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Advancing Cell-Based Cancer Immunotherapy Through Stem Cell Engineering

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SUMMARY

Advances in cell-based therapy, particularly CAR-T cell therapy, have transformed the treatment of hematological malignancies. Although an important step forward for the field, autologous CAR-T therapies are hindered by high costs, manufacturing challenges, and limited efficacy against solid tumors. With ongoing progress in gene editing and culture techniques, engineered stem cells and their application in cell therapy are poised to address some of these challenges. Here, we review stem-cell-based immunotherapy approaches, stem cell sources, gene engineering

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AUTHOR CONTRIBUTIONS

Y.-R.L. and Z.S.D. wrote the manuscript, with the assistance from Y.Y. and M.L.. L.Y. and P.W. reviewed and edited the manuscript.

DECLARATION OF INTERESTS

Y.-R.L., Z.S.D., P.W., and L.Y. are inventors on patents relating to this manuscript. Y.Y. is currently an employee of the Fate Therapeutics. P.W. is a co-founder, stockholder, consultant, and advisory board member of HRain Biotechnology, TCRCure Biopharma, and Appia Bio. L.Y. is a scientific advisor to AlzChem and Amberstone Biosciences, and a co-founder, stockholder, and advisory board member of Appia Bio. None of the declared companies contributed to or directed any of the writing of this manuscript. The remaining authors declare no competing interests.

and manufacturing strategies, therapeutic platforms and clinical trials, as well as challenges and future directions for the field.

Short Summary:

Stem cell engineering and its application to cell therapy hold immense potential for the future of cancer treatments. In this review, Li et al. describe the most relevant stem cell sources, therapeutic platforms and clinical trials, evaluate gene engineering and manufacturing strategies, and discuss current challenges and future directions.

INTRODUCTION

Two unique properties of stem cells, their ability to self-renew and differentiate into multiple cell types, make them an attractive source for cell-based therapies. Stem cells and stem-cell-derived products have been investigated for diseases such as muscular dystrophy, heart disease, Parkinson's disease, Alzheimer's disease, spinal cord injuries, diabetes, and cancer [1]. Even though much work remains to be done, recent advances in stem cell engineering underscore the promise of a new generation of stem-cell-based therapies to alter the treatment landscape for several clinical indications. One such area is cancer, where therapies that rely on genetically engineered stem cells are beginning to enter the clinic and show encouraging signs of safety and efficacy.

Cell therapy, in the form of non-genetically modified hematopoietic stem cell transplantation (HSCT), has been a mainstay of blood cancer treatment for decades [2]. In the late 1900s, as our understanding and acceptance of the relationship between the immune system and cancer matured, investigators began utilizing immune cells to fight cancer[3]. Several clinical trials reported the application of *ex vivo* expanded tumor infiltrating lymphocytes and lymphokine-activated killer cells to treat end-stage solid tumors, and noteworthy clinical responses were observed [4,5]. Soon after, developments in gene therapy ushered in another form of cell therapy for cancer, centered on tailor-made, genetically modified immune cells [6]. Chimeric antigen receptor (CAR)-T cell therapy has transformed the treatment of hematological malignancies, with six products approved by the FDA so far. While instrumental in treating liquid cancers and propelling the field of cell therapy forward, the current CAR-T cell therapies face several limitations [7–11]. The products are autologous, obtained from the patient itself, which impedes their scalability, affordability, and accessibility. The vein-to-vein manufacturing process also prevents the administration of CAR-T cell therapy to patients with rapidly progressing disease, and individualized starting material coupled with an *ex vivo* manufacturing process can result in variable and suboptimal final products [12–17]. For example, patient-derived T cells and current expansion protocols can cause the therapeutic cells to enter a later differentiation state and express inhibitory receptors. A growing body of work contends that T cell fitness impacts clinical activity, with "younger," less differentiated, less exhausted T cells correlating with improved responses [18–24]. In addition to production and quality control challenges, CAR-T cell therapies can cause serious adverse events, namely cytokine release syndrome (CRS) and neurotoxicity [25–27]. Despite outstanding response rates, the majority of CAR-T cell recipients relapse [28–30]. Lastly, CAR-T cell therapies have often failed against solid tumors,

except for the anti-glioma activity of GD2-targeting CAR-T cells [31,32] and the success of Claudin-18.2 CAR-T in gastric cancers [33].

