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Editor

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transformation by dysregulating the expression of key target genes.

- Epigenetic Therapy
- Fusion Genes
- BCR-ABL1

Chimeric T Cell Receptors

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Synonyms

Recombinant T cell receptor; Immunoreceptor; T-body

Definition

Chimeric T cell receptors are recombinant transmembrane receptor molecules, which are derived from the ►T cell receptor (TCR) complex and consist of an extracellular binding domain, a transmembrane domain, and an intracellular domain to initiate T cell activation upon engagement of the specific receptor ligand.

Characteristics

Recombinant TCRs provide the basis to engineer T cells with predefined specificity in order to redirect the ►T cell response toward defined target cells and to break tolerance for use in ►adoptive immunotherapy. Moreover, chimeric TCRs are valuable tools for the analysis of receptor driven activation of immune cells including T and NK cells. Several formats of recombinant TCRs have been designed during the last years, most of them are chimeric molecules composed of an extracellular binding and an intracellular signaling domain.

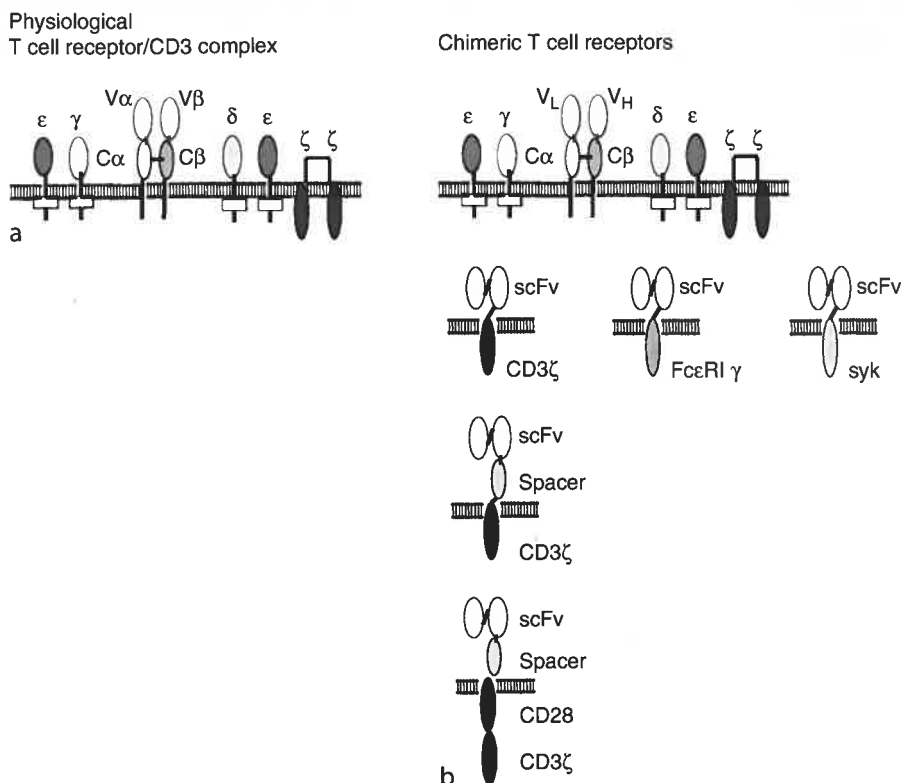
T cells exhibit their binding specificity via the TCR complex. For a number of applications in ►molecular therapy, T cells with predefined specificity would be desirable. By ►viral vector-mediated gene transfer, T cells can be grafted with a recombinant TCR of predefined specificity, which allows to redirect engineered T cells toward defined target cells. The approach, however, is hampered by the fact that the TCR of wanted specificity is frequently not available. In this situation, various formats of recombinant chimeric TCRs with predefined specificity were designed.

