

Glioma stem cells remodel immunotolerant microenvironment in GBM and are associated with therapeutic advancements

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Abstract. Glioma is the most common primary tumor of the central nervous system (CNS). Glioblastoma (GBM) is incurable with current treatment strategies. Additionally, the treatment of recurrent GBM (rGBM) is often referred to as terminal treatment, necessitating hospice-level care and management. The presence of the blood-brain barrier (BBB) gives GBM a more challenging or “cold” tumor microenvironment (TME) than that of other cancers and glioma stem cells (GSCs) play an important role in the TME remodeling, occurrence, development and recurrence of glioma. In this review, our primary focus will be on discussing the following topics: niche-associated GSCs and macrophages, new theories regarding GSC and TME involving pyroptosis and ferroptosis in GBM, metabolic adaptations of GSCs, the influence of the cold environment in GBM on immunotherapy, potential strategies to transform the cold GBM TME into a hot one, and the advancement of GBM immunotherapy and GBM models.

Keywords: Glioma stem cells, niche, glioma cold environment, immunotherapy, GBM models

1. Introduction

Glioblastoma multiforme (GBM) is the most common intracranial malignant tumor, and its prognosis has not made significant progress, despite the advances in treatments. In the 2021 edition of the WHO classification, gliomas lacking IDH mutations that have concomitant +7/–10 chromosome copy number changes, EGFR gene amplification, or TERT promoter mutations

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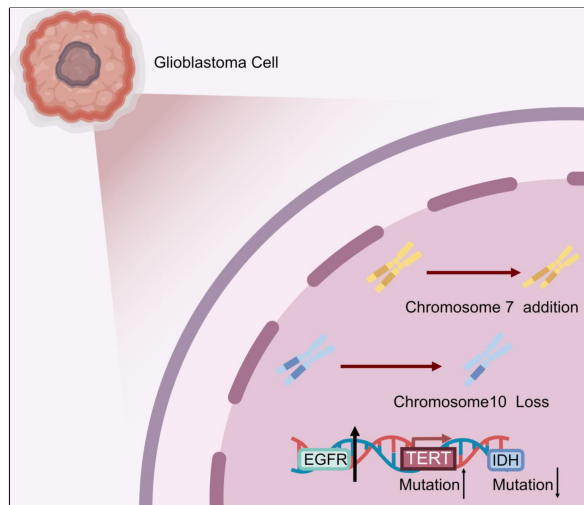


Fig. 1. All tumors lacking IDH mutations with concomitant gain of chromosome 7 and loss of chromosome 10, EGFR amplification, or TERT promoter mutations are referred to as glioblastomas.

are called glioblastoma and are given a WHO grade of 4 [1] (Fig. 1). Glioma stem cells (GSCs) in GBM are a small group of cells with low proliferative activity and drug resistance that are associated with tumor recurrence and are at the root of GBM refractoriness and recurrence. In most instances, these GSCs may be already progenitor cells for differentiation when they remodel the host tissues, and we refer to them as glioma stem/progenitor cells (GSPCs) [2]. The incidence of most cancers, including GBM, rose between 2018 and 2020 [3], outstripping increases in survival rates, and with only few cancers, such as melanoma, showing improvement due to immunotherapy [4,5]. In contrast to the “hot” melanoma tumor microenvironment (TME), the “cold” GBM TME and the presence of the blood-brain barrier (BBB) which limits drug passage [6,7], and, complicate treatment advances. Recent studies show that neuroinflammation creates an immunomodulatory niche in the meningeal lymphatic vessel system close to the cribriform plate in which cerebrospinal fluid drainage kinetics are reduced with aging [8,9,10] and the immune cells contained in the lymphatic fluid are currently the focus of attention. Current research is focused on enhancing pyroptosis and ferroptosis in GBM cells as a strategy to convert the cold GBM tumor microenvironment into a hot one. Then with the help of single-cell sequencing to screen regulatory molecules, study prognosis and develop targeted therapies to improve the efficacy of GBM immunotherapy [11,12,13,14,15]. Although immunotherapy shows some advantages to improve the quality of life and survival prog-

nosis of GBM patients, much work is necessary to optimize immunotherapy for GBM patients. While we have briefly outlined these issues, we will now delve into a more detailed description of the molecular support and regulatory mechanisms involved in the immunotolerant microenvironment remodeled by GSCs in GBM.

2. Glioma stem cells and immune-related niches

2.1. TME and glioma stem cells

A tumor is a complex system comprising both tumor cells and various non-tumor cells, and the TME is a direct representation of this intricate system. The TME consists of cancer cells surrounded by diverse non-malignant cell types, such as cancer-associated fibroblasts, endothelial cells, pericytes, and other cell types that can differ based on the tissue, like adipocytes and neurons. Throughout various stages of tumor development, including initiation, progression, invasion, intravasation, metastatic dissemination, and outgrowth, the TME and its cells play a crucial role. Immune tolerance in the tumor microenvironment leads to immune escape from therapy, which is mainly due to the ability of tumor stem cells to remodel the tumor’s immune microenvironment [16]. Interaction of CSCs with their niche is critical for tumor immunosuppression and tumor recurrence. Moreover, it was demonstrated that a high-stemness signature related to a poor immunogenic response across 21 solid malignancies. Most notably, CSCs are able to recruit tumor-associated immune cells such as monocytes and macrophages, and these immune cells can play a role in promoting tumor progression due to the remodeling of the tumor microenvironment [17]. As a result, conducting systematic research on cancer stem cells and other related cells within the TME will be a vital approach in identifying new targets for treating malignant tumors [18].

In glioma, the TME includes not only tumor cells but also immune cells, endothelial cells, glial cells, and neuronal cells. GSCs can remodel the immune-tolerant microenvironment of gliomas regardless of tissue cell type, and immune-inflammatory cells in the tumor microenvironment are even capable of undergoing malignant transformation through the remodeling of glioma stem cells, which leads to changes in immune tolerance and heterogeneity of tumors by a mechanism that may be related to cell fusion [19]. Furthermore GSCs promote tumor angiogenesis and remodel the microenvironment of GBM by secreting histamine [20]. GBM has the ability to recruit normal cells from its surroundings

to support its growth, maintenance, and invasion into the brain. Studies have demonstrated that the microenvironment in GBM varies depending on factors such as the isocitrate dehydrogenase status (mutated/wild type), the presence or absence of codeletion, and the expression of specific alterations like H3K27 and/or other gene mutations [21]. Recent investigations using Single-cell RNA sequencing (scRNA-seq) in high- and low-grade gliomas have revealed that intratumoral heterogeneity and dynamic plasticity across different cellular states are characteristic features of malignant brain tumors. As the tumor grade increases, there is an observed increase in the proliferation of malignant cells, larger populations of undifferentiated glioma cells, and a shift towards a higher expression of macrophage programs in the tumor microenvironment, compared to microglia expression programs [22].

Human GSCs in adult and child were first reported in 2003 by Singh SK [23], and in 2006 by Quanbin Zhang, respectively [24], and their mysteries have not yet been fully unveiled. The existence of GSCs can be a subject of debate, and the answer to whether they exist or not depends on various factors and perspectives. The stem cell marker CD133 expressing cells which are identified as GSCs in experiments tend to express the progenitor marker Nestin simultaneously [24], thus they are actually progenitor cells that have initiated the differentiation process. Real GSCs are treatment-resistant, quiescent and pluripotent and reside in a niche determined by the adaptive GBM immune microenvironment (Fig. 2A and 1B). The mystery lies in the fact that if the same cells are traced by only CD133 single positive fluorescent staining but not by CD133 and Nestin double staining, they may be GSPCs, rather than GSCs [2,25]. As of today, there are still cells that are discreetly referred to as GSC-like cells, rather than being explicitly labeled as GSCs. This distinction reflects ongoing debates and complexities in the field of glioma research [26]. In fact, as early as 2011, GSCs were defined as those cells capable of driving tumor formation and spreading by differentially labeling human GBM cell components in a xenograft model and following tumor development using a living microscope [27]. GSCs have also been reported as capable of differentiation into offspring cells which may reverse-differentiate into stem cells [24] (Fig. 2D). This is not consistent with the view of Singh SK [28], who cloned GSCs from pediatric GBM and stated that GSCs originated from resident neural stem cells (NSCs) of the host hippocampus or under ependyma and differentiate irreversibly [23]. Subsequent research appeared to provide evidence support-

ing the concept of reverse-differentiation in GSCs [24]. This suggests that GSCs may possess the ability to revert back to a less differentiated state, adding further complexity to our understanding of these cells and their role in glioma. Furthermore, new CD133⁺ cells were detected in the *in vitro* cell cultures of rat glioma C6 after all CD133⁺ had been removed and defined most C6 cells as GSCs [29]. The potential for C6 cells to reverse differentiate into GSCs now seems a more realistic possibility. Under the conditions at the time, this reverse differentiation observation was not comprehensive enough, and the potential stem cell microenvironment, especially the Niche, was proposed later and is still a hot topic today.

2.2. Stem cell niche

Studies conducted on *Drosophila* have contributed to the introduction of the concept of the niche [30], and in many instances, niches have been observed to be located in close proximity to the endothelium of blood vessels [31]. The understanding of its function has improved with the deeper research. Our research of GSCs transdifferentiating into vascular endothelial cells [25,32] was published in 2011, ahead of similar reports by Wang R [33] and Ricci-Vitiani L [34], and exciting commentary by Victoria L Baultch [35]. Nowadays, it is understood that this transdifferentiation process may occur within the hypoxic periarterial niche of GSCs [36]. The GSC niche may also be subdivided into perivascular, peri-hypoxic, immune extracellular matrix and GBM peri-invasive sectors [37,38,39,40, 41], the functions of which remain obscure except as an adaptive GBM immune microenvironment. The niche regulates angiogenesis and protects the GSC from radiotherapy and chemotherapy, driving recurrent GBM (rGBM) [42,43]. Macrophage niches are similar to the adaptive immune microenvironment of GBM.

2.3. Macrophage niche and tumor-associated macrophages

Researchers believe that the macrophage niche (mNiche) can be characterized by four fundamental functions: (1) providing a physical foundation or scaffold for the macrophage; (2) supplying nutritional factors to support the macrophage's self-maintenance ability; (3) imparting the tissue-specific identity to the resident macrophage within the niche; and (4) the macrophages, in turn, should provide benefits to their niche. The mNiche plays an important role in tumor

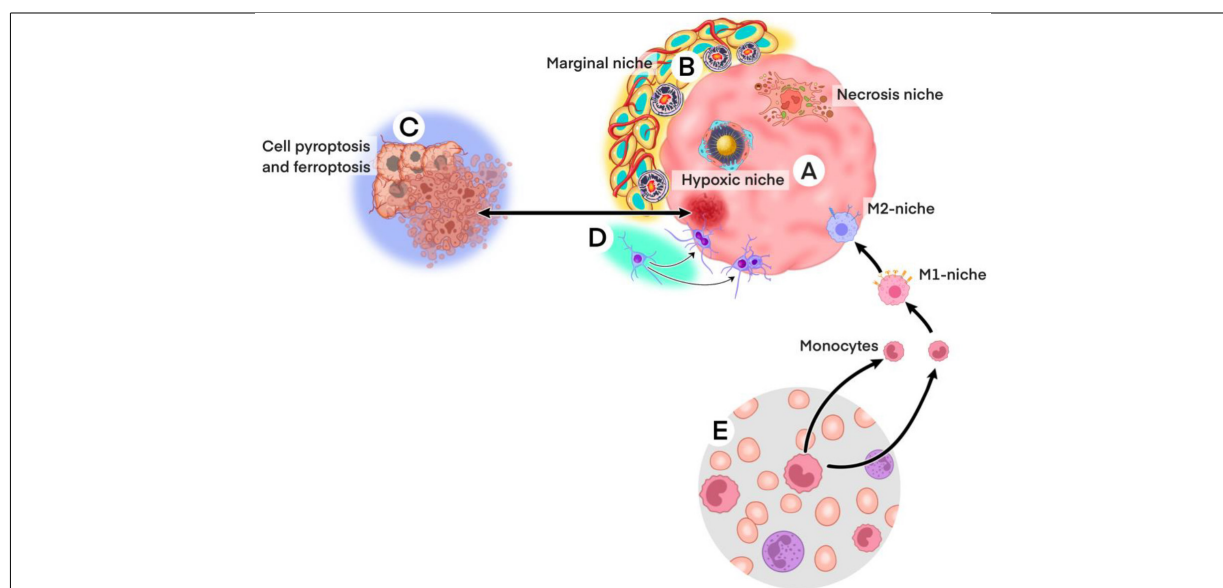


Fig. 2. Schematic diagram of GSCs and immune-related mechanisms: A. Tumor entities, including the hypoxic niche and cell necrosis niches caused by tumor cell pyroptosis and ferroptosis and the macrophage niche, involved in adaptive immunity in the tumor microenvironment. B. The jagged and vague tumor periphery mediates tumor cell invasion and dissemination and marginal ecological niches are colonized here. C. Inflammatory necrotic cells located in the tumor necrosis zone caused by pyroptosis and ferroptosis. D. Hippocampus-subependymal neural stem cell niche: Maintenance and expansion of hippocampal- and subventricular-derived neural stem cells follow both symmetric and asymmetric disaggregation patterns to maintain homeostasis of glial-associated downstream cells in normal brain tissue, which in the case of GBM are largely replaced by the associated tumor stem cell niche. At this point, tumor cells may reverse-differentiate into GSCs.

186 progression. mNiche is found throughout all mam- 212
 187 malian organs. In addition to their role as immunesen- 213
 188 tinels, macrophages perform day-to-day functions es- 214
 189 sential to tissue homeostasis. mNiche maintains tissue 215
 190 homeostasis of macrophage, controls the macrophage 216
 191 population size and imprints their tissue-specific iden- 217
 192 tity [41]. The mNiche has attracted attention for its po- 218
 193 tential therapeutic value. Previously, competition be- 219
 194 tween macrophage precursors was proposed for devel- 220
 195 opment into resident macrophages in a limited number 221
 196 of niches [44]. Tight regulation ensures that monocytes 222
 197 differentiate into multiple heterogeneous macrophages 223
 198 only when niche space is available.

199 Nevertheless, the study of mNiche in tumors is still 224
 200 in its early stages, but significant progress has been 225
 201 made in understanding tumor-associated macrophages 226
 202 (TAMs). TAMs are the most abundant immune cells 227
 203 present in tumor tissues and are typically classified 228
 204 into two distinct subtypes: M1 macrophages and M2 229
 205 macrophages [45].

206 M1 macrophages are known for their anti-tumor 230
 207 functions, whereas M2 macrophages have the opposite 231
 208 effect, promoting tumor development, metastasis, and 232
 209 inhibiting the anti-tumor immune response mediated by 233
 210 T cells. Additionally, M2 macrophages facilitate tumor 234
 211 angiogenesis and contribute to tumor progression. As a

212 result, TAMs have become a promising target for tumor 213
 214 therapy [45].

215 In gliomas, similar to other solid tumors, the infiltra- 216
 217 tion of TAMs is a notable characteristic. In GBM, TAMs 218
 219 are significantly elevated, as confirmed through bioin- 220
 221 formatics studies. Higher levels of TAMs are associated 222
 223 with a decreased overall survival rate in glioma patients, 224
 225 suggesting that increased TAMs may be one of the 226
 227 mechanisms involved in immune escape in GBM. These 228
 229 findings indicate that TAMs-related signatures can serve 230
 231 as valuable prognostic biomarkers in GBM [46].

In addition to the presence of mNiche, the immune 232
 233 microenvironment of GBM is more complicated than 234
 235 in that of extracranial cancers such as the cold immune 236
 237 microenvironment.

3. The cold GBM immune microenvironment 227 228 resists the immune response

3.1. Cold immune microenvironment of GBM 229

230 Cancers may be classified as “hot” when there is a 231
 232 large T cell and inflammatory response after immune

checkpoint inhibitor treatment, “warm” or “cold” when there is little response to treatment [47]. For example, approximately 50% of melanoma patients respond to the combined blockade of the immune checkpoint PD-1 and CTLA-4, 75% of whom have a long-lasting response [48]. Thus, melanoma is a hot tumor type. Conversely, Glioblastoma is a cold tumor, mainly because of immune tolerance in the GBM microenvironment. Compared to other tumor types, glioblastomas have relatively few tumor-infiltrating lymphocytes (TILs), and those that are present have been shown to be highly expressive of exhaustion markers. The glioblastoma microenvironment is characterized by the presence of a large number of myeloid cells, such as microglia and macrophages, which have immunosuppressive activity. In addition, defects in antigen-presenting mechanisms can make the tumor cold in response to T-cell-dependent immunity. Finally, necrosis in glioblastoma plays an important role in weakening the anti-tumor immune response [47]. Only 10% of GBM patients have a short-lived response to immunotherapy [49,50]. The concept of transforming a “Cold” tumor into a “Hot” one is a novel area of research in tumor immunotherapy (IO). However, the impact of intratumoral injection of tilosotolimod, an oligodeoxynucleotide Toll-like receptor 9 (TLR9) agonist, in patients with advanced melanoma has not been conclusively determined [51], suggesting that traditional research approaches still have limitations. Fortunately, quantitative systems pharmacology modeling in cancer immunotherapy holds great promise in addressing major challenges in the IO field [52].

3.2. Exploration for GBM cold environment

In the case of GBM, immunotherapy research has not stopped because of the cold immune microenvironment. Preclinical GBM models suggest Antigen-primed T cells could accumulate in brain tumors through healthy tissue tracking [53], and execute cytotoxic function with cellular precision [54], as well as adapt to a tumor’s evolving molecular profile via epitope spreading. Antitumor CD8 T cells can be controlled by PD-1/PD-L1 interactions [55]. PD-1 blockade augmented the anti-tumor CD8 T cell response, allowing the formation of memory T cells with the ability to prevent delayed tumor outgrowth [56]. In summary, data from preclinical models indicated the potential for GBM immunotherapy [56,57,58,59,60] but clinical trials have proved unsuccessful [61]. The phase III clinical trial of the anti-PD-1 monoclonal antibody, nivolumab, and the anti-growth factor VEGF-A monoclonal antibody, bevaciz-

umab, for rGBM was terminated. However, Jackson, et al. considered that the cold nature of GBM may be converted into hot [62]. Recently, GBM cold tumors were divided into two subtypes with immune tolerance or immunodeficiency from data in the TCGA-GBM transcription database and the GEO dataset [63]. Tumor-associated macrophages were indicated as promising new therapeutic targets and GIPS as a biomarker for assessing the immune evasion mechanism, immunotherapy response and patient prognosis.

3.3. Can microglia/macrophages turn cold GBM hot?

Resident tissue macrophages (RTMs) proposed by Blériot C [64] appear to be much more reasonable than those of macrophages in the tumor tissue microenvironment simply divided into M1 and M2 proposed earlier [50,65]. The heterogeneity of RTMs includes four characteristics: cell origin, local environment, inflammatory state and residence time in tissues that contributes to the resilient adaptation of macrophages to their dynamic environment [64]. Brain RTMs also present these characteristics, in addition to the blood-brain barrier [66,67,68] and the cerebral lymphatic system [69,70,71]. Microglia are a unique tissue-resident macrophage population that plays an important role in maintaining the tissue homeostasis of the CNS [72]. Its characteristics and functions are mediated by Sall1, SMAD2/3, IRF8, Nr4a1 (Nur77), Nr4a2 (Nurr1) and Nr4a3 (Nor1). Nr4a1 (Nur77) can downregulate the transcription of thyroxine-hydroxylase by recruiting the CoREST complex involving HDAC1 and HDAC2 enzymes in the TH promoter region [73,74,75,76]. Mice lacking Nr4a1 had poor prognosis and had high concentrations of norepinephrine (NE), pro-inflammatory IL-6, and autoimmune effector T cells at the site of the affected tissue area in the CNS, which was also necessary for GBM to switch from cold to hot. Thus, we may deduce that if a similar experiment is performed in a GBM mouse model, transcriptomic sequencing of the tumor and myeloid precursor derived macrophages may enable identification of factors responsible for turning cold GBM into a hot tumor. Appropriate sequencing targets would be those concerned with initiation of pyroptosis or ferroptosis, which can trigger an acute inflammatory response. Hence, there is a reason to be optimistic about the search for regulatory molecules that could potentially transform GBM from a cold tumor microenvironment to a hot one.

