

Chimeric antigen receptors: Leading the next frontier of anti-cancer therapy

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Abstract

Background: T cell engineering has redefined personalized medicine as a novel facet of synthetic biology through its ability to individually harness and redirect the power of a patient's immune system to combat cancer.

Body: Chimeric Antigen Receptor (CAR) T cells, which possess synthetic receptors against CD19, a cell surface molecule found in most hematological malignancies, has yielded high remission rates and even cures in relapsed and/or refractory hematological malignancies, including Non-Hodgkin's lymphomas, chronic lymphocytic leukemia, and acute lymphoblastic leukemia. Although toxicities, such as cytokine release syndrome and neurotoxicity exist, recent developments have resulted in effective protocols to both manage and possibly prevent them through pharmaceutical methods and novel approaches to CAR delivery. The recent United States Food and Drug Administration approval of Novartis's Kymriah and Kite's Yescarta have justified the therapeutic successes of this technology in blood cancers, but efficacy in solid tumors remains limited. Several theories hope to explain this limitation with possible solutions entering clinical trials very soon.

Conclusion: As CARs have gained mainstream attention, more institutions worldwide are opening clinical trials against a vast array of malignant cancers, including GI, brain, pancreatic and lung, in hopes of tapping into their therapeutic potential to drive durable remissions.

Abbreviations: ALL: Acute Lymphoblastic Leukemia; CAR: Chimeric Antigen Receptor; CD: Cluster of Differentiation; CRS: Cytokine Release Syndrome; IL: interleukin; MSKCC: Memorial Sloan Kettering Cancer Center

Background and history

Scientists and physicians have amassed a greater understanding of the molecular mechanisms behind the pathogenesis of most cancers since the late 1800s, leading to the development of novel therapies to combat this detrimental malady. However, current medications are mostly limited to providing patients with respite in the early stages of disease progression and possess limited efficacy in the latter stages as more than 600,000 Americans continue to die from cancer each year [1]. As a result, the field of Chimeric Antigen Receptor (CAR) T cell immunotherapy was created and exemplifies the very definition of personalized medicine, where each patient's immune cells act as a living drug that can rev up the immune response to fight disease.

CAR T cells are a unique facet of modern day synthetic biology that redirect the function and specificity of T cells to combat disease, whereby upon binding to a cancer cell, they activate downstream T cell signaling pathways to yield cytotoxic killing through granzymes and perforins and a more efficacious immune response through cytokine release [2]. Initially comprised of an antibody-binding HLA-independent extracellular domain fused to an intracellular T cell signaling receptor (typically the zeta chain of the CD3 complex), first generation CARs exhibited limited efficacy due to their inability to produce sustained T cell responses [3]. Second generation CARs rectified this through the addition of a costimulatory domain to enhance proliferation, provide antiapoptotic function and enhance overall T cell function [4]. Several subsequent generations of CARs were later produced which differ in the number of co-stimulatory domains to induce T cell activation and an

enhanced antitumor response [5]. CD19 was chosen as the first target for CARs because of its prevalence in B-cell lymphomas and leukemias and higher overall expression compared to other cell surface markers [6]. Demonstrating efficacy in lymphoma and leukemia mouse models to eradicate disease after a single infusion, CARs were taken into clinical trials in the late 2000's. In this one-time treatment, a patient's T cells were harvested, expanded *in vivo* with antibody-coated beads, transduced with a retroviral vector bearing the CAR, reexpanded and subsequently reinfused into the patient post-conditional chemotherapy [7]. Physicians at the National Cancer Institute were the first to demonstrate clinical efficacy of this therapy in 2010, when they successfully induced regression of advanced lymphoma in one patient [8]. The following year, Memorial Sloan Kettering Cancer Center (MSKCC) was the first institution to demonstrate complete remission of relapsed or refractory B-cell Acute Lymphoblastic Leukemia (ALL) in their 8 patient Phase I clinical trial [9]. When this trial was expanded to 38 patients, the overall complete remission rate was 87% with a median time to complete remission lasting 23 days. Patients achieved complete bone marrow recovery within two months [10]. In 2012, the University of Pennsylvania's Children's Hospital demonstrated similar results for relapsed B-cell ALL as MSKCC when Emily Whitehead, a 7-year-old girl, achieved complete remission [11].