The TCR Format

Based on the similarity of the primary structure and the spatial conformation of the variable regions of immunoglobulin (Ig) and TCR α and β chain molecules, antibody derived binding regions V_H and V_L for antigen were grafted on to the constant domain of the TCR α and β chains, respectively (Fig. 1). Thereby, the TCR $V\alpha$ and $V\beta$ domains are replaced resulting in chimeric TCR molecules with antibody directed T cell recognition. The strategy, however, requires simultaneous expression of two modified TCR chains in engineered T cells. Moreover, the transfected TCR α and β chains frequently heterodimerize with the corresponding chains of the endogenous TCR resulting in chain mispairing and reduced expression of the recombinant TCR.

The Single Chain Format

The alternative format of a chimeric TCR (immunoreceptor, T-body) consists of one polypeptide chain which is composed of an extracellular binding and an intracellular signaling domain (Fig. 1). The single chain format avoids a number of difficulties that rise with the TCR format including simultaneous expression of the two recombinant TCR chains and mispairing with the endogenous TCR. The antigen binding domain is derived from a single chain fragment of variable regions (►scFv) antibody, the signaling moiety is preferentially derived from the CD3 ζ chain of the TCR/CD3 complex or, alternatively, from the γ chain of the high affinity IgE Fc receptor (Fc ϵ RI). The scFv moiety used for binding is generated from an antibody molecule by genetically joining the V_H and V_L immunoglobulin regions via a flexible peptide linker, e.g., (Gly₄Ser)₃, resulting in a continuous polypeptide chain of the V_H -linker- V_L or V_L -linker- V_H type. ►Phage display techniques are applied to isolate scFv antibodies with high binding affinity. Due to the antibody derived binding domain, chimeric immunoreceptors can be generated that bind antigen of any chemical composition or conformation including classical and nonclassical T cell targets like carbohydrates, as far as an antibody exists. Thereby, a broad variety of chimeric TCRs were generated against tumor associated antigens like ►HER-2/neu, or ►cancer germline antigens. Instead of an scFv domain, any polypeptide with specific binding properties, e.g., receptor ligands, may be suitable as targeting domain as well. The majority of chimeric TCRs of the immunoreceptor type harbor antibody derived binding domains that mediate T cell activation independently of MHC. However, antibodies are available that recognize the processed peptide in the context of MHC. MHC/peptide recognizing antibodies when integrated into a chimeric TCR mediate T cell recognition in a MHC dependent fashion. It is so far not elucidated whether MHC dependent or independent, chimeric TCRs are



Chimeric T Cell Receptors. Figure 1 Modular composition of chimeric T cell receptors (TCRs). The physiological TCR complex consists of the α and β chain for antigen recognition and the CD3 complex to initiate intracellular signaling. Fusion of the antibody derived binding domains V_H and V_L to the α and β chains of the TCR creates a chimeric TCR. In an alternative format the chimeric TCR consists of one polypeptide chain with the single chain fragment of variable region (scFv) antibody as antigen binding domain which is fused via spacer and transmembrane (TM) domain to the intracellular signaling domain which is preferentially derived from the CD3 ζ chain or the Fc ϵ RI γ chain, or alternatively from lck, syk, or CD3 ϵ . Chimeric TCRs of the second generation harbor in addition to the CD3 ζ chain a costimulatory domain, e.g., the CD28 domain.

superior in redirecting T cells *in vivo*. Some advantages of MHC independent chimeric receptors, however, are obvious, including targeting of unconventional T cell antigens like carbohydrates and targeting of cells even when their target antigen is not properly processed or presented in the MHC. Both CD8⁺ and CD4⁺ T cells can be redirected by the same immunoreceptor. The latter is of clinical relevance since redirected T cells of both CD4⁺ and CD8⁺ subpopulations execute granule-dependent cytotoxicity of target cells when stimulated via a chimeric TCR that circumvents MHC restriction.

The intracellular signaling domain of the chimeric TCR is most frequently derived from the CD3 ζ or the Fc ϵ RI γ chain. Both signaling chains contain immunoreceptor tyrosine activation motifs (ITAMs), the CD3 ζ chain three, the Fc ϵ RI γ chain one ITAM, which become phosphorylated upon receptor crosslinking. The signaling domains thereby serve as specific adaptors for downstream signaling proteins of the TCR complex. Alternatively, signaling moieties of

downstream kinases like lck can be used as activation domains in chimeric TCRs.

