The Clinical Effects and Application of CAR-T Therapy in Cancer Biology

Baixi He[1, *, a, †], Chenfei Lv[2, *, b, †], Jingru Zhang[3, *, c, †]

[1]Molecular, Cellular and Developmental Biology, University of California, Santa Cruz, United States

[2]College of Life Sciences, Anhui Medical University, China

[3]Health and medical science (advanced), University of Adelaide, Australia

*Corresponding author: [a]Bhe43@ucsc.edu, [b]1913060030@stu.ahmu.edu.cn, [c]a1806870@student.Adelaide.edu.au

Keywords: CAR T therapy, adoptive cellular immunotherapy, cancer.

Abstract: From the public health perspective, the trend of cancer prevalence increased significantly every year, which is a sufficient reason for immunotherapy emergence and application. CAR T-cell is one of the emerging immunotherapies which have already made several effective achievements at this stage. This article aims to mainly illustrate the application of CAR T-cell therapy. In this article, five FDA-approved CAR T-cell therapies also some related information will be introduced. Based on the research, the consequences of these treatments all showed effectiveness to their target cancer based on the overall response rate, complete response rate, and duration of response evaluation.

1. Introduction

Immunotherapy is slowly overtaking other forms of traditional cancer treatments like chemotherapy and radiotherapy in the recent past. Immunotherapy is an innovative remedy that modulates the immune system to destroy cancer cells that exist in multiple directions and targets. Immunotherapy primarily works by enhancing the strength and regulating the microenvironment of the immune system. By doing this, the immune cells can effectively destroy tumor cells at various essential nodes. With the combination of standard anti-tumor therapy or numerous Immune-checkpoint inhibitors (ICI), most of the impacts will be vitally improved. However, the situation is still under further research [1]. In addition, the study of the connection between anti-tumor drugs is slowly growing, and the subsequent market and use of drugs are also on the rise.

Researchers in van den Bulk's lab claimed that the immune cells can identify and destroy tumors by using a method of stimulation of immune cells. In addition, the immune cells can use tumor-specific responses to inhibit the escape of the tumor. Also, immune cells check and clear tumors [1]. Cell immunotherapy is therefore efficient in hematologic tumors. Nonetheless, heterogeneity in the external microenvironment and solid tumors there reduces the efficacy of solid tumors.

Adoptive cellular immunotherapy (ACI) is a method of injecting immune effector cells that have been genetically modified and cultured in vitro into patients to kill tumor cells. This topic is becoming an extensive research area and a vital way of remedying tumors due to its easy preservation, strong specificity, and non-resistance to drugs. ACI is categorized into specific and non-specific [2]. Nonspecific ACI has immune cells that are made active by cytokines or lymphocytes existing in the peripheral blood. They pose the capability to destroy several tumors.

Nevertheless, they are only preferred for adjuvant therapy because of the week targeting and destroying capability [2]. Nonspecific ACI also has immunologic effector cells that include natural killer cells (NKC), dendritic cells (DC), cytokine-induced killer cells (CIK), lymphocyte-activated killer cells (LAK), tumor-infiltrating lymphocyte (TIL), and macrophages activated killer cells (MAK). Immune cells that belong to ACI are induced with the help of certain stimulating factors and exceptional tumor antigen stimulation [2]. The vital factors include Chimeric Antigen Receptor

T-Cell immunotherapy (CAR-T), T cell receptor-T (TCR-T). The primary effector cells include CD4+T cells and CD4+8 cells. Due to the difficulty in collecting and separating, TIL therapy is often used for melanoma. On the other hand, CAR-T therapy is mainly used for the treatment of blood tumors.

CAR-T therapy is an efficient adoptive cell therapy whose primary function is to extract the patient's body T cells by the leukocyte reduction procedure. Tan et al. reiterate that "It mainly extracts the patient's body T cells through leukocyte reduction procedures and transformed into the surface CAR-T by genetic engineering means, and then transferred to the tumor of patients to achieve tumor-specific killing." This type of therapy is mainly known for its strong remission rate for tumors that are known to express CD19 proteins like sizeable B-cell lymphoma and B-cell large leukemia. This review mainly focused on the clinical effects and application of CAR T-cell therapy.

