Proceedings of ICBioMed 2025 Symposium: AI for Healthcare: Advanced Medical Data Analytics and Smart Rehabilitation
DOI: 10.54254/2753-8818/2025.AU24769

Study on the Correlation Between the Microenvironment of Glioblastoma and CAR-T Cell Therapy

Guozhi Zhang

Clinical Medical College, Shaanxi University of Chinese Medicine, Xi'an, China
a1530544173@outlook.com

Abstract. Among brain tumors in adults, Glioblastoma (GBM) stands as the most frequently occurring and most aggressive form. GBM exhibits aggressive invasiveness and a poor survival prognosis. Without clinical intervention, the median survival duration remains approximately 6 months, whereas combined modalities (surgery, radiotherapy, and chemotherapy) prolong this to roughly 15 months. As a cutting-edge immunotherapeutic approach, chimeric antigen receptor T cell (CAR-T) therapy has shown significant efficacy in a variety of malignant tumors. However, the tumor microenvironment (TME) unique to GBM is rich in immunosuppressive factors, which severely limits the function and persistence of CAR-T cells. This article systematically reviews the bidirectional mechanism of CAR-T cells in the treatment of GBM, focusing on the effects of T cell exhaustion, PI3K/Akt/mTOR (PAM) signaling pathway dysregulation, M2 tumor-associated macrophages (TAMs) and immunosuppressive molecules. Studies have shown that T cell function exhaustion and PAM pathway inhibition inhibit the killing activity of CAR-T; M2 TAMs exacerbate immune escape by secreting anti-inflammatory factors; and cytokines released by CAR-T cells themselves enhance anti-tumor effects but may induce systemic inflammatory responses. This article provides a theoretical basis for optimizing GBM immunotherapy and proposes future research directions, encompassing the advancement of multi-target CAR-T, combined immunomodulators, and the application of single-cell sequencing and organoid models, in order to break through the key bottlenecks of CAR-T treatment for GBM and promote its clinical transformation.

Keywords: CAR-T, tumor microenvironment, glioblastoma, exhaustion, tumor-associated macrophages.

1. Introduction

Glioblastoma (GBM) represents the most prevalent and highly malignant brain tumor in adults. In the absence of clinical intervention, the average survival duration of patients is approximately 6 months, whereas multimodal treatments (including surgery, radiotherapy, and chemotherapy) can extend this to 15 months. GBM cells depend on a local microenvironment for growth, proliferation, and metastasis—the tumor microenvironment (TME)characterized by components such as the extracellular matrix and chemokines, both of which are intimately linked to the efficacy of chimeric antigen receptor T cell (CAR-T) therapy [1]. As a leading immunotherapeutic technology, this

© 2025 The Authors. This is an open access article distributed under the terms of the Creative Commons Attribution License 4.0 (https://creativecommons.org/licenses/by/4.0/).

DOI: 10.54254/2753-8818/2025.AU24769

immunotherapeutic approach has shown significant efficacy in treating specific malignant tumors. The core workflow entails isolating autologous T cells, genetically modifying them to express chimeric antigen receptors (CARs) for tumor antigen recognition and enhancing T cell cytotoxicity to selectively target and eradicate cancer cells [2]. Current research on CAR-T cell therapy within the GBM microenvironment focuses on their bidirectional interaction: on one hand, the GBM microenvironment harbors multiple immunosuppressive factors that impede CAR-T cell function; on the other hand, CAR-T cell therapy exerts reciprocal effects on the GBM microenvironment [3].

This review synthesizes and analyzes factors including T cell exhaustion, The signaling route known as PI3K/Akt/mTOR (PAM), and immunosuppressive factors, with the aim of elucidating the adaptability and bidirectional interactions of CAR-T cell therapy within the GBM microenvironment and offering novel insights for clinical GBM treatment.

2. CAR - t cell adaptation in the GBM microenvironment

The underlying principle of CAR-T cell therapy centers on the isolation of autologous T cells from patients. Subsequently, these cells are genetically engineered to express chimeric antigen receptors, conferring upon them the ability to selectively recognize tumor - associated antigens located on cancer cell surfaces. Nonetheless, the therapy's efficacy is impacted due to the innate heterogeneity of GBM and the distinctive traits of its microenvironment, resulting in variable degrees of influence.