Expressed in cytotoxic T lymphocytes (CTLs), crosslinking of the chimeric receptor by antigen engagement initiates T cell activation resulting in proliferation, cytokine secretion, and specific cytotoxicity of antigen expressing target cells. Chimeric TCRs define a new specificity of the grafted effector cell. This is indicated by the fact that (i) T cells with immunoreceptor are activated upon coincubation with target cells that express the defined antigen on the cell surface whereas antigen-negative target cells do not initiate receptor signaling in receptor grafted T cells, (ii) T cells without immunoreceptor or with immunoreceptor of other specificity are not activated upon coincubation with antigen expressing target cells, and (iii) receptor triggered activation can be blocked by antibodies directed toward the scFv domain of the immunoreceptor. The efficacy of T cell activation depends on various parameters including the expression level of the

receptor on the effector cell surface, the density of antigen on the target cell, the targeted epitope and the position of the epitope within the antigen.

Chimeric T Cell Receptors with Primary and Costimulatory Signal

Naive T cells, in contrast to preactivated T cells, are not fully activated when stimulated via CD3 ζ but require ►costimulation via CD28. When T cells encounter antigen but lack costimulatory signals antigen-specific tolerance or anergy is induced. Current chimeric TCR strategies therefore aim to provide appropriate costimulatory signals on effector T cells in order to avoid induction of anergy. This is done by simultaneous expression of two recombinant immunoreceptors, one with the CD3 ζ and the other with the CD28 signaling domain or, technically more convenient, by combining the intracellular CD3 ζ chain together with the CD28 signaling domain into one polypeptide chain receptor molecule (Fig. 1). Different costimulatory domains result in different activation profiles. T cells triggered by the combined CD28-CD3 ζ signaling chimeric receptor exhibit increased IFN- γ secretion and secretion of substantial amounts of IL-2 whereas T cells equipped with the CD3 ζ chimeric receptor secrete lower amounts of IFN- γ and do not secrete IL-2. The specific cytolytic capacity, noteworthy, is not dramatically altered when preactivated T cells are stimulated via the CD28-CD3 ζ compared to the CD3 ζ signaling receptor.

Clinical Aspects

The major application of chimeric TCR strategy is to redirect cytotoxic T cells (CTLs) toward defined tumor cells or virus infected cells. T cells equipped with a tumor specific, chimeric TCR break tolerance toward autologous tumor cells as demonstrated in mouse models in vivo. Moreover, patient's T cells from the peripheral blood can be redirected in vitro toward the autologous tumor cells isolated from a tumor biopsy of the same patient. The approach is universal since by inserting the appropriate binding domain into the chimeric TCR, T cells can be redirected toward a broad variety of tumor associated antigens expressed on tumor cells of different entities, including ►gastrointestinal tumors and ►pancreas carcinoma (CEA, CA19-9, B72-4), ►melanoma (HMW-MAA, p97, GD3, Mage-1), breast carcinoma and ►epithelial tumors (►Her2/neu ErbB2, Muc1), renal cell carcinoma (G250), ►prostate carcinoma (PSMA), ►ovarian cancer (FBP), ►hematological malignancies including ►multiple myeloma and ►Hodgkin's lymphoma (CD19, CD20, CD30, CD33), and, moreover, toward virus infected cells, including hepatitis B virus infected ►hepatocellular carcinoma cells that express virus encoded proteins on the cell surface.

The strategy of redirecting T cells by chimeric TCRs has advantages over antibody-based therapies. In particular, the strategy makes use of the autologous cellular defense system represented by immunological effector cells that actively penetrate tissues. In contrast to antibodies, the engineered effector cells persist and circulate over long periods of time, i.e., until up to 1 year or even longer, amplify by proliferation upon antigen encounter and interact with numerous target cells by executing their cytolytic attack. Thereby, the chimeric TCR strategy is anticipated to be most powerful in eliminating (micro-) ►metastasis and ►circulating tumor cells. Since T cells in patients with advanced stages of the disease are frequently defective in TCR signaling, the chimeric receptor approach will require repeated administration of "freshly" grafted T cells to substitute burn-out or anergized CTLs. Once initiated the immune reaction is thought to be specifically sustained by chimeric receptor signaling upon repetitive stimulation with antigen. Having target cells eliminated, the immune reaction is supposed to be self-limiting since T cells that do not furthermore interact with their antigen enter apoptosis. It is furthermore anticipated that some engineered T cells convert into memory cells that will be active in ►immunoprevention of tumor relapse.