2. Cancer Biology

Various changes in cell metabolism contribute to tumor transformation and progression. In other instances, metabolic phenotypes have the potential of being exploited to form image tumors and treat cancer after providing an appropriate prognosis. Therefore, it is also important to understand cancer metabolism, clinical oncology, and primary cancer pathphysiology when understanding cancer metabolism.

Research studies have unearthed the autonomous effects that influence the formation of cancer mutations. In this light, there have been numerous discovered principles concerning metabolic regulations and subsequent crosstalk occurring between signaling and metabolic networks. One pathway that has received much attention is aerobic attention [3]. It comprises the propensity that concerns proliferating cells such as cancer cells so that they respire and secrete carbon in the form of lactate even in the presence of oxygen. Furthermore, these articles explored the regulation of glycolysis principle that occur in cancer cells profoundly.

Experiments rule out the possibility of defective respiration to be the cause of malignant cells. Conversely, tumor growth requires mitochondrial activities and respiration [3]. Reaffirms that, for non-transformed cells, the Warburg effect is a phenomenon which is reversible and relevant to cell proliferation. This effect reports proliferation-related variation of metabolic and not only limited to malignant cells. Proliferating cells tend to express glycolytic enzymes and glucose transporters in more significant proportions than the machine responsible for pyruvate oxidization. Also, it must be consistent with no loss of respiration and the process of changing glucose to lactate [3]. The differentiation is vital since the tricarboxylic acid cycle generates intermediaries for nucleotides, lipids, and amino acids. These precursors do not only complement pathways and metabolites from glycolysis but also complement precursor metabolites.

Besides glucose, other forms of fuel also stimulate the growth of cancer cells in the form of biomass assimilation, energy formation, and redox control. Recently, scientists have realized that pathways and diversity of nutrients are also responsible for the above functions. Some of the nutrients needed for metabolism include branched-chain amino acids, fatty acids, lactate, serine, and glycine. Notably, oncogenic signaling imposes a strong dependence on particular nutrients. Cancer cells have an inclined tendency to compete for nutrients wherever they are few nutrients. June et al. opine that several types of cancer cells can cause autophagic degradation and scavenge macromolecules as their energy resources, which is highly efficient for cell replication. Noteworthy, only a few reprogrammed metabolic activities influence the multiplication of cancer cells.

3. Immune system

The development of tumor or cancer cells results from auto-unregulated immune mechanisms. Tumors do not grow instantly but grow from one step to the next since they involve an evolutionary process that alleviates cells from circumstances. Just as in Devil Facial Tumor Disease (DFTD), tumors can be contagious, in addition to being either malignant or benign. However, it is a paradox

to understand the role of immune cells concerning the development of tumors. Robert and Rosalie point out that tumors can craft protection for themselves while utilizing the host's immune system and other killing factors such as inflammation to support their growth [4]. Malignancy and angiogenesis will also be built up among these processes. If factors are kept constant, the immune system should be able to eradicate the tumor and subsequently maintain homeostasis [4]. However, some cells have genome plasticity that enables them to proliferate constantly to establish an equilibrium phase. This characteristic enables the host to escape the immune attack and install a clinically foreseeable tumor, then metastasis.

Regarding tumor development, immuno-escaping is the immune-corporation or immunosuppression, meaning tumor cells take advantage of the immunity to grow in strength and progress the development of cancer. After the initial development of the tumor cell and stromal, manipulate the activity of immune cells by modulation, thereby inducing metastasis, angiogenesis, intravasation, and suppressing antitumor response [5]. First, chemokine-regulated immune cells are recruited, then cultivated and later used to provide a suitable environment for the progression of the tumor. One of the most prominent features noticed in lung cancer cells was the suppression of adaptive and innate immune responses by human leukocyte antigens.