Our research aims to investigate the adaptation mechanisms of engineered T cells in CAR-T cell therapy. We focus on the phenotypic, functional, and signaling pathway modifications these cells undergo within the GBM microenvironment, with the objective of offering fresh insights into the treatment. Here, we predominantly display how well CAR-T cell therapy can adapt to the microenvironment of GBM, highlighting its potential and challenges in this specific context.

2.1. Dynamic adjustments in phenotype and function

Experimental evidence demonstrates that upon infiltration into the GBM microenvironment, CAR-T cells exhibit substantial phenotypic alterations. A subset of CAR-T cells shifts from an effector memory T cell (TEM) phenotype to terminally differentiated effector T cells (TEFF) or exhausted T cells (TEX), a process termed T cell exhaustion—a pathological state induced by chronic antigen stimulation during viral infections and neoplastic diseases [4]. While TEFF exhibits potent cytotoxic activity, their short lifespan renders them prone to exhaustion; conversely, TEX cells overexpress multiple immune checkpoint molecules, leading to functional impairment and diminished antitumor efficacy [5]. Concurrently, CAR-T cell functionality is compromised, evidenced by diminished proliferative capacity, dysregulated cytokine secretion (e.g., decreased generation of proinflammatory cytokines like interferon-γ and TNF-α, and increased secretion of immunosuppressive cytokines like IL-10—collectively attenuating their cytolytic efficacy against GBM cells [6].

To raise the therapeutic effectiveness of CAR-T cell therapy for GBM, it is imperative to thoroughly explore the constantly changing phenotype and function of CAR - T cells within the GBM microenvironment. This exploration serves as a fundamental strategy for optimizing treatment outcomes.

DOI: 10.54254/2753-8818/2025.AU24769

2.2. Adaptive alterations in signaling pathways

CAR-T cell therapy encounters significant challenges in the treatment of GBM. To accommodate the hostile conditions of the GBM microenvironment, CAR-T cells undergo adaptive alterations in their intracellular signaling pathways. Notably, the PI3K/Akt/mTOR (PAM) signaling pathway is pivotal for metabolic regulation and functional maintenance in CAR-T cells [6]. In recent years, while numerous targeted therapies against the PAM signaling pathway have been developed, their clinical efficacy remains significantly limited. The underlying causes of this phenomenon are multifaceted. Such challenges involve the presence of the blood - brain barrier (BBB), a crucial structure that restricts the passage of substances, along with the natural and extensive heterogeneity within GBM tumors. Within the GBM microenvironment, the PAM signaling pathway is potentially suppressed by nutrient scarcity and immune signaling disruptions. Inhibition of this pathway disrupts metabolic reprogramming in CAR-T cells, compromising their survival and functionality, and ultimately diminishing treatment efficacy [1,7].

3. Interactions between CAR-T cell therapy and the GBM microenvironment

3.1. Inhibitory effects of the GBM microenvironment on CAR-T cell therapy

3.1.1. Immune cell-mediated inhibitory effects

Within the GBM microenvironment, tumor-associated macrophages (TAMs) serve a pivotal immunosuppressive role. TAMs can polarize into two distinct subtypes in response to different stimuli: classically activated M1 macrophages and selectively activated M2 macrophages. n the GBM microenvironment, TAMs predominantly polarize into the immunosuppressive M2 phenotype. M2 macrophages inhibit CAR-T cell proliferation, activation, and cytotoxicity by secreting anti-inflammatory cytokines like interleukin-10 (IL-10). Besides, numerous factors from particular macrophages play crucial roles in carcinogenesis and the growth of new blood vessels through various molecular routes. Figure 1 illustrates the functions of these macrophages in the tumor - surrounding environment, emphasizing their comprehensive effects on diverse tumor - related biological activities.