4. Pyroptosis and ferroptosis

4.1. Pyroptosis, PP

Thornberry NA [77] observed cysteine aspartase [caspase]-1-mediated programmed cell death, of a form morphologically distinct from apoptosis [AP], but of unknown mechanism in 1992. By 2015, PP effect is initially understood after gasdermin D (GSDMD) cleavage target of caspases-1 and -11 was discovered [78,79]. PP was shown to be mediated by a pro-inflammatory caspase effect which caused cell death by cell membrane rupture and cell disintegration and was an anti-infective mode of inflammatory cell death against pathogens [63, 80,81,82,83,84,85,86,87]. Chemical disruption of GSDMD was found to inhibit inflammatory cell death and activate IL-1 secretion by macrophages [88,89]. More recently, methods to regulate its activity have recently been investigated. Succinate and disulfiram have been found to inactivate GSDMD to control PP and Ragulator-Rag complex has been found to be necessary for GSDMD pore formation and pyroptosis in macrophages [90,91,92]. Thus, mediation of PP centers around the inflammatory caspase substrate, GSDMD, which releases GSDMD-N and GSDMD-C domains on lysis, leading to PP by forming membrane pores. The extensive gasdermin family is composed of GSDMA, GSDMB, GSDMC, GSDMD, GSDME/DNFA5 and PVJK/GSDMF of which Gasdermin E shows promise as a potential target for disease therapy [93,94].

4.2. Glioma pyroptosis (GPP)

Recent interest in GPP [95,96,97,98,99] has focused on TCGA and CCGA database bio-informatics-selection of genes and non-coding RNA (ncRNAs) associated with GPP and glioma prognosis [100,101,102]. Indeed, copy number variation and somatic mutation of 33 PP-related genes have been associated with GBM survival prognosis and a prognostic model constructed from 7 PP-related genes for validation in the CGGA cohort [95]. Moreover, CASP8, CASP4, CASP1, NLRP3, NLRP1 and NLRC4 have been identified as hub genes that divide gliomas into two subtypes with good and poor prognoses [96]. Fifteen scorch-death-related genes predicted overall glioma survival and nine pairs of target genes and drugs were identified. Genes encoding caspase 3 and IL-18 have been suggested as a potential prognostic biomarkers for overall survival of patients with diffuse gliomas [97]. Patients in the high-risk subgroup had shorter survival times than those in the low-

risk subgroup. GSEA and ssGSEA showed the activation of immune-related pathways and the extensive infiltration of immune cells in high-risk subgroup. The prognostic value of PP-related gene expression in infiltrating immune cells has been indicated [98] in addition to glioma prognosis models of PP-related genes [99] and PP-related ncRNAs, including miRNA, lncRNA and circRNA, have also been implicated [100]. Most circRNAs are highly conserved and exon-derived with a few arising from intron cyclization. They may be classified as follows: exon circRNA (ecRNA), cyclic intron RNA (ciRNA), exon-intron circRNA (EIciRNA) and tRNA intron cyclic RNA (tri RNA) [103]. Expression of circRNA varies with developmental stage and is tissue-specific. Because circRNA is insensitive to nuclease and more stable than linear RNA, circRNA has obvious advantages in the development and application of new clinical diagnostic markers, such as the autophagy-associated circRNA, circCDYL [104] and other circRNAs have been linked to cancer cell ferroptosis [105], tumorigenesis [106], tumor metabolism [107] and drug resistance [108].

4.3. Ferroptosis and glioma immunity

Ferroptosis, similar to PP described above, is different from AP, but rather a recently highly concerned, new form of cell death that plays an important role in the occurrence and development of many diseases. The comprehensive introduction from the past, present and future of ferroptosis research written in 2020 lacked relevance to glioma [109] However, by 2021, Fe deficiency-related genes was proved to predict prognosis and immunotherapy in glioma., and the prognostic ferroptosis-related lncRNAs in glioma were associated with the immune landscape of glioma microenvironment and radiotherapy response [110,111]. Furthermore, the characterization of a ferroptosis signature has been employed to assess the predictive prognosis and potential effectiveness of immunotherapy in glioblastoma [112]. Additionally, a prognostic risk model has been developed using seven Fe deficiency-related genes for low-grade glioma (LGG), considering their implications for immunotherapy [113]. The utility of ferroptosis for GBM and LGG research is thus demonstrated.

Ferroptosis has also been shown to be responsible for glioma-associated immunogenic cell death [114,115, 116]. The immunogenicity of ferroptosis *in vitro* and *in vivo* was first demonstrated by the induction of ferroptosis by RAS-selective lethal compound 3 (RSL3) in mouse fibrosarcoma MCA205 or glioma GL261

424 cells. Ironophils promoted bone marrow-derived den- 474
425 dritic cell (BMDC) phenotype maturation and elicited a
426 vaccination-like effect in immunocompetent mice sug- 475
427 gesting that the mechanism of immunogenicity is very 476
428 tightly regulated by the adaptive immune system and 477
429 is time dependent [117]. RNA-sequencing was used 478
430 to construct a prognostic risk score model (FRGPRS) 479
431 related to GBM overall survival from Fe deficiency re- 480
432 lated genes. Further comparison of genomic and clinical 481
433 features, immune infiltration, enrichment pathways, 482
434 pan-cancer, drug resistance and immune checkpoint in- 483
435 hibitor therapy in different FRGPRS subgroups showed 484
436 that five Hub genes in the FRGPRS could be used to 485
437 predict overall and progression-free survival of GBM 486
438 patients. High FRGPRS was associated with strong im- 487
439 munity, higher tumor tissue ratio, good cytotoxic immu- 488
440 nity and chemotherapy response in GBM patients [118]. 489
441 The utility of ferroptosis for GBM treatment was also 490
442 reported, and combination of Onofen and cold atmo- 491
443 spheric plasmas could trigger AP, ferroptosis and im- 492
444 munogenic responses in GBM [119,120]. Temozolo- 493
445 mide was found to precipitate ferroptosis through dmt1- 494
446 dependent pathways [121] and the ferroptosis inducer, 495
447 disulfiram, could trigger lysosomal membrane perme- 496
448 ability by upregulating ROS and enhanced the radiosensi- 497
449 tivity of GBM cells [122]. Recently, scholars redis- 498
450 covered from transcriptomic data that CYBB and SOD2 499
451 genes were significantly up-regulated in the mesenchy- 500
452 mal subtype of GBM. In GBM cells that are resistant to 501
453 the chemotherapy drug TMZ, they exhibit mesenchy- 502
454 mal and stemness characteristics while also displaying 503
455 resistance to ferroptosis, a type of cell death caused 504
456 by iron-dependent oxidative stress. This resistance to 505
457 ferroptosis is achieved through the activation of the 506
458 CYBB/Nrf2/SOD2 axis. As a result, CYBB plays a 507
459 crucial role in conferring ferroptosis resilience in mes- 508
460 enchymal GBM. The downstream compensatory activ- 509
461 ity of CYBB, achieved through the Nrf2/SOD2 axis, 510
462 presents an opportunity for exploiting a potential strat- 511
463 egy to overcome TMZ resistance by modulating ferro- 512
464 ptosis. This finding holds promise for the develop- 513
465 ment of new approaches to tackle drug resistance in 514
466 mesenchymal GBM [123]. 515

467 In summary, PP and ferroptosis in GBM are con- 516
468 fined to the cell necrosis region, followed by immune 517
469 adaptation (Fig. 2C). However, the immune cells come 518
470 from the CNS lymphatic system (Fig. 2E), and the brain 519
471 has traditionally been regarded as immune-exempt and 520
472 lacking a lymphatic system, a view that may require 521
473 updating. 522
523

5. Metabolic adaptations of GBM

474 The metabolic abnormalities in glioma involve dis- 475
476 ruptions in sugar, protein, and fat metabolism. Recently, 477
478 more attention has been directed towards studying the 479
480 glycosylation of post-translational modifications of pro- 481
482 teins. The differential expression of glycosyltransferase 483
484 genes determines the type of glycosylation and epige- 485
486 netically regulates the progression of glioma. Hypoxia, 487
488 a well-known factor in gliomas, has been found to in- 489
490 duce GLT8D1, which enhances stem cell maintenance 491
492 in glioma by inhibiting CD133 degradation through N- 493
494 linked glycosylation [124]. As a result of these findings, 495
496 various changes in the biology, biomarkers, and targeted 497
498 therapies for glioma have emerged [125]. Comprehen- 499
500 sive analyses have identified glycosyltransferase sig- 501
502 natures and prognostic long non-coding RNAs (lncR- 503
504 NAs) related to glycosylation from databases such as 504
505 TCGA and CGGA [126]. These analyses can be used to 506
507 evaluate the prognosis of glioma patients and construct 507
508 prognostic models for overall survival [127]. 508
509

494 GSC-specific histamine secretion has been found to 494
495 drive proangiogenic tumor microenvironment remodel- 495
496 ing. Histamine, a metabolite secreted by GSCs, is 496
497 produced due to MYC-mediated transcriptional up- 497
498 regulation of histidine decarboxylase (HDC) through 498
499 GSC-specific H3K4me3 modification. GSC-secreted 499
500 histamine promotes angiogenesis and GBM progression 500
501 by activating endothelial cells through the histamine H1 501
502 receptor (H1R)-Ca²⁺-NFκB axis [128]. Interestingly, 502
503 the role of histamine in the GBM microenvironment is 503
504 opposite to that in the peripheral blood, where histamine 504
505 triggers a positive immune response. The blood-brain 505
506 barrier limits the entry of peripheral blood histamine 506
507 into the GBM microenvironment, making the role of 507
508 histamine-driven pro-angiogenic tumor microenviron- 508
509 ment remodeling particularly noteworthy. Another im- 509
510 portant factor of concern is the MYC oncogene, which 510
511 is often referred to as a “Superoncogene” due to its 511
512 powerful role in regulating GBM metabolism [129]. 512
513 The understanding of MYC has evolved over the years, 513
514 and it is now known to control gene expression at mul- 514
515 tiple levels, including directly binding to chromatin and 515
516 recruiting transcriptional coregulators, regulating RNA 516
517 polymerase activity, and more. GBM is characterized by 517
518 Myc deregulation and undergoes significant metabolic 518
519 changes to meet the increased energy demand. Con- 519
520 versely, cancer metabolism disorders also impact MYC 520
521 expression and function, making MYC a crucial link 521
522 between metabolic pathway activation and gene expres- 522
523 sion. Ongoing and future studies will focus on control-

ling the Myc oncogene and exploring new treatments for GBM by targeting metabolic pathways to deprive tumor cells of nutrients through inhibiting MYC expression [129]. In summary, metabolic adaptations in GBM play a vital role in its malignant progression.

6. The immune system in the normal brain and the lymphatic system in GBM

Lymphatic vessels do not exist in human brain in medical cognition for a long time. However, as early as 2015, discharge of cerebral interstitial fluid and macromolecules by the dural lymphatic system and structure and function of CNS lymphatic vessels were described [130,131]. Meningeal lymphatic vessels at the skull base were proved to involve in the clearance of cerebrospinal fluid (CSF) and neuroinflammation-induced lymphangiogenesis near the cribriform plate was showed to contribute to drainage of CNS-derived antigens and immune cells in 2019 [132,133]. Furthermore, until 2021, meningeal lymphatic vessels were found to regulate lymphatic drainage and immunity in brain tumors [134] and VEGF-c-dependent lymphatic drainage to participate in immune surveillance [135]. Finally, a complete CNS lymphatic system, encompassing arachnoid villi, periarangial pathways and dural lymphatic vessels and communicating with the cerebrospinal fluid has been proposed [136]. The view of immune exemption for the CNS has thus been considerably revised.

The situation is more complex in GBM and lymphatic outflow of cerebrospinal fluid in glioma is decreased [137]. Indeed, GBM cells inoculation proximal to the left ventricle (LV) in a mouse model disrupted the ependymal barrier and increased tumor-CSF interaction, negatively impacting immunotherapy. The author considered the occurrence of therapeutic targets in cerebrospinal fluid only if healthy ependymal membrane cells were present [138].

7. GBM immunotherapy

The failure of phase III GBM immunotherapy clinical trials has been attributed to the targeting of a single anti-tumor component, ignoring the acknowledged heterogeneity of the environment [139]. Further research progress has been widely concerned. Successful advances in immune checkpoint blockade therapy and targeting immunosuppressive proteins, such as pro-

grammed cell death protein-1(PD-1) and/or cytotoxic T lymphocyte-associated antigen-4 (CTLA-4), have been reviewed [140], Initiating a paradigm shift in clinical and preclinical research and applied immunotherapy to solid tumors, which will be a potential breakthrough in the field of GBM drug treatment. However, resistance to GBM therapy has been ascribed to cancer stem cells (CSCs) and the inability of immunotherapy (IT) to completely eliminate CSCs results in failure to universally prolong patient survival [141]. A systematic IT approach to CSC elimination may provide a solution and progress has been made in CAR-T, immune checkpoint inhibitors, vaccination and oncolytic virus therapies for GBM (Fig. 3 and Table 1).

7.1. CAR-T for gliomas

Chimeric antigen receptors (CAR) engineered T cell mediated adoptive immunotherapy (CAR-T) has made great progression in the treatment of hematological malignancies [142]. As far as GBM is concerned, as the peculiarities of the immune microenvironment described above, CAR-T has been of limited benefit for GBM, although preclinical models have furnished hope [143]. More research continues with the aim of improving CAR efficacy in GBM [144,145]. The following three research approaches have been described.

7.1.1. IL13 α 2 specific CAR-T

Interleukin 13 receptor subunit α -2 (IL13R α 2) is present in 60 percent of GBMs and is associated with pro-inflammatory and immune pathway activation [146, 147]. Overexpression of IL13R α 2 in GBM patients results in the activation of phosphatidylinositol-3 kinase/AKT/rapamycin pathway, thereby leading to poor prognosis and increased tumor aggressiveness [148, 149]. Intracranial injection of IL13-zetakine CAR-T into tumor-bearing animals significantly prolonged survival [150] and the brain inflammation, grade 3 headache and transient grade 3 neurological events were controllable by infusion of IL13 α 2-directed CAR-T cells through implanted container/catheter system into the tumor resection stumps. Decreased IL13 R α 2 tumor expression, persistently increased tumor necrosis volume observed during MRI and improved overall survival resulted from treatment [150]. Second-generation IL13-zetakine CAR-T cells for 6-cycle tumor residual infusion and 10-cycle ventricular system infusion (via lumbar puncture) were developed to treat one patient of rGBM. Residual intraluminal perfusion inhibited local tumor progression but extraluminal intracranial tumor

Immunotherapy modalities	Description
CAR-T cell therapy	Including IL13 α 2 specific CAR-T, EGFRvIII CAR-T, HER2 specific CAR-T, B7-H3 specific CAR-T and CAR-NK immunotherapy
Immune checkpoint inhibitor therapy	PD-1/PD-L1 blocking therapy
Tumor vaccination	Including cell vaccines, synthetic peptide vaccine, and nucleic acid vaccine
Oncolytic virus therapy	Using intratumoral delivery of virus to TME for treatment, or causes direct cytotoxicity through viral infection and replication

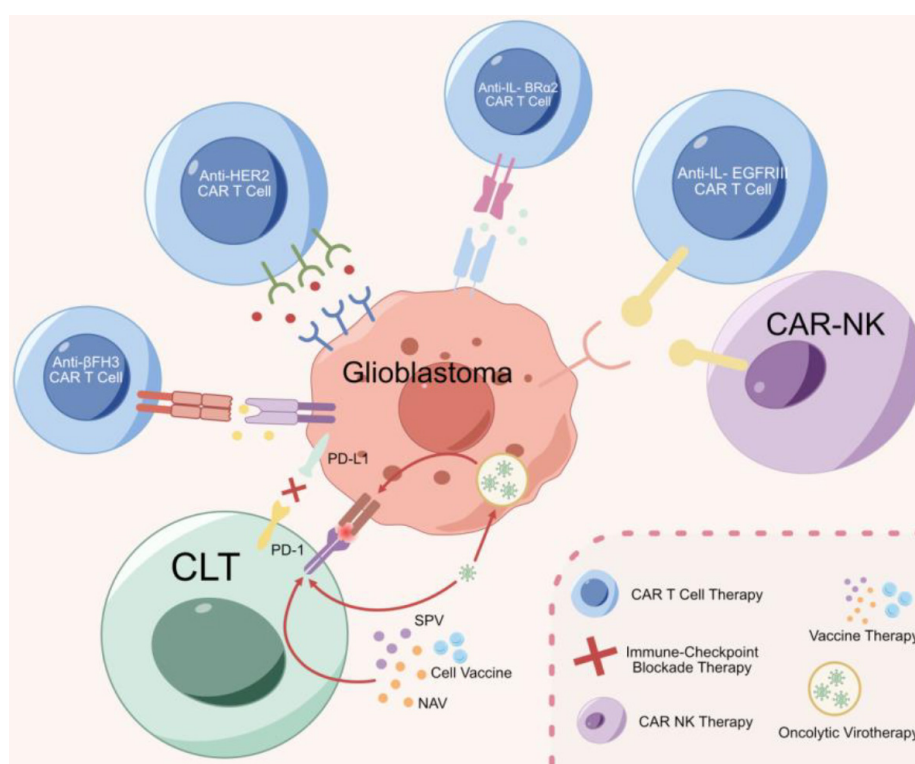


Fig. 3. Current immunotherapy modalities for the treatment of glioblastoma: 1. CAR T-cell therapy such as anti-IL-13R α 2 CART cell therapy, anti-EGFRvIII CART cell therapy, anti-HER2 CART cell therapy, anti-BFH3 CART cell therapy, and the relatively specific CAR-NK cell therapy; 2. Immune checkpoint inhibitor therapy, the most important of which is to inhibit the binding of PD-1 and PD-L1, thus restoring the tumor cell killing effect of CTL; 3. Vaccine therapies, including cellular vaccines, SPV and NAV, which can promote the tumor-killing effect of CTL; 4. Oncolytic virus therapies, are viruses that can selectively infect or replicate in tumor cells, which not only directly kill infected tumor cells, but also promote the tumor-killing effect of CTL.

617 progression and new spinal cord lesions were discovered.
 618 Although, the fifth ventricular infusion reduced
 619 intracranial and spinal cord tumors by 77–100% but
 620 only lasted 7.5 months. Recently, a novel TanCAR,
 621 comprising the tandem arrangement of IL13 (4MS) and
 622 EphA2 scFv, was reported to selectively kill GBM tumor
 623 cells, but did not kill normal cells bearing only the
 624 IL13R α 1/IL4R α receptor. TanCAR T cells have proved
 625 more effective in glioma reduction than single IL13
 626 CAR or EphA2 scFv CARs and prevent antigen escape

reducing off-target cytotoxicity in a xenograft mouse
 model [151].

7.1.2. EGFRvIII CAR-T and CAR-NK immunotherapy

The antitumor effects of EGFRvIII-specific CAR-T
 in *in vitro* and *in vivo* models of U87 cells were reported
 in 2013 [152]. It was later discovered that Infusion
 of CAR-modified T cell (CART)-EGFRvIII cells into
 ten recurrent GBM patients produced off-tumor
 toxicity or cytokine release syndrome and significant

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EGFRvIII -mediated CAR-T cells were found in peripheral blood [153]. Third generation EGFRvIII CAR-T (G3-EGFRvIII) increased IFN- γ levels on co-culture with glioma cells *in vitro* and prolonged survival in tumor-bearing mice [154] but controversies remain over clinical treatments based on EGFRvIII CAR-T due to EGFRvIII do not represent prognostic keys in EGFR-amplified glioma patients [155].