Importantly, tumor-specific antigens, which are exceptionally expressed by tumors, result from translocations or mutation that occurs to normal genes such as Ras, CDK4, and B-Catenin [6]. They also result when some proteins are overexpressed or invasion of viral antigens. Yang explains that. With regard to destruction of target by immune system's different branches [6], Major Histocompatibility molecules and antigens are presented as well as processed together on the surfaces which contains different antigen-presenting cells (dendritic cells, activated macrophages, and B cells). Therefore, APCs eliminate tumor neoantigens are cross presented to the adaptive immune system. Sometimes antigens escape immune cells by antigen tolerance or when immune responses are inefficiently stimulated. Impaired antigen presentation is responsible for escaping the adaptive immunity.

While the concept of immunotherapy traces back to 1893, it had experienced slow development until the recent past when advancements in the medical field went a notch higher. As a result, researchers are coming up with new ways of detecting ways in which cancer cells escape detection by the immune system. The success of clinical trials has led to the invention of cytotoxic T lymphocyte antigen-4 (CTLA-4), programmed death-1 (PD-1), and CAR T cells [6]. In addition, it was discovered that immune cells could also lead to the progression and growth of tumors. This mechanism of immune checkpoint therapy to block the immune system from progressing the growth of tumors is known as immunotherapy. Vital antibodies that help to prevent this occurrence are CTLA-4 and PD-1.

4. CAR-T therapy

4.1 CAR T-cell therapies application in cancer treatment

Normally the immune system is designed to identify foreign bodies by locating the antigens found on the exterior part of those cells [6]. The T-cells in the immune system possess receptors that attach to foreign antigens and assist in triggering the rest parts of the immune system to terminate the foreign antigen. For instance, cancer or tumor cells have antigens, but unfortunately, the immune system does not have the right receptor to fight the foreign antigen. Therefore, genetically engineered CAR T cells are specifically fashioned to detect cancer antigens called CD19 [7]. Once the CAR T-cells are introduced into an individual bloodstream, they amplify and find target tumor cells by releasing cytokines that destroy identified cancer cells.

1) Brexucabtagene autoleucel (Tecartus).

This is approved by the FDA for patients with relapsing mantle cell lymphoma. Though its approval is based on response durability and the overall rate of the response.

2) Axicabtagene ciloleucel (Yescarta)

This treatment applies among patients with reoccurring large B-cell lymphoma following more than two consecutive therapies [8]. It is also administered to patients with reoccurring follicular lymphoma after more than two systemic therapies. However, it is not indicated to be administered to patients with central nervous system lymphoma.

3) Tocilizumab (Actemra)

The treatment applies to pediatric patients (2 years) and adults. On the same note, patients with severe CRS symptoms can be treated using Actemra.

4) Lisocabtagene maraleucel (Breyanzi)

Patient suffering from relapsing large B-cell lymphoma with two or more systemic therapies qualify for this type of treatment. The treatment also includes diffuse large B cell lymphoma (DLBCL) [2]. However, it limits patients with the primary central nervous system from using this category of treatment.

5) Idecabtagene violence (Abecma)

Patients suffering from multiple myeloma after more than three or four therapy lines qualify for this treatment.

6) Tisagenlecleucel (Kymriah)

Patients 25 years of age with B-cell precursor acute lymphoblastic leukemia (ALL) receive this kind of treatment. It is also applicable to patients with large B-cell lymphoma following two or more systemic lines of therapy.

Table.1. Efficacy of FDA-approved CAR T cell therapy

CAR T cell	Name of the product	Directed By	Overall response rate for each targeted disease.
Brexucabtagene autoleucel	Tecartus	CD-19	87% for Relapsed/Refractory MCL
Lisocabtagene Maraleucel	Breyanzi	CD-19	73% for follicular lymphoma and relapsed or refractory large B-cell lymphoma
Idecabtagene Vicleucel	Abecma	B-cell maturation antigen	72% for refectory or relapsed multiple myeloma
Axicabtagene ciloleucel	Yescarta	CD-19	72% for Large B-cell and Follicular Lymphoma
Tisagenlecleucel	Kymriah	CD-19	83% for relapsed or refractory B-cell acute lymphoblastic leukemia, and recurrent or refractory Diffuse Large B-cell lymphoma

4.2 The effects of CAR-T therapy in clinical trials

One of the greatest benefits of CAR T cell immunotherapy is that it is a targeted therapy. The chimeric receptors monitor the targeted cancer cell population as well as multiply to promote immune memory for the treatment of lesions [2]. Immunotherapy is efficacy occurs for a longer period with a high level of accuracy targeting and specificity. CAR T cell therapy is broadly adaptable because of its ability to destroy and control multiple categories of tumors. When the therapy is done with thoroughness is can eradicate microscopic lesions and cancer cells from the body [2]. Therefore, using this approach of treating hematological malignancies stimulates the body's immune system that restores its functionality for a long time.