DOI: 10.54254/2753-8818/2025.AU24769

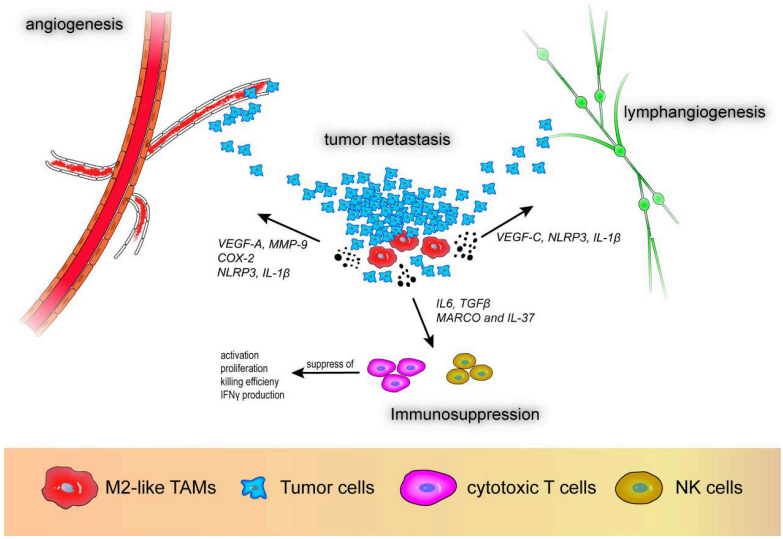

Figure 1: M2 – like TAMs are involved in driving angiogenesis, lymphatic vessel formation, immune evasion, and cancer metastasis, as indicated by the research in reference [4]

Tumor angiogenesis and metastasis are complex processes, and VEGF-A emerges as a key factor in facilitating the formation of blood vessels that tumors rely on. Research findings indicate that within non-small cell lung cancer (NSCLC), an elevated level of VEGF-A expressed by M2-like macrophages triggers angiogenesis, and this is closely associated with an unfavorable prognosis for patients.

From a cellular and molecular perspective, M2 - polarized TAMs exert a substantial influence on the formation of the TME. The secretory factors, namely heme oxygenase 1 (HO - 1) and COX - 2, released by these TAMs function as indispensable drivers that facilitate tumor progression. HO-1, a protein that responds to stress and is involved in oxidative-inflammatory processes, is regulated by an intricate network that includes IL-10 and Bach1. Due to this regulatory complexity, HO-1 represents a target with great potential for immunomodulation. When utilizing the LL2/OVA cancer cell line model, macrophages with high expressions of FAP-α and F4/80 display M2-like malignant features, and this is accompanied by increased HO-1 expression. Additional investigations have revealed that a high level of HO-1 expression results in tumor immunosuppression.COX-2 is also of equal importance in tumor progression, as it contributes to both inflammatory responses and the invasion and metastasis of tumors. When macrophages express COX-2, a series of downstream products, such as thromboxane A2, are generated. These products have a dual effect: on one side, they stimulate angiogenesis in solid tumors, thereby supplying adequate nutrients and creating pathways for the growth and spread of tumor cells; on the other side, they disrupt the normal functioning of immune cells and undermine the body's immune defense mechanisms. Significantly, COX-2 inhibitors have shown the ability to counteract these harmful effects, suggesting a possible therapeutic approach for cancer treatment. Apart from their role in angiogenesis and metastasis, the reducing immune activity function of M2-like macrophages significantly contributes to tumor malignancy. These macrophages are capable of suppressing the activity and multiplication and propagation of T cells, and also the propagation of natural killer (NK) cells. Studies show that certain stimuli make macrophages adopt a specific function. These macrophages create an anti - inflammatory setting and undermine the body's defenses against tumors. They hinder the activation and multiplication of immune cells that attack tumors, reducing the production of key immune molecules. They also suppress the function of other immune cells. Additionally, specific substances

DOI: 10.54254/2753-8818/2025.AU24769

can directly weaken the tumor - killing power of engineered immune cells and block their activation, allowing tumors to avoid detection and spread [4].