CAR-NK, a development based on CAR-T, is already a fourth-generation engineered cell, which has received as much attention as CAR-T. Fourth generation EGFRvIII specific CAR-NKs have been engineered [156]. Since EGFRvIII specific CAR-NK has been reported, a number of researchers [157,158,159,160,161] have demonstrated their results from different perspectives such as molecular mechanism and efficacy. Especially, MSCs can be home to GBM and not healthy brain cells, hence it serves as a tumour-specific drug-delivery system, including pro-apoptotic factors and tumor necrosis factor-related apoptosis-inducing ligands (TRAIL) [162]. Furthermore, the design of bifunctional MSCs expressing high levels of TRAIL and GD2 tCAR, which is associated with a robust anti-tumor activity against GD2-positive GBM cells, shows promise [163,164].

7.1.3. HER2 or B7-H3 specific CAR-T therapy

HER2 is highly expressed on GBM ependymoma and medulloblastoma, but not in normal CNS tissues [165]. HER2-specific T cells, which target primary glioblastoma stem cells, have demonstrated promising preclinical effects in 10 GBM patients [166]. In clinical treatment of 17 HER2-positive, progressive GBM patients, there were no dose-limiting toxic effects, and CAR-T cells were detected in the peripheral blood for up to 12 months after infusion. However, despite these findings, there was no notable expansion of CAR-T cells or significant survival benefit observed in these patients [167].

B7-H3 (also known as CD276) is a newly found molecule of B7 family. B7-H3 could promote the activation of T cells and the proliferation of IFN- γ . It is highly expressed in all most human cancers, associated with undesirable treatment outcomes and survival time, due to function of the immune checkpoint molecule. B7-H3 is frequently overexpressed in GBM patients, and its expression levels were correlated to the malignancy grade and poor survival in both low-grade glioma (LGG) and GBM patients. Therefore, it may serve as a valuable target for CAR-T therapy [168,169,170,171,172].

CAR-T research on both hematological and solid tumors has increased between 2009–2021 [173]. When it comes to GBM, including targets such as IL13Ra2, EGFRvIII, and HER2, there are challenges that need to be addressed. However, obstacles still exist, such as the high investment costs and a lack of cooperation among research units.

7.2. Immune checkpoint inhibitor therapy

Immunotherapy, involved in various immune checkpoint inhibitor molecules, has improved patients' survival in different types of cancers. This is one of the most hopeful approaches for antitumor therapy. Glioma immune checkpoints including PD-1/PDL-1, Tim-3/Galectin-9, CTLA4, LAG3 and TIGIT/CD96, are targets for immune checkpoint inhibitor therapy [174]. The anti-PD-1 and anti-PD-L1 monoclonal antibodies approved by the US FDA- block distinct inhibitory signals that unleash T cells to aid tumor eradication. T cells, B cells, TAMs, myeloid stem cells (MDSCs) and natural killer cells (NK) all target the PD-1/PD-L1 pathway in GBM to trigger an anti-tumor immune response. Tumor that has been immunosuppressed is removed first and then immunotherapy is used to enhance the functions of the tumor infiltrating lymphocytes (TILs). Unfortunately, the administration of checkpoint inhibitor therapy has shown limited success in GBM clinical trials, primarily due to the challenges of successfully delivering the drugs across the BBB. Some progress has been made since PD-1/PD-L1 blocking therapy was predicted to be the future for cancer immunotherapy in 2019 [175]. PD-L1-mediated GBM immunosuppression has been reported to be related with infiltration and M2 polarization of TAM [176], suggesting targeting both TAMs and mNiche as a promising strategy [44]. Indeed, CD137 and PD-L1 targeted immunoviral therapy has been shown to induce a lasting anti-tumor immune response in a malignant glioma model [177]. Follicular helper T cells have been found to restore CD8⁺-dependent anti-tumor immunity and anti-PD-L1/PD-1 activity [178]. For gliomas, the PD-1/PD-L1 axis and adenosine pathways have been found to be immunosuppressive [179] and TIGIT and PD-1 immune checkpoint pathways to be associated with prognosis and anti-tumor immunity [180]. Despite these promising results, we are still far from resolving the clinical challenges posed by the disease. Indeed, the prognostic value of bioinformatics in relation to immune checkpoint inhibition for GBM has been extensively studied [181,182,183]. Additionally, the inhibitory impact

of engineered extracellular vesicle irradiation on GBM immune checkpoints has been reported [184], and all of these findings hold promise for potential clinical applications.

7.3. Vaccination: Cell, peptide and mRNA vaccines for glioma

Cell vaccines: In addition to CAR-T and CAR-NK regarded as T and NK cell vaccines [185], Dendritic cell (DC) fusion vaccine is the most important cell vaccine. Bone marrow-derived DC fusion vaccines have been given to tumor-bearing mice, alone or in combination with telimazolid, to prolong survival time [186,187]. Glioma stem cell-targeted dendritic cells as a tumor vaccine against malignant glioma and DC glioma cell fusion as an antitumor vaccine in vitro culture have also been studied respectively [188,189]. In a large phase III clinical trial of DC vaccine for GBM, 331 patients with GBM after standardized treatment were included, patients were randomized to receive temozolomide plus DC vaccine ($n = 232$) or temozolomide and placebo ($n = 99$). The results showed that the addition of DC vaccine to standard therapy is both feasible and safe for patients, and it has the potential to extend survival. Only 2.1% of patients experienced a grade 3 or 4 adverse event [190]. Indeed, an almost complete response of GBM patients to treatment with an allogeneic dendritic cell-based vaccine was an encouraging outcome of a 2022 trial [191].

Synthetic peptide vaccine (SPV): TollR-3/poly-ICLC and TGF- β improved the therapeutic efficacy of glioma-associated antigen peptide vaccines on tumor-bearing mice [192,193] and patients with WHO grade II gliomas produced a strong CD8⁺ T cell response after receiving peptide vaccine combined with polyurethrasome (iclc) [194]. Following these encouraging outcomes, VEGF receptor 1 and 2 peptide vaccine was investigated [195], peptide vaccines (ICT-107), autologous dendritic cells (DC) pulsed with six synthetic peptide epitopes targeting GBM tumor/stem cell-associated antigens MAGE-1, HER-2, AIM-2, TRP-2, gp100, and IL13R α 2, was proposed [196], multiple glioma tumor antigens/glioma angiogenesis-related antigen peptide vaccine was evaluated [197], neoantigen vaccine using multi-epitope, personalized neoantigen vaccination strategies was created [198], and mass cytometry for detecting H3.3K27M-specific vaccine mutant IDH1 vaccine were developed [199,200]. These vaccines have been tested in newly diagnosed and relapsed GBM diffuse midline glioma respectively, and the results show

that they are well tolerated and have good curative effect. However, they all belong to single-center phase I/II clinical trials and need to be further studied.

Nucleic acid vaccine (NAV): Both DNAV and mRNAV are safe and more easily manufactured than SPVs and aim to transmit genetic information encoding tumor antigens (Tas) to the host to generate an anticancer immune response [201,202]. Although NAV is safe and easy to manufacture compared to SPVs, they have so far not been considered a viable alternative to SPVs. Judging from the situation that has been carried out, DNAV for cervical cancer, prostate cancer and breast cancer and mRNAV for melanoma, GBM and prostate cancer have been investigated. A DNA vaccine with a glioma antigen, SOX6 and a vaccine targeting IL13R α 2 have been shown to induce therapeutic anti-tumor immunity in 2008 [203,204]. Thirteen years later, an immune response of a new DNA-based immunotherapy and increased survival times in different tumor models have also been reported [205]. Between 2021 and 2022, 6 studies used information in the TCGA and/or CGGA databases to screen for suitable tumor-associated or tumor-specific antigen candidates for mRNAV in gliomas but no mRNAVs were synthesized [206,207,208,209,210,211]. Therefore, the use of mRNAV as a specific prophylactic vaccine for clinical trials still appears to be distant or not yet feasible at present.

7.4. Oncolytic virus therapy

Oncolytic viruses (OVs) can replicate in cancer cells but not in normal cells, leading to death of the tumor cells. Oncolytic viruses therapy (OVT) uses intratumoral delivery of virus to TME for treatment, or causes direct cytotoxicity through viral infection and replication [212,213]. The treatment induces immunogenic cell death (ICD) in infected tumor cells when destruction of tumor cells by OVT releases antigens into the TME, recruiting and activating local dendritic cells and specific T cells [213]. The research on oncolytic virus has never ceased. Earlier regimens involving the HSV1-tk gene with the antiviral drug acyclovir [212,214] suffered from poor vector delivery and poor efficacy. However, HSV1G207, developed later, has been shown to be safe and effective in clinical trials. The advantage is that it allows conditional replication in tumor cells while preventing infection of normal cells [215], phase I clinical trials have been conducted, whether alone or in combination with radiotherapy GBM is effective and safe [216,217,218]. Furthermore, the new drug,

833 HSV-rQnestin34.5v.2, is currently undergoing clinical
834 trials, and it has demonstrated low toxicity to human
835 beings [219,220].

836 8. Summary and outlook

837 8.1. Plasticity of the GSC niche

838 The aforementioned GSCs Niche are almost ubiq-
839 uitous in and around GBM entities, and their func-
840 tion has not been fully demonstrated. The perivascu-
841 lar niche (PVN) is considered to be a complex mi-
842 croenvironment containing endothelial cells plus astro-
843 cytes, pericytes, immune cells and other stromal cells
844 that regulate GSC biology [221,222,223]. It is not clear
845 how the various cellular components of PVN change
846 GSC behavior, such as proliferation, quiescence, in-
847 vasive dissemination, homing and chemoradiation re-
848 sistance. Previous 2D and 3D *in vitro* cultures and
849 tumor-bearing mouse models have inevitable limita-
850 tions, and bionic models have received great attention
851 and shown a bright future [224,225,226,227,228,229,
852 230]. However, it seems that there are still many diffi-
853 culties whether the wish of using bionic model to com-
854 pletely replace clinical cases can be achieved. Single-
855 cell sequencing has been used to detect the interactions
856 between GSCs and immune cells during tumorigenes-
857 is [13], analyze the inhibition of CD161 receptor by
858 GBM infiltrating T cells [12], reveal functional hetero-
859 geneity of glioma-associated brain macrophages [11],
860 and reveal the role of m6A-modified RNA in the
861 glioblastoma microenvironment [231]. Single cell se-
862 quencing can detect the molecules of all single cell
863 components from clinical specimens. In biomimetic
864 models, the cells are often artificially introduced or
865 stocked to mimic the natural environment, ranging from
866 biomimicry to simulation, and even high simulation,
867 eventually forming a realistic landscape resembling
868 clinical GBM. However, such models come with po-
869 tential risks that are difficult to achieve or replicate in
870 reality.

871 The dynamic nature of CSCs implies plasticity of
872 GSCs [232], reinforcing the message of our recently
873 published review “GSCs and Their Microenvironments:
874 Docking and Transformation” [233]. In short, GSCs
875 change according to the microenvironment and thera-
876 peutic signals.

877 8.2. A cure for GBM

878 Standard care for GBM only prolongs the patient’s
879 very short lifespan and the prognosis is particularly

880 severe for unresectable GBM [234,235,236,237,238].
881 Immunotherapy promises to be less than ideal [239,
882 240,241]. Future treatment direction pays more atten-
883 tion to combination strategies. For example, the bis-
884 specific antibodies targeting two different antigens has
885 proven to be a valuable approach, [242,243] but the
886 BBB excludes most macromolecular monoclonal anti-
887 bodies [244,245]. Fortunately, novel cyclic peptides that
888 modulate BBB functions have been reported to enhance
889 monoclonal antibody delivery to the brain [244] and
890 focused ultrasound-mediated BBB disruption has been
891 showed to improve the delivery of anti-CD47 mono-
892 clonal antibodies [246]. Alternatively, intratumoral ad-
893 ministration is very valuable for improving drug dis-
894 tribution and sustained release. For example, PLGA
895 nanoparticles which have been found to enhance the
896 penetration of paclitaxel in brain tissue, including some
897 other implants, can improve the therapeutic effect [247,
898 248,249,250,251]. In addition, nanoformulation has
899 been used to transform “cold” GBM tumors into “hot”
900 and promote immune cell infiltration [252,253]. In-
901 tranasal administration has also been proposed as a po-
902 tential delivery method [254,255]. However, most of the
903 mentioned approaches are still in the preclinical stage,
904 and more research is needed to explore their potential
905 effectiveness and safety for further investigation.

906 Botanical medicines, such as leaf extract of *Termin-
907 alia catappa* L. inhibited tumor cell migration and in-
908 vasion in a human GBM PDX [256,257], *artemisia an-
909 nua* had an *in vitro* anti-cancer effect and resveratrol
910 inhibited the proliferation of dendritic cells induced by
911 human GBM GSCs [258].

912 In short, there is hope to improve GBM, especially
913 the survival prognosis of rGBM, which is currently in
914 the stage of *in vitro* or *in vivo* experiments in animals,
915 and there is still a painstaking research process on when
916 incurable GBM can be turned into a treatable one.

917 8.3. A new model of GBM immunotherapy

918 GBM heterogeneity of cell composition, gene expres-
919 sion and phenotype means that some experimental mod-
920 els involved in the above preclinical studies are over-
921 simplified, such as spheroids which represent a random
922 aggregations of cells without a tissue-like structure, ex-
923 tracellular matrix or neighboring non-tumor cells. Het-
924 erogeneous tumor spheres that better meet the require-
925 ments of clinical research are being studied, including
926 heterospheres from co-culture of cancer and stromal
927 cells, producing spheroids containing NK cells [259]
928 or grown in the presence of osteoclasts and probiotics,

929 increased cytotoxicity to CSCs [260]. Moreover, an
930 immunocompetent cancer stem cell model that reca-
931 pitulates tumor heterogeneity, invasiveness, vascular-
932 ity, and immunosuppressive microenvironment in syn-
933 geneic immunocompetent mice was developed and used
934 for tested a genetically engineered oncolytic herpes
935 simplex virus that is armed with interleukin 12 (G47-
936 mIL12). The results showed G47 Δ -mIL12 could pro-
937 vide a multifaceted approach to targeting GSCs, tumor
938 microenvironment, and the immune system [261].

939 Organotype tissue sectioning models involve cul-
940 ture of surgically removed tumor tissue, maintaining
941 inter- and intra-tumor heterogeneity and tumor struc-
942 ture [262,263,264]. This technique does not involve se-
943 lective growth of tumor cells may be used for person-
944 alized treatments and to evaluate individual sensitivity
945 to invasive and patient-specific effects of anti-invasive
946 drugs [263]. An *in vitro* brain slice model for targeting
947 of brain metastases of breast cancer has also been con-
948 structed [265]. Such a model is expected to contribute
949 to immunotherapy studies of solid tumors, including
950 GBM.

951 Currently, one of the most cutting-edge areas of re-
952 search is focused on organoid models. Organoid mod-
953 els have the ability to replicate the structure and func-
954 tion of original organs, and in the long-term, they hold
955 the potential to replace patient-based studies [266,267].
956 They have potential for basic cancer research, drug
957 screening and personalized susceptibility studies and
958 may bridge the gap between *in vitro* and *in vivo* cancer
959 models [266,268]. The GBM organoid model, gener-
960 ated by traditional 3D culture, genetic engineering and
961 co-culture, shows promise, preserving the phenotype
962 and 3D TME of the original tumor [269,270,271,272,
963 273,274,275,276,277,278]. These methods can also be
964 used to produce other organoid models of brain tumor
965 such as medulloblastoma and brain metastasis. It has
966 been widely used in basic research and clinical trans-
967 formation research, especially in immunotherapy re-
968 search, which has considerable potential. Combining
969 innovative technologies, such as 3D bioprinting and 4D
970 real-time imaging, are likely to produce realistic mod-
971 eling of brain tumor organoids although structural and
972 genetic fidelity aspects remain unclear [279].

973 In summary, the path towards transforming incurable
974 GBM into a curable condition has come closer, but there
975 is still a considerable distance to cover. Nevertheless,
976 there is hope as a recent seminar, co-organized by the
977 National Brain Tumor Society and the Parker Institute
978 of Cancer Immunotherapy, has brought together experts
979 who have highlighted potential future directions for
980 GBM therapy [280,281,282].

9. Conclusions

981 Glioma microenvironment, which is remodeled by
982 GSCs, is different from other cancers. In addition to
983 the unique characteristics mentioned above, the hetero-
984 geneity of GSCs and TME is the key to be clarified in
985 the future. For example, Driving factors of GSC plas-
986 ticity and heterogeneity (such as reprogramming tran-
987 scription factors and epigenetic modifications) has been
988 proved to be related to the induction of immunosuppres-
989 sive cell states, which may lead to therapeutic oppor-
990 tunities for GSC-intrinsic mechanisms [283]. Another
991 example is the interaction between tumor-associated
992 microglia/macrophages and GSCs in TME [284]. We
993 have only verified that SU3 (GSCs) can trigger the
994 malignant transformation of macrophages into cancer
995 cells [285]. However, if we can elucidate the molecular
996 mechanisms underlying this transformation, we may be
997 able to manipulate the related molecules and revert the
998 transformed macrophages back to the M1 state, which
999 could potentially inhibit GSCs.
1000

Data availability

1001 No underlying data was collected or produced in this
1002 study.
1003

Author contributions

1004 Conception: HQ, WAM, ZWY, WY.
1005 Interpretation or analysis of data: FXF, WJ, THY,
1006 YK, ZYD, JDY, CHC, CH, XXT.
1007 Preparation of the manuscript: FXF, WJ, THY, HQ.
1008 Revision for important intellectual content: HQ,
1009 WAM, ZWY, WY.
1010 Supervision: HQ, WAM, ZWY, JDY, CHC.
1011 All authors agree to be accountable for the content
1012 of the work.
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Conflict of interest

The authors declare that they have no competing interests.