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Side effects & future solutions

Despite these promising results, CARs are not without their respective side effects. Cytokine Release Syndrome (CRS), defined by fever, hypotension, and respiratory insufficiency, arises when CARs stimulate the immune system via cytokines to induce an enhanced antitumor response [9]. Clinically, this has been controlled with Tocilizumab (IL-6 inhibitor) and high dose steroids if refractory, until researchers earlier this year showed that macrophages producing IL-1 cytokines are responsible for the symptoms and can be effectively managed with readily available IL-1 inhibitors, such as Anakinra [10,12]. Subsequent studies will aim to develop IL-1 receptor blocking CAR T cells to better control this toxicity. Neurological changes are another side effect that can present with delirium, global encephalopathy, aphasia, and seizures [10]. Although scientists are unsure of the exact cause for these toxicities, it has been speculated that they may arise from semi-random insertional mutagenesis when using a retroviral vector to transduce a patient's T cells [13]. Applying this therapy, researchers at MSKCC were able to successfully target a defined region of the genome using CRISPR/CAS9 to induce more uniform CAR expression with enhanced T cell potency in an ALL mouse model with minimal side effects [14]. Scientists are hopeful that this modification in CAR design will yield less complications and further enhance outcomes when it enters clinical trials.

Comparison of current CAR Products on the market

The promise of CAR T therapy became a reality in August 2017 when Novartis's CAR T product, Kymriah, for pediatric relapsed/refractory B-cell ALL became the first to receive FDA approval. Subsequently, Kite's Yescarta, now owned by Gilead, received FDA approval for the treatment of Diffuse Large B cell lymphoma in October 2017 [15]. Juno Therapeutics' Liso-cel, indicated for Diffuse Large B cell, Non-Hodgkin's, follicular, mantle cell, and primary mediastinal B cell lymphomas, will be submitted for FDA approval before the end of 2018 [15]. When compared over a 6-month time period, Liso-cel demonstrated a 50% complete remission with only 1% of patients experiencing severe CRS and 15% severe neurotoxicity in a cohort of 67 patients, while Yescarta showed a 36% complete remission in 101 patients with 13% experiencing CRS and 31% severe neurotoxicity [16]. Kymriah, in a cohort of 81 patients, showed a 30% complete response with 23% experiencing CRS and 12% severe neurotoxicity [16].

Application to solid tumors

Although CARs have demonstrated clinical efficacy in hematological malignancies, their utility has been limited in solid tumors. This may be due to a heterogeneous tumor microenvironment coupled with a lack of specific cell surface antigens on the tumor and an overall an inhospitable microenvironment for CAR T cells to act [17]. Additionally, solid tumors are known to exhibit antigenic escape, whereby the tumor can alter how much of a target molecule is expressed on the cell surface with potential for target loss over time [17]. These hurdles have required creative thinking to overcome them. Scientists at the University of Pennsylvania have proposed designing CARs that target the unique O-linked glycosylation sites exhibited by cancer cells, which act like a molecular signature [18,19]. Meanwhile, researchers at MSKCC have designed "Armored CARs", which express a co-stimulatory molecule with an IL-12 secretable cytokine. The benefits of armored CARs include enhanced cytotoxicity with resistance to the inhibitory tumor microenvironment, decreased exhaustion, enhanced trafficking to solid tumors and greater persistence past the current 3-month time period [20]. In another study, researchers developed armored CAR T cells targeting MUC-16, a marker found on some

ovarian cancers, co-expressing PD-1 checkpoint inhibiting antibodies, similar to Nivolumab and Pembrolizumab, resulted in longer CAR persistence, enhanced T cell activation, stronger anti-tumor responses, and overall improved longevity in mouse models giving greater credence to the potential of this new class of CARs to improve solid tumor outcomes [21].

Conclusion and perspectives

Despite these challenges, CAR T therapy is a novel treatment approach whose utility and prevalence will only continue to grow. Although this technology is still in its infancy, it has already demonstrated profound efficacy in inducing complete remission in certain hematological malignancies with a potential to offer treatments and possibly cures to many more. CAR T cell therapy is undergoing rapid worldwide expansion as more clinical targets are discovered and institutions and industry continue to open new clinical trials for such malignant cancers as glioblastomas, neuroblastomas, prostate, pancreatic, lung, liver and breast [22]. It remains to be seen whether CAR T cell therapy will circumvent standard approaches for treating certain cancers, but the future looks bright now that this potent new weapon exists in the war against cancer.

Declarations

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