Myeloid-derived suppressor cells (MDSCs) are known for their strong immunosuppressive power, mainly targeting T cells. They use multiple mechanisms to suppress immunity, like through certain enzymes, cytokines, and by sequestering molecules or downregulating cell markers. There are two main subsets of MDSCs. One subset suppresses T cell responses either in a way that depends on the antigen or not, through producing nitric oxide and cytokines. The other subset mainly causes antigen - specific immune suppression by making T cells tolerant, using reactive oxygen species. Peroxynitrite directly inhibits T cells by modifying their receptors, reducing their response to antigen complexes. It also decreases the binding of antigens to tumor cells and disrupts T cell movement by modifying specific signaling molecules [8].

3.1.2. Inhibitory effects of immunosuppressive molecules and metabolic environment

The elevated expression of immunosuppressive molecules within the GBM microenvironment profoundly suppresses CAR-T cell functionality. As a member of the immunoglobulin superfamily, Programmed death 1 (PD - 1) is present on T cells, B cells, natural killer (NK) cells that have been activated, as well as on specific subpopulations of monocytes. PD-1 is critical for downregulating immune responses, suppressing T cell-mediated inflammation, and promoting self-tolerance. Upon binding of PD - 1 to its ligands, specifically, programmed cell death ligand 1 (PD - L1) and, separately, programmed cell death ligand 2 (PD - L2), it sparks a series of downstream signaling pathways that inhibit the activity of cytotoxic T lymphocytes (CTLs). These signaling events diminish T cell proliferation in lymph nodes and impair immune effector functions in peripheral tissues. Modulations in CTLs activity exert dual effects on host immune surveillance. On the one hand, moderate regulation of CTLs activity helps reduce the risk of immune responses against self-antigens, thereby decreasing the occurrence of autoimmune diseases; on the other hand, excessive inhibition of CTLs activation may provide favorable conditions for tumor cell growth, allowing tumor cells to escape host immune surveillance and thus promoting tumor development [9].

Moreover, the hypermetabolic nature of GBM cells results in nutrient scarcity and metabolic byproduct accumulation within the microenvironment. Hypoglycemic and hypoxic conditions impose an energy crisis on CAR-T cells, disrupting their metabolic processes and functional capacity. Concurrently, aerobic glycolysis in tumor cells generates lactic acid, acidifying the microenvironment and exacerbating impairments in CAR-T cell activity and proliferation [9,10].

3.2. The reciprocal effects of CAR-T cells on the GBM microenvironment

3.2.1. Microenvironmental changes induced by cytotoxic effects

CAR - T cells specifically identify antigens on the surfaces of glioblastoma multiforme (GBM) cells, which triggers their cytotoxic actions against tumor cells. When eliminating GBM cells, CAR - T cells discharge cytotoxic substances like perforin and granzyme. These molecules directly act on tumor cells, leading to their lysis. The demise of tumor cells sets off a series of immune reactions. Tumor - associated antigens (TAAs) are released, then internalized, processed, and presented by antigen - presenting cells (APCs). As a result, the body's endogenous immune system gets activated. This activation causes the recruitment of immune cells to the tumor location. Consequently, the cellular makeup and immune state within the GBM microenvironment are transformed [6].

DOI: 10.54254/2753-8818/2025.AU24769

3.2.2. Cytokine secretion regulates the immune microenvironment

During activation and tumor cell lysis, CAR-T cells secrete multiple cytokines that modulate the GBM immune microenvironment. IFN-γ, a key cytokine secreted by CAR-T cells, promotes M1 macrophage polarization to enhance phagocytic and cytolytic functions while suppressing immunosuppressive activity of M2 macrophages. IFN-γ further upregulates major histocompatibility complex molecule expression on GBM cells, enhancing tumor immunogenicity and facilitating immune recognition and elimination [7]. Additionally, cytokines like TNF-α secreted by CAR-T cells induce tumor apoptosis, recruit and activate immune cells, promote inflammation, and improve the GBM immune microenvironment to enhance antitumor immunity [6].

