Similarly, the CTLA-4 pathway involves the inhibitory receptor Cytotoxic T-Lymphocyte Antigen 4 (CTLA-4), which competes with CD28 for binding to B7 molecules on antigen-presenting cells. Blocking CTLA-4 with ipilimumab has been effective in other cancers, but clinical studies in GBM have shown minimal activity, likely due to poor BBB penetration and profound immunosuppression in the TME [65–67]. To overcome these limitations, researchers are pursuing combination immunotherapy approaches, pairing ICIs with other modalities to amplify anti-tumor immunity:

Checkpoint–Checkpoint Combinations: Dual blockade of PD-1 and CTLA-4 has shown synergistic effects in melanoma and is under evaluation in GBM. Preclinical glioma models demonstrate that combined anti–PD-1 + anti–CTLA-4 therapy elicits more robust T-cell infiltration and tumor control than either agent alone [68, 69]. Early-phase GBM trials of nivolumab + ipilimumab indicate feasibility but also increased immune-related toxicities, underscoring the challenge of balancing efficacy with safety [70, 71].

Checkpoint + Vaccine or Agonist Strategies: In preclinical models, adding Smac mimetics (IAP antagonists) to PD-1 or CTLA-4 blockade markedly enhanced anti-GBM immunity. In murine GBM models (CT-2A, GL261), Smac mimetics synergized

Table 1 Mechanisms of immunosuppression and metabolic checkpoints in GBM

Pathway / factor	Mechanism of action	Effect on immune cells	Therapeutic implications	Challenges / limitations	References
TGF-β	Induces FOXP3⁺ Tregs; inhibits perforin, granzymes, IFN-γ; downregulates NKG2D on NK cells; reduces HLA-DR expression	Promotes Treg expansion, suppresses CD8⁺ T cells and NK activity	Anti–TGF-β antibodies, TGF-βR inhibitors in trials	Pleiotropic effects; systemic blockade risks autoimmunity	[43, 44]
IL-10	Inhibits DC maturation; blocks antigen presentation; expands Tregs	Dendritic cell dysfunction; Treg proliferation	IL-10/IL-10R blockade under preclinical study	Broad immunoregulatory role may impair homeostasis	[45]
PGE_2	Promotes MDSC expansion; synergizes with IL-10 and TGF-β	Expands immunosuppressive myeloid cells	COX-2/PGE_2 inhibitors may reduce MDSCs	Widespread systemic effects limit specificity	[46]
PD-1/PD-L1 axis	PD-L1 on GBM/myeloid cells binds PD-1, recruiting SHP2 to block TCR/CD28 signaling (PI3K–Akt, NF-κB, NFAT)	T-cell exhaustion, anergy, loss of cytotoxicity	Checkpoint inhibitors (nivolumab, pembrolizumab) tested in GBM	Limited efficacy as monotherapy; antigen-poor TME	[43]
CTLA-4	Competes with CD28 for CD80/CD86; suppresses IL-2 production	Blocks T-cell priming; promotes Treg suppression	Anti–CTLA-4 (ipilimumab, tremelimumab) explored in GBM combos	High toxicity; limited brain penetration	[47]
Fas/FasL	FasL on GBM endothelium induces apoptosis in Fas⁺ TILs	Eliminates tumor-specific T cells at invasive margins	Fas/FasL blockade considered in preclinical models	Risk of interfering with normal apoptosis pathways	[48]
IDO1–Kynurenine	Catabolizes tryptophan, depleting it and producing kynurenine (activates AhR)	T-cell apoptosis, reduced NK cytotoxicity, Treg and MDSC expansion	IDO1 inhibitors (indoximod, epacadostat) under evaluation	Resistance via redundant metabolic pathways	[48, 49]
Extracellular vesicles (EVs)	EVs carry PD-L1, FasL, IL-10, TGF-β, microRNAs	Inhibit CD4⁺/CD8⁺ proliferation, reprogram monocytes into suppressive phenotypes	EV-targeted therapies and EV PD-L1 blockade explored	EV heterogeneity and systemic spread complicate targeting	[44, 50, 51]
Lactate (Warburg effect)	Excess lactate acidifies TME, reduces TCR signaling	M2 macrophage skewing, impaired T-cell proliferation, suppressed NK function	LDHA or MCT4 inhibitors reduce lactate; metabolic reprogramming	Systemic blockade risks affecting normal metabolism	[52]
Adenosine (CD39/CD73 → A2A receptor)	Hypoxia-induced ATP hydrolysis generates adenosine, suppressing MAPK, NF-κB, NFAT pathways	Reduced cytokine secretion; T-cell exhaustion; Treg and M2 macrophage expansion	A2A antagonists (ciforadenant), anti-CD73 antibodies in early trials	Redundant adenosine pathways, systemic side effects	[53–57]

with anti–PD-1 or anti–CTLA-4 to produce durable cures, an effect dependent on CD8⁺ T cells and TNF-α signaling, and accompanied by a reduction of immunosuppressive Tregs [72]. Strikingly, triple therapy with a Smac mimetic, anti–PD-1, and anti–CTLA-4 achieved 100% durable cures in mice, demonstrating the potential of IAP-targeting drugs to potentiate checkpoint blockade. Similarly, anti–PD-1 combined with a TLR3

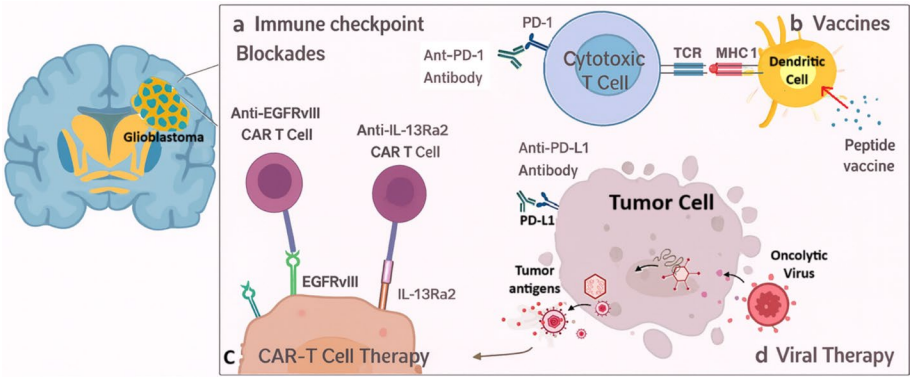


Fig. 2 Immunotherapeutic approaches for GBM treatment include several strategies. **a** Immune checkpoint receptors and ligands, such as PD-1 on T cells and PD-L1 on tissue cells, can suppress the adaptive immune response in healthy tissues. Tumors can also express PD-L1, further preventing T cell activation within the tumor environment. Immune checkpoint inhibitors are antibodies that block these receptor-ligand interactions, like those between PD-1 and PD-L1, thereby counteracting their immunosuppressive effects. **b** Vaccines work by presenting GBM-specific antigens to antigen-presenting cells (APCs), including dendritic cells, and depend on MHC-mediated presentation to activate T cells for a targeted immune response against GBM. **c** CAR-T cell therapy involves using a patient's own T cells, which are genetically engineered to recognize specific surface antigens associated with GBM, such as EGFRvIII and IL-13Rα2. Unlike vaccines, CAR-T cells function independently of MHC-dependent antigen presentation. **d** Viral therapy utilizes oncolytic viruses and retroviruses to induce tumor cell death and release tumor antigens or to incorporate therapeutic genes that can be expressed by the tumor cells

agonist (poly(I:C)) and a dendritic cell vaccine induced stronger tumor-specific T-cell responses than any monotherapy [73]. Clinically, pembrolizumab combined with the oncolytic virus DNX-2401 increased long-term responders in recurrent GBM.

Checkpoint + Standard Therapy: Radiation synergizes with PD-1 blockade by increasing antigen release and T-cell trafficking. Preclinical and early clinical studies of neoadjuvant PD-1 blockade with radiation have shown augmented immune gene expression in GBM tumors [74, 75]. Chemotherapy, however, presents a double-edged sword: temozolomide (TMZ) can cause lymphopenia, which may facilitate adoptive cell therapy engraftment but also increases Tregs and PD-L1 expression. In murine studies, concurrent high-dose TMZ reduced anti–PD-1 efficacy due to T-cell depletion and elevated Tregs, highlighting the need for careful sequencing of TMZ with immunotherapy [60].

Novel Combinations: Several innovative pairings are entering trials, including atezolizumab (anti–PD-L1) with tocilizumab (IL-6 blockade) to mitigate steroid-induced immunosuppression, and pembrolizumab with a CD73 inhibitor plus stereotactic radiation to counteract adenosine-mediated T-cell suppression. Preclinical work also highlights checkpoint blockade combined with STING agonists as a promising approach, leveraging innate immune activation to improve adaptive responses [76, 77].

In summary, While single-agent checkpoint blockade has shown limited efficacy in GBM, combination strategies that incorporate Smac mimetics, innate immune agonists, or rational sequencing with radiation and chemotherapy have produced encouraging preclinical results and are now the focus of multiple ongoing trials. The optimization of BBB penetration, biomarker selection, and toxicity management will be crucial for successful clinical translation.

Combination strategies involving ICIs and other therapeutic modalities, such as CAR-T cell therapy, cytokine-based treatments, and radiotherapy, are currently under investigation to enhance their efficacy against GBM [78, 79]. Improving delivery systems

to penetrate the BBB and identifying biomarkers to predict response to ICIs are critical areas of ongoing research [80].

4.2 Vaccine-Based approaches

Vaccine-based immunotherapy, which stimulates active immune responses against GBM, is currently being investigated using cell-based and peptide-based vaccines. Among these, dendritic cell (DC) vaccination has been the most extensively studied. Over 20 phase I and II clinical trials have evaluated DC vaccines for adult GBM patients. In most studies, DCs are generated by differentiating peripheral blood monocytes with IL-4 and GM-CSF. Other agents, such as IL1B, TNF, TLR agonists, and IFN-γ, have also been used to enhance DC maturation [81].

Antigen sources vary across trials, but commonly include: (1) Autologous tumor lysates, (2) Tumor-specific peptides, (3) mRNA extracted from tumor lysates, (4) Peptides derived from autologous tumor cells [82].

Vaccines are generally administered subcutaneously, intradermally, or intracranially. DC-based vaccines are generally well-tolerated, with only mild side effects such as flu-like symptoms, headaches, and minor inflammation reported [13, 83]. Severe adverse effects are rare, with only one documented case of peritumoral edema and sensory loss. Most clinical studies suggest that DC immunotherapy benefits some GBM patients.

Yamanaka and colleagues conducted a phase I/II trial involving 24 patients with newly diagnosed or recurrent GBM. These patients received autologous tumor cells combined with DCs and keyhole limpet hemocyanin (KLH). The DCs were administered either intradermally alone or with additional intracranial injections. Results showed improved survival in patients receiving both subcutaneous and intracranial injections, as well as in those vaccinated with mature DCs [84]. Liau and colleagues reported that immune responses involving T cell infiltration into tumor tissues and decreased TGF-β levels were associated with better overall survival in patients [85]. While vaccines and checkpoint inhibitors remain at the forefront of conventional immunotherapy, their limited efficacy highlights the urgent need for more sophisticated approaches. This has paved the way for advanced molecular and cellular strategies, which seek to overcome the barriers of GBM by reprogramming immune responses at the genetic, epigenetic, and nanoscale levels.

5 Advanced and molecular strategies

5.1 Gene therapy

Gene therapy vectors have been extensively explored as innovative tools to enhance the clinical efficacy of GBM treatment. These vectors are typically engineered to promote localized release of pro-inflammatory cytokines that attract immune cells to the tumor microenvironment (e.g., Flt3L), stimulate the host immune response (e.g., interleukin-12), or exert direct anti-tumor effects (e.g., interferon-alpha) [86, 87].

Among these strategies, local expression of Flt3L, a growth factor essential for the proliferation and differentiation of dendritic cells, has received particular attention. Intracranial administration of adenoviral vectors encoding Flt3L (Ad-Flt3L) has been shown to induce dendritic cell proliferation and migration into the brain parenchyma [88]. Preclinical studies demonstrated that intratumoral injection of Ad-Flt3L significantly increased dendritic cell infiltration and enhanced the presence of other

antigen-presenting cells within rodent GBM models [43]. However, despite robust immune cell recruitment, Ad-Flt3L alone did not produce consistent tumor inhibition, suggesting that combination approaches may be required to achieve therapeutic benefit [87].

5.2 RNA-based therapy

RNA-based immunotherapies have gained significant attention as versatile tools to reprogram immune responses against GBM. Unlike conventional therapies, RNA therapeutics can directly encode tumor antigens or immunomodulatory molecules within target cells, enabling rapid and highly specific immune activation [89].

One of the most advanced approaches is mRNA vaccines, which deliver tumor-specific antigens into dendritic cells or other antigen-presenting cells. These cells then translate the encoded antigens and stimulate strong cytotoxic T-lymphocyte responses [90]. Advances in lipid nanoparticle (LNP) technology have improved RNA stability and intracellular delivery, making systemic administration feasible [91]. Preclinical orthotopic GBM models confirmed that LNP-mRNA vaccines achieve protein expression within tumors and trigger robust immune activation [92]. Early clinical efforts using mRNA vaccines encoding GBM-associated antigens, such as EGFRvIII or patient-specific neoantigens, have demonstrated immunogenicity [93]. A particularly noteworthy translational finding came from a veterinary trial in pet dogs with spontaneous gliomas, where a multi-antigen mRNA vaccine elicited measurable immune responses and prolonged survival, supporting feasibility in human applications [94].