Even though it has a beneficial effect on a patient, there are several side effects of the therapy.

This is one of the greatest known side effects of CAR T cell therapy. For the T cells to function adequately, then cytokines are released to act as chemical messengers. The cytokines are released by the CAR T cells to recognize and destroy cell malignancies [9]. Some of the effects of CRS occur as mild symptoms, including headache, fatigue, nausea, chills. However, the effects can be more severe, leading to symptoms such as Cardiac failure, Cardiac arrest, Low blood pressure Multiple failure Cardiac arrhythmias Hypoxia (insufficient oxygen in tissue) Capillary seepage (further results

in low blood pressure due to leakage of fluids from minute blood vessel to other tissues) Reduced lung oxygenation Tachycardia Kidney problems (renal insufficiency) Hemophagocytic lymph histiocytosis.

In the treatment of some lymphomas and leukemia, patient risk of infection is increased using this therapy. CAR T cells are made to target CD19 proteins that are found on the surface of B cells. B cells, just like T cells are also other kinds of white blood cells that perform a fundamental part in protecting the body against infection [2]. When the CD 19 protein is destroyed by the CAR T cells, the B-cells have also destroyed; hence immunity of an individual goes down. Although this side effect is manageable using immunoglobulin therapy containing antibodies that fight infections caused by pathogens.

CAR T cells may cause problems to the brain. These neurological toxicities include confusion or disorientation, modified consciousness, seizure, speech problems, involuntary muscle twitching, and delirium [4]. However, these symptoms are reversible without long-term effects or intervention.

The immune response toward the CAR T cell therapy can be overwhelming. Symptoms connected to anaphylaxis include low blood pressure, facial swelling, and respiratory problems [7].

When CAR T cells destroy tumor cells, the rapid possibility of an increased uric acid level in the blood happens because when the cancer cells are broken down by the CAR T cells, they release a chemical known as uric acid into the bloodstream [2]. High uric acid levels may lead to renal inadequacy because the kidney cannot handle high uric acid levels.

This setback is almost connected with CRS severity. MAS occurs due to high multiplication and activation of macrophages and T-cells [3]. This condition is common among rheumatic patients or patients with chronic autoimmune diseases. This condition can be intervened by introducing monoclonal antibody tocilizumab. In cases of severity, anti- cytokine therapy or corticosteroid treatment options can be used for MAS.

Targeting and choosing antigens associated with tumors is a fundamental factor in making the therapy safe and successful. However, it is challenging to identify an ideal target. This is because most of the tumor antigens also express themselves similarly to healthy cells in organ tissue [4]. Since it is difficult to identify such cell destruction, healthy cells can pose life-threatening risks to an individual, especially when the destruction has occurred in vital organs such as the lungs, kidneys, heart, and lungs.

The administration of T-cell therapy has increased the specificity, accuracy, and target of immunotherapy. It has considerably increased the efficiency of immunotherapy regarding the long-term effect. This type of immunotherapy has wide adaptability given that the treatment can control and destroy various types of tumors. The administration of CAR T- cell is persistent since it initiates the body’s immunity to activate the immune function and consequently destroys cancer cells for a long time [2]. It is comprehensive because it can restore and enhance the body's immunity by exhaustively identifying, searching, and destroying cancer cells. Then, it prevents metastasis and tumor recurrence. It exhibits thoroughness when applied since it eliminates microscopic lesions and all residual tumor cells from the body. Last, the side effects of using this type of immunotherapy are less severe than the traditional treatments.