References

- [1] D.N. Louis, A. Perry, P. Wesseling, D.J. Brat, I.A. Cree, D. Figarella-Branger, C. Hawkins, H.K. Ng, S.M. Pfister, G. Reifenberger, R. Soffietti, A. von Deimling and D.W. Ellison, The 2021 WHO classification of tumors of the central nervous system: a summary, *Neuro Oncol* **23** (2021), 1231–1251.
- [2] Y. Zhao, J. Dong, Q. Huang, M. Lou, A. Wang and Q. Lan, Endothelial cell transdifferentiation of human glioma stem progenitor cells in vitro, *Brain Res Bull* **82** (2010), 308–12.
- [3] H. Sung, J. Ferlay, R.L. Siegel, M. Laversanne, I. Soerjomataram, A. Jemal and F. Bray, Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries, *CA Cancer J Clin* **71** (2021), 209–249.
- [4] A.M. Menzies, R.A. Scolyer and G.V. Long, Neoadjuvant Immunotherapy in Melanoma – The New Frontier, *Clin Cancer Res* **27** (2021), 4133–4135.
- [5] P.A. Ascierto, A.B. Warner, C. Blank, C. Caracò, S. Demaria, J.E. Gershenwald, N.I. Khushalani, G.V. Long, J.J. Luke, J.M. Mehnert, C. Robert, P. Rutkowski, H.A. Tawbi, I. Osman and I. Puzanov, The “Great Debate” at Melanoma Bridge 2021, December 2nd–4th, 2021, *J Transl Med* **20** (2022), 200.
- [6] W. Wu, J.L. Klockow, M. Zhang, F. Lafortune, E. Chang, L. Jin, Y. Wu and H.E. Daldrup-Link, Glioblastoma multiforme (GBM): An overview of current therapies and mechanisms of resistance, *Pharmacol Res* **171** (2021), 105780.
- [7] R. Abounader and D. Schiff, The blood-brain barrier limits the therapeutic efficacy of antibody-drug conjugates in glioblastoma, *Neuro Oncol* **23** (2021), 1993–1994.
- [8] M. Hsu, C. Laaker, A. Madrid, M. Herbath, Y.H. Choi, M. Sandor and Z. Fabry, Neuroinflammation creates an immune regulatory niche at the meningeal lymphatic vasculature near the cribriform plate, *Nat Immunol* **23** (2022), 581–593.
- [9] M. Brady, A. Rahman, A. Combs, C. Venkatraman, R.T. Kasper, C. McQuaid, W.E. Kwok, R.W. Wood and R. Deane, Cerebrospinal fluid drainage kinetics across the cribriform plate are reduced with aging, *Fluids Barriers CNS* **17** (2020), 71.
- [10] N.H. Mehta, J. Sherbansky, A.R. Kamer, R.O. Carare, T. Butler, H. Rusinek, G.C. Chiang, Y. Li, S. Strauss, L.A. Saint-Louis, N.D. Theise, R.A. Suss, K. Blennow, M. Kaplitt and M.J. de Leon, The brain-nose interface: A potential cerebrospinal fluid clearance site in humans, *Front Physiol* **12** (2021), 769948.
- [11] N. Ochocka, P. Segit, K.A. Walentynowicz, K. Wojnicki, S. Cyranowski, J. Swatler, J. Mieczkowski and B. Kaminska, Single-cell RNA sequencing reveals functional heterogeneity of glioma-associated brain macrophages, *Nat Commun* **12** (2021), 1151.
- [12] N.D. Mathewson, O. Ashenberg, I. Tirosh, S. Gritsch, E.M. Perez, S. Marx, L. Jerby-Aron, R. Chanoch-Myers, T. Hara, A.R. Richman, Y. Ito, J. Pyrdol, M. Friedrich, K. Schumann, M.J. Poitras, P.C. Gokhale, L.N. Gonzalez Castro, M.E. Shore, C.M. Hebert, B. Shaw, H.L. Cahill, M. Drummond, W. Zhang, O. Olawoyin, H. Wakimoto, O. Rozenblatt-Rosen, P.K. Brastianos, X.S. Liu, P.S. Jones, D.P. Cahill, M.P. Frosch, D.N. Louis, G.J. Freeman, K.L. Ligon, A. Marson, E.A. Chiocca, D.A. Reardon, A. Regev, M.L. Suvà and K.W. Wucherpfennig, Inhibitory CD161 receptor identified in glioma-infiltrating T cells by single-cell analysis, *Cell* **184** (2021), 1281–1298e26.
- [13] Y. Zhai, G. Li, R. Li, Y. Chang, Y. Feng, D. Wang, F. Wu and W. Zhang, Single-cell RNA-sequencing shift in the interaction pattern between glioma stem cells and immune cells during tumorigenesis, *Front Immunol* **11** (2020), 581209.
- [14] Z. Yang, Z. Chen, Y. Wang, Z. Wang, D. Zhang, X. Yue, Y. Zheng, L. Li, E. Bian and B. Zhao, A novel defined pyroptosis-related gene signature for predicting prognosis and treatment of glioma, *Front Oncol* **12** (2022), 717926.
- [15] K. Yang, Z. Wu, H. Zhang, N. Zhang, W. Wu, Z. Wang, Z. Dai, X. Zhang, L. Zhang, Y. Peng, W. Ye, W. Zeng, Z. Liu and Q. Cheng, Glioma targeted therapy: insight into future of molecular approaches, *Mol Cancer* **21** (2022), 39.
- [16] L. Li and R.A. Jensen, Understanding and overcoming immunosuppression shaped by cancer stem cells, *Cancer Res* **83** (2023), 2096–2104.
- [17] A. Miranda, P.T. Hamilton, A.W. Zhang, S. Pattnaik, E. Becht, A. Mezheyeuski, J. Bruun, P. Micke, A. de Reynies and B.H. Nelson, Cancer stemness, intratumoral heterogeneity, and immune response across cancers, *Proc Natl Acad Sci USA* **116** (2019), 9020–9029.
- [18] K.E. de Visser and J.A. Joyce, The evolving tumor microenvironment: From cancer initiation to metastatic outgrowth, *Cancer Cell* **41** (2023), 374–403.
- [19] T. Xie, J.W. Ma, B. Liu, J. Dong and Q. Huang, [Experimental study of glioma stem cell-mediated immune tolerance in tumor microenvironment], *Zhonghua Zhong Liu Za Zhi* **39** (2017), 808–813.
- [20] J. Chen, G. Liu, X. Wang, H. Hong, T. Li, L. Li, H. Wang, J. Xie, B. Li, T. Li, D. Lu, Y. Zhang, H. Zhao, C. Yao, K. Wen, T. Li, J. Chen, S. Wu, K. He, W.N. Zhang, J. Zhao, N. Wang, Q. Han, Q. Xia, J. Qi, J. Chen, T. Zhou, J. Man, X.M. Zhang, A.L. Li and X. Pan, Glioblastoma stem cell-specific histamine secretion drives pro-angiogenic tumor microenvironment remodeling, *Cell Stem Cell* **29** (2022), 1531–1546e7.
- [21] V. Di Nunno, E. Franceschi, A. Tosoni, L. Gatto, S. Bartolini and A.A. Brandes, Tumor-associated microenvironment of adult gliomas: A review, *Front Oncol* **12** (2022), 891543.
- [22] A.S. Venteicher, I. Tirosh, C. Hebert, K. Yizhak, C. Neftel, M.G. Filbin, V. Hovestadt, L.E. Escalante, M.L. Shaw, C. Rodman, S.M. Gillespie, D. Dionne, C.C. Luo, H. Ravichandran, R. Mylvaganam, C. Mount, M.L. Onozato, B.V. Nahed, H. Wakimoto, W.T. Curry, A.J. Iafrate, M.N. Rivera, M.P. Frosch, T.R. Golub, P.K. Brastianos, G. Getz, A.P. Patel, M. Monje, D.P. Cahill, O. Rozenblatt-Rosen, D.N. Louis, B.E. Bernstein, A. Regev and M.L. Suvà, Decoupling genetics, lineages, and microenvironment in IDH-mutant gliomas by single-cell RNA-seq, *Science* **355** (2017).
- [23] S.K. Singh, I.D. Clarke, M. Terasaki, V.E. Bonn, C. Hawkins, J. Squire and P.B. Dirks, Identification of a cancer stem cell in human brain tumors, *Cancer Res* **63** (2003), 5821–8.
- [24] Q.B. Zhang, X.Y. Ji, Q. Huang, J. Dong, Y.D. Zhu and Q.

- 1144 Lan, Differentiation profile of brain tumor stem cells: a comparative study with neural stem cells, *Cell Res* **16** (2006), 909–15.
- 1145
- 1146
- 1147 [25] J. Dong, Y. Zhao, Q. Huang, X. Fei, Y. Diao, Y. Shen, H. Xiao, T. Zhang, Q. Lan and X. Gu, Glioma stem/progenitor cells contribute to neovascularization via transdifferentiation, *Stem Cell Rev Rep* **7** (2011), 141–52.
- 1148
- 1149
- 1150
- 1151 [26] J. Feldheim, A.F. Kessler, J.J. Feldheim, E. Schulz, D. Wend, L. Lazaridis, C. Kleinschnitz, M. Glas, R.I. Ernestus, S. Brandner, C.M. Monoranu, M. Löhr and C. Hagemann, Effects of long-term temozolomide treatment on glioblastoma and astrocytoma WHO grade 4 stem-like cells, *Int J Mol Sci* **23** (2022).
- 1152
- 1153
- 1154
- 1155
- 1156
- 1157 [27] J.D. Lathia, J. Gallagher, J.T. Myers, M. Li, A. Vasanthi, R.E. McLendon, A.B. Hjelmeland, A.Y. Huang and J.N. Rich, Direct in vivo evidence for tumor propagation by glioblastoma cancer stem cells, *PLoS One* **6** (2011), e24807.
- 1158
- 1159
- 1160
- 1161 [28] S.K. Singh, I.D. Clarke, T. Hide and P.B. Dirks, Cancer stem cells in nervous system tumors, *Oncogene* **23** (2004), 7267–73.
- 1162
- 1163
- 1164 [29] X. Zheng, G. Shen, X. Yang and W. Liu, Most C6 cells are cancer stem cells: evidence from clonal and population analyses, *Cancer Res* **67** (2007), 3691–7.
- 1165
- 1166
- 1167 [30] H. Lin, The stem-cell niche theory: lessons from flies, *Nat Rev Genet* **3** (2002), 931–40.
- 1168
- 1169 [31] L. Li and T. Xie, Stem cell niche: structure and function, *Annu Rev Cell Dev Biol* **21** (2005), 605–31.
- 1170
- 1171 [32] R. Wang, K. Chadalavada, J. Wilshire, U. Kowalik, K.E. Hovinga, A. Geber, B. Fligelman, M. Leversha, C. Brennan and V. Tabar, Glioblastoma stem-like cells give rise to tumour endothelium, *Nature* **468** (2010), 829–33.
- 1172
- 1173
- 1174
- 1175 [33] L. Ricci-Vitiani, R. Pallini, M. Biffoni, M. Todaro, G. Invernici, T. Cenci, G. Maira, E.A. Parati, G. Stassi, L.M. Larocca and R. De Maria, Tumour vascularization via endothelial differentiation of glioblastoma stem-like cells, *Nature* **468** (2010), 824–8.
- 1176
- 1177
- 1178
- 1179
- 1180 [34] X.F. Fei, Q.B. Zhang, J. Dong, A.D. Wang, Z.M. Wang and Q. Huang, [Preliminary studies on cell derivation of neovascularization in human glioma and its functional evaluation], *Zhonghua Zhong Liu Za Zhi* **33** (2011), 726–31.
- 1181
- 1182
- 1183
- 1184 [35] V.L. Bautch, Cancer: Tumour stem cells switch sides, *Nature* **468** (2010), 770–1.
- 1185
- 1186 [36] D.A. Aderetti, V.V.V. Hira, R.J. Molenaar and C.J.F. van Noorden, The hypoxic peri-arteriolar glioma stem cell niche, an integrated concept of five types of niches in human glioblastoma, *Biochim Biophys Acta Rev Cancer* **1869** (2018), 346–354.
- 1187
- 1188
- 1189
- 1190
- 1191 [37] L.J. Brooks, M.P. Clements, J.J. Burden, D. Kocher, L. Richards, S.C. Devesa, L. Zakka, M. Woodberry, M. Ellis, Z. Jaunmuktane, S. Brandner, G. Morrison, S.M. Pollard, P.B. Dirks, S. Marguerat and S. Parrinello, The white matter is a pro-differentiative niche for glioblastoma, *Nat Commun* **12** (2021), 2184.
- 1192
- 1193
- 1194
- 1195
- 1196
- 1197 [38] D. Schiffer, L. Annovazzi, C. Casalone, C. Corona and M. Mellai, Glioblastoma: Microenvironment and Niche Concept, *Cancers (Basel)* **11** (2018).
- 1198
- 1199
- 1200 [39] Q. Wang, Z. He, M. Huang, T. Liu, Y. Wang, H. Xu, H. Duan, P. Ma, L. Zhang, S.S. Zamvil, J. Hidalgo, Z. Zhang, D.M. O'Rourke, N. Dahmane, S. Brem, Y. Mou, Y. Gong and Y. Fan, Vascular niche IL-6 induces alternative macrophage activation in glioblastoma through HIF-2 α , *Nat Commun* **9** (2018), 559.
- 1201
- 1202
- 1203
- 1204
- 1205
- 1206 [40] I.A.W. Ho and W.S.N. Shim, Contribution of the microenvironmental niche to glioblastoma heterogeneity, *Biomed Res Int* **2017** (2017), 9634172.
- 1207
- 1208
- 1209 [41] T. Hide, I. Shibahara and T. Kumabe, Novel concept of the border niche: glioblastoma cells use oligodendrocytes progenitor cells (GAOs) and microglia to acquire stem cell-like features, *Brain Tumor Pathol* **36** (2019), 63–73.
- 1210
- 1211
- 1212
- 1213 [42] E. Jung, M. Osswald, M. Ratliff, H. Dogan, R. Xie, S. Weil, D.C. Hoffmann, F.T. Kurz, T. Kessler, S. Heiland, A. von Deimling, F. Sahm, W. Wick and F. Winkler, Tumor cell plasticity, heterogeneity, and resistance in crucial microenvironmental niches in glioma, *Nat Commun* **12** (2021), 1014.
- 1214
- 1215
- 1216
- 1217 [43] T. Taga and K. Tabu, Glioma progression and recurrence involving maintenance and expansion strategies of glioma stem cells by organizing self-advantageous niche microenvironments, *Inflamm Regen* **40** (2020), 33.
- 1218
- 1219
- 1220
- 1221
- 1222 [44] M. Guilliams, G.R. Thierry, J. Bonnardel and M. Bajenoff, Establishment and maintenance of the macrophage niche, *Immunity* **52** (2020), 434–451.
- 1223
- 1224
- 1225 [45] Y. Pan, Y. Yu, X. Wang and T. Zhang, Tumor-associated macrophages in tumor immunity, *Front Immunol* **11** (2020), 583084.
- 1226
- 1227
- 1228 [46] L.J. Wang, Y. Xue and Y. Lou, Tumor-associated macrophages related signature in glioma, *Aging (Albany NY)* **14** (2022), 2720–2735.
- 1229
- 1230
- 1231 [47] M. Lim, Y. Xia, C. Bettegowda and M. Weller, Current state of immunotherapy for glioblastoma, *Nat Rev Clin Oncol* **15** (2018), 422–442.
- 1232
- 1233
- 1234 [48] J. Larkin, F.S. Hodi and J.D. Wolchok, Combined nivolumab and ipilimumab or monotherapy in untreated melanoma, *N Engl J Med* **373** (2015), 1270–1.
- 1235
- 1236
- 1237 [49] M. Weller, N. Butowski, D.D. Tran, L.D. Recht, M. Lim, H. Hirte, L. Ashby, L. Mechtler, S.A. Goldlust, F. Iwamoto, J. Drappatz, D.M. O'Rourke, M. Wong, M.G. Hamilton, G. Finocchiaro, J. Perry, W. Wick, J. Green, Y. He, C.D. Turner, M.J. Yellin, T. Keler, T.A. Davis, R. Stupp and J.H. Sampson, Rindopepimut with temozolomide for patients with newly diagnosed, EGFRvIII-expressing glioblastoma (ACT IV): a randomised, double-blind, international phase 3 trial, *Lancet Oncol* **18** (2017), 1373–1385.
- 1238
- 1239
- 1240
- 1241
- 1242
- 1243 [50] A. Omuro, G. Vlahovic, M. Lim, S. Sahebjam, J. Baehring, T. Cloughesy, A. Voloschin, S.H. Ramkissoon, K.L. Ligon, R. Latek, R. Zvirtes, L. Strauss, P. Paliwal, C.T. Harbison, D.A. Reardon and J.H. Sampson, Nivolumab with or without ipilimumab in patients with recurrent glioblastoma: results from exploratory phase I cohorts of CheckMate 143, *Neuro Oncol* **20** (2018), 674–686.
- 1244
- 1245
- 1246
- 1247 [51] S.R. Punekar and J.S. Weber, Intratumoral therapy to make a “Cold” tumor “Hot”: The jury is still out, *Clin Cancer Res* **28** (2022), 5007–5009.
- 1248
- 1249
- 1250
- 1251
- 1252 [52] V. Lemaire, D. Bassen, M. Reed, R. Song, S. Khalili, Y.T.K. Lien, L. Huang, A.P. Singh, S. Stamatelos, D. Bottino and F. Hua, From cold to hot: Changing perceptions and future opportunities for quantitative systems pharmacology modeling in cancer immunotherapy, *Clin Pharmacol Ther* **113** (2023), 963–972.
- 1253
- 1254
- 1255
- 1256 [53] C.M. Jackson, J. Choi and M. Lim, Mechanisms of immunotherapy resistance: lessons from glioblastoma, *Nat Immunol* **20** (2019), 1100–1109.
- 1257
- 1258
- 1259
- 1260 [54] J.S. O'Donnell, M.W.L. Teng and M.J. Smyth, Cancer immunoeediting and resistance to T cell-based immunotherapy, *Nat Rev Clin Oncol* **16** (2019), 151–167.
- 1261
- 1262
- 1263
- 1264 [55] A. Memarnejadian, C.E. Meilleur, C.R. Shaler, K. Khazaei, J.R. Bennink, T.D. Schell and S.M.M. Haeryfar, PD-1 blockade promotes epitope spreading in anticancer CD8(+) T Cell responses by preventing fratricidal death of subdominant
- 1265
- 1266
- 1267
- 1268
- 1269
- 1270
- 1271