Parallel strategies employ small interfering RNAs (siRNAs) and microRNAs (miRNAs) to suppress immunosuppressive signaling in the GBM microenvironment. For instance, siRNAs targeting PD-L1, IDO1, or STAT3 reprogrammed TAMs from an M2 tumor-supportive phenotype to an M1 pro-inflammatory state in preclinical studies [95]. Likewise, miR-124 delivered via polymeric carriers downregulated immune checkpoint molecules and significantly extended survival in murine GBM models [96].

RNA therapies present distinct advantages, including rapid design for novel targets, high specificity, and favorable safety profiles due to transient expression. Nevertheless, challenges remain. RNA molecules are inherently unstable, can trigger unintended innate immune activation through Toll-like receptors, and display limited penetration across the blood–brain barrier. Off-target accumulation in the liver and spleen also poses risks. Current research focuses on refining delivery platforms through advanced LNPs, targeted ligands, biomaterial scaffolds, and nanoparticles [97]. Furthermore, combining RNA-based therapies with immune checkpoint inhibitors or radiotherapy is under investigation as a strategy to maximize efficacy [98].

5.3 CAR-T and CAR-NK Therapy

Chimeric antigen receptor (CAR) technologies represent a major advance in cellular immunotherapy for GBM, enabling MHC-independent recognition of tumor-associated antigens [99].

CAR-T cells engineered to target EGFRvIII, a tumor-specific EGFR variant, showed potent preclinical efficacy but only transient responses in a phase I trial due to antigen loss and tumor heterogeneity [100]. To overcome this, alternative targets such as IL13Rα2, HER2, EphA2, and B7-H3 are under investigation [99]. IL13Rα2-directed

CAR-T therapy delivered locoregionally demonstrated dramatic responses in individual cases, and a subsequent phase I trial confirmed safety and transient tumor regression [101]. Delivery route has proven critical: intravenous infusion is feasible but limited by BBB penetration, while intracavitary or intraventricular administration enhances local distribution and tumor infiltration. Comparative trials suggest intraventricular delivery provides broader CNS coverage, whereas intratumoral delivery yields higher localized concentrations [101, 102]. Combination strategies are also being explored, such as CAR-T cells with checkpoint inhibitors, radiotherapy, or oncolytic viruses, as well as "armored" CARs engineered to secrete cytokines or T-cell engagers [102, 103].

CAR-NK cells offer complementary advantages, as they can mediate tumor killing via both CAR-directed recognition and innate NK pathways (e.g., NKG2D, DNAM-1, antibody-dependent cytotoxicity) [98–100]. Unlike CAR-T cells, CAR-NK therapies can be generated from allogeneic sources without risk of graft-versus-host disease and display a lower incidence of cytokine release syndrome or neurotoxicity [104, 105]. Preclinical studies demonstrated effective tumor clearance in GBM models using EGFRvIII- or B7-H3-targeted CAR-NK cells. However, their limited persistence remains a challenge, prompting engineering strategies such as IL-15 expression to prolong survival. These features suggest CAR-NK cells may serve as safer, "off-the-shelf" options or in sequential use with CAR-T therapies [106].

5.4 Nanoparticles and exosome-based delivery

5.4.1 Nanoparticle systems

Nanoparticles (NPs) provide a platform to improve GBM immunotherapy delivery by enhancing BBB penetration, sustaining drug release, and enabling targeted delivery [107, 108]. Lipid nanoparticles have successfully transported mRNA encoding tumor antigens or cytokines into GBM models, while polymeric nanoparticles have delivered siRNAs targeting STAT3 or S100A4 in TAMs to reprogram them toward pro-inflammatory phenotypes [109, 110]. Inorganic particles such as iron oxide and gold nanoparticles can also be guided or activated externally to disrupt the blood–tumor barrier or provide adjuvant hyperthermia [107].

Innovative designs include macrophage-coated nanoparticles that evade immune clearance and preferentially home to GBM tissue, as well as nanoparticles loaded with STING agonists or TLR ligands, which induced strong innate responses and extended survival in GL261 and CT-2A models [111, 112]. Challenges include off-target accumulation, limited penetration through heterogeneous BBB regions, and variability in tumor uptake. One promising alternative is using immune cells, particularly monocytes and macrophages, as delivery vectors. These cells naturally migrate to GBM and can transport therapeutic payloads, including oncolytic viruses or engineered exosomes, directly into the TME [113].

5.4.2 Exosome-based immune modulation and delivery

Exosomes are endogenous vesicles capable of crossing the BBB, offering low toxicity and stability in circulation [114]. They have been engineered to deliver tumor antigens, cytokines, or oligonucleotides to immune or tumor cells [112]. Exosomes carrying therapeutic oligonucleotides have also been shown to modulate TAMs, supporting their role as versatile immune modulators [115].

A notable translational effort involved canine spontaneous GBM, where personalized mRNA vaccines and tumor-lysate–based immunotherapies were tested in pet dogs. These studies demonstrated feasibility, induced tumor-specific immune responses, and extended survival, supporting their role as an intermediate step toward human trials [116]. Despite promise, clinical-scale exosome manufacturing and concerns regarding tumor-derived exosome immunosuppression remain significant challenges [117]. Building upon these molecular and delivery-based strategies, another promising approach involves bispecific antibodies that can directly link T cells to tumor cells.

5.5 Bispecific antibodies

Bispecific antibodies (BsAbs) and multi-specific formats have emerged as highly promising immunotherapeutic strategies for GBM. These engineered molecules are designed to simultaneously recognize two or more antigens, thereby enhancing immune system precision and cytotoxicity. A key mechanism of BsAbs is the redirection of T lymphocytes toward tumor cells by engaging a tumor-associated antigen (TAA) on GBM cells and the CD3 receptor on T cells, which fosters direct immune–tumor interactions and facilitates tumor eradication [118]. Preclinical studies have demonstrated that BsAbs targeting EGFRvIII, HER2, and B7-H3 can elicit potent antitumor responses in GBM models, particularly where tumor-specific mutations are present [119, 120].

To overcome the heterogeneity and adaptive resistance that characterize GBM, multi-specific strategies are also under development. These approaches aim to expand immune engagement by targeting multiple TAAs concurrently or by integrating tumor-directed recognition with immune checkpoint blockade [121]. Such combinatorial designs may prove particularly effective in tumors like GBM, where therapeutic resistance and antigenic diversity remain significant challenges.

Despite encouraging results, the clinical translation of BsAbs faces major obstacles, especially limited penetration across the blood–brain barrier (BBB) and the need to minimize off-target effects on healthy tissues. Advances in antibody engineering, along with optimized carrier systems such as nanoparticles and viral vectors, are expected to improve safety, specificity, and delivery efficiency [122, 123]. Collectively, bispecific and multi-specific antibodies represent a rapidly evolving class of immunotherapeutics with strong potential for future GBM treatment (Table 2). Beyond antibody engineering and exosome-based platforms, innate immune sensing has emerged as another promising avenue. In particular, activation of the STING pathway provides a unique means of converting GBM into an immune-responsive tumor and has gained growing attention in both preclinical and translational studies.

5.6 STING pathway activation

The cyclic GMP–AMP synthase (cGAS)–stimulator of interferon genes (STING) pathway is a central component of innate immunity that detects cytosolic DNA and triggers the production of type I interferons together with inflammatory cytokines [124]. In GBM, which is generally considered an immunologically "cold" tumor with low basal STING activity, activating this pathway has emerged as a promising strategy to reshape the TME into an immune-infiltrated "hot" state [125]. When STING is activated in dendritic cells it promotes type I interferon signaling, enhances antigen presentation, and

Table 2 Advanced and molecular strategies in GBM immunotherapy

Therapy type	Mechanism / target	Preclinical / clinical evidence	Advantages / clinical promise	Challenges / limitations	References
Gene therapy (Ad-Flt3L, IL-12, IFN-α vectors)	Local cytokine expression to recruit dendritic cells and stimulate T cell priming	Ad-Flt3L increased dendritic cell infiltration in rodent GBM; IL-12 vectors enhanced intratumoral lymphocyte recruitment	Strong local immune activation; enhances antigen presentation	Limited efficacy as monotherapy; invasive intracranial delivery	[43, 86–88]
RNA-based therapy (mRNA vaccines, siRNA, miRNA)	mRNA vaccines encode tumor antigens (e.g., EGFRvIII, neoantigens); siRNA/miRNA suppress immune checkpoints (PD-L1, STAT3, IDO1)	mRNA vaccines induced CTL responses in preclinical GBM and canine glioma trials; siRNA against PD-L1/STAT3 reprogrammed TAMs; miR-124 prolonged survival in mice	Rapidly adaptable, transient expression improves safety; LNP technology supports systemic delivery	RNA instability, innate immune activation via TLRs, limited BBB penetration, off-target accumulation	[89–97]
CAR-T therapy	MHC-independent T cell recognition of TAAs (EGFRvIII, IL13Rα2, HER2, EphA2, B7-H3)	EGFRvIII CAR-T showed safety but transient benefit in phase I; IL13Rα2 CAR-T induced dramatic regression in case studies; intraventricular delivery expanded CNS coverage	Potent tumor-specific killing; multiple delivery routes (IV, intratumoral, intraventricular); combinable with checkpoints or radiotherapy	Antigen loss, heterogeneity, limited persistence, toxicity risk	[99–103]
CAR-NK therapy	NK cells engineered with CARs for dual recognition via CAR and innate NK pathways (e.g., NKG2D, DNAM-1)	Preclinical GBM models using EGFRvIII or B7-H3 CAR-NKs showed effective tumor clearance; engineered IL-15 CAR-NKs prolonged persistence	Allogeneic "off-the-shelf" use; reduced CRS/neurotoxicity risk; innate + CAR killing	Limited persistence; clinical trials still early; large-scale NK expansion challenging	[98–106]
Nanoparticles (NPs)	LNPs deliver mRNA; polymeric NPs carry siRNAs; inorganic NPs enable targeted BBB disruption or hyperthermia	LNP-mRNA achieved protein expression in orthotopic GBM; polymeric siRNA NPs reprogrammed TAMs; macrophage-coated NPs homed to GBM	Enhanced BBB penetration; sustained drug release; tunable targeting	Off-target accumulation in liver/spleen; heterogeneous BBB penetration; variable tumor uptake	[107–113]
Exosome-based delivery	Natural vesicles engineered to carry antigens, cytokines, or oligonucleotides	Engineered exosomes reprogrammed TAMs; canine glioma trials with exosome/mRNA vaccines improved survival	Cross BBB naturally; low toxicity; versatile immune modulation	Clinical-scale production difficult; tumor-derived exosomes may suppress immunity	[112–117]

Table 2 (continued)

Therapy type	Mechanism / target	Preclinical / clinical evidence	Advantages / clinical promise	Challenges / limitations	References
Bispecific antibodies (BsAbs, multi-specifics)	Engage T cells (CD3) with tumor antigens (EGFRvIII, HER2, B7-H3); multi-specific formats address heterogeneity	Preclinical BsAbs induced potent T cell–mediated killing of GBM; multi-specifics explored for checkpoint + TAA targeting	Redirects host T cells to tumor; can target multiple TAAs simultaneously; modular engineering	Limited BBB penetration; potential off-target toxicity; still early-phase development	[118–123]

drives the priming and recruitment of cytotoxic T lymphocytes and natural killer (NK) cells, thereby linking innate and adaptive immunity [124, 126].

Preclinical studies using murine glioma models such as GL261 and CT-2A have demonstrated that intratumoral administration of STING agonists, including cyclic dinucleotides (CDNs) like 2′3′-cGAMP and synthetic small molecules, can markedly improve survival [125, 127]. The efficacy of these treatments was completely abolished in STING-deficient mice, confirming that the therapeutic effects are pathway-specific [128]. In addition to immune cell recruitment, STING activation disrupted tumor vasculature and induced a pro-inflammatory state that opposed tumor progression [129]. More recently, next-generation agonists such as compound 8803 achieved complete responses in resistant QPP8 glioma models, further highlighting the therapeutic potential of optimized STING stimulation [130].

Beyond monotherapy, combination approaches have produced encouraging results. Incorporating STING agonists with immune checkpoint inhibitors, cancer vaccines, or CAR-T therapies led to synergistic outcomes, as STING-induced interferon signaling created a favorable context for adaptive immune responses [125, 131]. Because this mechanism does not depend on MHC presentation or pre-existing T cell repertoires, it has broad potential across patients with highly heterogeneous GBM [132].

Translational research has also reached canine spontaneous gliomas. In these studies, intratumoral delivery of STING agonists led to measurable tumor regression in several animals, although severe inflammatory toxicity was observed in others, underscoring the need for careful dosing and safety monitoring [133]. Early clinical trials in non-CNS solid tumors have reported immune activation but variable efficacy, reflecting the challenges of delivery and immune regulation that must still be addressed for GBM [134].

Major practical barriers remain. Systemic administration is restricted by the blood–brain barrier and carries a significant risk of systemic inflammation, while intratumoral injection requires invasive neurosurgical procedures and limits distribution to diffuse infiltrative tumor regions [135]. To overcome these limitations, new strategies such as convection-enhanced delivery, implantable biomaterial scaffolds, and nanoparticle-based carriers are being developed to improve brain-specific targeting, retention, and safety [131, 136].