Clinical trials have been and are currently initiated using engineered T cells equipped with chimeric TCRs in order to eliminate tumor cells, e.g., of ►ovarian carcinoma, renal cell carcinoma, lymphoma, and melanoma. The safety of the immunotherapeutic strategies to redirect T cells by chimeric TCRs is currently explored. One of the major risks of redirecting T cells with chimeric TCRs, however, is the risk of autonomous amplification of engineered T cells independently of antigen. Secondly, there is the risk of an unwanted autoimmune reaction toward healthy tissues that express the same antigen as the target cell, however at lower levels. Costimulatory chimeric TCRs may harbor a higher risk of autoaggression compared to CD3 ζ signaling receptors. Crucial questions in the treatment of malignant diseases, however, are whether effector cells with chimeric TCRs have retained their homing capabilities after genetic manipulation in order to accumulate at the tumor site, whether they execute their effector functions at the tumor site and are resistant toward suppression by ►T regulatory cells, and whether they remain silent upon contact with healthy tissues with physiological expression of target antigen.

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Chimeric Transcripts

► Fusion Genes

Chinese Medicine, Traditional Chinese Medicine, Oriental Medicine

► Chinese *versus* Western Medicine

Chinese *versus* Western Medicine

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Synonyms

Chinese medicine, Traditional Chinese medicine, Oriental medicine; Western medicine, Modern medicine, Conventional medicine, Mainstream medicine, Orthodox medicine, Biomedicine, Allopathic medicine

Definition

Chinese medicine is a patient-oriented medical system that treats the ►cancer patients instead of the malignant diseases. It is believed that ►qi (the Chinese term for

vital energy) supports the functional activities and blood supplies nutriment for the whole body. There exists a system of channels within the human body, through which the vital energy and blood circulate, and by which the internal organs are connected with superficial organs and tissues, and the body is made an organic whole. Using these holistic and harmonic approaches, Chinese medicine emphasizes to strengthen the body resistance. It attaches importance to the self-healing ability of human body to remove pathogenic factors and recover health. Many of its cancer therapies, such as Chinese medication (including medicinal decoction, patent medicine, and proprietary medicine), medicated diet, acupuncture, and moxibustion, as well as ►qigong and massage, are employed for enhancing this power.

Western medicine is an evidence-based medical system, in which authorized medical and healthcare professionals struggle to avert the destruction of cancer patients by the unrelenting displacement of normal homeostasis accompanying neoplastic growth. The clinical problems faced by oncologists include overcoming the inherent or acquired resistance of the malignant cell to therapy, ameliorating the toxicities of aggressively applied therapies, as well as exploiting the synergistic potency of surgery, radiotherapy, and chemotherapy. In the last few decades, the limited clinical skills demand the use of destructive agent for cancer therapy. In the post-genomic era, ►targeted therapy and novel therapeutic strategy are applied to complement the conventional treatment for an achievement of optimal anticancer results.

Characteristics

Developing History

The Yellow Emperor's Canon of Internal Medicine is believed to be the earliest medical monograph in China, which appeared during the Warring States period (475–221 BC), first defined the etiology of tumor. Since the ancient times, Chinese medicine has made a great contribution to the health of the Asian people. Based on empirical and clinical experience, Chinese medicine has been systematized and theorized in complex practice. Many safe and effective methods have been developed to diagnose and treat cancer over the past thousands of years.

Hippocrates (460–375 BC), the father of Western medicine, first attributed the origin of cancer to natural causes. Improved microscopes, stimulated cancer researches, and important discoveries in human and animal studies have resulted in a better understanding of neoplasia. The last few decades has witnessed spectacular progress in describing the fundamental molecular basis of cancer following the advent of molecular biology and genetics, which allows the device of advanced or targeted therapy for cancer.