- clones to relieve immunodomination, *J Immunol* **199** (2017), 3348–v3359.
- [56] D. Mathios, J.E. Kim, A. Mangraviti, J. Phallen, C.K. Park, C.M. Jackson, T. Garzon-Muvdi, E. Kim, D. Theodoros, M. Polanczyk, A.M. Martin, I. Suk, X. Ye, B. Tyler, C. Bettegowda, H. Brem, D.M. Pardoll and M. Lim, Anti-PD-1 antitumor immunity is enhanced by local and abrogated by systemic chemotherapy in GBM, *Sci Transl Med* **8** (2016), 370ra180.
- [57] A.L. Hung, R. Maxwell, D. Theodoros, Z. Belcaid, D. Mathios, A.S. Luksik, E. Kim, A. Wu, Y. Xia, T. Garzon-Muvdi, C. Jackson, X. Ye, B. Tyler, M. Selby, A. Korman, B. Barnhart, S.M. Park, J.I. Youn, T. Chowdhury, C.K. Park, H. Brem, D.M. Pardoll and M. Lim, TIGIT and PD-1 dual checkpoint blockade enhances antitumor immunity and survival in GBM, *Oncoimmunology* **7** (2018), e1466769.
- [58] J.E. Kim, M.A. Patel, A. Mangraviti, E.S. Kim, D. Theodoros, E. Velarde, A. Liu, E.W. Sankey, A. Tam, H. Xu, D. Mathios, C.M. Jackson, S. Harris-Bookman, T. Garzon-Muvdi, M. Sheu, A.M. Martin, B.M. Tyler, P.T. Tran, X. Ye, A. Olivi, J.M. Taube, P.C. Burger, C.G. Drake, H. Brem, D.M. Pardoll and M. Lim, Combination therapy with anti-PD-1, Anti-TIM-3, and focal radiation results in regression of murine gliomas, *Clin Cancer Res* **23** (2017), 124–136.
- [59] J. Zeng, A.P. See, J. Phallen, C.M. Jackson, Z. Belcaid, J. Ruzevick, N. Durham, C. Meyer, T.J. Harris, E. Albesiano, G. Pradilla, E. Ford, J. Wong, H.J. Hammers, D. Mathios, B. Tyler, H. Brem, P.T. Tran, D. Pardoll, C.G. Drake and M. Lim, Anti-PD-1 blockade and stereotactic radiation produce long-term survival in mice with intracranial gliomas, *Int J Radiat Oncol Biol Phys* **86** (2013), 343–9.
- [60] A. Wu, R. Maxwell, Y. Xia, P. Cardarelli, M. Oyasu, Z. Belcaid, E. Kim, A. Hung, A.S. Luksik, T. Garzon-Muvdi, C.M. Jackson, D. Mathios, D. Theodoros, J. Cogswell, H. Brem, D.M. Pardoll and M. Lim, Combination anti-CXCR4 and anti-PD-1 immunotherapy provides survival benefit in glioblastoma through immune cell modulation of tumor microenvironment, *J Neurooncol* **143** (2019), 241–249.
- [61] R. Medikonda, G. Dunn, M. Rahman, P. Fecci and M. Lim, A review of glioblastoma immunotherapy, *J Neurooncol* **151** (2021), 41–53.
- [62] C.M. Jackson and M. Lim, Immunotherapy for glioblastoma: playing chess, not checkers, *Clin Cancer Res* **24** (2018), 4059–4061.
- [63] W. Xiong, C. Li, G. Kong, B. Wan, S. Wang and J. Fan, Glioblastoma: two immune subtypes under the surface of the cold tumor, *Aging (Albany NY)* **14** (2022), 4357–4375.
- [64] C. Blériot, S. Chakarov and F. Ginhoux, Determinants of resident tissue macrophage identity and function, *Immunity* **52** (2020), 957–970.
- [65] C.D. Mills, K. Kincaid, J.M. Alt, M.J. Heilman and A.M. Hill, Pillars Article: M-1/M-2 Macrophages and the Th1/Th2 Paradigm, *J Immunol* **164** (2000) : 6166–6173; *J Immunol* **199** (2017), 2194–2201.
- [66] K. Beccaria, M. Canney, G. Bouchoux, C. Desseaux, J. Grill, A.B. Heimberger and A. Carpentier, Ultrasound-induced blood-brain barrier disruption for the treatment of gliomas and other primary CNS tumors, *Cancer Lett* **479** (2020), 13–22.
- [67] A. Sabbagh, K. Beccaria, X. Ling, A. Marisetty, M. Ott, H. Caruso, E. Barton, L.Y. Kong, D. Fang, K. Latha, D.Y. Zhang, J. Wei, J. DeGroot, M.A. Curran, G. Rao, J. Hu, C. Desseaux, G. Bouchoux, M. Canney, A. Carpentier and A.B. Heimberger, Opening of the blood-brain barrier using low-intensity pulsed ultrasound enhances responses to immunotherapy in preclinical glioma models, *Clin Cancer Res* **27** (2021), 4325–4337.
- [68] C. Kim, M. Lim, G.F. Woodworth and C.D. Arvanitis, The roles of thermal and mechanical stress in focused ultrasound-mediated immunomodulation and immunotherapy for central nervous system tumors, *J Neurooncol* **157** (2022), 221–236.
- [69] M. Toriello, V. González-Quintanilla and J. Pascual, The glymphatic system and its involvement in disorders of the nervous system, *Med Clin (Barc)* **156** (2021), 339–343.
- [70] G. Yankova, O. Bogomyakova and A. Tulupov, The glymphatic system and meningeal lymphatics of the brain: new understanding of brain clearance, *Rev Neurosci* **32** (2021), 693–705.
- [71] S. Naganawa and T. Taoka, The glymphatic system: A review of the challenges in visualizing its structure and function with MR imaging, *Magn Reson Med* **21** (2022), 182–194.
- [72] K. Bene, L. Halasz and L. Nagy, Transcriptional repression shapes the identity and function of tissue macrophages, *FEBS Open Bio* **11** (2021), 3218–3229.
- [73] Y. Lavin, D. Winter, R. Blecher-Gonen, E. David, H. Keren-Shaul, M. Merad, S. Jung and I. Amit, Tissue-resident macrophage enhancer landscapes are shaped by the local microenvironment, *Cell* **159** (2014), 1312–26.
- [74] A. Buttgerit, I. Lelios, X. Yu, M. Vrohligs, N.R. Krakoski, E.L. Gautier, R. Nishinakamura, B. Becher and M. Greter, Sall1 is a transcriptional regulator defining microglia identity and function, *Nat Immunol* **17** (2016), 1397–1406.
- [75] C. Minten, R. Terry, C. Deffrasnes, N.J. King and I.L. Campbell, IFN regulatory factor 8 is a key constitutive determinant of the morphological and molecular properties of microglia in the CNS, *PLoS One* **7** (2012), e49851.
- [76] S. Chen, J. Yang, Y. Wei and X. Wei, Epigenetic regulation of macrophages: from homeostasis maintenance to host defense, *Cell Mol Immunol* **17** (2020), 36–49.
- [77] N.A. Thornberry, H.G. Bull, J.R. Calaycay, K.T. Chapman, A.D. Howard, M.J. Kostura, D.K. Miller, S.M. Molineaux, J.R. Weidner, J. Aunins et al., A novel heterodimeric cysteine protease is required for interleukin-1 beta processing in monocytes, *Nature* **356** (1992), 768–74.
- [78] J. Shi, Y. Zhao, K. Wang, X. Shi, Y. Wang, H. Huang, Y. Zhuang, T. Cai, F. Wang and F. Shao, Cleavage of GSDMD by inflammatory caspases determines pyroptotic cell death, *Nature* **526** (2015), 660–5.
- [79] N. Kayagaki, I.B. Stowe, B.L. Lee, K. O'Rourke, K. Anderson, S. Warming, T. Cuellar, B. Haley, M. Roose-Girma, Q.T. Phung, P.S. Liu, J.R. Lill, H. Li, J. Wu, S. Kummerfeld, J. Zhang, W.P. Lee, S.J. Snipas, G.S. Salvesen, L.X. Morris, L. Fitzgerald, Y. Zhang, E.M. Bertram, C.C. Goodnow and V.M. Dixit, Caspase-11 cleaves gasdermin D for non-canonical inflammasome signalling, *Nature* **526** (2015), 666–71.
- [80] X. Liu, Z. Zhang, J. Ruan, Y. Pan, V.G. Magupalli, H. Wu and J. Lieberman, Inflammasome-activated gasdermin D causes pyroptosis by forming membrane pores, *Nature* **535** (2016), 153–8.
- [81] J. Shi, W. Gao and F. Shao, Pyroptosis: Gasdermin-mediated programmed necrotic cell death, *Trends Biochem Sci* **42** (2017), 245–254.
- [82] E.A. Miao, J.V. Rajan and A. Aderem, Caspase-1-induced pyroptotic cell death, *Immunol Rev* **243** (2011), 206–14.
- [83] S.B. Kovacs and E.A. Miao, Gasdermins: Effectors of pyroptosis, *Trends Cell Biol* **27** (2017), 673–684.
- [84] Y. Wang, B. Yin, D. Li, G. Wang, X. Han and X. Sun, GS-DME mediates caspase-3-dependent pyroptosis in gastric

- cancer, *Biochem Biophys Res Commun* **495** (2018), 1418–1425.
- [85] Y.Y. Wang, X.L. Liu and R. Zhao, Induction of pyroptosis and its implications in cancer management, *Front Oncol* **9** (2019), 971.
- [86] J. Yu, S. Li, J. Qi, Z. Chen, Y. Wu, J. Guo, K. Wang, X. Sun and J. Zheng, Cleavage of GSDME by caspase-3 determines lobaplatin-induced pyroptosis in colon cancer cells, *Cell Death Dis* **10** (2019), 193.
- [87] Y. Fang, S. Tian, Y. Pan, W. Li, Q. Wang, Y. Tang, T. Yu, X. Wu, Y. Shi, P. Ma and Y. Shu, Pyroptosis: A new frontier in cancer, *Biomed Pharmacother* **121** (2020), 109595.
- [88] J.K. Rathkey, J. Zhao, Z. Liu, Y. Chen, J. Yang, H.C. Kondolf, B.L. Benson, S.M. Chirieleison, A.Y. Huang, G.R. Dubyak, T.S. Xiao, X. Li and D.W. Abbott, Chemical disruption of the pyroptotic pore-forming protein gasdermin D inhibits inflammatory cell death and sepsis, *Sci Immunol* **3** (2018).
- [89] C.L. Evavold, J. Ruan, Y. Tan, S. Xia, H. Wu and J.C. Kagan, The pore-forming protein gasdermin D regulates interleukin-1 secretion from living macrophages, *Immunity* **48** (2018), 35–44e6.
- [90] F. Humphries, L. Shmuel-Galia, N. Ketelut-Carneiro, S. Li, B. Wang, V.V. Nemmara, R. Wilson, Z. Jiang, F. Khalighinejad, K. Muneeruddin, S.A. Shaffer, R. Dutta, C. Ionete, S. Pesiridis, S. Yang, P.R. Thompson and K.A. Fitzgerald, Succination inactivates gasdermin D and blocks pyroptosis, *Science* **369** (2020), 1633–1637.
- [91] J.J. Hu, X. Liu, S. Xia, Z. Zhang, Y. Zhang, J. Zhao, J. Ruan, X. Luo, X. Lou, Y. Bai, J. Wang, L.R. Hollingsworth, V.G. Magupalli, L. Zhao, H.R. Luo, J. Kim, J. Lieberman and H. Wu, FDA-approved disulfiram inhibits pyroptosis by blocking gasdermin D pore formation, *Nat Immunol* **21** (2020), 736–745.
- [92] C.L. Evavold, I. Hafner-Bratkovič, P. Devant, J.M. D’Andrea, E.M. Ngwa, E. Boršič, J.G. Doench, M.W. LaFleur, A.H. Sharpe, J.R. Thiagarajah and J.C. Kagan, Control of gasdermin D oligomerization and pyroptosis by the Ragulator-RagmTORC1 pathway, *Cell* **184** (2021), 4495–4511e19.
- [93] X.X. Liao, Y.Z. Dai, Y.Z. Zhao and K. Nie, Gasdermin E: A prospective target for therapy of diseases, *Front Pharmacol* **13** (2022), 855828.
- [94] J. Zou, Y. Zheng, Y. Huang, D. Tang, R. Kang and R. Chen, The versatile gasdermin family: Their function and roles in diseases, *Front Immunol* **12** (2021), 751533.
- [95] B. Chao, F. Jiang, H. Bai, P. Meng, L. Wang and F. Wang, Predicting the prognosis of glioma by pyroptosis-related signature, *J Cell Mol Med* **26** (2022), 133–143.
- [96] P. Chen, Y. Li, N. Li, L. Shen and Z. Li, Comprehensive analysis of pyroptosis-associated in molecular classification, immunity and prognostic of glioma, *Front Genet* **12** (2021), 781538.
- [97] Y. Zhang, F. Xi, Q. Yu, W. Lou, Z. Zeng, N. Su, J. Gao, S. Duan, Y. Deng, S. Guo, S. Lai, X. Tang and J. Zhang, Identification of a novel pyroptosis-related gene signature correlated with the prognosis of diffuse glioma patients, *Ann Transl Med* **9** (2021), 1766.
- [98] M. Zhang, Y. Cheng, Z. Xue, Q. Sun and J. Zhang, A novel pyroptosis-related gene signature predicts the prognosis of glioma through immune infiltration, *BMC Cancer* **21** (2021), 1311.
- [99] X. Niu, R. Cheng, Y. Wang, J. Chen, C. Wang and H. Ji, Development of a prognostic model of glioma based on pyroptosis-related genes, *World Neurosurg* **158** (2022), e929–e945.
- [100] L. Gao, Z. Jiang, Y. Han, Y. Li and X. Yang, Regulation of Pyroptosis by ncRNA: A Novel Research Direction, *Front Cell Dev Biol* **10** (2022), 840576.
- [101] Y. Liu, H. Wu, J. Jing, H. Li, S. Dong and Q. Meng, Down-regulation of hsa_circ_0001836 induces pyroptosis cell death in glioma cells via epigenetically upregulating NLRP1, *Front Oncol* **11** (2021), 622727.
- [102] T. Jin, M. Liu, Y. Liu, Y. Li, Z. Xu, H. He, J. Liu, Y. Zhang and Y. Ke, Lcn2-derived Circular RNA (hsa_circ_0088732) Inhibits Cell Apoptosis and Promotes EMT in Glioma via the miR-661/RAB3D Axis, *Front Oncol* **10** (2020), 170.
- [103] L. Chen, C. Wang, H. Sun, J. Wang, Y. Liang, Y. Wang and G. Wong, The bioinformatics toolbox for circRNA discovery and analysis, *Brief Bioinform* **22** (2021), 1706–1728.
- [104] G. Liang, Y. Ling, M. Mehrpour, P.E. Saw, Z. Liu, W. Tan, Z. Tian, W. Zhong, W. Lin, Q. Luo, Q. Lin, Q. Li, Y. Zhou, A. Hamai, P. Codogno, J. Li, E. Song and C. Gong, Autophagy-associated circRNA circCDYL augments autophagy and promotes breast cancer progression, *Mol Cancer* **19** (2020), 65.
- [105] Z. Liu, Q. Wang, X. Wang, Z. Xu, X. Wei and J. Li, Circular RNA ciARS regulates ferroptosis in HCC cells through interacting with RNA binding protein ALKBH5, *Cell Death Discov* **6** (2020), 72.
- [106] J. Guarnerio, Y. Zhang, G. Cheloni, R. Panella, J. Mae Katon, M. Simpson, A. Matsumoto, A. Papa, C. Loretelli, A. Petri, S. Kauppinen, C. Garbutt, G.P. Nielsen, V. Deshpande, M. Castillo-Martin, C. Cordon-Cardo, S. Dimitrios, J.G. Clohessy, M. Batish and P.P. Pandolfi, Intragenic antagonistic roles of protein and circRNA in tumorigenesis, *Cell Res* **29** (2019), 628–640.
- [107] T. Yu, Y. Wang, Y. Fan, N. Fang, T. Wang, T. Xu and Y. Shu, CircRNAs in cancer metabolism: a review, *J Hematol Oncol* **12** (2019), 90.
- [108] J. Liu, X. Dou, C. Chen, C. Chen, C. Liu, M.M. Xu, S. Zhao, B. Shen, Y. Gao, D. Han and C. He, N (6)-methyladenosine of chromosome-associated regulatory RNA regulates chromatin state and transcription, *Science* **367** (2020), 580–586.
- [109] J. Li, F. Cao, H.L. Yin, Z.J. Huang, Z.T. Lin, N. Mao, B. Sun and G. Wang, Ferroptosis: past, present and future, *Cell Death Dis* **11** (2020), 88.
- [110] R.J. Wan, W. Peng, Q.X. Xia, H.H. Zhou and X.Y. Mao, Ferroptosis-related gene signature predicts prognosis and immunotherapy in glioma, *CNS Neurosci Ther* **27** (2021), 973–986.
- [111] J. Zheng, Z. Zhou, Y. Qiu, M. Wang, H. Yu, Z. Wu, X. Wang and X. Jiang, A prognostic ferroptosis-related lncRNAs signature associated with immune landscape and radiotherapy response in glioma, *Front Cell Dev Biol* **9** (2021), 675555.
- [112] X. Zhu, Y. Zhou, Y. Ou, Z. Cheng, D. Han, Z. Chu and S. Pan, Characterization of ferroptosis signature to evaluate the predict prognosis and immunotherapy in glioblastoma, *Aging (Albany NY)* **13** (2021), 17655–17672.
- [113] L. Sun, B. Li, B. Wang, J. Li and J. Li, Construction of a risk model to predict the prognosis and immunotherapy of low-grade glioma ground on 7 ferroptosis-related genes, *Int J Gen Med* **15** (2022), 4697–4716.
- [114] D. Tang, R. Kang, T.V. Berge, P. Vandenabeele and G. Kroemer, The molecular machinery of regulated cell death, *Cell Res* **29** (2019), 347–364.
- [115] W. Wang, M. Green, J.E. Choi, M. Gijón, P.D. Kennedy, J.K. Johnson, P. Liao, X. Lang, I. Kryczek, A. Sell, H. Xia, J. Zhou, G. Li, J. Li, W. Li, S. Wei, L. Vatan, H. Zhang, W. Szeliga, W. Gu, R. Liu, T.S. Lawrence, C. Lamb, Y. Tanno, M. Cieslik, E. Stone, G. Georgiou, T.A. Chan, A. Chinnaiyan and

- W. Zou, CD8(+) T cells regulate tumour ferroptosis during cancer immunotherapy, *Nature* **569** (2019), 270–274.
- [116] C. Wan, Y. Sun, Y. Tian, L. Lu, X. Dai, J. Meng, J. Huang, Q. He, B. Wu, Z. Zhang, K. Jiang, D. Hu, G. Wu, J.F. Lovell, H. Jin and K. Yang, Irradiated tumor cell-derived microparticles mediate tumor eradication via cell killing and immune reprogramming, *Sci Adv* **6** (2020), eaay9789.
- [117] I. Efimova, E. Catanzaro, L. Van der Meer, V.D. Turubanova, H. Hammad, T.A. Mishchenko, M.V. Vedunova, C. Fimognari, C. Bachert, F. Coppieters, S. Lefever, A.G. Skirtach, O. Krysko and D.V. Krysko, Vaccination with early ferroptotic cancer cells induces efficient antitumor immunity, *J Immunother Cancer* **8** (2020).
- [118] D. Xiao, Y. Zhou, X. Wang, H. Zhao, C. Nie and X. Jiang, A ferroptosis-related prognostic risk score model to predict clinical significance and immunogenic characteristics in glioblastoma multiforme, *Oxid Med Cell Longev* **2021** (2021), 9107857.
- [119] A.O. Mitre, A.I. Florian, A. Buruiana, A. Boer, I. Moldovan, O. Soritau, S.I. Florian and S. Susman, Ferroptosis involvement in glioblastoma treatment, *Medicina (Kaunas)* **58** (2022).
- [120] J. Van Loenhout, L. Freire Boulosa, D. Quatannens, J. De Waele, C. Merlin, H. Lambrechts, H.W. Lau, C. Hermans, A. Lin, F. Lardon, M. Peeters, A. Bogaerts, E. Smits and C. Deben, Auranofin and cold atmospheric plasma synergize to trigger distinct cell death mechanisms and immunogenic responses in glioblastoma, *Cells* **10** (2021).
- [121] Q. Song, S. Peng, Z. Sun, X. Heng and X. Zhu, Temozolomide drives ferroptosis via a DMT1-dependent pathway in glioblastoma cells, *Yonsei Med J* **62** (2021), 843–849.
- [122] C. Qiu, X. Zhang, B. Huang, S. Wang, W. Zhou, C. Li, X. Li, J. Wang and N. Yang, Disulfiram, a ferroptosis inducer, triggers lysosomal membrane permeabilization by Up-Regulating ROS in glioblastoma, *Oncotargets Ther* **13** (2020), 10631–10640.
- [123] I.C. Su, Y.K. Su, S.A. Setiawan, V.K. Yadav, I.H. Fong, C.T. Yeh, C.M. Lin and H.W. Liu, NADPH Oxidase Subunit CYBB Confers Chemotherapy and Ferroptosis Resistance in Mesenchymal Glioblastoma via Nrf2/SOD2 Modulation, *Int J Mol Sci* **24** (2023).
- [124] K. Liu, L. Jiang, Y. Shi, B. Liu, Y. He, Q. Shen, X. Jiang, Z. Nie, J. Pu, C. Yang and Y. Chen, Hypoxia-induced GLT8D1 promotes glioma stem cell maintenance by inhibiting CD133 degradation through N-linked glycosylation, *Cell Death Differ* **29** (2022), 1834–1849.
- [125] J. Yue, R. Huang, Z. Lan, B. Xiao and Z. Luo, Abnormal glycosylation in glioma: related changes in biology, biomarkers and targeted therapy, *Biomark Res* **11** (2023), 54.
- [126] Y. Qi, W. Lv, X. Liu, Q. Wang, B. Xing, Q. Jiang, Z. Wang, Y. Huang, K. Shu and T. Lei, Comprehensive analysis identified glycosyltransferase signature to predict glioma prognosis and TAM phenotype, *Biomed Res Int* **2023** (2023), 6082635.
- [127] X. Wu, H. Wang, S. Li, H. Luo and F. Liu, Mining glycosylation-related prognostic lncRNAs and constructing a prognostic model for overall survival prediction in glioma: A study based on bioinformatics analysis, *Medicine (Baltimore)* **102** (2023), e33569.
- [128] S.K. Natarajan and S. Venneti, Glioblastoma stem cell H3TARionics, *Cell Stem Cell* **29** (2022), 1509–1510.
- [129] C. Cencioni, F. Scagnoli, F. Spallotta, S. Nasi and B. Illi, The “superoncogene” myc at the crossroad between metabolism and gene expression in glioblastoma multiforme, *Int J Mol Sci* **24** (2023).
- [130] A. Aspelund, S. Antila, S.T. Proulx, T.V. Karlsen, S. Karman, M. Detmar, H. Wiig and K. Alitalo, A dural lymphatic vascular system that drains brain interstitial fluid and macromolecules, *J Exp Med* **212** (2015), 991–9.
- [131] A. Louveau, I. Smirnov, T.J. Keyes, J.D. Eccles, S.J. Rouhani, J.D. Peske, N.C. Derecki, D. Castle, J.W. Mandell, K.S. Lee, T.H. Harris and J. Kipnis, Structural and functional features of central nervous system lymphatic vessels, *Nature* **523** (2015), 337–41.
- [132] J.H. Ahn, H. Cho, J.H. Kim, S.H. Kim, J.S. Ham, I. Park, S.H. Suh, S.P. Hong, J.H. Song, Y.K. Hong, Y. Jeong, S.H. Park and G.Y. Koh, Meningeal lymphatic vessels at the skull base drain cerebrospinal fluid, *Nature* **572** (2019), 62–66.
- [133] M. Hsu, A. Rayasam, J.A. Kijak, Y.H. Choi, J.S. Harding, S.A. Marcus, W.J. Karpus, M. Sandor and Z. Fabry, Neuroinflammation-induced lymphangiogenesis near the cribriform plate contributes to drainage of CNS-derived antigens and immune cells, *Nat Commun* **10** (2019), 229.
- [134] X. Hu, Q. Deng, L. Ma, Q. Li, Y. Chen, Y. Liao, F. Zhou, C. Zhang, L. Shao, J. Feng, T. He, W. Ning, Y. Kong, Y. Huo, A. He, B. Liu, J. Zhang, R. Adams, Y. He, F. Tang, X. Bian and J. Luo, Meningeal lymphatic vessels regulate brain tumor drainage and immunity, *Cell Res* **30** (2020), 229–243.
- [135] E. Song, T. Mao, H. Dong, L.S.B. Boisserand, S. Antila, M. Bosenberg, K. Alitalo, J.L. Thomas and A. Iwasaki, VEGF-C-driven lymphatic drainage enables immunosurveillance of brain tumours, *Nature* **577** (2020), 689–694.
- [136] S.T. Proulx, Cerebrospinal fluid outflow: a review of the historical and contemporary evidence for arachnoid villi, perineural routes, and dural lymphatics, *Cell Mol Life Sci* **78** (2021), 2429–2457.
- [137] Q. Ma, F. Schlegel, S.B. Bachmann, H. Schneider, Y. Decker, M. Rudin, M. Weller, S.T. Proulx and M. Detmar, Lymphatic outflow of cerebrospinal fluid is reduced in glioma, *Sci Rep* **9** (2019), 14815.
- [138] E.S. Norton, L.A. Whaley, M.J. Ulloa-Navas, P. García-Tárraga, K.M. Meneses, M. Lara-Velazquez, N. Zarco, A. Carrano, A. Quiñones-Hinojosa, J.M. García-Verdugo and H. Guerrero-Cázares, Glioblastoma disrupts the ependymal wall and extracellular matrix structures of the subventricular zone, *Fluids Barriers CNS* **19** (2022), 58.
- [139] T. McGranahan, K.E. Therkelsen, S. Ahmad and S. Nagpal, Current state of immunotherapy for treatment of glioblastoma, *Curr Treat Options Oncol* **20** (2019), 24.
- [140] N. Zhang, L. Wei, M. Ye, C. Kang and H. You, Treatment progress of immune checkpoint blockade therapy for glioblastoma, *Front Immunol* **11** (2020), 592612.
- [141] I. Bryukhovetskiy, Cell-based immunotherapy of glioblastoma multiforme, *Oncol Lett* **23** (2022), 133.
- [142] S. Rohit Reddy, A. Llukmani, A. Hashim, D.R. Haddad, D.S. Patel, F. Ahmad, M. Abu Sneineh and D.K. Gordon, The role of chimeric antigen Receptor-T Cell therapy in the treatment of hematological malignancies: advantages, trials, and tribulations, and the road ahead, *Cureus* **13** (2021), e13552.
- [143] C.T. Petersen and G. Krenciute, Next generation CAR T cells for the immunotherapy of High-Grade glioma, *Front Oncol* **9** (2019), 69.
- [144] H.J. Yoo and B.N. Harapan, Chimeric antigen receptor (CAR) immunotherapy: basic principles, current advances, and future prospects in neuro-oncology, *Immunol Res* **69** (2021), 471–486.
- [145] B. Yuan, G. Wang, X. Tang, A. Tong and L. Zhou, Immunotherapy of glioblastoma: Recent advances and future prospects, *Hum Vaccin Immunother* **18** (2022), 2055417.