In conclusion, activating the STING pathway offers a unique opportunity to initiate upstream immune signaling in GBM, converting tumors with weak immunogenicity into highly immune-active lesions [137]. If the challenges of delivery and toxicity can be overcome, STING agonists are likely to become a valuable component of future combination immunotherapy strategies for GBM [138] (Table 3).

Table 3 Therapeutic targeting of the STING pathway in GBM

Strategy / approach	Model / setting	Key findings	Advantages / clinical promise	Challenges / limitations	References
STING activation with cyclic dinucleotides (e.g., 2′3′-cGAMP)	Murine GBM models (GL261, CT-2A)	Improved survival; efficacy lost in STING-deficient mice	Pathway-specific immune activation; promotes cytotoxic T cell priming	Limited brain penetration; local injection required	[125, 127, 128]
Synthetic small-molecule agonists	Murine GBM models	Induced immune infiltration, vascular disruption, and tumor control	Strong innate immune activation, potential for systemic use	Risk of systemic inflammation; short half-life	[129]
Next-generation agonists (e.g., compound 8803)	Resistant QPP8 glioma model	Achieved complete responses in resistant tumors	Potential to overcome therapy resistance	Still preclinical; unknown human safety	[130]
Combination with checkpoint inhibitors	Preclinical GBM models	Enhanced response vs monotherapy; synergistic antitumor effect	Creates favorable context for adaptive immunity	Increased risk of toxicity; variable synergy	[125, 131]
Combination with vaccines or CAR-T	Preclinical GBM models	Improved vaccine-induced T cell activity; enhanced CAR-T efficacy	Broad application across heterogeneous tumors	Complex trial design; manufacturing challenges	[131, 132]
Canine glioma studies	Spontaneous gliomas in dogs	Tumor regression in some animals; severe toxicity in others	Proof-of-concept in large spontaneous models	Dosing and safety remain unresolved	[133]
Human early-phase trials (non-CNS tumors)	Solid tumors	Immune activation observed; mixed efficacy	Translational feasibility demonstrated	Delivery to brain remains unsolved	[134]
Novel delivery strategies (nanoparticles, scaffolds, convection-enhanced delivery)	Preclinical models	Improved targeting and retention; reduced systemic toxicity	Potential for localized and sustained release in brain	Still experimental; regulatory barriers	[131, 135, 136]

6 Personalized immunotherapy

6.1 Neoantigen vaccines

Neoantigens are tumor-specific antigens derived from somatic mutations that are absent from normal tissues. Due to their unique tumor-specific nature, neoantigens present attractive targets for the immune system [13]. The identification of these antigens through whole-exome sequencing and mass spectrometry enables the design of personalized vaccines capable of eliciting robust T cell responses. Several studies have demonstrated the potential of neoantigen-based vaccines to induce effective anti-tumor immunity in GBM models [139]. However, challenges remain in identifying high-quality neoantigens and developing standardized protocols for vaccine preparation and administration.

6.2 TCR engineering

T cell receptor (TCR) engineering involves modifying autologous T cells to express receptors that specifically recognize antigens presented by tumor cells. Unlike CAR T cells, TCR-engineered cells can target intracellular antigens processed and presented via major histocompatibility complex (MHC) molecules [140]. This feature makes TCR therapy particularly valuable for targeting GBM-specific antigens such as EGFRvIII and other mutation-derived peptides [141]. Early-phase clinical trials have demonstrated promising results, but challenges related to MHC restriction and antigen heterogeneity remain [102].

6.3 TIL therapy

Tumor-infiltrating lymphocyte (TIL) therapy involves isolating lymphocytes from the patient's tumor, expanding them ex vivo, and reintroducing them to enhance immune-mediated tumor destruction. While TIL therapy has demonstrated efficacy in melanoma, its application in GBM has been limited by the immunosuppressive tumor microenvironment and the challenge of obtaining sufficient viable T cells from the tumor. However, ongoing research is focused on improving TIL isolation techniques and optimizing expansion protocols to enhance therapeutic efficacy [142].

6.4 Biomarker-guided therapy

Biomarkers play a critical role in guiding personalized immunotherapy by predicting patient responses to specific treatments. For example, MGMT promoter methylation status, IDH mutation status, and PD-L1 expression are well-established biomarkers that influence treatment outcomes in GBM [143]. Patients with MGMT promoter methylation tend to respond better to temozolomide, while those with high PD-L1 expression may benefit from immune checkpoint inhibitors. Personalized immunotherapy approaches that incorporate biomarker-guided therapy are expected to enhance treatment efficacy by identifying patients most likely to benefit from specific interventions [144].

6.5 Epigenetic factors

Epigenetic silencing of the MGMT gene promoter is a molecular marker with significant prognostic implications. The MGMT gene encodes a DNA repair protein responsible for removing alkyl groups from the O6 position of guanine. Promoter methylation causes epigenetic silencing and impairs DNA repair mechanisms [145].

Patients with methylated MGMT promoters treated with temozolomide and radiotherapy demonstrated a median survival of 21.7 months, whereas patients without MGMT promoter methylation exhibited a median survival of 15.3 months under the same treatment regimen [146]. Beyond genetic and epigenetic markers, the impact of standard chemotherapies such as temozolomide on the efficacy of immunotherapy also warrants consideration.

6.6 Impact of temozolomide and chemotherapy on immunotherapy efficacy

Temozolomide (TMZ), the standard chemotherapeutic agent for GBM, exerts complex immunomodulatory effects that influence the outcome of immunotherapy. While TMZ can induce immunogenic cell death of tumor cells and transiently deplete lymphocytes to create "space" for adoptive T-cell therapies, its lymphodepletion is often profound and persistent at standard dosing, resulting in reduced effector T-cell numbers and increased relative frequencies of Tregs and myeloid-derived suppressor cells (MDSCs) [147]. In murine GBM models, the combination of anti-PD-1 therapy with TMZ produced superior short-term tumor control compared to monotherapy, yet it abolished the establishment of durable antitumor immunological memory, which was preserved only with anti-PD-1 alone [148]. Likewise, Karachi et al. demonstrated that standard high-dose TMZ combined with anti-PD-1 increased Treg frequency and upregulated PD-L1 expression on tumor cells, thereby attenuating checkpoint blockade efficacy, whereas metronomic low-dose TMZ was less suppressive but still promoted Treg expansion

[149]. Clinically, these observations help explain why concurrent TMZ has not consistently enhanced immunotherapy outcomes and suggest alternative strategies such as scheduling immunotherapy during TMZ breaks, employing shorter or dose-dense regimens to trigger immunogenic cell death without long-term lymphopenia, or supporting lymphocyte recovery with hematopoietic growth factors [150]. Other alkylating agents like lomustine (CCNU) similarly cause lymphocyte nadirs that can blunt vaccine or adoptive cell therapy responses [151]. Furthermore, chemotherapy-induced DNA damage can increase PD-L1 expression on tumor and myeloid cells, providing a rationale for rational sequencing with checkpoint blockade only when sufficient immune effector cells remain present [152]. In summary, standard cytotoxic regimens act as a double-edged sword: they may promote antigen release and transiently reset immunosuppressive circuits, but they also deplete the very immune populations required for sustained tumor control [153]. Future GBM protocols may therefore need to use chemotherapy in immunomodulatory doses or carefully timed schedules to maximize synergy with immunotherapy rather than antagonize it [154].

6.7 Clinical and preclinical trials of immunotherapy in GBM

Over the past decade, numerous clinical and preclinical studies have evaluated immunotherapeutic strategies for GBM. These include checkpoint inhibitors, dendritic cell and peptide vaccines, CAR-T and CAR-NK therapies, oncolytic viruses, gene therapies, and novel modalities such as exosome-based delivery and STING pathway activation [43]. Table 4 summarizes selected trials from 2014 to 2025, highlighting their phase, target, key outcomes, and translational relevance (Table 4).

6.7.1 Critical appraisal of immunotherapy trials (Table 4)

The trials summarized in Table 4 illustrate both the progress and limitations of immunotherapy in GBM. While checkpoint blockade and peptide vaccines have shown minimal benefit as monotherapies, vaccines such as DCVax-L and SurVaxM suggest that durable survival is achievable in selected patients, particularly in the newly diagnosed setting. Oncolytic viruses, especially PVSRIPO and DNX-2401, have also provided encouraging survival signals in subsets of recurrent GBM patients. Cell-based therapies such as CAR-T have proven safe but are hampered by antigen loss and the highly suppressive tumor microenvironment, highlighting the need for multi-target or combination strategies. Preclinical advances, including STING agonists and exosomal delivery platforms, point to novel ways of reshaping immune responses but remain early in development. Overall, the clinical evidence to date indicates that no single immunotherapy is sufficient in GBM. The next phase of research should focus on rational combinations, patient stratification using biomarkers, and integration of translational endpoints to guide mechanistic understanding. For clinical practice, these therapies remain investigational, but they provide a foundation for designing the next generation of trials aimed at making GBM an immunologically manageable disease.

Although personalization of immunotherapy through biomarkers, neoantigen vaccines, and TCR-engineered cells offers a tailored approach, additional systemic and technological innovations are needed to maximize treatment outcomes. Two emerging frontiers, namely the gut microbiome and artificial intelligence, illustrate how cross-disciplinary advances can further reshape GBM immunotherapy.

Table 4 Summary of clinical and preclinical studies from 2014 to 2025

Therapy type	Phase	Subjects	Target	Key findings	Status	Reference
PD-1 inhibitor (Nivolumab, CheckMate 143)	Phase III	369 (rGBM patients)	PD-1/PD-L1	No OS benefit vs bevacizumab	Completed	[64]
DCVax-L (autologous tumor lysate-loaded DC)	Phase III	331 (ND & rGBM)	Tumor antigens	Improved OS in ND GBM (19.3 vs ~ 16 mo); 5-yr survival 13%	Completed	[155]
Rindopepimut (EGFRvIII vaccine)	Phase III	745 (ND EGFRvIII + GBM)	EGFRvIII	No OS benefit; trial stopped for futility	Terminated	[156]
SurVaxM (survivin-targeting peptide vaccine)	Phase IIa	63 (ND GBM)	Survivin	Improved OS (25.9 mo); robust immune response	Completed	[157]
CAR-T therapy (EGFRvIII)	Phase I	10 (rGBM)	EGFRvIII	Safe, but transient effect; antigen loss observed	Completed	[158]
CAR-T therapy (IL13Rα2)	Phase I	65 (across studies)	IL13Rα2	Safe with some responses; limited OS benefit	Completed	[101]
Oncolytic adenovirus (DNX-2401) + anti–PD-1	Phase I/II	49 (rGBM)	Adenovirus + PD-1	52.7% 12-mo OS; durable responses in subset	Completed	[159]
Gene therapy (Ad–RTS-hIL-12) + veledimex (with/without nivolumab)	Phase I/IB	31 (rGBM)	IL-12	Controllable intratumoral IL-12 expression; OS ~ 12.7 mo (high dose); lymphocyte infiltration noted	Completed	[160]
Oncolytic poliovirus (PVSRIPO)	Phase I	61 (rGBM)	Poliovirus	21% 3-year survival; safe with some long-term benefit	Completed	[161]
Indoximod (IDO inhibitor) + Temozolomide ± Bevacizumab / Radiotherapy	Phase I/II	160 patients (primary malignant brain tumor)	IDO	Ongoing trial assessing MTD and efficacy of indoximod + TMZ. Includes 3 cohorts (with/without bevacizumab or radiosurgery); correlative immune studies planned.	A-NR (Active, Not Recruiting)	[162]
Exosomal STAT6-ASO delivery	Preclinical	Murine GBM models	STAT6 (TAM reprogramming)	Reprogrammed M2 to M1 macrophages; enhanced antitumor immunity	Preclinical	[163]
STING agonist + anti–PD-1	Preclinical	Murine GBM models	STING pathway	Increased immune infiltration; prolonged survival	Preclinical	[130]

7 Systemic and digital enhancements

7.1 Microbiome–Gut–Brain–Immune axis

An unexpected player in GBM treatment outcomes has recently emerged: the gut microbiome. The composition of the gut microbiota has been shown to profoundly influence systemic immunity and the efficacy of immunotherapies in multiple cancers, and mounting evidence suggests a similar connection exists for brain tumors [164]. The concept of a gut–brain–immune axis implies that microbes in the gastrointestinal tract can modulate immune cell development and function peripherally, which in turn can affect the brain's immune environment and tumor behavior.

Several clinical and preclinical observations support the microbiome's role in GBM. Notably, an analysis from a Phase I/II trial (evaluating PD-L1 blockade in newly diagnosed GBM) found that patients who had longer survival on the checkpoint inhibitor had distinct gut microbiome signatures compared to those with shorter survival [165–167].

Mechanistically, the gut microbiota can impact GBM through various pathways [168]. (1) Microbial metabolites: Commensal bacteria produce metabolites (such as short-chain fatty acids, tryptophan metabolites, or polyamines) that enter circulation and can cross into the brain or act on immune cells in lymphoid organs. These metabolites can influence the maturation and polarization of immune cells. For example, certain SCFAs promote anti-tumor cytotoxic T cells, whereas others might promote Tregs; the net effect can alter the immune readiness to attack a brain tumor [168, 169]. (2) Immune conditioning: The gut microbiome continually educates the host immune system. A diverse and "healthy" microbiome tends to prime the immune system for robust responses (enhancing dendritic cell function and effector T cell generation), whereas dysbiosis might lead to a predominance of suppressive immune cells or chronic inflammatory states that paradoxically exhaust the immune system. In GBM models, antibiotics that disrupt the microbiome have been shown to reduce the efficacy of immune therapies, indicating the microbiome's supportive role in treatment response (observed in other cancers, now being examined in GBM specifically). (3) Microbial antigens: There is a hypothesis of molecular mimicry – certain microbial antigens could induce T cells that cross-react with brain tumor antigens, thereby helping immune surveillance of the tumor. (4) Gut–brain communication: Beyond humoral factors, neural pathways (like the vagus nerve) can transmit signals from gut microbes that influence brain glial cells and cytokine levels in the CNS, potentially affecting TME [168, 170, 171].