4. Conclusion

This review systematically investigates the mechanisms governing interactions between the TME and CAR-T cell therapy, focusing on immunotherapeutic challenges in GBM. Key findings indicate that T cell exhaustion within the TME, adaptive alterations in the PAM signaling pathway, immunosuppressive functions of M2 tumor-associated macrophages (TAMs), and elevated immunosuppressive molecules represent major barriers to CAR-T therapy efficacy. Specifically, T cell exhaustion impairs CAR-T cell cytotoxicity against tumor cells; the PAM pathway is suppressed in the microenvironment by immune signaling perturbations; M2 TAMs exacerbate immunosuppression through secretion of anti-inflammatory cytokines. Additionally, while cytokines released by CAR-T cells enhance antitumor activity, they may induce systemic inflammatory responses.

This study explains the dynamic interaction between the GBM immune microenvironment and CAR-T therapy, providing a theoretical basis for further optimizing CAR-T therapy. Current limitations include reliance on limited sample sizes, incomplete modeling of human TME heterogeneity, and unresolved questions regarding CAR-T cell penetration of the blood-brain barrier (BBB). Future directions may include developing multi-target CAR-T cells to counter antigen escape, combining immunomodulatory inhibitors for synergistic effects, and using single-cell sequencing and organoid models to dynamically characterize TME evolution.

References

[1] Liu G, Rui W, Zhao X, Lin X. (2021) Enhancing CAR-T cell efficacy in solid tumors by targeting the tumor microenvironment. Cell Mol Immunol, 18, 1085-1095.
[2] Zhang X, Zhu L, Zhang H, Chen S, Xiao Y. (2022) CAR-T Cell Therapy in Hematological Malignancies: Current Opportunities and Challenges. Front Immunol, 13, 927153.
[3] Dapash M, Hou D, Castro B, Lee-Chang C, Lesniak MS. (2021) The Interplay between Glioblastoma and Its Microenvironment. Cells, 10, 2257.
[4] Gao J, Liang Y, Wang L. (2022) Shaping Polarization Of Tumor-Associated Macrophages In Cancer Immunotherapy. Front Immunol, 13, 888713.
[5] Dimitri AJ, Baxter AE, Chen GM, Hopkins CR, Rouin GT, Huang H, Kong W, Holliday CH, Wiebking V, Bartoszek R, Drury S, Dalton K, Koucky OM, Chen Z, Giles JR, Dils AT, Jung IY, O'Connor R, Collins S, Everett JK, Amses K, Sherrill-Mix S, Chandra A, Goldman N, Vahedi G, Jadlowsky JK, Young RM, Melenhorst JJ, Maude SL, Levine BL, Frey NV, Berger SL, Grupp SA, Porter DL, Herbst F, Porteus MH, Carty SA, Bushman FD, Weber EW, Wherry EJ, Jordan MS, Fraietta JA. (2024) TET2 regulates early and late transitions in exhausted CD8+ T cell differentiation and limits CAR T cell function. Sci Adv, 10, 9371.
[6] Luksik AS, Yazigi E, Shah P, Jackson CM. (2023) CAR T Cell Therapy in Glioblastoma: Overcoming Challenges Related to Antigen Expression. Cancers (Basel), 15, 1414.

DOI: 10.54254/2753-8818/2025.AU24769

[7] Ramapriyan R, Caetano MS, Barsoumian HB, Mafra ACP, Zambalde EP, Menon H, Tsouko E, Welsh JW, Cortez MA. (2019) Altered cancer metabolism in mechanisms of immunotherapy resistance. Pharmacol Ther, 195, 162-171.
[8] Gabrilovich DI. (2017) Myeloid-Derived Suppressor Cells. Cancer Immunol Res, 5, 3-8.
[9] Rodriguez SMB, Tataranu LG, Kamel A, Turliuc S, Rizea RE, Dricu A. (2024) Glioblastoma and Immune Checkpoint Inhibitors: A Glance at Available Treatment Options and Future Directions. Int J Mol Sci, 25, 10765.
[10] Liang Qiaoyan, Yu Min. (2021) Metabolic changes in the tumor microenvironment and tumor immunotherapy. Chinese Journal of Oncology, 30, 459-464.