- 1656 [146] A. Mintz, D.M. Gibo, B. Slagle-Webb, N.D. Christensen and
1657 W. Debinski, IL-13R α 2 is a glioma-restricted receptor
1658 for interleukin-13, *Neoplasia* **4** (2002), 388–99.
- 1659 [147] S. Sengupta, B. Thaci, A.C. Crawford and P. Sampath,
1660 Interleukin-13 receptor alpha 2-targeted glioblastoma im-
1661 munotherapy, *Biomed Res Int* **2014** (2014), 952128.
- 1662 [148] M. Tu, W. Wange, L. Cai, P. Zhu, Z. Gao and W. Zheng, IL-13
1663 receptor α 2 stimulates human glioma cell growth and metas-
1664 tasis through the Src/PI3K/Akt/mTOR signaling pathway,
1665 *Tumour Biol* **37** (2016), 14701–14709.
- 1666 [149] C.E. Brown, C.D. Warden, R. Starr, X. Deng, B. Badie, Y.C.
1667 Yuan, S.J. Forman and M.E. Barish, Glioma IL13R α 2 is
1668 associated with mesenchymal signature gene expression and
1669 poor patient prognosis, *PLoS One* **8** (2013), e77769.
- 1670 [150] S. Kong, S. Sengupta, B. Tyler, A.J. Bais, Q. Ma, S. Doucette,
1671 J. Zhou, A. Sahin, B.S. Carter, H. Brem, R.P. Junghans and
1672 P. Sampath, Suppression of human glioma xenografts with
1673 second-generation IL13R-specific chimeric antigen receptor-
1674 modified T cells, *Clin Cancer Res* **18** (2012), 5949–60.
- 1675 [151] N. Muhammad, R. Wang, W. Li, Z. Zhang, Y. Chang, Y. Hu,
1676 J. Zhao, X. Zheng, Q. Mao and H. Xia, A novel TanCAR
1677 targeting IL13R α 2 and EphA2 for enhanced glioblastoma
1678 therapy, *Mol Ther Oncolytics* **24** (2022), 729–741.
- 1679 [152] C.J. Shen, Y.X. Yang, E.Q. Han, N. Cao, Y.F. Wang, Y. Wang,
1680 Y.Y. Zhao, L.M. Zhao, J. Cui, P. Gupta, A.J. Wong and S.Y.
1681 Han, Chimeric antigen receptor containing ICOS signaling
1682 domain mediates specific and efficient antitumor effect of T
1683 cells against EGFRvIII expressing glioma, *J Hematol Oncol*
1684 **6** (2013), 33.
- 1685 [153] D.M. O'Rourke, M.P. Nasrallah, A. Desai, J.J. Melenhorst, K.
1686 Mansfield, J.J.D. Morrisette, M. Martinez-Lage, S. Brem, E.
1687 Maloney, A. Shen, R. Isaacs, S. Mohan, G. Plesa, S.F. Lacey,
1688 J.M. Navenot, Z. Zheng, B.L. Levine, H. Okada, C.H. June,
1689 J.L. Brogdon and M.V. Maus, A single dose of peripherally
1690 infused EGFRvIII-directed CAR T cells mediates antigen
1691 loss and induces adaptive resistance in patients with recurrent
1692 glioblastoma, *Sci Transl Med* **9** (2017).
- 1693 [154] A. Sahin, C. Sanchez, S. Bullain, P. Waterman, R. Weissleder
1694 and B.S. Carter, Development of third generation anti-
1695 EGFRvIII chimeric T cells and EGFRvIII-expressing arti-
1696 ficial antigen presenting cells for adoptive cell therapy for
1697 glioma, *PLoS One* **13** (2018), e0199414.
- 1698 [155] A. Oprita, S.C. Baloi, G.A. Staicu, O. Alexandru, D.E. Tache,
1699 S. Danoiu, E.S. Micu and A.S. Sevastre, Updated Insights on
1700 EGFR signaling pathways in glioma, *Int J Mol Sci* **22** (2021).
- 1701 [156] N. Müller, S. Michen, S. Tietze, K. Töpfer, A. Schulte, K.
1702 Lamszus, M. Schmitz, G. Schackert, I. Pastan and A. Temme,
1703 Engineering NK cells modified with an EGFRvIII-specific
1704 chimeric antigen receptor to overexpress CXCR4 improves
1705 immunotherapy of CXCL12/SDF-1 α -secreting glioblastoma,
1706 *J Immunother* **38** (2015), 197–210.
- 1707 [157] K. Töpfer, M. Cartellieri, S. Michen, R. Wiedemuth, N.
1708 Müller, D. Lindemann, M. Bachmann, M. Füssel, G. Schack-
1709 ert and A. Temme, DAP12-based activating chimeric antigen
1710 receptor for NK cell tumor immunotherapy, *J Immunol* **194**
1711 (2015), 3201–12.
- 1712 [158] J. Han, J. Chu, W. Keung Chan, J. Zhang, Y. Wang, J.B.
1713 Cohen, A. Victor, W.H. Meisen, S.H. Kim, P. Grandi, Q.E.
1714 Wang, X. He, I. Nakano, E.A. Chiocca, J.C. Glorioso Iii, B.
1715 Kaur, M.A. Caligiuri and J. Yu, CAR-Engineered NK cells
1716 targeting Wild-Type EGFR and EGFRvIII enhance killing of
1717 glioblastoma and patient-derived glioblastoma stem cells, *Sci*
1718 *Rep* **5** (2015), 11483.
- 1719 [159] K. Schönfeld, C. Sahm, C. Zhang, S. Naundorf, C. Bren-
del, M. Odendahl, P. Nowakowska, H. Bönig, U. Köhl, S.
Kloess, S. Köhler, H. Holtgreve-Grez, A. Jauch, M. Schmidt,
R. Schubert, K. Köhlcke, E. Seifried, H.G. Klingemann, M.A.
Rieger, T. Tonn, M. Grez and W.S. Wels, Selective inhi-
bition of tumor growth by clonal NK cells expressing an
ErbB2/HER2-specific chimeric antigen receptor, *Mol Ther*
23 (2015), 330–8.
- [160] A. Jamali, J. Hadjati, Z. Madjd, H.R. Mirzaei, F.B. Thal-
heimer, S. Agarwal, H. Bonig, E. Ullrich and J. Hartmann,
Highly efficient generation of transgenically augmented CAR
NK cells overexpressing CXCR4, *Front Immunol* **11** (2020),
2028.
- [161] G.H. Ran, Y.Q. Lin, L. Tian, T. Zhang, D.M. Yan, J.H. Yu
and Y.C. Deng, Natural killer cell homing and trafficking
in tissues and tumors: from biology to application, *Signal*
Transduct Target Ther **7** (2022), 205.
- [162] L.Y. Chan, S.A. Dass, G.J. Tye, S.A.M. Imran, W.S. Wan
Kamarul Zaman and F. Nordin, CAR-T cells/-NK Cells in
Cancer Immunotherapy and the Potential of MSC to Enhance
Its Efficacy: A Review, *Biomedicines* **10** (2022).
- [163] G. Golinelli, G. Grisendi, M. Prapa, M. Bestagno, C. Spano,
F. Rossignoli, F. Bambi, I. Sardi, M. Cellini, E.M. Horwitz, A.
Feletti, G. Pavesi and M. Dominici, Targeting GD2-positive
glioblastoma by chimeric antigen receptor empowered mes-
enchymal progenitors, *Cancer Gene Ther* **27** (2020), 558–
570.
- [164] T.D. Traylor and E.L. Hogan, Gangliosides of human cerebral
astrocytomas, *J Neurochem* **34** (1980), 126–31.
- [165] J.G. Zhang, C.A. Kruse, L. Driggers, N. Hoa, J. Wisoff, J.C.
Allen, D. Zagzag, E.W. Newcomb and M.R. Judas, Tumor
antigen precursor protein profiles of adult and pediatric brain
tumors identify potential targets for immunotherapy, *J Neu-
roncol* **88** (2008), 65–76.
- [166] N. Ahmed, V.S. Salsman, Y. Kew, D. Shaffer, S. Powell,
Y.J. Zhang, R.G. Grossman, H.E. Heslop and S. Gottschalk,
HER2-specific T cells target primary glioblastoma stem cells
and induce regression of autologous experimental tumors,
Clin Cancer Res **16** (2010), 474–85.
- [167] N. Ahmed, V. Brawley, M. Hegde, K. Bielamowicz, M. Kalra,
D. Landi, C. Robertson, T.L. Gray, O. Diouf, A. Wakefield,
A. Ghazi, C. Gerken, Z. Yi, A. Ashoori, M.F. Wu, H. Liu,
C. Rooney, G. Dotti, A. Gee, J. Su, Y. Kew, D. Baskin, Y.J.
Zhang, P. New, B. Grilley, M. Stojakovic, J. Hicks, S.Z.
Powell, M.K. Brenner, H.E. Heslop, R. Grossman, W.S. Wels
and S. Gottschalk, HER2-specific chimeric antigen receptor-
modified virus-specific T cells for progressive glioblastoma:
A phase 1 dose-escalation trial, *JAMA Oncol* **3** (2017), 1094–
1101.
- [168] S. Yang, W. Wei and Q. Zhao, B7-H3, a checkpoint molecule,
as a target for cancer immunotherapy, *Int J Biol Sci* **16** (2020),
1767–1773.
- [169] R.G. Majzner, J.L. Theruvath, A. Nellan, S. Heitzeneder,
Y. Cui, C.W. Mount, S.P. Rietberg, M.H. Linde, P. Xu, C.
Rota, E. Sotillo, L. Labanieh, D.W. Lee, R.J. Orentas, D.S.
Dimitrov, Z. Zhu, B.S. Croix, A. Delaidelli, A. Sekunova, E.
Bonvini, S.S. Mitra, M.M. Quezado, R. Majeti, M. Monje,
P.H.B. Sorensen, J.M. Maris and C.L. Mackall, CAR T cells
Targeting B7-H3, a Pan-Cancer Antigen, Demonstrate Potent
Preclinical Activity Against Pediatric Solid Tumors and Brain
Tumors, *Clin Cancer Res* **25** (2019), 2560–2574.
- [170] Z. Zhou, N. Luther, G.M. Ibrahim, C. Hawkins, R. Vibhakar,
M.H. Handler and M.M. Souweidane, B7-H3, a potential
therapeutic target, is expressed in diffuse intrinsic pontine
glioma, *J Neurooncol* **111** (2013), 257–64.

- 1784 [171] X. Tang, S. Zhao, Y. Zhang, Y. Wang, Z. Zhang, M. Yang, Y. Zhu, G. Zhang, G. Guo, A. Tong and L. Zhou, B7-H3 as a novel CAR-T therapeutic target for glioblastoma, *Mol Ther Oncolytics* **14** (2019), 279–287.
- 1785
1786
1787
1788 [172] D. Nehama, N. Di Ianni, S. Musio, H. Du, M. Patané, B. Pollo, G. Finocchiaro, J.J.H. Park, D.E. Dunn, D.S. Edwards, J.S. Damrauer, H. Hudson, S.R. Floyd, S. Ferrone, B. Savoldo, S. Pellegatta and G. Dotti, B7-H3-redirected chimeric antigen receptor T cells target glioblastoma and neurospheres, *EBioMedicine* **47** (2019), 33–43.
- 1789
1790
1791
1792
1793
1794 [173] L. Miao, J. Zhang, Z. Zhang, S. Wang, F. Tang, M. Teng and Y. Li, A bibliometric and knowledge-map analysis of CAR-T cells from 2009 to 2021, *Front Immunol* **13** (2022), 840956.
- 1795
1796
1797 [174] A. Ghouzliani, S. Kandoussi, M. Tall, K.P. Reddy, S. Rafii and A. Badou, Immune Checkpoint Inhibitors in Human Glioma Microenvironment, *Front Immunol* **12** (2021), 679425.
- 1798
1799
1800 [175] Y. Jiang, M. Chen, H. Nie and Y. Yuan, PD-1 and PD-L1 in cancer immunotherapy: clinical implications and future considerations, *Hum Vaccin Immunother* **15** (2019), 1111–1122.
- 1801
1802
1803
1804 [176] Z. Zhu, H. Zhang, B. Chen, X. Liu, S. Zhang, Z. Zong and M. Gao, PD-L1-Mediated Immunosuppression in Glioblastoma Is Associated With the Infiltration and M2-Polarization of Tumor-Associated Macrophages, *Front Immunol* **11** (2020), 588552.
- 1805
1806
1807
1808 [177] M. Puigdelloses, M. Garcia-Moure, S. Labiano, V. Laspi-dea, M. Gonzalez-Huarriz, M. Zalacain, L. Marrodan, N. Martinez-Velez, D. De la Nava, I. Ausejo, S. Hervás-Stubbs, G. Herrador, Z. Chen, D. Hambardzumyan, A. Patino Garcia, H. Jiang, C. Gomez-Manzano, J. Fueyo, J. Gállego Pérez-Larraya and M. Alonso, CD137 and PD-L1 targeting with immunovirotherapy induces a potent and durable antitumor immune response in glioblastoma models, *J Immunother Cancer* **9** (2021).
- 1809
1810
1811
1812
1813
1814
1815
1816
1817 [178] J. Niogret, H. Berger, C. Rebe, R. Mary, E. Ballot, C. Truntzer, M. Thibaudin, V. Derangère, C. Hibos, L. Hampe, D. Rageot, T. Accogli, P. Joubert, B. Routy, J. Harker, F. Vegran, F. Ghiringhelli and F. Chalmin, Follicular helper-T cells restore CD8(+)-dependent antitumor immunity and anti-PD-L1/PD-1 efficacy, *J Immunother Cancer* **9** (2021).
- 1818
1819
1820
1821
1822
1823
1824 [179] T.B. Scheffel, N. Grave, P. Vargas, F.M. Diz, L. Rockenbach and F.B. Morrone, Immunosuppression in gliomas via PD-1/PD-L1 axis and adenosine pathway, *Front Oncol* **10** (2020), 617385.
- 1825
1826
1827
1828 [180] I. Raphael, R. Kumar, L.H. McCarl, K. Shoger, L. Wang, P. Sandlesh, C.T. Sneiderman, J. Allen, S. Zhai, M.L. Campagna, A. Foster, T.C. Bruno, S. Agnihotri, B. Hu, B.A. Castro, F.S. Lieberman, A. Broniscer, A.A. Diaz, N.M. Amankulor, D. Rajasundaram, I.F. Pollack and G. Kohanbash, TIGIT and PD-1 Immune checkpoint pathways are associated with patient outcome and anti-tumor immunity in glioblastoma, *Front Immunol* **12** (2021), 637146.
- 1829
1830
1831
1832
1833
1834
1835
1836 [181] S. Zhang, X. Xiao, Y. Wang, T. Song, C. Li, H. Bao, Q. Liu, G. Sun, X. Sun, T. Su, T. Fu, Y. Wang and P. Liang, Developing an immune-related signature for predicting survival rate and the response to immune checkpoint inhibitors in patients with glioma, *Front Genet* **13** (2022), 899125.
- 1837
1838
1839
1840
1841 [182] R. Liu, W. Liang, Q. Hua, L. Wu, X. Wang, Q. Li, F. Zhong, B. Li and Z. Qiu, Fatty acid metabolic signaling pathway alternation predict prognosis of immune checkpoint inhibitors in glioblastoma, *Front Immunol* **13** (2022), 819515.
- 1842
1843
1844
1845 [183] G. Lai, K. Li, J. Deng, H. Liu, B. Xie and X. Zhong, Identification and validation of a gene signature for lower-grade gliomas based on pyroptosis-related genes to predict survival and response to immune checkpoint inhibitors, *J Healthc Eng* **2022** (2022), 8704127.
- 1846
1847
1848
1849
1850 [184] T. Tian, R. Liang, G. Erel-Akbaba, L. Saad, P.J. Obeid, J. Gao, E.A. Chioocca, R. Weissleder and B.A. Tannous, Immune checkpoint inhibition in GBM Primed with radiation by engineered extracellular vesicles, *ACS Nano* **16** (2022), 1940–1953.
- 1851
1852
1853
1854
1855 [185] Y. Yan, S. Zeng, Z. Gong and Z. Xu, Clinical implication of cellular vaccine in glioma: current advances and future prospects, *J Exp Clin Cancer Res* **39** (2020), 257.
- 1856
1857
1858 [186] O. Inslug, G. Ku, H.C. Ertl and M. Blaszczyk-Thurin, A dendritic cell vaccine induces protective immunity to intracranial growth of glioma, *Anticancer Res* **22** (2002), 613–21.
- 1859
1860
1861 [187] C.H. Kim, S.J. Woo, J.S. Park, H.S. Kim, M.Y. Park, S.D. Park, Y.K. Hong and T.G. Kim, Enhanced antitumor immunity by combined use of temozolomide and TAT-survivin pulsed dendritic cells in a murine glioma, *Immunology* **122** (2007), 615–22.
- 1862
1863
1864
1865 [188] B. Ji, Q. Chen, B. Liu, L. Wu, D. Tian, Z. Guo and W. Yi, Glioma stem cell-targeted dendritic cells as a tumor vaccine against malignant glioma, *Yonsei Med J* **54** (2013), 92–100.
- 1866
1867
1868 [189] F. Tian, C. Dou, S. Qi, B. Chen, L. Zhao and X. Wang, Dendritic cell-glioma fusion activates T lymphocytes by elevating cytotoxic efficiency as an antitumor vaccine, *Cent Eur J Immunol* **39** (2014), 265–70.
- 1869
1870
1871
1872 [190] L.M. Liao, K. Ashkan, D.D. Tran, J.L. Campian, J.E. Trusheim, C.S. Cobbs, J.A. Heth, M. Salacz, S. Taylor, S.D. D’Andre, F.M. Iwamoto, E.J. Dropcho, Y.A. Moshel, K.A. Walter, C.P. Pillainayagam, R. Aiken, R. Chaudhary, S.A. Guldust, D.A. Bota, P. Duic, J. Grewal, H. Elinzano, S.A. Toms, K.O. Lillehei, T. Mikkelsen, T. Walbert, S.R. Abram, A.J. Brenner, S. Brem, M.G. Ewend, S. Khagi, J. Portnow, L.J. Kim, W.G. Loudon, R.C. Thompson, D.E. Avigan, K.L. Fink, F.J. Geoffroy, S. Lindhorst, J. Lutzky, A.E. Sloan, G. Schackert, D. Krex, H.J. Meisel, J. Wu, R.P. Davis, C. Duma, A.B. Etame, D. Mathieu, S. Kesari, D. Piccioni, M. Westphal, D.S. Baskin, P.Z. New, M. Lacroix, S.A. May, T.J. Pluard, V. Tse, R.M. Green, J.L. Villano, M. Pearlman, K. Petrecca, M. Schulder, L.P. Taylor, A.E. Maida, R.M. Prins, T.F. Cloughesy, P. Mulholland and M.L. Bosch, First results on survival from a large Phase 3 clinical trial of an autologous dendritic cell vaccine in newly diagnosed glioblastoma, *J Transl Med* **16** (2018), 142.
- 1873
1874
1875
1876
1877
1878
1879
1880
1881
1882 [191] M.P. Pinho, G.A. Lepski, R. Rehder, N.E. Chauca-Torres, G.C.M. Evangelista, S.F. Teixeira, E.A. Flatow, J.V. de Oliveira, C.S. Fogolin, N. Peres, A. Arévalo, V. Alves, J.A.M. Barbuto and P.C. Bergami-Santos, Near-complete remission of glioblastoma in a patient treated with an allogenic dendritic cell-based vaccine: The role of tumor-specific CD4+T-cell cytokine secretion pattern in predicting response and recurrence, *Int J Mol Sci* **23** (2022).
- 1883
1884
1885
1886
1887
1888
1889
1890
1891 [192] X. Zhu, F. Nishimura, K. Sasaki, M. Fujita, J.E. Dusak, J. Eguchi, W. Fellows-Mayle, W.J. Storkus, P.R. Walker, A.M. Salazar and H. Okada, Toll like receptor-3 ligand poly-ICLC promotes the efficacy of peripheral vaccinations with tumor antigen-derived peptide epitopes in murine CNS tumor models, *J Transl Med* **5** (2007), 10.
- 1892
1893
1894
1895
1896
1897
1898
1899
1900 [193] K. Ueda, M. Fujita, X. Zhu, K. Sasaki, E.R. Kasthuber, G. Kohanbash, H.A. McDonald, J. Harper, S. Lonning and H. Okada, Systemic inhibition of transforming growth factor-beta in glioma-bearing mice improves the therapeutic efficacy of glioma-associated antigen peptide vaccines, *Clin Cancer Res* **15** (2009), 6551–9.
- 1901
1902
1903
1904
1905
1906
1907
1908
1909
1910 [194] H. Okada, L.H. Butterfield, R.L. Hamilton, A. Hoji, M.