Capitalizing on this axis, researchers are exploring interventions such as probiotics, diet modulation, or fecal microbiota transplantation (FMT) in GBM. Early preclinical work suggests that colonizing mice with specific beneficial bacteria (like those from the *Bifidobacterium* or *Akkermansia* genera, known to boost checkpoint inhibitor efficacy in other cancers) can improve anti-GBM immune activity. In parallel, FMT from long-term GBM survivors or responders into germ-free or antibiotic-treated mice is being tested to see if it can transfer a pro-immunotherapy phenotype. Given the importance of gut microbes, some have proposed that GBM patients might be stratified by their microbiome profiles in the future, or even conditioned with microbiome therapy before starting immunotherapy [165, 168, 172]. Indeed, there are case reports where compassionate-use FMT was associated with unexpected tumor control in glioma, though it's far from proven.

The advantages of targeting the microbiome are that it is relatively accessible and malleable. Altering a patient's microbiome via diet or FMT is non-invasive compared to altering their immune cells or brain directly. It could also have a broad effect, enhancing multiple arms of the immune system simultaneously (innate and adaptive). Moreover, microbiome profiling might serve as a biomarker to predict who will respond to a given immunotherapy, a tool for personalization [168].

However, challenges abound. The microbiome is incredibly complex and individual-specific; it's difficult to pinpoint which microbes or metabolites are truly key for anti-tumor immunity versus which are bystanders. There is also a risk that changing the microbiome (especially via FMT) could have unintended consequences, like infections or immune-mediated diseases. In the context of brain tumors, an added complexity is that GBM itself can perturb the gut microbiome through tumor-induced systemic effects (stress hormones, etc.), creating a bidirectional puzzle [173]. Rigorous clinical trials will be needed to determine if deliberate microbiome modulation can reproducibly improve GBM patient outcomes. As of now, several centers are initiating pilot trials of FMT in GBM patients receiving immunotherapy, and others are testing diets high in fermentable fiber (to promote beneficial SCFA-producing gut bacteria) during chemo-radiation and immunotherapy [168].

In conclusion, the gut–brain–immune axis represents a frontier in GBM therapy. While not a direct "drug" in the traditional sense, the microbiome's influence is such that it could be considered an adjuvant target: an integral part of a patient's makeup that we can optimize to tip the balance in favor of anti-tumor immunity. Future GBM immunotherapy protocols may include a regimen for the microbiome, just as they include lymphodepletion or adjuvant cytokines today.

While microbiome modulation focuses on systemic immune conditioning, computational modeling and artificial intelligence provide complementary tools that directly optimize the design, delivery, and monitoring of GBM immunotherapies.

7.2 Computational modeling and AI-guided immunotherapy

To overcome GBM's complexity, computational modeling and artificial intelligence (AI) are increasingly being used to optimize immunotherapy design. Computational models can simulate how immune cells traffic, interact, and kill tumor cells in the brain, offering insights that guide delivery strategies [174]. For instance, mathematical models of CAR T-cell distribution predicted that intraventricular infusion would enable broader cerebrospinal fluid (CSF) dispersion, whereas intratumoral injection would achieve higher local concentrations but limited spread. These predictions aligned with clinical observations in an IL13Rα2 CAR trial, where dual intratumoral plus intraventricular delivery led to robust local control and suppression of leptomeningeal disease [175, 176]. Modeling CAR T-cell kinetics has also informed dose selection by predicting non-monotonic dose–response relationships; lower initial doses may suffice because CAR T-cells can proliferate in vivo, thereby avoiding excessive immune activation [174, 177]. Furthermore, "virtual clinical trials" incorporating T-cell expansion rates, antigen heterogeneity, and tumor immunosuppression have been conducted in silico, testing thousands of dose, schedule, and combination permutations to identify optimal regimens for experimental validation [178].

Case studies of computationally guided CARs illustrate the translational power of these approaches. Structure-based design of IL13 muteins produced CARs selective for IL13Rα2, reducing off-target effects on IL13Rα1 while maintaining efficacy in orthotopic GBM models, where 0.3–0.36 × 10^6 CAR T cells were delivered intratumorally or 10 × 10^6 intravenously [179]. Similarly, de novo protein binder–based CARs targeting EGFR and CD276 achieved superior persistence and functionality in xenograft models when 1–2 × 10^6 CAR T cells were administered intracranially, outperforming conventional scFv CARs by enhancing surface stability and reducing exhaustion marker expression. Together, these examples demonstrate how computational design can refine CAR binding domains, improve delivery route selection, and calibrate cell dosing to maximize safety and efficacy [180, 181].

Beyond CAR T-cell therapy, AI is also uncovering novel therapeutic angles. A machine learning–driven analysis of GBM gene regulatory networks identified "fate-determination" genes that, when modulated, reprogrammed GBM cells into dendritic-like antigen-presenting cells. In mouse models, combining this AI-guided reprogramming with checkpoint blockade or dendritic cell vaccination improved survival by approximately 75% compared to controls, exemplifying AI's potential to reveal non-intuitive therapeutic strategies [182].

Clinical applications of AI are beginning to inform real-world decision-making. In a multicenter phase II study of PD-L1 inhibition (durvalumab) in GBM, radiomics-based machine learning on the first on-treatment MRI predicted overall and progression-free survival with external validation (concordance index ≈0.69–0.75), supporting AI as a tool for response prediction and patient stratification [183]. In parallel, multicenter radiomics analyses have shown that MR-perfusion–derived features can distinguish pseudoprogression from true progression in GBM, reporting around 90% accuracy with strong AUCs, which directly addresses a common diagnostic dilemma during immunotherapy and chemoradiation follow-up [184]. Complementary work with [^{18}F]FET PET radiomics has also demonstrated improved discrimination and prognostication in newly diagnosed IDH-wildtype GBM, highlighting multimodal imaging pipelines for clinical deployment [185].

AI-enabled adaptive trial methodologies are also entering GBMb Bayesian response-adaptive randomization to evaluate multiple regimens under a seamless phase II/III master protocol, dynamically enriching patients to better-performing arms and expediting go/no-go decisions [186]. Within cell-therapy development more broadly, a subset of early CAR-T trials have already implemented model-based or model-assisted Bayesian dose-finding designs (for example, EffTox, mCRM, BOIN), illustrating how adaptive statistics can identify an "optimal biological dose" rather than the traditional maximum tolerated dose, although uptake remains limited to date [187].

In summary, computational simulations and AI not only accelerate therapeutic discovery but also optimize delivery, dosing, and personalization of GBM immunotherapies. These approaches offer tangible clinical utility today in tasks such as response prediction and PsP discrimination, and they are beginning to shape adaptive trial platforms. Nonetheless, broader validation, standardization, and regulatory alignment are still required before widespread adoption [184]. To provide a structured overview of these systemic and digital strategies, Table 5 summarizes the emerging roles of the microbiome–gut–brain–immune axis and artificial intelligence in GBM immunotherapy. The

Table 5 Systemic and digital enhancements in GBM immunotherapy: mechanisms, evidence, and clinical translation

Domain	Specific mechanism / approach	Representative evidence (preclinical/clinical)	Clinical status / translation	Advantages / clinical promise	Challenges / limitations	Key references
Microbiome–Gut–Brain–Immune Axis	Microbial metabolites (SCFAs, tryptophan derivatives, polyamines)	SCFAs modulate Treg vs. cytotoxic T-cell polarization; tryptophan metabolites influence microglia activation	Pilot dietary fiber interventions under study in GBM patients on chemoradiation	Easily modified by diet; broad systemic and CNS immune effects	Difficult to isolate key metabolites; individual microbiome variability	[168, 169]
	Immune conditioning by commensal diversity	Antibiotic-induced dysbiosis reduces immunotherapy efficacy; healthy microbiome enhances dendritic and T-cell priming	Observed in other cancers, exploratory analyses in GBM immunotherapy	Shapes systemic immunity; potential to stratify patients	Microbiome signatures not standardized; dysbiosis risk	[165–168]
	Microbial antigens (molecular mimicry)	Hypothesized cross-reactive T-cell responses between gut microbes and GBM antigens	Preclinical only, no clinical confirmation	May enhance anti-tumor surveillance	Hypothetical; requires antigen mapping	[170, 171]
	Gut–brain neural signaling (vagus-mediated)	Gut microbial activity influences CNS cytokines and glial cells	Preclinical models, limited direct GBM data	Novel neuromodulation angle for GBM	Mechanistic links poorly mapped; indirect evidence only	[171]
	FMT and probiotics	FMT from long-term GBM survivors enhanced response in murine models; probiotics (Bifidobacterium, Akkermansia) boost checkpoint efficacy in other cancers	Pilot FMT trials in GBM immunotherapy ongoing	Non-invasive, patient-friendly, potentially low-cost	Safety concerns (infection, autoimmunity); lack of controlled GBM data	[165, 172, 173]
Computational Modeling	In silico CAR T-cell trafficking/dosing	Predicted intraventricular + intratumoral infusion superior to single-route; non-linear dose–response relationships identified	Models validated against IL13Rα2 CAR trial	Accelerates dosing/delivery optimization	Relies on assumptions; tumor heterogeneity difficult to capture	[174–177]
	Virtual clinical trials	Simulated thousands of permutations for dose, schedule, and combinations in silico	Preclinical tool; not yet clinical practice	Reduces cost/time of early-phase design	Translation limited; regulatory acceptance lacking	[178]
AI-Guided Immunotherapy Design	AI-driven reprogramming of GBM fate	ML identified "fate-determination" genes; reprogramming GBM → dendritic-like cells boosted survival in mice	Preclinical	Reveals novel therapeutic angles; synergies with checkpoint blockade	Unproven in humans; genetic manipulation complexity	[182]
	Computational CAR engineering (IL13 muteins, EGFR/CD276 binders)	Designed CARs showed improved persistence, reduced exhaustion, and reduced off-target toxicity in GBM models	Preclinical to early-phase preclinical validation	Enhances CAR stability, specificity, durability	Protein design complexity; manufacturing hurdles	[179–181]

Table 5 (continued)

Domain	Specific mechanism / approach	Representative evidence (preclinical/clinical)	Clinical status / translation	Advantages / clinical promise	Challenges / limitations	Key references
AI in Clinical Decision-Making	Radiomics for response prediction	Radiomics on early MRI predicted OS/PFS in PD-L1 (durvalumab) GBM trial	Multicenter phase II, external validation achieved	Supports patient stratification; early treatment adaptation	Requires harmonization of imaging protocols; moderate accuracy (~0.7 C-index)	[183]
	PsP vs true progression discrimination	MR-perfusion radiomics achieved ~90% accuracy; [18F]FET PET radiomics improved prognostication in IDH-wildtype GBM	Multiple multicenter retrospective studies	Addresses a key clinical dilemma in immunotherapy monitoring	Validation needed in larger prospective trials	[184, 185]
	Adaptive AI-enabled trial design	GBM AGILE uses Bayesian randomization across multiple regimens	Actively enrolling global trial platform	Efficient drug screening; enriches patient allocation	Complexity in regulatory oversight; limited uptake in early CAR-T studies	[186, 187]

table highlights their underlying mechanisms, representative evidence, clinical implications, and current challenges, offering a concise framework that complements the narrative discussion and illustrates translational opportunities (Table 5).

8 Challenges and future directions

Over the past 10 years, Immunotherapy is increasingly recognized as an effective means of mobilizing immune defenses against aggressive brain tumors, such as GBM [43]. The overall effect of these therapies on patient survival is still unclear, and additional clinical and biological obstacles persist, limiting their effectiveness [188]. Genetic and phenotypic variation within tumors helps to create GBM, which presents a major difficulty for therapy advancement [43, 189]. This variation makes it more difficult to find reliable therapeutic targets and causes different patient treatment reactions. The lack of consistent treatment strategies across different research studies impairs repeatability and weakens the strength of clinical findings [189]. The infrequency and rapid advancement of GBM limit patient participation in clinical trials, leading to small study cohorts that are inadequate for comprehensive statistical evaluation [188, 190]. Despite promising results from preclinical studies, only a limited number have progressed to large-scale clinical trials with human participants [190].

For instance, despite compelling preclinical data, IDO inhibitors ultimately failed to improve survival in large phase III trials [191, 192], underscoring the challenge of translating metabolic checkpoint blockade into clinical benefit. Similarly, rindopepimut, an EGFRvIII-targeted peptide vaccine, elicited strong immunogenicity in early-phase studies but did not demonstrate survival advantage in a definitive phase III trial in newly diagnosed GBM [193, 194]. These examples highlight the translational gap between laboratory efficacy and clinical outcomes, emphasizing the need for carefully designed trials and patient stratification to bridge this divide.