4.3 The Cost of CAR-T therapy

CAR T-cell immunotherapy is an expensive and complicated therapy for individuals who are entitled to the treatment. For instance, approximately EUR 320,000 is needed for a single patient [10]. This is an amount the manufacturers charge for the manufacture of CAR T cells. Apart from the production of the immune cells, the treatment is associated with the additional cost such as consultation fee before the therapy, chemotherapy to eliminate residual lymphocytes, and doctors’ visits, CT scans, and X-rays [10]. All these expenses amount to 400,000 dollars which is so expensive for any ordinary individual to afford to get treated. Surveys show that post-procedure and treatments are high, which is a financial burden. In a certain scenario, investigators assessed patients receiving treatment for B-cell lymphoma before the administration of the therapy and post-treatment outcomes of the therapy. It was found that the patient incurred costs amounting to almost 2 million

dollars [10]. Even though the procedure is effective, some patients will incur more costs if the malignancy relapses, requiring systemic therapy lines.

4.4 The Prognosis of CAR-T therapy

Prognosis refers to the approximate outcome or the likely cause of a disease. Regarding cancer, the prognosis of cancer patients means the chance that the condition will be remedied successfully, making the patient recover. Myriad factors can affect the prognosis of a cancer patient [11]. The most important reason is the location and type of cancer, grade of cancer, and stage of cancer. Also, other factors that influence prognosis are genetic and biological characteristics of tumors or cancer cells. The properties of cancer cells are often referred to as biomarkers and are mainly determined by laboratory experiments using imaging tests. Besides, the prognosis can be determined by the patient's age, overall health condition, and the patient's responsiveness to treatment.

Prognosis can often be predicted using statistics, making the treatment and diagnosis process easier for medical assistants. First, the doctor will consider the nature of the disease, underlying health problems, and the current treatment options. Then, the doctor will analyze how these factors can potentially affect the cause and nature of the disease. Most importantly, the doctor bases his diagnosis on the research that has been done over the past years on to particular disease to draw insightful decisions. The most popular statistics are cancer-specific survival, relative survival, overall survival, and disease-free survival. Immunotherapy using T-cell therapy has significantly improved the outcome of the treatment of “immunoinflammatory” and subsequently significantly increased the long- term survival rate—hence reduction in the mortality rates of cancer patients [2].

4.5 Cancer Treatable for T cell-related Treatment

For a long time, traditional methods such as chemotherapy and radiotherapy have been used to treat cancer before the innovation of the immune efficiency method. Healthcare professionals may use CAR T cell therapy if the traditional method fails or if the cancerous cells return. Under the approval of the FDA (Food and Drug Administration), several blood cancers can be treated using the immunotherapy method [7]. The following types of cancers can be treated using CAR T cell therapy. This includes mantle cell lymphoma occurring among the adults, follicular lymphoma, diffuse large B-cell lymphoma (DLBCL), B- cell acute lymphoblastic leukemia (ALL) among the children and young adult patients with 25 years of age and multiple myeloma. However, over CAR T cell clinical trials have been performed on over 600 types of cancer such as liver cancer, colon cancer, lung cancer, pancreatic cancer, ovarian cancer, acute lymphoblastic leukemia, and colorectal cancer, among other types of cancer.

5. The Challenges and Future Trends

To date, several hematological malignancies have been treated using the CAR T-cell approach. For instance, anti-CD19 CAR-T therapy's effectiveness in Phase 1 and 2 clinical studies for malignancies such as leukemia, neuroblastoma, and lymphoma look to be paving the way for FDA approval [5]. The CAR-T platform's advantages might be extended to a variety of other disorders. This is because these advancements are encouraging for these categories of diseases with significant morbidity and incidence. It is evident that CAR has the ability to treat other diseases. An explanation for this statement is that the antigen specificity does not only rely on the protein properties but also covers lipid, carbohydrates. The most important aspect about the CAR T cells is that it is the first therapy to be commercially approved by Food and Drug Administration in the United States [7]. Even though CAR T cell immunotherapy is highly connected to neurotoxicity, Cytokine Release Syndrome increased infection risk, and tumor lysis, FDA still approves the therapy with the intervention strategies and evaluation of risks assured. In this sense, the FDA ensures that physicians, specialists, and other health care providers undergo thorough training to manage the surging effects of the therapy.