- 1912 Sakaki, B.J. Ahn, G. Kohanbash, J. Drappatz, J. Engh, N.
1913 Amankulor, M.O. Lively, M.D. Chan, A.M. Salazar, E.G.
1914 Shaw, D.M. Potter and F.S. Lieberman, Induction of robust
1915 type-I CD8+ T-cell responses in WHO grade 2 low-grade
1916 glioma patients receiving peptide-based vaccines in combi-
1917 nation with poly-ICLC, *Clin Cancer Res* **21** (2015), 286–94.
1918 [195] S. Shibao, R. Ueda, K. Saito, R. Kikuchi, H. Nagashima,
1919 A. Kojima, H. Kagami, E.S. Pareira, H. Sasaki, S. Noji, Y.
1920 Kawakami, K. Yoshida and M. Toda, A pilot study of pep-
1921 tide vaccines for VEGF receptor 1 and 2 in patients with re-
1922 current/progressive high grade glioma, *Oncotarget* **9** (2018),
1923 21569–21579.
1924 [196] P.Y. Wen, D.A. Reardon, T.S. Armstrong, S. Phuphanich,
1925 R.D. Aiken, J.C. Landolfi, W.T. Curry, J.J. Zhu, M. Glantz,
1926 D.M. Peereboom, J.M. Markert, R. LaRocca, D.M. O'Rourke,
1927 K. Fink, L. Kim, M. Gruber, G.J. Lesser, E. Pan, S. Kesari,
1928 A. Muzikansky, C. Pinilla, R.G. Santos and J.S. Yu, A ran-
1929 domized double-blind placebo-controlled phase II trial of
1930 dendritic cell vaccine ICT-107 in newly diagnosed patients
1931 with glioblastoma, *Clin Cancer Res* **25** (2019), 5799–5807.
1932 [197] R. Kikuchi, R. Ueda, K. Saito, S. Shibao, H. Nagashima, R.
1933 Tamura, Y. Morimoto, H. Sasaki, S. Noji, Y. Kawakami, K.
1934 Yoshida and M. Toda, A pilot study of vaccine therapy with
1935 multiple glioma oncoantigen/glioma angiogenesis-associated
1936 antigen peptides for patients with recurrent/progressive high-
1937 grade glioma, *J Clin Med* **8** (2019).
1938 [198] D.B. Keskin, A.J. Anandappa, J. Sun, I. Tirosh, N.D. Math-
1939 ewson, S. Li, G. Oliveira, A. Giobbie-Hurder, K. Felt, E.
1940 Gjini, S.A. Shukla, Z. Hu, L. Li, P.M. Le, R.L. Allesøe, A.R.
1941 Richman, M.S. Kowalczyk, S. Abdelrahman, J.E. Geduldig,
1942 S. Charbonneau, K. Pelton, J.B. Iorgulescu, L. Elagina, W.
1943 Zhang, O. Olive, C. McCluskey, L.R. Olsen, J. Stevens, W.J.
1944 Lane, A.M. Salazar, H. Daley, P.Y. Wen, E.A. Chiocca, M.
1945 Harden, N.J. Lennon, S. Gabriel, G. Getz, E.S. Lander, A.
1946 Regev, J. Ritz, D. Neuberger, S.J. Rodig, K.L. Ligon, M.L.
1947 Suvà, K.W. Wucherpfeffnig, N. Hacohen, E.F. Fritsch, K.J. Li-
1948 vak, P.A. Ott, C.J. Wu and D.A. Reardon, Neoantigen vaccine
1949 generates intratumoral T cell responses in phase Ib glioblas-
1950 toma trial, *Nature* **565** (2019), 234–239.
1951 [199] S. Mueller, J.M. Taitt, J.E. Villanueva-Meyer, E.R. Bonner,
1952 T. Nejo, R.R. Lulla, S. Goldman, A. Banerjee, S.N. Chi,
1953 N.S. Whipple, J.R. Crawford, K. Gauvain, K.J. Nazemi, P.B.
1954 Watchmaker, N.D. Almeida, K. Okada, A.M. Salazar, R.D.
1955 Gilbert, J. Nazarian, A.M. Molinaro, L.H. Butterfield, M.D.
1956 Prados and H. Okada, Mass cytometry detects H3.3K27M-
1957 specific vaccine responses in diffuse midline glioma, *J Clin*
1958 *Invest* **130** (2020), 6325–6337.
1959 [200] M. Platten, L. Bunse, A. Wick, T. Bunse, L. Le Cornet, I.
1960 Harting, F. Sahm, K. Sanghvi, C.L. Tan, I. Poschke, E. Green,
1961 S. Justesen, G.A. Behrens, M.O. Breckwoldt, A. Freitag, L.M.
1962 Rother, A. Schmitt, O. Schnell, J. Hense, M. Misch, D. Krex,
1963 S. Stevanovic, G. Tabatabai, J.P. Steinbach, M. Bendszus, A.
1964 von Deimling, M. Schmitt and W. Wick, A vaccine targeting
1965 mutant IDH1 in newly diagnosed glioma, *Nature* **592** (2021),
1966 463–468.
1967 [201] Z. Jahanafrooz, B. Baradaran, J. Mosafer, M. Hashemzaei,
1968 T. Rezaei, A. Mokhtarzadeh and M.R. Hamblin, Comparison
1969 of DNA and mRNA vaccines against cancer, *Drug Discov*
1970 *Today* **25** (2020), 552–560.
1971 [202] M.A. McNamara, S.K. Nair and E.K. Holl, RNA-based vac-
1972 cines in cancer immunotherapy, *J Immunol Res* **2015** (2015),
1973 794528.
1974 [203] R. Ueda, E. Kinoshita, R. Ito, T. Kawase, Y. Kawakami and
1975 M. Toda, Induction of protective and therapeutic antitumor
immunity by a DNA vaccine with a glioma antigen, SOX6,
Int J Cancer **122** (2008), 2274–9.
[204] A. Mintz, D.M. Gibo, A.B. Madhankumar, N.M. Cladel, N.D.
Christensen and W. Debinski, Protein- and DNA-based ac-
tive immunotherapy targeting interleukin-13 receptor alpha2,
Cancer Biother Radiopharm **23** (2008), 581–9.
[205] A. Lopes, C. Bastiancich, M. Bausart, S. Ligoit, L. Lambrecht,
K. Vanvarenberg, B. Ucakar, B. Gallez, V. Pr at and G. Van-
dermeulen, New generation of DNA-based immunotherapy
induces a potent immune response and increases the survival
in different tumor models, *J Immunother Cancer* **9** (2021).
[206] L. Ye, L. Wang, J. Yang, P. Hu, C. Zhang, S. Tong, Z. Liu
and D. Tian, Identification of tumor antigens and immune
subtypes in lower grade gliomas for mRNA vaccine develop-
ment, *J Transl Med* **19** (2021), 352.
[207] Q. Zhou, X. Yan, H. Zhu, Z. Xin, J. Zhao, W. Shen, W. Yin,
Y. Guo, H. Xu, M. Zhao, W. Liu, X. Jiang and C. Ren, Ident-
ification of three tumor antigens and immune subtypes for
mRNA vaccine development in diffuse glioma, *Theranostics*
11 (2021), 9775–9790.
[208] H. Zhong, S. Liu, F. Cao, Y. Zhao, J. Zhou, F. Tang, Z. Peng,
Y. Li, S. Xu, C. Wang, G. Yang and Z.Q. Li, Dissecting tumor
antigens and immune subtypes of glioma to develop mRNA
vaccine, *Front Immunol* **12** (2021), 709986.
[209] S. Ma, Y. Ba, H. Ji, F. Wang, J. Du and S. Hu, Recognition of
Tumor-Associated Antigens and Immune Subtypes in Glioma
for mRNA Vaccine Development, *Front Immunol* **12** (2021),
738435.
[210] H. Lin, K. Wang, Y. Xiong, L. Zhou, Y. Yang, S. Chen, P. Xu,
Y. Zhou, R. Mao, G. Lv, P. Wang and D. Zhou, Identifica-
tion of tumor antigens and immune subtypes of glioblastoma
for mRNA vaccine development, *Front Immunol* **13** (2022),
773264.
[211] Z. Chen, X. Wang, Z. Yan and M. Zhang, Identification of
tumor antigens and immune subtypes of glioma for mRNA
vaccine development, *Cancer Med* **11** (2022), 2711–2726.
[212] E.A. Chiocca, F. Nassiri, J. Wang, P. Peruzzi and G. Zadeh,
Viral and other therapies for recurrent glioblastoma: is a 24-
month durable response unusual? *Neuro Oncol* **21** (2019),
14–25.
[213] G. Marelli, A. Howells, N.R. Lemoine and Y. Wang, On-
colytic viral therapy and the immune system: A double-edged
sword against cancer, *Front Immunol* **9** (2018), 866.
[214] N.G. Rainov, A phase III clinical evaluation of herpes simplex
virus type 1 thymidine kinase and ganciclovir gene therapy as
an adjuvant to surgical resection and radiation in adults with
previously untreated glioblastoma multiforme, *Hum Gene*
Ther **11** (2000), 2389–401.
[215] T. Mineta, S.D. Rabkin, T. Yazaki, W.D. Hunter and R.L.
Martuza, Attenuated multi-mutated herpes simplex virus-1
for the treatment of malignant gliomas, *Nat Med* **1** (1995),
938–43.
[216] J.M. Markert, S.N. Razdan, H.C. Kuo, A. Cantor, A. Knoll,
M. Karrasch, L.B. Nabors, M. Markiewicz, B.S. Agee, J.M.
Coleman, A.D. Lakeman, C.A. Palmer, J.N. Parker, R.J.
Whitley, R.R. Weichselbaum, J.B. Fiveash and G.Y. Gillespie,
A phase 1 trial of oncolytic HSV-1, G207, given in combi-
nation with radiation for recurrent GBM demonstrates safety
and radiographic responses, *Mol Ther* **22** (2014), 1048–55.
[217] G.K. Friedmann, J.M. Johnston, A.K. Bag, J.D. Bernstock, R.
Li, I. Aban, K. Kachurak, L. Nan, K.D. Kang, S. Totsch, C.
Schlappi, A.M. Martin, D. Pastakia, R. McNall-Knapp, S.
Farouk Sait, Y. Khakoo, M.A. Karajannis, K. Woodling, J.D.
Palmer, D.S. Osorio, J. Leonard, M.S. Abdelbaki, A. Madan-

- Swain, T.P. Atkinson, R.J. Whitley, J.B. Fiveash, J.M. Markert and G.Y. Gillespie, Oncolytic HSV-1 G207 immunovirotherapy for pediatric high-grade gliomas, *N Engl J Med* **384** (2021), 1613–1622.
- [218] J.M. Markert, M.D. Medlock, S.D. Rabkin, G.Y. Gillespie, T. Todo, W.D. Hunter, C.A. Palmer, F. Feigenbaum, C. Tornatore, F. Tufaro and R.L. Martuza, Conditionally replicating herpes simplex virus mutant, G207 for the treatment of malignant glioma: results of a phase I trial, *Gene Ther* **7** (2000), 867–74.
- [219] E.A. Chiocca, H. Nakashima, K. Kasai, S.A. Fernandez and M. Oglesbee, Preclinical toxicology of rQNestin34.5v.2: An oncolytic herpes virus with transcriptional regulation of the ICP34.5 neurovirulence gene, *Mol Ther Methods Clin Dev* **17** (2020), 871–893.
- [220] Y. Otani, J.Y. Yoo, C.T. Lewis, S. Chao, J. Swanner, T. Shimizu, J.M. Kang, S.A. Murphy, K. Rivera-Caraballo, B. Hong, J.C. Glorioso, H. Nakashima, S.E. Lawler, Y. Banasavadi-Siddegowda, J.D. Heiss, Y. Yan, G. Pei, M.A. Caligiuri, Z. Zhao, E.A. Chiocca, J. Yu and B. Kaur, NOTCH-Induced MDSC RECRUITment after oHSV virotherapy in CNS cancer models modulates antitumor immunotherapy, *Clin Cancer Res* **28** (2022), 1460–1473.
- [221] E.A. Adjei-Sowah, S.A. O'Connor, J. Veldhuizen, C. Lo Cascio, C. Plaisier, S. Mehta and M. Nikkha, Investigating the interactions of glioma stem cells in the perivascular niche at single-cell resolution using a microfluidic tumor microenvironment model, *Adv Sci (Weinh)* **9** (2022), e2201436.
- [222] D. Hambarzumyan and G. Bergers, Glioblastoma: Defining tumor niches, *Trends Cancer* **1** (2015), 252–265.
- [223] A. Filatova, T. Acker and B.K. Garvalov, The cancer stem cell niche(s): the crosstalk between glioma stem cells and their microenvironment, *Biochim Biophys Acta* **1830** (2013), 2496–508.
- [224] P.S. Nakod, Y. Kim and S.S. Rao, Biomimetic models to examine microenvironmental regulation of glioblastoma stem cells, *Cancer Lett* **429** (2018), 41–53.
- [225] Y. Chonan, S. Taki, O. Sampetean, H. Saya and R. Sudo, Endothelium-induced three-dimensional invasion of heterogeneous glioma initiating cells in a microfluidic coculture platform, *Integr Biol (Camb)* **9** (2017), 762–773.
- [226] Y. Xiao, D. Kim, B. Dura, K. Zhang, R. Yan, H. Li, E. Han, J. Ip, P. Zou, J. Liu, A.T. Chen, A.O. Vortmeyer, J. Zhou and R. Fan, Ex vivo dynamics of human glioblastoma cells in a microvasculature-on-a-chip system correlates with tumor heterogeneity and subtypes, *Adv Sci (Weinh)* **6** (2019), 1801531.
- [227] M.T. Ngo and B.A. Harley, The influence of hyaluronic acid and glioblastoma cell coculture on the formation of endothelial cell networks in gelatin hydrogels, *Adv Healthc Mater* **6** (2017).
- [228] C. Wang, J. Li, S. Sinha, A. Peterson, G.A. Grant and F. Yang, Mimicking brain tumor-vasculature microanatomical architecture via co-culture of brain tumor and endothelial cells in 3D hydrogels, *Biomaterials* **202** (2019), 35–44.
- [229] X. Cui, R.T. Morales, W. Qian, H. Wang, J.P. Gagner, I. Dolgalev, D. Placantonakis, D. Zagzag, L. Cimmino, M. Snuderl, R.H.W. Lam and W. Chen, Hacking macrophage-associated immunosuppression for regulating glioblastoma angiogenesis, *Biomaterials* **161** (2018), 164–178.
- [230] J.M. Ayuso, R. Monge, A. Martínez-González, M. Virumbrales-Muñoz, G.A. Llamazares, J. Berganzo, A. Hernández-Laín, J. Santolaria, M. Doblaré, C. Hubert, J.N. Rich, P. Sánchez-Gómez, V.M. Pérez-García, I. Ochoa and L.J. Fernández, Glioblastoma on a microfluidic chip: Generating pseudopalisades and enhancing aggressiveness through blood vessel obstruction events, *Neuro Oncol* **19** (2017), 503–513.
- [231] F. Yuan, X. Cai, Z. Cong, Y. Wang, Y. Geng, Y. Aili, C. Du, J. Zhu, J. Yang, C. Tang, A. Zhang, S. Zhao and C. Ma, Roles of the m(6)A modification of RNA in the glioblastoma microenvironment as revealed by single-cell analyses, *Front Immunol* **13** (2022), 798583.
- [232] R.C. Gimple, K. Yang, M.E. Halbert, S. Agnihotri and J.N. Rich, Brain cancer stem cells: resilience through adaptive plasticity and hierarchical heterogeneity, *Nat Rev Cancer* **22** (2022), 497–514.
- [233] W. Zhu, H. Chen, K. Yan, J. Wu, Y. Zhao and Q. Huang, Glioma stem cells and their microenvironment: A narrative review on docking and transformation, *Glioma* **5** (2022), 12–19.
- [234] R. Stupp, W.P. Mason, M.J. van den Bent, M. Weller, B. Fisher, M.J.B. Taphoorn, K. Belanger, A.A. Brandes, C. Marosi, U. Bogdahn, J. Curschmann, R.C. Janzer, S.K. Ludwin, T. Gorlia, A. Allgeier, D. Lacombe, J.G. Cairncross, E. Eisenhauer and R.O. Mirimanoff, Radiotherapy plus Concomitant and Adjuvant Temozolomide for Glioblastoma, *New England Journal of Medicine* **352** (2005), 987–996.
- [235] R. Stupp, M. Brada, M.J. van den Bent, J.C. Tonn and G. Pentheroudakis, High-grade glioma: ESMO clinical practice guidelines for diagnosis, treatment and follow-up, *Ann Oncol* **25**(Suppl 3) (2014), iii93–101.
- [236] B. Fazeny-Dörner, C. Wenzel, M. Veitl, M. Piribauer, K. Rössler, K. Dieckmann, K. Ungersböck and C. Marosi, Survival and prognostic factors of patients with unresectable glioblastoma multiforme, *Anticancer Drugs* **14** (2003), 305–312.
- [237] P. Lesueur, J. Lequesne, J.M. Grellard, A. Dugué, E. Coquan, P.E. Brachet, J. Geffrelot, W. Kao, E. Emery, D.H. Berro, L. Castera, N. Goardon, J. Lacroix, M. Lange, A. Capel, A. Leconte, B. Andre, A. Léger, A. Lelaidier, B. Clarisse and D. Stefan, Phase I/IIa study of concomitant radiotherapy with olaparib and temozolomide in unresectable or partially resectable glioblastoma: OLA-TMZ-RTE-01 trial protocol, *BMC Cancer* **19** (2019), 198.
- [238] K.R. Yabroff, L. Harlan, C. Zeruto, J. Abrams and B. Mann, Patterns of care and survival for patients with glioblastoma multiforme diagnosed during 2006, *Neuro Oncol* **14** (2012), 351–9.
- [239] D.A. Reardon, A.A. Brandes, A. Omuro, P. Mulholland, M. Lim, A. Wick, J. Baehring, M.S. Ahluwalia, P. Roth, O. Bähr, S. Phuphanich, J.M. Sepulveda, P. De Souza, S. Sahebjam, M. Carleton, K. Tatsuoka, C. Taitt, R. Zwirtes, J. Sampson and M. Weller, Effect of nivolumab vs bevacizumab in patients with recurrent glioblastoma: The CheckMate 143 phase 3 randomized clinical trial, *JAMA Oncol* **6** (2020), 1003–1010.
- [240] T. Cloughesy, G. Finocchiaro, C. Belda-Iniesta, L. Recht, A.A. Brandes, E. Pineda, T. Mikkelsen, O.L. Chinot, C. Balana, D.R. Macdonald, M. Westphal, K. Hopkins, M. Weller, C. Bais, T. Sandmann, J.M. Bruey, H. Koeppe, B. Liu, W. Verret, S.C. Phan and D.S. Shames, Randomized, double-blind, placebo-controlled, multicenter phase ii study of onartuzumab plus bevacizumab versus placebo plus bevacizumab in patients with recurrent glioblastoma: efficacy, safety, and hepatocyte growth factor and O(6)-methylguanine-DNA methyltransferase biomarker analyses, *J Clin Oncol* **35** (2017), 343–351.
- [241] A.F. Haddad, J.S. Young, D. Amara, M.S. Berger, D.R.