The complexity of TME presents a significant challenge to the efficacy of immunotherapy [43, 189]. Immunosuppressive cells and cytokines, along with the protective characteristics of the BBB, hinder the infiltration and function of immune cells in the tumor environment [43, 189, 195]. The collective influence of these factors establishes a hostile environment for immunotherapeutic agents, thereby diminishing their efficacy [43]. Ongoing research is enhancing our understanding of the interactions between gliomas and the immune system, suggesting potential for more effective interventions [189, 195]. Future investigations should concentrate on several critical areas to address these limitations. The main objective is to examine combination strategies that integrate immunotherapy with conventional treatments, such as radiotherapy, chemotherapy, or molecularly targeted therapies [43, 195]. These combinations may produce synergistic effects and improve patient outcomes. Improving the delivery of immunotherapeutic agents across the BBB is essential, as enhanced delivery systems can significantly increase treatment efficacy [188, 189]. Moreover, customizing immunotherapy to the particular molecular and genetic characteristics of each patient could help to solve GBM's natural diversity [195]. Personalized strategies could enhance accuracy and treatment results. Validating results from experimental investigations and creating uniform treatment procedures depend on carefully planned large-scale, multicenter clinical trials [188, 190]. Addressing these challenges is crucial for the effective translation of immunotherapy from laboratory research to clinical application [43, 189, 195]. Through ongoing research and innovation, immunotherapy may become a crucial component of GBM treatment. Nevertheless, significant barriers remain, and translating these advances into consistent clinical benefit is still the key challenge ahead.

9 Conclusion

Glioblastoma (GBM) remains one of the most aggressive and treatment-resistant primary brain tumors, with standard therapies such as surgery, radiotherapy, and chemotherapy offering only limited survival benefits. Despite intensive research, major barriers including tumor heterogeneity, the immunosuppressive tumor microenvironment, and restricted drug penetration across the blood–brain barrier continue to undermine therapeutic progress. Recent advances in immunotherapy have introduced diverse approaches, including immune checkpoint inhibitors, CAR-T and CAR-NK cells, cytokine-based therapies, bispecific antibodies, RNA-based platforms, and nanoparticle-mediated delivery systems. Personalized strategies guided by molecular and immunological biomarkers have also gained momentum. However, the translation of these modalities to consistent clinical benefit remains challenging. Past failures, such as the lack of efficacy of IDO inhibitors in phase III trials and the discontinuation of the EGFRvIII vaccine rindopepimut despite strong early immunogenicity, underscore that single-agent immunotherapies are rarely sufficient and that rational combinations with careful patient selection are essential. Emerging systemic and digital innovations, including modulation of the gut–brain–immune axis and AI-guided adaptive trial designs, are beginning to complement traditional immunotherapy approaches. These novel dimensions may accelerate trial efficiency, refine patient stratification, and uncover non-intuitive therapeutic synergies. Moving forward, the most promising path lies in integrating immunotherapies with existing standard treatments in well-designed, biomarker-driven clinical trials, while simultaneously improving delivery systems and addressing the

barriers of the GBM microenvironment. Through these combined efforts, immunotherapy holds the potential not only to extend survival but also to gradually transform GBM into a more manageable disease.

Abbreviations

GBM	Glioblastoma
BBB	Blood-brain barrier
WHO	World Health Organization
TME	The tumor microenvironment
ECM	Extracellular matrix
TAMs	Tumor-associated macrophages
Tregs	Regulatory T cells
MDSCs	Myeloid-derived suppressor cells
CAR	Chimeric antigen receptor
IDO	Indoleamine 2,3-dioxygenase 1

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References

1. Lechpammer M, Rao R, Shah S, Mirheydari M, Bhattacharya D, Koehler A, et al. Advances in immunotherapy for the treatment of adult glioblastoma: overcoming chemical and physical barriers. Cancers. 2022. https://doi.org/10.3390/cancers14071627.
2. Au TH, Willis C, Reblin M, Peters KB, Nghiemphu PL, Taylor JW, et al. Caregiver burden by treatment and clinical characteristics of patients with glioblastoma. Support Care Cancer. 2022;30(2):1365–75.
3. You H, Geng S, Li S, Imani M, Brambilla D, Sun T, et al. Recent advances in biomimetic strategies for the immunotherapy of glioblastoma. Biomaterials. 2024;311:122694.
4. Robert T. Immune privilege in the central nervous system: implications for neuroinflammatory diseases. Research Invention Journal of Biological and Applied Sciences. 2024.
5. Zhou C, Xu H, Luo J. Meningeal lymphatic vasculature, a general target for glioblastoma therapy? Fundam Res. 2023;4:267–9.
6. Frederico S, Hancock J, Brettschneider E, Ratnam N, Gilbert M, Terabe M. Making a cold tumor hot: the role of vaccines in the treatment of glioblastoma. Front Oncol. 2021. https://doi.org/10.3389/fonc.2021.672508.
7. Schulte SC, Peter W, Rosenberger G, Schäfer M, Maire CL, Rünger A, et al. Somatic mutations in HLA class genes and antigen-presenting molecules in malignant glioma. Cancer Immunol Res. 2025;13(7):1111–23.
8. Kawengian KJ, Wanandi S. Evasion of the immune system by glioblastoma multiforme: an obstacle to achieving effective therapies. Mol Cell Biomed Sci. 2024. https://doi.org/10.21705/mcbs.v8i2.434.
9. Low JJW, Sulaiman S, Johdi NA, Abu N. Immunomodulatory effects of extracellular vesicles in glioblastoma. Front Cell Dev Biol. 2022. https://doi.org/10.3389/fcell.2022.996805.
10. Ratnam N, Sonnemann H, Frederico S, Chen H, Hutchinson M-K, Dowdy T, et al. Reversing epigenetic gene silencing to overcome immune evasion in CNS malignancies. Front Oncol. 2021. https://doi.org/10.3389/fonc.2021.719091.
11. Gangoso E, Southgate B, Bradley L, Rus S, Gálvez-Cancino F, McGivern N, et al. Glioblastomas acquire myeloid-affiliated transcriptional programs via epigenetic immunoediting to elicit immune evasion. Cell. 2021;184:2454–70.
12. Cao TQ, Wainwright DA, Lee-Chang C, Miska J, Sonabend AM, Heimberger AB, et al. Next steps for immunotherapy in glioblastoma. Cancers. 2022. https://doi.org/10.3390/cancers14164023.

13. Zhu P, Li SY, Ding J, Fei Z, Sun SN, Zheng ZH, et al. Combination immunotherapy of glioblastoma with dendritic cell cancer vaccines, anti-PD-1 and poly I:C. J Pharm Anal. 2023;13(6):616–24.
14. Xu X, Chen W, Zhu W, Chen J, Ma B, Ding J, et al. Adeno-associated virus (AAV)-based gene therapy for glioblastoma. Cancer Cell Int. 2021;21(1):76.
15. Price M, Ballard C, Benedetti J, Neff C, Cioffi G, Waite KA, et al. CBTRUS statistical report: primary brain and other central nervous system tumors diagnosed in the United States in 2017–2021. Neuro Oncol. 2024;26(Supplement_6):vi1–85.
16. Jovanovich N, Habib A, Chilukuri A, Hameed NUF, Deng H, Shanahan R, et al. Sex-specific molecular differences in glioblastoma: assessing the clinical significance of genetic variants. Front Oncol. 2024. https://doi.org/10.3389/fonc.2023.1340386.
17. Goyani P, Christodoulou R, Vassiliou E. Immunosenescence: aging and immune system decline. Vaccines. 2024. https://doi.org/10.3390/vaccines12121314.
18. Liu Z, Liang Q, Ren Y, Guo Cc, Ge X, Wang L, et al. Immunosenescence: molecular mechanisms and diseases. Signal Transduct Target Ther. 2023. https://doi.org/10.1038/s41392-023-01451-2.
19. Salminen A. Immunosuppressive network promotes immunosenescence associated with aging and chronic inflammatory conditions. J Mol Med. 2021;99:1553–69.
20. Salminen A, Kaarniranta K, Kauppinen A. Immunosenescence: the potential role of myeloid-derived suppressor cells (MDSC) in age-related immune deficiency. Cell Mol Life Sci. 2019;76:1901–18.
21. Salminen A. Activation of immunosuppressive network in the aging process. Age Res Rev. 2019. https://doi.org/10.1016/j.arr.2019.100998.
22. Santoro A, Bientinesi E, Monti D. Immunosenescence and inflammaging in the aging process: age-related diseases or longevity? Ageing Res Rev. 2021. https://doi.org/10.1016/j.arr.2021.101422.
23. Sundaram T, Ahmed S. Age, autoimmunity, and inflammation: the curious case of immunosenescence and inflammaging. Anti Aging East Eur. 2022. https://doi.org/10.56543/aaeeu.2022.1.1.04.
24. Guégan M, Bichon M, Chaput N, Houot R, Lemoine J. Cancer immunotherapy in elderly patients: the concept of immune senescence challenged by clinical experience. Eur J Cancer. 2024;214:115145.
25. Ziogas D, Theocharopoulos C, Aravantinou K, Boukouris A, Stefanou D, Anastasopoulou A, et al. Clinical benefit of immune checkpoint inhibitors in elderly cancer patients: current evidence from immunosenescence pathophysiology to clinical trial results. Crit Rev Oncol Hematol. 2025. https://doi.org/10.1016/j.critrevonc.2025.104635.
26. Ladomersky E, Scholtens D, Kocherginsky M, Hibler E, Bartom E, Otto-Meyer S, et al. The coincidence between increasing age, immunosuppression, and the incidence of patients with glioblastoma. Front Pharmacol. 2019. https://doi.org/10.3389/fphar.2019.00200.
27. Ladomersky E, Otto-Meyer S, Zhai L, Savoor R, Lauing K, Lenzen A, et al. IMMU-46. glioblastoma patient diagnoses and immunosuppression are maximal during old age: a random coincidence, or cause and effect? Neuro-Oncol. 2018. https://doi.org/10.1093/neuonc/noy148.549.
28. Ukibe N, Dike CR, Ihim A, Ukibe E, Ukibe B, Ukibe V, et al. Immunological changes with age and innovative approaches to bolster immune function in older adults. IDOSR J Sci Res. 2024. https://doi.org/10.59298/IDOSRJSR/2024/1.1.11.100.
29. Fekete B, Werlenius K, Tisell M, Pivodic A, Smits A, Jakola AS, et al. What predicts survival in glioblastoma? A population-based study of changes in clinical management and outcome. Front Surg. 2023. https://doi.org/10.3389/fsurg.2023.1249366.
30. Luckett PH, Olufawo M, Lamichhane B, Park KY, Dierker D, Verastegui GT, et al. Predicting survival in glioblastoma with multimodal neuroimaging and machine learning. J Neurooncol. 2023;164(2):309–20.
31. Deshpande R, K C, Panigrahi M, Babu P. Prognostic significance of anatomic origin and evaluation of survival statistics of Astrocytoma patients—a tertiary experience. Indian J Surg Oncol. 2018;10:55–60.
32. Padwal J, Dong X, Hirshman B, Hoi-Sang U, Carter B, Chen C. Superior efficacy of gross total resection in anaplastic astrocytoma patients relative to glioblastoma patients. World Neurosurg. 2015;90:186–93.
33. Belyaev A, Kobyakov G, Shmakov P, Efremov K, Pronin I, Usachev D. [Prognosis of overall and disease-free survival in patients with grade 3 astrocytomas (anaplastic astrocytoma, WHO 2016)]. Burdenko J Neurosurg. 2023;87(4):46–57.
34. Shonka N, Theeler B, Cahill D, Yung A, Smith L, Lei X, et al. Outcomes for patients with anaplastic astrocytoma treated with chemoradiation, radiation therapy alone or radiation therapy followed by chemotherapy: a retrospective review within the era of temozolomide. J Neuro-Oncol. 2013;113:305–11.
35. Rousseau J, Desforges S-M, Jabbour G, Lemieux B, Lapointe S, Bélanger K, et al. BIOS-02. Clinical outcomes of over 600 patients with glioblastoma treated at a Canadian tertiary center in the past 15 years: a comparative analysis with the pivotal Stupp trial. Neuro-oncol. 2022. https://doi.org/10.1093/neuonc/noac209.078.
36. Lin H, Liu C, Hu A, Zhang D, Yang H, Mao Y. Understanding the immunosuppressive microenvironment of glioma: mechanistic insights and clinical perspectives. J Hematol Oncol. 2024. https://doi.org/10.1186/s13045-024-01544-7.
37. Chen J, Gao Y, Zhong J, Wu X, Leng Z, Liu M, et al. Lnc-H19-derived protein shapes the immunosuppressive microenvironment of glioblastoma. Cell Rep Med. 2024. https://doi.org/10.1016/j.xcrm.2024.101806.
38. Liu J, Piranlioglu R, Ye F, Shu K, Lei T, Nakashima H. Immunosuppressive cells in oncolytic virotherapy for glioma: challenges and solutions. Front Cell Infect Microbiol. 2023. https://doi.org/10.3389/fcimb.2023.1141034.
39. Tianfei SG, Wu Q, Shen M, Feng X, Zhang Z-C. Tumor-derived extracellular vesicles: how they mediate glioma immunosuppression. Mol Biol Rep. 2024;51(1):235.
40. Yin B, Cai Y, Chen L, Li Z, Li X. Immunosuppressive MDSC and Treg signatures predict prognosis and therapeutic response in glioma. Int Immunopharmacol. 2024;141:112922.
41. Haist M, Stege H, Grabbe S, Bros M. The functional crosstalk between Myeloid-Derived Suppressor Cells and Regulatory T Cells within the immunosuppressive tumor microenvironment. Cancers. 2021. https://doi.org/10.3390/cancers13020210.
42. Wang H, Zhou F, Qin W, Yang Y, Li X, Liu R. Metabolic regulation of myeloid-derived suppressor cells in tumor immune microenvironment: targets and therapeutic strategies. Theranostics. 2025;15:2159–84.
43. Liu Y, Zhou F, Ali H, Lathia JD, Chen P. Immunotherapy for glioblastoma: current state, challenges, and future perspectives. Cell Mol Immunol. 2024;21(12):1354–75.
44. Golán-Cancela I, Caja L. The TGF-β family in glioblastoma. Int J Mol Sci. 2024. https://doi.org/10.3390/ijms25021067.
45. Liu Y, Su C, Wei X, Wei N, Qian Q, Xu Z. Prospects and applications of NK therapy in the treatment of gliomas (review). Oncol Rep. 2025. https://doi.org/10.3892/or.2025.8921.