Allogeneic cell therapies have the potential to overcome some of the hurdles faced by autologous therapies but are not without challenges [34,35]. Many concerns about safety and potency are not readily addressed and will remain important considerations as allogeneic cell immunotherapies advance further. Healthy donor-derived cells can improve manufacturability and standardization of products. They provide readily administrable "off-the-shelf" cell therapies and can be pre-screened for desired characteristics. However, they also pose the risk of graft-versus-host-disease (GvHD) and are subject to host cell-mediated allorejection. GvHD is mediated by alloreactive donor T cells and can be avoided by TCR editing or the use of immune cells with known specificities or a lack of response to peptide-MHC mismatches [36–38]. Examples are virus-specific T cells, tumor-antigen-specific T cells, innate-like T cells, natural killer (NK) cells, and macrophages [39–42]. Through innate-like TCRs and/or NK receptors, gamma delta (γδ) T, invariant natural killer T (iNKT), mucosal-associated invariant T (MAIT), and NK cells exhibit intrinsic cancer cytotoxicity against many liquid and solid cancer types [43–46]. Equally, macrophages can display innate cancer phagocytosis [44,47]. Multiple tumor-targeting mechanisms are necessary for durable remission to oppose tumor antigen escape and antigen heterogeneity. Although it is possible to engineer dual-targeting CARs and other means of tumor recognition onto conventional T cells, the limits of current techniques for genomic alterations must be considered. Lentiviral and retroviral vectors remain the most common gene engineering methods. Given their relatively small gene payload capacity, the number of genes introduced in a single production round for mature immune cells is limited[48]. Between CARs, safety switches, immunomodulatory proteins, and other enhancements, selecting the optimal combination for transduction into mature immune cells is challenging and viral transduction and genomic editing affect cell yield and quality [49]. Potentially desirable immune cell populations, such as iNKT and γδ T cells, have low frequencies in peripheral blood, requiring extensive expansion for clinical usage. This expansion can introduce variability, limit manufacturability, and produce highly differentiated cell products.

Regardless of the allogeneic cell type, recognition and elimination by the host immune system may limit the persistence and therapeutic efficacy of allogeneic cells [50]. Interestingly, targeting B cell malignancies is a unique scenario, as B cell aplasia is a common sequelae of CD19 CAR-T cell treatment and thus humoral-mediated immunorejection of an allogeneic cell therapy will be diminished [51,52]. Cord blood-derived, HLA-mismatched NK cells transduced to express CD19 CAR and IL-15 were administered to 11 patients with CD19-positive lymphoid tumors and resulted in a 73% response rate [53]. The infused CAR-NK cells expanded and persisted at low levels for at least 12 months [53]. This long-term persistence was attributed to a permissive environment created by the lymphodepleting regimen combined with the ectopic expression of IL-15 by the CAR-NK cells [53]. However, the presence of therapeutic cells was not sufficient to prevent relapse [53]. Particularly for treating solid tumors, the rational design of allogeneic cells resistant to host rejection may be necessary to create a therapeutic window. Immunoevasion can be achieved by HLA "cloaking," in which HLA genes are inactivated using gene editing techniques [54]. Cells

that lack HLA, however, will be subject to NK "missing self-recognition," and will require addition genetic modifications, such as the overexpression of HLA-E, to resist NK killing [55].

Despite rapid progress in the development of cell therapies, the question remains: can we produce off-the-shelf, safe, scalable cell products that lead to durable responses in cancers, including solid tumors? In this review, we discuss the potential of stem-cell-based cell therapies to achieve this lofty goal. We start by reviewing stem cell sources, engineering strategies, and manufacturing details, and then examine stem cell engineering challenges, solutions, and optimizations as well as current stem-cell-based therapeutic platforms and clinical trials.

Stem Cell Sources

The challenges faced by autologous therapy led us to consider alternative master stocks as therapeutic cell sources. Two major stem cell resources, hematopoietic stem cells (HSCs) and pluripotent stem cells (PSCs), have been used to develop therapeutic immune cells and could provide a sustained supply and bypass the unavailability of autologous cell materials [56,57]. Compared to HSCs, PSCs could be utilized as an "unlimited" cell source to derive immune cells for the generation of therapeutic products. However, generating cells from human PSCs has typically been less efficient[58]. Several strategies, such as co-culturing PSCs with stromal cells (e.g., S17 or OP9 cells) and producing embryoid bodies, can improve PSC differentiation and immune cell production [59–62]. In addition, healthy donor periphery blood mononuclear cell (PBMC)-derived immune cells, such as conventional αβ T, iNKT, MAIT, γδ T, and NK cells, could be reprogrammed to pluripotency and then re-differentiated into rejuvenated immune cells [63–68]. These PBMC-derived induced PSCs (iPSCs) can be further engineered with CARs for enhanced antitumor efficiency. However, it is necessary to explore the usage of the endogenous promoter to drive transgene expression in PBMC-derived iPSCs, as the endogenous regulatory elements could still be activated in iPSCs, thereby affecting differentiation. Inducible expression systems could be incorporated to address this concern [58].