- 2168 Raleigh, M.K. Aghi and N.A. Butowski, Mouse models of
2169 glioblastoma for the evaluation of novel therapeutic strate-
2170 gies, *Neurooncol Adv* **3** (2021), vdab100.
- 2171 [242] M. Bausart, V. Pr at and A. Malfanti, Immunotherapy for
2172 glioblastoma: the promise of combination strategies, *J Exp*
2173 *Clin Cancer Res* **41** (2022), 35.
- 2174 [243] H. Luo, R. Hernandez, H. Hong, S.A. Graves, Y. Yang, C.G.
2175 England, C.P. Theuer, R.J. Nickles and W. Cai, Noninvasive
2176 brain cancer imaging with a bispecific antibody fragment,
2177 generated via click chemistry, *Proc Natl Acad Sci USA* **112**
2178 (2015), 12806–11.
- 2179 [244] K.R. Ulapane, B.M. Kopec and T.J. Siahaan, Improving in
2180 vivo brain delivery of monoclonal antibody using novel cyclic
2181 peptides, *Pharmaceutics* **11** (2019).
- 2182 [245] K.E. Warren, Beyond the Blood: Brain Barrier: The impor-
2183 tance of central nervous system (CNS) pharmacokinetics for
2184 the treatment of CNS tumors, including diffuse intrinsic pon-
2185 tine glioma, *Front Oncol* **8** (2018), 239.
- 2186 [246] N.D. Sheybani, V.R. Breza, S. Paul, K.S. McCauley,
2187 S.S. Berr, G.W. Miller, K.D. Neumann and R.J. Price,
2188 ImmunoPET-informed sequence for focused ultrasound-
2189 targeted mCD47 blockade controls glioma, *J Control Release*
2190 **331** (2021), 19–29.
- 2191 [247] E.A. Nance, G.F. Woodworth, K.A. Sailor, T.Y. Shih, Q. Xu,
2192 G. Swaminathan, D. Xiang, C. Eberhart and J. Hanes, A
2193 dense poly(ethylene glycol) coating improves penetration of
2194 large polymeric nanoparticles within brain tissue, *Sci Transl*
2195 *Med* **4** (2012), 149ra119.
- 2196 [248] E. Nance, C. Zhang, T.Y. Shih, Q. Xu, B.S. Schuster and J.
2197 Hanes, Brain-penetrating nanoparticles improve paclitaxel
2198 efficacy in malignant glioma following local administration,
2199 *ACS Nano* **8** (2014), 10655–64.
- 2200 [249] L.C.S. Erthal, O.L. Gobbo and E. Ruiz-Hernandez, Biocom-
2201patible copolymer formulations to treat glioblastoma multi-
2202forme, *Acta Biomater* **121** (2021), 89–102.
- 2203 [250] C. Bastiancich, A. Malfanti, V. Pr at and R. Rahman, Rati-
2204onally designed drug delivery systems for the local treatment
2205of resected glioblastoma, *Adv Drug Deliv Rev* **177** (2021),
2206113951.
- 2207 [251] C.T. Tsao, F.M. Kievit, A. Ravanpay, A.E. Erickson, M.C.
2208Jensen, R.G. Ellenbogen and M. Zhang, Thermoreversible
2209poly(ethylene glycol)-g-chitosan hydrogel as a therapeutic T
2210lymphocyte depot for localized glioblastoma immunotherapy,
2211*Biomacromolecules* **15** (2014), 2656–62.
- 2212 [252] C. He, H. Ding, J. Chen, Y. Ding, R. Yang, C. Hu, Y. An, D.
2213Liu, P. Liu, Q. Tang and Z. Zhang, Immunogenic cell death
2214induced by chemoradiotherapy of novel pH-sensitive cargo-
2215loaded polymersomes in glioblastoma, *Int J Nanomedicine*
2216**16** (2021), 7123–7135.
- 2217 [253] J. Zhang, C. Chen, A. Li, W. Jing, P. Sun, X. Huang, Y. Liu,
2218S. Zhang, W. Du, R. Zhang, Y. Liu, A. Gong, J. Wu and
2219X. Jiang, Immunostimulant hydrogel for the inhibition of
2220malignant glioma relapse post-resection, *Nat Nanotechnol* **16**
2221(2021), 538–548.
- 2222 [254] F.A. Bruinsmann, G. Richter Vaz, A. de Cristo Soares Alves,
2223T. Aguirre, A. Raffin Pohlmann, S. Stanis uski Guterres
2224and F. Sonvico, Nasal drug delivery of anticancer drugs for
2225the treatment of glioblastoma: Preclinical and clinical trials,
2226*Molecules* **24** (2019).
- 2227 [255] W. Wang, S. Swenson, H.Y. Cho, F.M. Hofman, A.H. Sch on-
2228thal and T.C. Chen, Efficient brain targeting and therapeutic
2229intracranial activity of bortezomib through intranasal co-
2230delivery with NEO100 in rodent glioblastoma models, *J Neuro-
2231surg* **132** (2019), 959–967.
- [256] H.H. Chung, M.J. Hsieh, Y.S. Hsieh, P.N. Chen, C.P. Ko,
N.Y. Yu, C.W. Lin and S.F. Yang, The Inhibitory Effects of
Terminalia catappa L. Extract on the Migration and Invasion
of Human Glioblastoma Multiforme Cells, *Pharmaceutics*
(Basel) **14** (2021).
- [257] V. Bordoni, L. Sanna, W. Lyu, E. Avitabile, S. Zoroddu,
S. Medici, D.J. Kelvin and L. Bagella, Silver nanoparti-
cles derived by artemisia arborescens reveal anticancer and
apoptosis-inducing effects, *Int J Mol Sci* **22** (2021).
- [258] X. Fei, A. Wang, D. Wang, X. Meng, J. Ma, L. Hong, R. Qin,
A. Wang, J. Dong, Q. Huang and Z. Wang, Establishment
of malignantly transformed dendritic cell line SU3-ihDCTC
induced by Glioma stem cells and study on its sensitivity to
resveratrol, *BMC Immunol* **19** (2018), 7.
- [259] B. Majc, M. Novak, N. Kopitar-Jerala, A. Jewett and B.
Breznik, Immunotherapy of glioblastoma: Current strategies
and challenges in tumor model development, *Cells* **10** (2021).
- [260] K. Kaur, P. Topchyan, A.K. Kozłowska, N. Ohanian, J. Chi-
ang, P.O. Maung, S.H. Park, M.W. Ko, C. Fang, I. Nishimura
and A. Jewett, Super-charged NK cells inhibit growth and
progression of stem-like/poorly differentiated oral tumors in
vivo in humanized BLT mice; effect on tumor differentiation
and response to chemotherapeutic drugs, *Oncimmunology* **7**
(2018), e1426518.
- [261] T.A. Cheema, H. Wakimoto, P.E. Fecci, J. Ning, T. Kuroda,
D.S. Jeyaretna, R.L. Martuza and S.D. Rabkin, Multifaceted
oncolytic virus therapy for glioblastoma in an immunocom-
petent cancer stem cell model, *Proc Natl Acad Sci USA* **110**
(2013), 12006–11.
- [262] F. Merz, F. Gaunitz, F. Dehghani, C. Renner, J. Meixens-
berger, A. Gutenber, A. Giese, K. Schopow, C. Hellwig, M.
Sch afer, M. Bauer, H. St ocker, G. Taucher-Scholz, M. Du-
rante and I. Bechmann, Organotypic slice cultures of human
glioblastoma reveal different susceptibilities to treatments,
Neuro Oncol **15** (2013), 670–81.
- [263] J.J. Parker, M. Lizarraga, A. Waziri and K.M. Foshay, A
human glioblastoma organotypic slice culture model for study
of tumor cell migration and patient-specific effects of anti-
invasive drugs, *J Vis Exp* (2017).
- [264] X. Jiang, Y.D. Seo, K.M. Sullivan and V.G. Pillarisetty, Es-
tablishment of slice cultures as a tool to study the cancer
immune microenvironment, *Methods Mol Biol* **1884** (2019),
283–295.
- [265] L. Ciraku, R.A. Moeller, E.M. Esquea, W.A. Gocal, E.J.
Hartsough, N.L. Simone, J.G. Jackson and M.J. Reginato, An
ex vivo brain slice model to study and target breast cancer
brain metastatic tumor growth, *J Vis Exp* (2021).
- [266] D. Truong, R. Fiorelli, E.S. Barrientos, E.L. Melendez, N.
Sanai, S. Mehta and M. Nikkha, A three-dimensional (3D)
organotypic microfluidic model for glioma stem cells – Vas-
cular interactions, *Biomaterials* **198** (2019), 63–77.
- [267] M.A. Koppens, G. Bounova, P. Cornelissen-Steijger, N. de
Vries, O.J. Sansom, L.F. Wessels and M. van Lohuizen, Large
variety in a panel of human colon cancer organoids in re-
sponse to EZH2 inhibition, *Oncotarget* **7** (2016), 69816–
69828.
- [268] J. Yu, B. Qin, A.M. Moyer, S. Newshean, T. Liu, S. Qin, Y.
Zhuang, D. Liu, S.W. Lu, K.R. Kalari, D.W. Visscher, J.A.
Copland, S.A. McLaughlin, A. Moreno-Aspitia, D.W. North-
felt, R.J. Gray, Z. Lou, V.J. Suman, R. Weinsilboum, J.C.
Boughey, M.P. Goetz and L. Wang, DNA methyltransferase
expression in triple-negative breast cancer predicts sensitivity
to decitabine, *J Clin Invest* **128** (2018), 2376–2388.
- [269] F. Jacob, R.D. Salinas, D.Y. Zhang, P.T.T. Nguyen, J.G.

- 2296 Schnoll, S.Z.H. Wong, R. Thokala, S. Sheikh, D. Saxena, S. Prokop, D.A. Liu, X. Qian, D. Petrov, T. Lucas, H.I. Chen, J.F. Dorsey, K.M. Christian, Z.A. Binder, M. Nasrallah, S. Brem, D.M. O'Rourke, G.L. Ming and H. Song, A patient-derived glioblastoma organoid model and biobank recapitulates inter- and intra-tumoral heterogeneity, *Cell* **180** (2020), 188–204e22..
- 2297
- 2298
- 2299
- 2300
- 2301
- 2302
- 2303 [270] M.J. Rybin, M.E. Ivan, N.G. Ayad and Z. Zeier, Organoid models of glioblastoma and their role in drug discovery, *Front Cell Neurosci* **15** (2021), 605255.
- 2304
- 2305
- 2306 [271] C. Zhang, M. Jin, J. Zhao, J. Chen and W. Jin, Organoid models of glioblastoma: advances, applications and challenges, *Am J Cancer Res* **10** (2020), 2242–2257.
- 2307
- 2308
- 2309 [272] R. Azzarelli, Organoid models of glioblastoma to study brain tumor stem cells, *Front Cell Dev Biol* **8** (2020), 220.
- 2310
- 2311 [273] A. Linkous, D. Balamatsias, M. Snuderl, L. Edwards, K. Miyaguchi, T. Milner, B. Reich, L. Cohen-Gould, A. Storaska, Y. Nakayama, E. Schenkein, R. Singhanian, S. Cirigliano, T. Magdeldin, Y. Lin, G. Nanjangud, K. Chadalavada, D. Pisapia, C. Liston and H.A. Fine, Modeling patient-derived glioblastoma with cerebral organoids, *Cell Rep* **26** (2019), 3203–3211e5..
- 2312
- 2313
- 2314
- 2315
- 2316
- 2317
- 2318 [274] J. Ogawa, G.M. Pao, M.N. Shokhirev and I.M. Verma, Glioblastoma model using human cerebral organoids, *Cell Rep* **23** (2018), 1220–1229.
- 2319
- 2320
- 2321 [275] F. Andreatta, G. Beccaceci, N. Fortuna, M. Celotti, D. De Felice, M. Lorenzoni, V. Foletto, S. Genovesi, J. Rubert and A. Alaimo, The organoid era permits the development of new applications to study glioblastoma, *Cancers (Basel)* **12** (2020).
- 2322
- 2323
- 2324
- 2325
- 2326 [276] T.G. Krieger, S.M. Tirier, J. Park, K. Jechow, T. Eisemann, H. Peterziel, P. Angel, R. Eils and C. Conrad, Modeling glioblastoma invasion using human brain organoids and single-cell transcriptomics, *Neuro Oncol* **22** (2020), 1138–1149.
- 2327
- 2328
- 2329
- 2330 [277] S. Bian, M. Repic, Z. Guo, A. Kavirayani, T. Burkard, J.A. Bagley, C. Krauditsch and J.A. Knoblich, Genetically engineered cerebral organoids model brain tumor formation, *Nat Methods* **15** (2018), 631–639.
- 2331
- 2332
- 2333
- 2334 [278] G. Goranci-Buzhala, A. Mariappan, E. Gabriel, A. Ramani, L. Ricci-Vitiani, M. Buccarelli, Q.G. D'Alessandris, R. Pallini and J. Gopalakrishnan, Rapid and efficient invasion assay of glioblastoma in human brain organoids, *Cell Rep* **31** (2020), 107738.
- 2335
- 2336
- 2337
- 2338
- 2339 [279] J.G. Camp, F. Badsha, M. Florio, S. Kanton, T. Gerber, M. Wilsch-Bräuninger, E. Lewitus, A. Sykes, W. Hevers, M. Lancaster, J.A. Knoblich, R. Lachmann, S. Pääbo, W.B. Huttner and B. Treutlein, Human cerebral organoids recapitulate gene expression programs of fetal neocortex development, *Proc Natl Acad Sci USA* **112** (2015), 15672–7.
- 2340
- 2341
- 2342
- 2343
- 2344
- 2345 [280] A.A. Pollen, A. Bhaduri, M.G. Andrews, T.J. Nowakowski, O.S. Meyerson, M.A. Mostajo-Radji, E. Di Lullo, B. Alvarado, M. Bedolli, M.L. Dougherty, I.T. Fiddes, Z.N. Kronenberg, J. Shuga, A.A. Leyrat, J.A. West, M. Bershteyn, C.B. Lowe, B.J. Pavlovic, S.R. Salama, D. Haussler, E.E. Eichler and A.R. Kriegstein, Establishing cerebral organoids as models of human-specific brain evolution, *Cell* **176** (2019), 743–756e17..
- 2346
- 2347
- 2348
- 2349
- 2350
- 2351
- 2352
- 2353 [281] S. Velasco, A.J. Kedaigle, S.K. Simmons, A. Nash, M. Rocha, G. Quadrato, B. Paulsen, L. Nguyen, X. Adiconis, A. Regev, J.Z. Levin and P. Arlotta, Individual brain organoids reproducibly form cell diversity of the human cerebral cortex, *Nature* **570** (2019), 523–527.
- 2354
- 2355
- 2356
- 2357
- 2358 [282] C.E. Brown, S. Bucktrout, L.H. Butterfield, O. Futer, E. Galanis, A. Hormigo, M. Lim, H. Okada, R. Prins, S.S. Marr and K. Tanner, The future of cancer immunotherapy for brain tumors: a collaborative workshop, *J Transl Med* **20** (2022), 236.
- 2359
- 2360
- 2361
- 2362
- 2363 [283] A.L. Johnson, J. Lathera and H. Lopez-Bertoni, Exploring glioblastoma stem cell heterogeneity: Immune microenvironment modulation and therapeutic opportunities, *Front Oncol* **12** (2022), 995498.
- 2364
- 2365
- 2366
- 2367 [284] X. Zhu, Y. Fang, Y. Chen, Y. Chen, W. Hong, W. Wei and J. Tu, Interaction of tumor-associated microglia/macrophages and cancer stem cells in glioma, *Life Sci* **320** (2023), 121558.
- 2368
- 2369
- 2370 [285] A. Wang, X. Dai, B. Cui, X. Fei, Y. Chen, J. Zhang, Q. Zhang, Y. Zhao, Z. Wang, H. Chen, Q. Lan, J. Dong and Q. Huang, Experimental research of host macrophage canceration induced by glioma stem progenitor cells, *Mol Med Rep* **11** (2015), 2435–42.
- 2371
- 2372
- 2373
- 2374