46. Lin H, Liu C, Hu A, Zhang D, Yang H, Mao Y. Understanding the immunosuppressive microenvironment of glioma: mechanistic insights and clinical perspectives. J Hematol Oncol. 2024;17(1):31.
47. Su H, Peng Y, Wu Y, Zeng X. Overcoming immune evasion with innovative multi-target approaches for glioblastoma. Front Immunol. 2025;16:1541467.
48. Li X, Meng H, Wang H, Zhang Y, Yu W. Indoleamine 2,3-dioxygenase 1 in cancer immunotherapy: from small-molecule inhibition to PROTAC-mediated degradation. Front Pharmacol. 2025. https://doi.org/10.3389/fphar.2025.1640073.
49. Liu M, Wang X, Wang L, Ma X, Gong Z, Zhang S, et al. Targeting the IDO1 pathway in cancer: from bench to bedside. J Hematol Oncol. 2018;11(1):100.
50. Ricklefs FL, Alayo Q, Krenzlin H, Mahmoud AB, Speranza MC, Nakashima H, et al. Immune evasion mediated by PD-L1 on glioblastoma-derived extracellular vesicles. Sci Adv. 2018;4(3):eaar2766.
51. Dai J, Jiang Y, Hu H, Zhang S, Chen Y. Extracellular vesicles as modulators of glioblastoma progression and tumor microenvironment. Pathol Oncol Res. 2024. https://doi.org/10.3389/pore.2024.1611549.
52. Wang S, Huang T, Wu Q, Yuan H, Wu X, Yuan F, et al. Lactate reprograms glioblastoma immunity through CBX3-regulated histone lactylation. J Clin Invest. 2024. https://doi.org/10.1172/JCI176851.
53. Xia C, Yin S, To K, Fu L. CD39/CD73/A2AR pathway and cancer immunotherapy. Mol Cancer. 2023. https://doi.org/10.1186/s12943-023-01733-x.
54. Leone R, Emens L. Targeting adenosine for cancer immunotherapy. J Immunother Cancer. 2018. https://doi.org/10.1186/s40425-018-0360-8.
55. Lupo K, Matosevic S. 123 Natural killer cells engineered with an inducible, responsive genetic construct targeting TIGIT and CD73 to relieve immunosuppression within the GBM microenvironment. Journal for ImmunoTherapy of Cancer. 2020
56. Zhang H, Yang L, Han M, Han Y, Jiang Z, Zheng Q, et al. Boost infiltration and activity of T cells via inhibiting ecto-5′-nucleotidase (CD73) immune checkpoint to enhance glioblastoma immunotherapy. ACS Nano. 2024. https://doi.org/10.1021/acsnano.4c04553.
57. Hatfield SM, Sitkovsky M. A2a adenosine receptor antagonists to weaken the hypoxia-HIF-1α driven immunosuppression and improve immunotherapies of cancer. Curr Opin Pharmacol. 2016;29:90–6.
58. Lupo K, Matosevic S. 130 Engineered natural killer cells reactively block TIGIT and CD73 in the GBM microenvironment. Journal for ImmunoTherapy of Cancer. 2021.
59. Bova V, Filippone A, Casili G, Lanza M, Campolo M, Capra A, et al. Adenosine targeting as a new strategy to decrease glioblastoma aggressiveness. Cancers. 2022. https://doi.org/10.3390/cancers14164032.
60. Wang X, Guo G, Guan H, Yu Y, Lu J, Yu J. Challenges and potential of PD-1/PD-L1 checkpoint blockade immunotherapy for glioblastoma. J Exp Clin Cancer Res. 2019. https://doi.org/10.1186/s13046-019-1085-3.
61. Mahoney K, Freeman G, McDermott D. The next immune-checkpoint inhibitors: PD-1/PD-L1 blockade in melanoma. Clin Ther. 2015;37(4):764–82.
62. Chen Y-M. Immune checkpoint inhibitors for nonsmall cell lung cancer treatment. J Chin Med Assoc. 2017;80(1):7–14.
63. Khasraw M, Reardon DA, Weller M, Sampson JH. PD-1 inhibitors: do they have a future in the treatment of glioblastoma? Clin Cancer Res. 2020;26(20):5287–96.
64. Reardon DA, Brandes AA, Omuro A, Mulholland P, Lim M, Wick A, et al. Effect of nivolumab vs bevacizumab in patients with recurrent glioblastoma: the CheckMate 143 phase 3 randomized clinical trial. JAMA Oncol. 2020;6(7):1003–10.
65. Agosti E, Zeppieri M, De Maria L, Tedeschi C, Fontanella MM, Panciani PP, et al. Glioblastoma immunotherapy: a systematic review of the present strategies and prospects for advancements. Int J Mol Sci. 2023. https://doi.org/10.3390/ijms242015037.
66. Chen CH, Chin RL, Hartley GP, Lea ST, Engel BJ, Hsieh CE, et al. Novel murine glioblastoma models that reflect the immunotherapy resistance profile of a human disease. Neuro Oncol. 2023;25(8):1415–27.
67. Buchbinder EI, Desai A. CTLA-4 and PD-1 pathways: similarities, differences, and implications of their inhibition. Am J Clin Oncol. 2016;39(1):98–106.
68. Willsmore ZN, Coumbe BGT, Crescioli S, Reci S, Gupta A, Harris RJ, et al. Combined anti-PD-1 and anti-CTLA-4 checkpoint blockade: treatment of melanoma and immune mechanisms of action. Eur J Immunol. 2021;51(3):544–56.
69. Wang K, Coutifaris P, Brocks D, Wang G, Azar T, Solis S, et al. Combination anti-PD-1 and anti-CTLA-4 therapy generates waves of clonal responses that include progenitor-exhausted CD8(+) T cells. Cancer Cell. 2024;42(9):1582-97.e10.
70. Markus L, Wen P, Sun L, Hugo W, Prins R. IMMU-23. Neoadjuvant anti-PD-1 and anti-CTLA-4 combination therapy induced intratumoral immunologic alterations in recurrent GBM patients. Neuro-oncol. 2024. https://doi.org/10.1093/neuonc/noae165.0616.
71. Sloan A, Gilbert M, Zhang P, Aldape K, Wu J, Rogers L, et al. NRG BN002: Phase I study of checkpoint inhibitors anti-CTLA-4, anti-PD-1, the combination in patients with newly diagnosed glioblastoma. J Clin Oncol. 2018;36:2053.
72. Beug ST, Beauregard CE, Healy C, Sanda T, St-Jean M, Chabot J, et al. Smac mimetics synergize with immune checkpoint inhibitors to promote tumour immunity against glioblastoma. Nat Commun. 2017. https://doi.org/10.1038/ncomms14278.
73. Liu C, Schaettler M, Bowman-Kirigin J, Kobayashi D, Miller C, Johanns T, et al. IMMU-09. Combination immune treatment of a highly aggressive orthotopic murine glioblastoma with checkpoint blockade and multi-valent neoantigen vaccination. Neuro Oncol. 2019. https://doi.org/10.1093/neuonc/noz175.503.
74. Kim J, Patel M, Mangraviti A, Kim E, Theodros D, Velarde E, et al. Combination therapy with Anti-PD-1, Anti-TIM-3, and focal radiation results in regression of murine gliomas. Clin Cancer Res. 2016;23:124–36.
75. Ladomersky E, Zhai L, Lenzen A, Lauing K, Qian J, Scholtens D, et al. IDO1 inhibition synergizes with radiation and PD-1 blockade to durably increase survival against advanced glioblastoma. Clin Cancer Res. 2018;24:2559–73.
76. Martino M, Daviaud C, Vanpouille-Box C. 457 Targeting FASN sensitizes irradiated glioblastoma to immune checkpoint blockade. Regular and Young Investigator Award Abstracts. 2023.
77. Martino M, Daviaud C, Vanpouille-Box C. 447 Dual targeting of PD-1 and CTLA-4 synergizes with focal radiation to durably increase survival against glioblastoma. Regular and Young Investigator Award Abstracts. 2022.
78. Ijaz M, Hasan I, Jiang Z, Ullah Z, Aslam B, Khurshid M, et al. Combinatory immunotherapy for glioblastoma treatment. Adv Ther. 2024. https://doi.org/10.1002/adtp.202400217.
79. Fu M, Xue B, Miao X, Gao Z. Overcoming immunotherapy resistance in glioblastoma: challenges and emerging strategies. Front Pharmacol. 2025. https://doi.org/10.3389/fphar.2025.1584688.

80. Hussein M, Ali E-S. Advancing glioblastoma treatment: challenges and opportunities of immune checkpoint inhibition. Asian J Med Health. 2025. https://doi.org/10.9734/ajmah/2025/v23i11160.
81. Rocha Pinheiro SL, Lemos FFB, Marques HS, Silva Luz M, de Oliveira Silva LG, Faria Souza Mendes Dos Santos C, et al. Immunotherapy in glioblastoma treatment: current state and future prospects. World J Clin Oncol. 2023;14(4):138–59.
82. Shireman JM, Ammanuel S, Eickhoff JC, Dey M. Sexual dimorphism of the immune system predicts clinical outcomes in glioblastoma immunotherapy: a systematic review and meta-analysis. Neuro-Oncol Adv. 2022;4(1):vdac082.
83. Zou Y, Li S, Li Y, Zhang D, Zheng M, Shi B. Glioblastoma cell derived exosomes as a potent vaccine platform targeting primary brain cancers and brain metastases. ACS Nano. 2025;19(18):17309–22.
84. Yamanaka R, Abe T, Yajima N, Tsuchiya N, Homma J, Kobayashi T, et al. Vaccination of recurrent glioma patients with tumour lysate-pulsed dendritic cells elicits immune responses: results of a clinical phase I/II trial. Br J Cancer. 2003;89:1172–9.
85. Liau LM, Ashkan K, Brem S, Campian JL, Trusheim JE, Iwamoto FM, et al. Association of autologous tumor lysate-loaded dendritic cell vaccination with extension of survival among patients with newly diagnosed and recurrent glioblastoma: a phase 3 prospective externally controlled cohort trial. JAMA Oncol. 2023;9(1):112–21.
86. Chiocca E, Yu J, Lukas R, Solomon I, Ligon K, Nakashima H, et al. Regulatable interleukin-12 gene therapy in patients with recurrent high-grade glioma: results of a phase 1 trial. Sci Transl Med. 2019. https://doi.org/10.1126/scitranslmed.aaw5680.
87. Chiocca E, Gelb A, Chen C, Rao G, Reardon D, Wen P, et al. Combined immunotherapy with controlled interleukin-12 gene therapy and immune checkpoint blockade in recurrent glioblastoma: an open-label, multi-institutional phase 1 trial. Neuro-oncol. 2021. https://doi.org/10.1093/neuonc/noab271.
88. Faisal SM, Castro MG, Lowenstein PR. Combined cytotoxic and immune-stimulatory gene therapy using Ad-TK and Ad-Flt3L: translational developments from rodents to glioma patients. Mol Ther. 2023;31(10):2839–60.
89. Lv Z, Dai Y. mRNA vaccines and SiRNAs targeting cancer immunotherapy: challenges and opportunities. Discov Oncol. 2025;16(1):1265.
90. Strika Z, Petković K, Likić R. Effectiveness and safety of mRNA vaccines in the therapy of Glioblastoma. J Pers Med. 2024. https://doi.org/10.3390/jpm14090993.
91. Ahmed T, Alam KT. Biomimetic nanoparticle based targeted mRNA vaccine delivery as a novel therapy for Glioblastoma multiforme. AAPS PharmSciTech. 2025;26(3):68.
92. Sayour E, Grippin A, Leon G, Stover B, Huang J, Deleyrolle L, et al. IMMU-67. Personalized tumor MRNA loaded lipid nanoparticles prime the systemic and intratumoral GBM MILIEU for response to immunotherapy. Neuro Oncol. 2018. https://doi.org/10.1093/neuonc/noy148.567.
93. Karimi-Sani I, Molavi Z, Naderi S, Mirmajidi SH, Zare I, Naeimzadeh Y, et al. Personalized mRNA vaccines in glioblastoma therapy: from rational design to clinical trials. J Nanobiotechnology. 2024;22(1):601.
94. Méndez-Gómez H, Devries A, Stover B, Zhang D, Grippin A, Von Roemeling C, et al. CTIM-28. Multilamellar MRNA lipid particles induce immunologic reprogramming in canine and human glioblastoma patients. Neuro Oncol. 2022. https://doi.org/10.1093/neuonc/noac209.260.
95. Mirzaei S, Mahabady M, Zabolian A, Abbaspour A, Fallahzadeh P, Noori M, et al. Small interfering RNA (siRNA) to target genes and molecular pathways in glioblastoma therapy: current status with an emphasis on delivery systems. Life Sci. 2021. https://doi.org/10.1016/j.lfs.2021.119368.
96. Liu Y, Zheng M, Jiao M, Yan C, Xu S, Du Q, et al. Polymeric nanoparticle mediated inhibition of miR-21 with enhanced miR-124 expression for combinatorial glioblastoma therapy. Biomaterials. 2021;276:121036.
97. Melnick K, Dastmalchi F, Mitchell D, Rahman M, Sayour E. Contemporary RNA therapeutics for glioblastoma. Neuromol Med. 2021;24:8–12.
98. Grippin A, Wummer B, Méndez-Gómez H, Stover B, Huang J, Rinaldi C, et al. IMMU-39. RNA loaded lipid-nanoparticles function as customizable immunotherapeutic vehicles against malignant gliomas. Neuro Oncol. 2019. https://doi.org/10.1093/neuonc/noz175.531.
99. Begley S, O'Rourke D, Binder Z. CAR t cell therapy for glioblastoma: a review of the first decade of clinical trials. Mol Ther. 2025. https://doi.org/10.1016/j.ymthe.2025.03.004.
100. Tanaka R, Caruso H, Ling X, Najjar A, Heimberger A. IMMU-17. Reducing the ex vivo manufacturing time of EGFRVIII-specific chimeric antigen receptor (CAR) T cells improves phenotype and demonstrates potent anti-glioma function in vivo. Neuro Oncol. 2017. https://doi.org/10.1093/neuonc/nox168.476.
101. Brown CE, Hibbard JC, Alizadeh D, Blanchard MS, Natri HM, Wang D, et al. Locoregional delivery of IL-13Rα2-targeting CAR-T cells in recurrent high-grade glioma: a phase 1 trial. Nat Med. 2024;30(4):1001–12.
102. Brown CE, Aguilar B, Starr R, Yang X, Chang WC, Weng L, et al. Optimization of IL13Rα2-targeted chimeric antigen receptor T cells for improved anti-tumor efficacy against glioblastoma. Mol Ther. 2018;26(1):31–44.
103. Shen S, Reedy J, Suryadevara C, Hotchkiss K, Snyder D, Sanchez-Perez L, et al. EXTH-18. Il-12 armored car T cell therapy for heterogeneous glioblastoma. Neuro-oncol. 2021. https://doi.org/10.1093/neuonc/noab196.657.
104. Ma R, Lu T, Li Z, Teng KY, Mansour AG, Yu M, et al. An oncolytic virus expressing IL15/IL15Rα combined with off-the-shelf EGFR-CAR NK cells targets glioblastoma. Cancer Res. 2021;81(13):3635–48.
105. Roller B, Reindl L, Strassheimer F, Ludmirski G, Elleringmann P, Kiefer A, et al. P06.10.A car-engineered primary NK cells show promising effects as targeted therapy against EGFR-positive glioblastoma. Neuro-oncol. 2023. https://doi.org/10.1093/neuonc/noad137.150.
106. Gong Y, Lin W, Fang X, Zhang R, Luo M, Wu H, et al. Advances in cellular immune theranostic approaches for glioblastoma: current trends and future directions. Cancer Innov. 2025;4(4):e70018.
107. Zhao C, Zhu X, Tan J, Mei C, Cai X, Kong F. Lipid-based nanoparticles to address the limitations of GBM therapy by overcoming the blood-brain barrier, targeting glioblastoma stem cells, and counteracting the immunosuppressive tumor microenvironment. Biomed Pharmacother. 2024;171:116113.
108. Tong H, Zesen M, Yu J, Li D, Zhu Q, Shi H, et al. Optimizing peptide-conjugated lipid nanoparticles for efficient sirna delivery across the blood-brain barrier and treatment of glioblastoma multiforme. ACS Chem Biol. 2025. https://doi.org/10.1021/acschembio.5c00039.
109. Monfaredan A, Şen S, Hosseininasab A, Taştekin D, Fazli G, Bozbey H, et al. Exosome enveloped by nano lipid particle a new model for signal transducer and activator of transcription 3 silencer ribonucleic acid delivery system to a glioblastoma mice model. Cancers. 2025. https://doi.org/10.3390/cancers17101648.

