CAR Evolution: Past, Present and Future

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Cell therapy based on a Chimeric Antigen Receptor (CAR) associated with immune cells, usually T lymphocytes, is known as CAR T-cell therapy. This therapy is revolutionizing the field of oncohaematology by directing the activity towards a specific target with the aim of eliminating it. However, there are still various limitations, such as a high percentage of relapse of the disease after this therapy, pathologies with a complex biology that hinders its use, long manufacturing times, and very high costs.

The activation of T cells triggered by antigen recognition by a chimeric receptor (HLA-independent recognition) was first described by Kuwana et al., at the Institute of Integral Medical Sciences of Aichi (Japan) in 1987 [1,2]. In 1989, a group of immunologists, including Zelig Eshhar and Gideon Gross, directed this research towards the field of oncology [3,4].

In 1991, Arthur Weiss of the University of California (San Francisco) reported that chimeric receptors containing the intracellular signalling domain of CD3ζ, activated T-cell signalling. This work resulted in the binding of the intracellular domains of CD3ζ to the chimeric receptors to transfer the activation signalling to the T cell [5].

Until then, the chimeric receptor (cTCR) was composed of the variable region domain of an antibody and the constant region domain of the TCR [1,2]. However, transduction had very low efficiency because two genes had to be introduced into the same lymphocyte using retroviral vectors to encode the chimeric receptor. In response to that question, in 1993, Zelig Eshhar, Gideon Gross et al., published the structure of a new chimeric receptor composed of a single-stranded variable fragment (scFv) derived from an antibody bound to the CD3ζ complex. Furthermore, the hypothesis of linking the chimeric receptor to other cells of the immune system such as Natural Killer (NK) lymphocytes is launched to explore its lytic activity [6].

Thus, was born the first T cell designed with the chimeric receptor structure of the products currently marketed, called “T body” [7]. In the mid-1990s, the first clinical trial was developed in patients with a first-generation CAR T (CD4 extracellular domain and CD3ζ intracellular domain) targeting HIV-infected cells [3]. However, the results of both this trial and other trials targeting solid tumours were not favourable as they did not have a robust response [8].

Between 1995-1998 several groups such as Sadelain et al., studied and introduced the role of co-stimulatory molecules in the amplification of lymphocyte activation and persistence of CAR T. Margo Roberts and Helene Finney were the first to develop a 2nd generation CAR T therapy (CD28-CARs). Also, Margo Roberts was the first to patent the concept of a co-stimulatory domain in the CAR construct in 1995 [3].

In the 2000s, this research led to the study of a CAR T-19 with the 4-1BB domain like co-stimulatory molecule and the first study on the efficacy of this therapy in mice with ALL published by Dr. Sadelain et al. [9]. These were the main foundation of today’s CAR T therapy [3,10,11].

The first clinical application of CAR T cells was performed in Rotterdam in 2005 for metastatic renal and ovarian carcinoma at the National Cancer Institute (NCI). In parallel, Brenner et al. studied third-generation CAR T consisting of two costimulatory molecules, CD28 and OX40 [12]. However, these studies did not achieve to demonstrate a therapeutic benefit, but warnings regarding the safety of its use have emerged [13]. Figure 1 shows the Evolution of the chimeric receptor structure.

Figure 1: Evolution of the chimeric receptor structure [1,7,14].
Indeed, the first truly favourable results came in 2010 from Steven Rosenberg at the NCI and Carl June and David Porter at the University of Pennsylvania, who used CAR T-19 in a patient with refractory follicular lymphoma and in patients with CLL (chronic lymphocytic leukaemia) and B-ALL, respectively [8].

This therapy experienced a revolution in 2012 in the case of a 7-year-old Emily Whitehead who had ALL refractory to all previous lines of treatment and became the first paediatric patient to receive this therapy. Dr. June’s team administered a CAR T-19 product, and the patient experienced severe side effects, predominantly a persistent fever with hemodynamic instability. These inflammatory characteristics were accompanied by very high IL-6 levels. The patient’s condition improved after administration of tocilizumab, an anti-IL6 monoclonal antibody recently approved by the FDA for childhood arthritis. The reaction described in these two patients is now called cytokine release syndrome (CRS) and is the most common adverse reaction [3,8,15].

Between 2013 and 2014, the first clinical trial results were published and it was not until 2017 that the Food and Drug Administration (FDA) approved Kymriah® (tisagenlecleucel) [16] for use in relapsed or refractory ALL after two prior lines of children and young adults up to 25 years old, and Yescarta® (axicabtagene ciloleucel) for relapsed mantle lymphoma [19], Breyzanix (lisocabtagene maraleucel) [20] or Abecma (idecabtagene vileucel) [21,22] and Carvykti (citacabtagene autoleucel) [9] as treatment for multiple myeloma in relapse or refractory to 2 or more lines of treatment.

However, the clinical outcomes in solid organ neoplasms as well as in T-lymphoid and myeloid neoplasms are poor. In solid organic neoplasms, the tumour microenvironment has been shown to play a critical role in tumour escape and resistance. In order to deal with CAR T cells with the ability to interact with microenvironment by producing cytokines (IL12, IL18 etc.) or CAR T cells that enhance their proliferation and activation by stimulating cytokines present in the tumour stroma are being studied. These CAR T cells are called TRUCKs or ‘fourth-generation CART cells and fifth-generation or next-generation CART cells’, respectively [23-25].

One of the barriers to overcome in T-lymphoid and myeloid neoplasms is the shared presence of tumour T cells and CAR T cells, which would lead to CAR T fratricide at the time of expansion. To avoid this, Li et al., designed an anti-CD7 CART, with a built-in command to retain its own CD7 in the endoplasmic reticulum so that it is not expressed on the membrane [26].

Another important limitation in the treatment of both T-lymphoid and myeloid neoplasms is the aplasia produced by a CAR T cell directed against antigens shared by T cells and healthy myeloid series and disease blasts. Allogeneic transplantation after CAR T cell therapy is the most studied option [26,27].

Although research in this field began in the 1980s, the boost came from the good results obtained in pivotal clinical trials of different CAR T-cell products in the field of oncohaematology. These scientific advances represent a historic moment in the treatment of cancer, as they make us dream of curing this disease with less toxicity soon.

**References**