Despite the achievements and contribution to solving oncological challenges, CAR–T cell immunotherapy faces challenges such as The low tumor heterogeneity and tumor antigens, targeting tumor-specific antigens, the problem of screening for the expression of the target antigen, Attempting to target numerous tumor antigens, Challenges relating to the enhancement of CART cells, Inefficient trafficking and minimal infiltration of various CART cells to various tumor sites

One of the setbacks of the therapy is the evaluation of the efficacy and safety of this therapeutic approach. Still, there are several drawbacks of handling solid tumors even though the CAR T-cell plays an important role in regulating cell malignancies [5]. Several studies have been done on how the CAR technology can be applied to manage type I human immune deficiency virus. Besides the continuing advances like cell manufacturing, genetic editing and engineering of the T-cells improve the hope of widening T-cell therapies. Moreover, advancement in oncology has enabled the feasibility of other treatment options. For example, pemphigus vulgaris, an autoimmune disease, uses CAR technology to develop CAR (chimeric autoantibody receptor), destroying autoimmune B-cells [5]. Moreover, CAR T cells for various autoantibody disorders appear to be a viable option that avoids some of the drawbacks of existing treatments, such as immune suppression and cancer CAR T cell therapies, such as target cell somatic mutations or cytokine release syndrome.

6. Conclusion

Due to the limitations of traditional cancer therapy, the emergence of CAR T-cell therapy provided more cancer patients with alternative options complementary to traditional therapies. Based on the analysis of CAR T-cell therapy, there are a variety of CAR T-cell related therapies that could effectively treat specific cancers such as lymphatic cancer and melanoma. However, there is still a large potential development space in the treatment of solid tumors using car T-cell technology. Moreover, the side effects of CAR T-cell therapy also need to be reasonably controlled.

References

[1] Wilkins, O., A. M. Keeler, and T. R. Flotte. "Car T-Cell Therapy: Progress and Prospects." Hum Gene Ther Methods 28.2, 2017, pp. 61-66.

[2] Tan, S., et al. "Cancer Immunotherapy: Pros, Cons and beyond." Biomedicine & Pharmacotherapy. Elsevier Masson Volume 124, 2020, p. 109821.

[3] Ma, S., et al. "Current Progress in Car-T Cell Therapy for Solid Tumors." Int J Biol Sci 15.12, 2019, pp. 2548-60.

[4] Sterner, R. C., and R. M. Sterner. "Car-T Cell Therapy: Current Limitations and Potential Strategies." Blood Cancer J 11.4, 2021, p. 69.

[5] Yang, Y. "Cancer Immunotherapy: Harnessing the Immune System to Battle Cancer." J Clin Invest 125.9, 2015, pp. 3335-3339.

[6] Sung, H., et al. "Global Cancer Statistics 2020: Globocan Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries." CA Cancer J Clin 71. 3, 2021, pp. 209-49.

[7] Miliotou, A. N., and L. C. Papadopoulou. "Car T-Cell Therapy: A New Era in Cancer Immunotherapy." Curr Pharm Biotechnol 19.1, 2018, pp. 5-18.

[8] Stern, L. A., V. D. Jonsson, and S. J. Priceman. "Car T Cell Therapy Progress and Challenges for Solid Tumors." Cancer Treat Res 180, 2020, pp. 297-326.

[9] June, C. H., et al. "Car T Cell Immunotherapy for Human Cancer." Science 359.6382, 2018, pp. 1361-65.

[10] Chadwick, Dara. "The Price of Hope: Weighing the Cost of Car-t Cell Therapy in Treating Blood Cancers." Curetoday. Hematology 2nd Special Issue 2020, Issue 2, Sept, 2020.

[11] Jindal, V., E. Arora, and S. Gupta. "Challenges and Prospects of Chimeric Antigen Receptor T Cell Therapy in Solid Tumors." Med Oncol 35.6, 2018, p. 87.