110. Liang S, Zuo F-F, Yin B, Ye B. Delivery of siRNA based on engineered exosomes for glioblastoma therapy by targeting STAT3. Biomater Sci. 2022. https://doi.org/10.1039/D1BM01723C.
111. Bao P, Gu HY, Ye J, He J, Zhong Z, Yu A, et al. Chimeric exosomes functionalized with STING activation for personalized glioblastoma immunotherapy. Adv Sci. 2023. https://doi.org/10.1002/advs.202306336.
112. Cui J, Wang X, Li J, Zhu A, Du Y, Zeng W, et al. Immune exosomes loading self-assembled nanomicelles traverse the blood-brain barrier for chemo-immunotherapy against glioblastoma. ACS Nano. 2023. https://doi.org/10.1021/acsnano.2c10219.
113. Yuan D, Zhao Y, Banks W, Bullock K, Haney M, Batrakova E, et al. Macrophage exosomes as natural nanocarriers for protein delivery to inflamed brain. Biomaterials. 2017;142:1–12.
114. Khatami S, Karami N, Taheri-Anganeh M, Taghvimi S, Tondro G, Khorsand M, et al. Exosomes: promising delivery tools for overcoming blood-brain barrier and glioblastoma therapy. Mol Neurobiol. 2023. https://doi.org/10.1007/s12035-023-03365-0.
115. Kim J-S, Zhu Y, Chen S, Wang D, Zhang S, Xia J, et al. Anti-glioma effect of ginseng-derived exosomes-like nanoparticles by active blood–brain-barrier penetration and tumor microenvironment modulation. J Nanobiotechnology. 2023. https://doi.org/10.1186/s12951-023-02006-x.
116. Afshar Y, Sharifi N, Kamroo A, Yazdanpanah N, Saleki K, Rezaei N. Implications of glioblastoma-derived exosomes in modifying the immune system: state-of-the-art and challenges. Rev Neurosci. 2024;36:315–25.
117. Benecke L, Coray M, Umbricht S, Chiang D, Figueiró F, Muller L. Exosomes: small EVs with large immunomodulatory effect in glioblastoma. Int J Mol Sci. 2021. https://doi.org/10.3390/ijms22073600.
118. Iurlaro R, Waldhauer I, Planas-Rigol E, Bonfill-Teixidor E, Arias A, Nicolini V, et al. A novel EGFRvIII-T cell bispecific antibody for the treatment of glioblastoma. Mol Cancer Ther. 2022. https://doi.org/10.1158/1535-7163.MCT-22-0201.
119. Park D, Liaw K, Bhojnagarwala P, Zhu X, Choi J, Ali A, et al. Multivalent in vivo delivery of DNA-encoded bispecific T cell engagers effectively controls heterogeneous GBM tumors and mitigates immune escape. Mol Ther Oncolytics. 2023;28:249–63.
120. Fan R, Chen C, Mu M, Chuan D, Liu H, Hou H, et al. Engineering MMP-2 activated nanoparticles carrying B7–H3 bispecific antibodies for ferroptosis-enhanced glioblastoma immunotherapy. ACS Nano. 2023. https://doi.org/10.1021/acsnano.2c12217.
121. Guan P, Jin F, Zhang A, Gao S, Liu Z. Rationally engineered bispecific nanoimmunoblocker restores anticancer immunity via dual immune checkpoint blockade. ACS Nano. 2025. https://doi.org/10.1021/acsnano.4c13463.
122. Wei R, Li J, Lin W, Pang X, Yang H, Lai S, et al. Nanoparticle-mediated blockade of CXCL12/CXCR4 signaling enhances glioblastoma immunotherapy: monitoring early responses with MRI radiomics. Acta Biomater. 2024. https://doi.org/10.1016/j.actbio.2024.02.007.
123. Wang H, Chao Y, Zhao H, Zhou X-X, Zhang F, Zhang Z, et al. Smart nanomedicine to enable crossing blood-brain barrier delivery of checkpoint blockade antibody for immunotherapy of glioma. ACS Nano. 2022. https://doi.org/10.1021/acsnano.1c08120.
124. Tabar MMM, Fathi M, Kazemi F, Bazregari G, Ghasemian A. STING pathway as a cancer immunotherapy: progress and challenges in activating anti-tumor immunity. Mol Biol Rep. 2024;51(1):487.
125. Berger G, Knelson E, Jimenez-Macias J, Nowicki M, Han S, Panagioti E, et al. STING activation promotes robust immune response and NK cell–mediated tumor regression in glioblastoma models. Proceedings of the National Academy of Sciences of the United States of America. 2021;119
126. Wang Q, Yu Y, Zhuang J, Liu R, Sun C. Demystifying the cGAS-STING pathway: precision regulation in the tumor immune microenvironment. Mol Cancer. 2025;24(1):178.
127. Boudreau CE, Najem H, Ott M, Horbinski C, Fang D, DeRay CM, et al. Intratumoral delivery of STING agonist results in clinical responses in canine glioblastoma. Clin Cancer Res. 2021;27(20):5528–35.
128. Deng L, Liang H, Xu M, Yang X, Burnette B, Arina A, et al. STING-dependent cytosolic DNA sensing promotes radiation-induced type I interferon-dependent antitumor immunity in immunogenic tumors. Immunity. 2014;41(5):843–52.
129. Jiang X, Luo T, Yang K, Lee MJ, Liu J, Tillman L, et al. STING activation disrupts tumor vasculature to overcome the EPR limitation and increase drug deposition. Sci Adv. 2024;10(29):eado0082.
130. Najem H, Lea ST, Tripathi S, Hurley L, Chen CH, William I, et al. STING agonist 8803 reprograms the immune microenvironment and increases survival in preclinical models of glioblastoma. J Clin Invest. 2024. https://doi.org/10.1172/JCI175033.
131. Sabahi M, Salehipour A, Bazl MSY, Rezaei N, Mansouri A, Borghei-Razavi H. Local immunotherapy of glioblastoma: a comprehensive review of the concept. J Neuroimmunol. 2023. https://doi.org/10.1016/j.jneuroim.2023.578146.
132. Tripathi S, Najem H, Mahajan A, Zhang P, Low J, Stegh A, et al. cGAS-STING pathway targeted therapies and their applications in the treatment of high-grade glioma. F1000Res. 2022. https://doi.org/10.12688/f1000research.125163.1.
133. Boudreau C, Najem H, Ott M, Horbinski C, Fang D, DeRay C, et al. Intratumoral delivery of STING agonist results in clinical responses in canine glioblastoma. Clin Cancer Res. 2021;27:5528–35.
134. Daei Sorkhabi A, Mohamed Khosroshahi L, Sarkesh A, Mardi A, Aghebati-Maleki A, Aghebati-Maleki L, et al. The current landscape of CAR T-cell therapy for solid tumors: mechanisms, research progress, challenges, and counterstrategies. Front Immunol. 2023;14:1113882.
135. Yu H, Liu W, Ding K, Wu J, Wang C, Wang S, et al. Sequential release hydrolipo system for STING gene epigenetic reprogramming and immune activation in glioblastoma. Adv Sci. 2024. https://doi.org/10.1002/advs.202408323.
136. Chellen T, Bausart M, Maus P, Vanvarenberg K, Limaye N, Préat V, et al. In situ administration of STING-activating hyaluronic acid conjugate primes anti-glioblastoma immune response. Mater Today Bio. 2024. https://doi.org/10.1016/j.mtbio.2024.101057.
137. Joseph J, Blaavand M, Cai H, Vernejoul F, Knopper R, Lindhardt T, et al. STING activation counters glioblastoma by vascular alteration and immune surveillance. Cancer Lett. 2023. https://doi.org/10.1016/j.canlet.2023.216480.
138. Lea S, Chen C-H, Wei J, William I, Del Castillo IL, Curran M. NLRP3 inflammasome activation expands the immunosuppressive myeloid stroma and antagonizes the therapeutic benefit of STING activation in glioblastoma. Cancer Res Commun. 2025;5:960–72.
139. Ott PA, Hu Z, Keskin DB, Shukla SA, Sun J, Bozym DJ, et al. An immunogenic personal neoantigen vaccine for patients with melanoma. Nature. 2017;547(7662):217–21.
140. Zhao Y, Song Y, Li W, Wu J, Zhao Z, Qu T, et al. Clofazimine enhances anti-PD-1 immunotherapy in glioblastoma by inhibiting Wnt6 signaling and modulating the tumor immune microenvironment. Cancer Immunol Immunother. 2025;74(4):137.

141. Chan HY, Choi J, Jackson C, Lim M. Combination immunotherapy strategies for glioblastoma. J Neurooncol. 2021;151(3):375–91.
142. Wang J, Shen F, Yao Y, Wang LL, Zhu Y, Hu J. Adoptive cell therapy: a novel and potential immunotherapy for glioblastoma. Front Oncol. 2020;10:59.
143. Dong L, Zhu Y, Zhang H, Gao L, Zhang Z, Xu X, et al. Open-source throttling of CD8(+) T cells in brain with low-intensity focused ultrasound-guided sequential delivery of CXCL10, IL-2, and aPD-L1 for glioblastoma immunotherapy. Adv Mater. 2024;36(44):e2407235.
144. Weller M, Le Rhun E. Immunotherapy for glioblastoma: Quo vadis? Nat Rev Clin Oncol. 2019;16(7):405–6.
145. Balaji EV, Kumar N, Satarker S, Nampoothiri M. Zinc as a plausible epigenetic modulator of glioblastoma multiforme. Eur J Pharmacol. 2020;887:173549.
146. Goh WC, Murali R, Shamsuddin S, Idris B, Idris Z, Ahmad F. Potential of epigenetic biomarker O6-methylguanine-DNA methyltransferase gene in glioma. Gulhane Med J. 2021;63(4):274–9.
147. Park J, Kim CG, Shim JK, Kim JH, Lee H, Lee JE, et al. Effect of combined anti-PD-1 and temozolomide therapy in glioblastoma. Oncoimmunology. 2019;8(1):e1525243.
148. Karachi A, Dastmalchi F, Yang C, Huang J, Mitchell D, Rahman M. Tmic-26. Immunomodulation with temozolomide to improve efficacy of immune checkpoint inhibition for the treatment of glioblastoma. Neuro Oncol. 2017. https://doi.org/10.1093/neuonc/nox168.1015.
149. Karachi A, Dastmalchi F, Yang C, Huang J, Sayour E, Mitchell D, et al. IMMU-45. Dose modulation of temozolomide have distinct effects on host response to PD-1 blockade. Neuro-oncol. 2018. https://doi.org/10.1093/neuonc/noy148.548.
150. Ehrlich N, Zhang R, Kuo J. Anti-programmed cell death protein-1 immunotherapy for glioblastoma is impaired by systemic chemotherapy but enhanced in combination with locally delivered chemotherapy. Neurosurgery. 2017;81:5.
151. Thomas A, Fisher J, Hampton T, Christensen B, Tsongalis G, Rahme G, et al. Immune modulation associated with vascular endothelial growth factor (VEGF) blockade in patients with glioblastoma. Cancer Immunol Immunother. 2017;66:379–89.
152. De Waele J, Marcq E, Van Audenaerde J, Van Loenhout J, Deben C, Zwaenepoel K, et al. Poly(I:C) primes primary human glioblastoma cells for an immune response invigorated by PD-L1 blockade. Oncoimmunology. 2018. https://doi.org/10.1080/2162402X.2017.1407899.
153. Simonds E, Lu E, Liu E, Tamaki W, Rancan C, Stultz J, et al. Deep immune profiling reveals targetable mechanisms of immune evasion in checkpoint blockade-refractory glioblastoma. bioRxiv. 2020. https://doi.org/10.1101/2020.12.01.404939.
154. Zhou Y, Shi F, Zhu J, Yuan Y. An update on the clinical trial research of immunotherapy for glioblastoma. Front Immunol. 2025. https://doi.org/10.3389/fimmu.2025.1582296/full.
155. Preusser M, van den Bent MJ. Autologous tumor lysate-loaded dendritic cell vaccination (DCVax-L) in glioblastoma: breakthrough or fata morgana? Neuro Oncol. 2023;25(4):631–4.
156. Weller M, Butowski N, Tran DD, Recht LD, Lim M, Hirte H, et al. Rindopepimut with temozolomide for patients with newly diagnosed, EGFRvIII-expressing glioblastoma (ACT IV): a randomised, double-blind, international phase 3 trial. Lancet Oncol. 2017;18(10):1373–85.
157. Ahluwalia MS, Reardon DA, Abad AP, Curry WT, Wong ET, Figel SA, et al. Phase IIa study of SurVaxM plus adjuvant temozolomide for newly diagnosed glioblastoma. J Clin Oncol. 2023;41(7):1453–65.
158. O'Rourke DM, Nasrallah MP, Desai A, Melenhorst JJ, Mansfield K, Morrissette JJD, et al. A single dose of peripherally infused EGFRvIII-directed CAR T cells mediates antigen loss and induces adaptive resistance in patients with recurrent glioblastoma. Sci Transl Med. 2017. https://doi.org/10.1126/scitranslmed.aaa0984.
159. Nassiri F, Patil V, Yefet LS, Singh O, Liu J, Dang RMA, et al. Oncolytic DNX-2401 virotherapy plus pembrolizumab in recurrent glioblastoma: a phase 1/2 trial. Nat Med. 2023;29(6):1370–8.
160. Chiocca EA, Gelb AB, Chen CC, Rao G, Reardon DA, Wen PY, et al. Combined immunotherapy with controlled interleukin-12 gene therapy and immune checkpoint blockade in recurrent glioblastoma: an open-label, multi-institutional phase I trial. Neuro Oncol. 2022;24(6):951–63.
161. Desjardins A, Gromeier M, Herndon JE 2nd, Beaubier N, Bolognesi DP, Friedman AH, et al. Recurrent glioblastoma treated with recombinant poliovirus. N Engl J Med. 2018;379(2):150–61.
162. Zakharia Y, Ts J, Colman H, Vahanian N, Link C, Kennedy E, et al. A phase I/II study of the combination of indoximod and temozolomide for adult patients with temozolomide-refractory primary malignant brain tumors. J Clin Oncol. 2014. https://doi.org/10.1200/jco.2014.32.15_suppl.tps2107.
163. Kamerkar S, Leng C, Burenkova O, Jang SC, McCoy C, Zhang K, et al. Exosome-mediated genetic reprogramming of tumor-associated macrophages by exoASO-STAT6 leads to potent monotherapy antitumor activity. Sci Adv. 2022;8(7):eabj7002.
164. Keane L, Cryan JF, Gleeson JP. Exploiting the gut microbiome for brain tumour treatment. Trends Mol Med. 2025;31(3):213–23.
165. BharathwajChetty B, Kumar A, Deevi P, Abbas M, Alqahtani A, Liang L, et al. Gut microbiota and their influence in brain cancer milieu. J Neuroinflammation. 2025;22(1):129.
166. Jiang H, Yang F, Zhang X, Fang H, Qiu T, Li Y, et al. Dysbiosis of the gut microbiota in glioblastoma patients and potential biomarkers for risk assessment. Microb Pathog. 2024;195:106888.
167. Weathers SP, Li X, Zhu H, Damania AV, Knafl M, McKinley B, et al. Improved overall survival in an anti-PD-L1 treated cohort of newly diagnosed glioblastoma patients is associated with distinct immune, mutation, and gut microbiome features: a single arm prospective phase I/II trial. Nat Commun. 2025;16(1):3950.
168. Zhang H, Hong Y, Wu T, Ben E, Li S, Hu L, et al. Role of gut microbiota in regulating immune checkpoint inhibitor therapy for glioblastoma. Front Immunol. 2024;15:1401967.
169. Aljarrah D, Chalour N, Zorgani A, Nissan T, Pranjol MZI. Exploring the gut microbiota and its potential as a biomarker in gliomas. Biomed Pharmacother. 2024;173:116420.
170. Dehhaghi M, Kazemi Shariat Panahi H, Heng B, Guillemin GJ. The gut microbiota, kynurenine pathway, and immune system interaction in the development of brain cancer. Front Cell Dev Biol. 2020;8:562812.
171. Lyu Y, Yang H, Chen L. Metabolic regulation on the immune environment of glioma through gut microbiota. Semin Cancer Biol. 2022;86(Pt 2):990–7.
172. Wang J, Liu Y, Zhang A, Yu W, Lei Q, Xiao B, et al. Investigational microbiological therapy for glioma. Cancers. 2022. https://doi.org/10.3390/cancers14235977.

173. Leigh J, Skidmore B, Wong A, Maleki Vareki S, Ng TL. Exploring the microbiome's impact on Glioma and brain metastases: insights into development, progression, and treatment response-a scoping review. Cancers. 2025. https://doi.org/10.3390/cancers17071228.
174. Sahoo P, Yang X, Abler D, Maestrini D, Adhikarla V, Brown C, et al. TMOD-18. Exploring the factors lead to success of car t-cell therapy in glioblastoma with computational modeling and in vitro data. Neuro Oncol. 2019. https://doi.org/10.1093/neuonc/noz175.1117.
175. Brown C, Blanchard M, Aftabizadeh M, Hibbard J, Dodia R, Lingaraju C, et al. CTIM-29. Clinical evaluation of chlorotoxin-directed car T cells for patients with recurrent glioblastoma. Neuro Oncol. 2021. https://doi.org/10.1093/neuonc/noab196.221.
176. Badie B, Barish M, Chaudhry A, D'Apuzzo M, Forman S, Portnow J, et al. A phase 1 study to evaluate chimeric antigen receptor (CAR) T cells incorporating a chlorotoxin tumor-targeting domain for patients with MMP2+ Recurrent or progressive glioblastoma (NCT04214392). 2021. J Clin Oncol. https://doi.org/10.1200/JCO.2021.39.15_suppl.TPS2662.
177. Qi T, McGrath K, Ranganathan R, Dotti G, Cao Y. Cellular kinetics: a clinical and computational review of CAR-T cell pharmacology. Adv Drug Deliv Rev. 2022;188:114421.
178. Singh S, Kumar S, Quadir SS, Bhandari S, Baniya B, Joshi G, et al. Artificial intelligence: preface, applications and future perspective in relation to pharmaceutical sector. J Pharm Innov. 2025. https://doi.org/10.1007/s12247-025-09940-3.
179. Stern LA, Gholamin S, Moraga I, Yang X, Saravanakumar S, Cohen JR, et al. Engineered IL13 variants direct specificity of IL13Rα2-targeted CAR T cell therapy. Proc Natl Acad Sci U S A. 2022;119(33):e2112006119.
180. Tian M, Wei JS, Cheuk AT, Milewski D, Zhang Z, Kim YY, et al. CAR t-cells targeting FGFR4 and CD276 simultaneously show potent antitumor effect against childhood rhabdomyosarcoma. Nat Commun. 2024;15(1):6222.
181. Chen T, Wang M, Chen Y, Liu Y. Current challenges and therapeutic advances of CAR-T cell therapy for solid tumors. Cancer Cell Int. 2024;24(1):133.
182. Liu T, Jin D, Le SB, Chen D, Sebastian M, Riva A, et al. Machine learning-directed conversion of glioblastoma cells to dendritic cell-like antigen-presenting cells as cancer immunotherapy. Cancer Immunol Res. 2024;12(10):1340–60.
183. George E, Flagg E, Chang K, Bai HX, Aerts HJ, Vallières M, et al. Radiomics-based machine learning for outcome prediction in a multicenter phase II study of programmed death-ligand 1 inhibition immunotherapy for glioblastoma. AJNR Am J Neuroradiol. 2022;43(5):675–81.
184. Elshafeey N, Kotrotsou A, Hassan A, Elshafei N, Hassan I, Ahmed S, et al. Multicenter study demonstrates radiomic features derived from magnetic resonance perfusion images identify pseudoprogression in glioblastoma. Nat Commun. 2019;10(1):3170.
185. Li Z, Holzgreve A, Unterrainer LM, Ruf VC, Quach S, Bartos LM, et al. Combination of pre-treatment dynamic [18F]FET PET radiomics and conventional clinical parameters for the survival stratification in patients with IDH-wildtype glioblastoma. Eur J Nucl Med Mol Imaging. 2023;50(2):535–45.
186. GBM AGILE: Global Adaptive Trial Master Protocol: An International, Seamless Phase II/III Response Adaptive Randomization Platform Trial Designed To Evaluate Multiple Regimens In Newly Diagnosed and Recurrent GBM. 2019. https://clinicaltrials.gov/study/NCT03970447
187. Bulsara S, Wu M, Wang T. Phase I CAR-T clinical trials review. Anticancer Res. 2022;42(12):5673–84.
188. Zhou Y, Shi F, Zhu J, Yuan Y. An update on the clinical trial research of immunotherapy for glioblastoma. Front Immunol. 2025;16:1582296.
189. Fu M, Xue B, Miao X, Gao Z. Overcoming immunotherapy resistance in glioblastoma: challenges and emerging strategies. Front Pharmacol. 2025;16:1584688.
190. Rodriguez SMB, Tataranu LG, Kamel A, Turliuc S, Rizea RE, Dricu A. Glioblastoma and immune checkpoint inhibitors: a glance at available treatment options and future directions. Int J Mol Sci. 2024. https://doi.org/10.3390/ijms251910765.
191. Wainwright D. IMMU-64. A decade of research targeting immunosuppressive IDO1 In Glioblastoma: nearing the finish line or just beginning the marathon? Neuro Oncol. 2018. https://doi.org/10.1093/neuonc/noy148.567.
192. Atkins MB, Abu-Sbeih H, Ascierto PA, Bishop MR, Chen DS, Dhodapkar M, et al. Maximizing the value of phase III trials in immuno-oncology: a checklist from the Society for Immunotherapy of Cancer (SITC). J Immunother Cancer. 2022. https://doi.org/10.1136/jitc-2022-005413.
193. Oster C, Lazaridis L, Feldheim J, Schmidt T, Kleinschnitz C, Kebir S, et al. P11.14. a systematic review of phase III trials in newly diagnosed glioblastoma 2005–2021. Neuro-oncol. 2022. https://doi.org/10.1093/neuonc/noac174.203.
194. Oster C, Schmidt T, Agkatsev S, Lazaridis L, Kleinschnitz C, Sure U, et al. Are we providing best-available care to newly diagnosed glioblastoma patients? Systematic review of phase III trials in newly diagnosed glioblastoma 2005–2022. Neuro Oncol Adv. 2023. https://doi.org/10.1093/noajnl/vdad105.
195. Obrador E, Moreno-Murciano P, Oriol-Caballo M, López-Blanch R, Pineda B, Gutiérrez-Arroyo JL, et al. Glioblastoma therapy: past, present and future. Int J Mol Sci. 2024. https://doi.org/10.3390/ijms25052529.

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