Stem-Cell-Derived Therapeutic Immune Cells

The transformative success of immune checkpoint inhibitors and CAR-T cells has reinforced the importance of T cells in cancer immunotherapy. It has further made the creation of stem-cell-derived T cells a primary focus in the field of stem cell engineering. Genetic engineering of autologous conventional αβ T cells to generate tumor-targeting T cells has been pursued for over two decades. Two categories of tumor antigen-specific receptors are applied to grant T cell specificity: physiological TCRs and synthetic chimeric antigen receptors CARs. CD19 CAR- and BCMA CAR-engineered T cells have been approved by the FDA to treat B-cell malignancies and multiple myeloma, respectively [15,69–73]. TCR-engineered T cells have shown promise in the treatment of melanoma, lung cancer, sarcoma, and multiple myeloma [74,75]. Nevertheless, several drawbacks exist for these approaches that may be addressed by stem-cell-derived products. Stem-cell-derived T cells have alternate cytokine profiles, which may reduce CRS and other safety risks [76]. Additionally, the composition of autologous T cells, such as the $CD4^+$:$CD8^+$ ratio, vary greatly between patients and may affect therapeutic outcomes [15,24]. The heterogeneity of starting materials

for CAR-T cell manufacturing may impede therapy efficacy. This issue could be resolved by adjusting a specific ratio/dose of $CD4^+$:$CD8^+$ cells [77,78] or by using allogeneic sources such as HSC or iPSC-derived off-the-shelf T cells. In these sources, theoretically, $CD4^+$:$CD8^+$ T cells can be tuned and additional genetic engineering performed on the small numbers of starting stem cells (Figure 1A). Notably, it is challenging to derive $CD4^+$ cells from current protocols that differentiate stem cells into T cell lineages [79–82]. One possible reason is that current stem cell differentiation systems utilize Notch signaling to induce T lineage development. Notch biases the CD4/CD8 T lineage decision, favoring CD8 over CD4 T cells [83–85].

NK cells, potent innate cytotoxic lymphocytes, effectively target virus-infected cells and cancer cells [45]. NK cell-based clinical trials attempt to treat cancers using a variety of NK cell sources, including NK cell line NK92, autologous patient-derived NK, allogeneic healthy donor-derived NK, and HSC and iPSC-derived NK cells (Figure 1A) [40,53,86–96]. Unlike conventional αβ T cells expressing rearranged TCRs, NK cells recognize target cells through the integration of signals from activating and inhibitory receptors [89,92,97,98]. NK cell recognition is independent of MHC restriction and prior sensitization, thereby free of GvHD risk [53,92,99]. The efficacy of NK cell-based immunotherapy could be potentially improved via multiple strategies, such as arming CARs on NK cells [45,86,89,100] engineering IL-2 or IL-15 to enhance NK cell antitumor activity and persistence *in vivo*[40,101], directing antibody-dependent cellular cytotoxicity (ADCC) through NK Fc receptor CD16 [102], and blocking NK inhibitory receptors such as NKG2A or killer cell immunoglobulin-like receptor (KIRs) [103].

Unconventional T cells, such as iNKT, MAIT, and γδ T cells, recognize tumor cells via TCRs and NK activating receptors, independent of MHC-restriction. These unique features allow innate T cells to target tumor cells without inducing GvHD. Generation of iNKT, MAIT and γδ cells through genetic engineering and differentiation of HSCs or iPSCs has been successful (Figure 1A). The resulting innate T cells respond to their agonist stimulation and display potent tumor killing abilities in leukemia, multiple myeloma and solid tumors [57,63,104–110]. These pre-clinical studies support the potential of developing off-the-shelf innate T cell-based cancer immunotherapy. Other innate immune cells, such as macrophages, dendritic cells (DCs) and myeloid cells, can also be generated from iPSCs (Figure 1A) [111–114]. These iPSC-derived innate immune cells display immunostimulatory function and may facilitate vaccination-based immunotherapy [112].

Stem Cell Engineering: Technologies and Manufacturing

Stem cell culture and differentiation—A variety of stem cell culture and differentiation systems have been developed in the past decades, as comprehensively described in earlier reviews (Figure 1B) [56,57]. These systems include humanized mouse models (e.g., bone marrow-liver-thymus mouse model) [106,107,115–117], feeder-dependent cultures (e.g., OP9-DL and artificial thymic organoid, ATO) [79–81], and feeder-free cultures (e.g., *Ex Vivo* HSC-iNKT culture) [105].

Considering the off-the-shelf purpose and safety profile of cellular products, the mouse origins of humanized mouse models severely limit their clinical application. Feeder-

dependent cultures containing mouse stromal cells have safety concerns which need to be addressed. One study reduced contamination by mouse feeder cells using a porous membrane, with feeder cells seeded at the bottom of the membrane and stem cells cultured on the other side of the membrane [118]. In addition, human-derived feeder cells such as foreskin fibroblasts, mesenchymal stem cells and adipose-derived stromal cells were utilized as feeders for human ESC and iPSC cultures [119–123]. Alternatively, feeder-free systems were developed to meet the clinically applicable and scalable requirements. For instance, an *ex vivo* feeder-free HSC-iNKT culture allowed to generate allogeneic iNKT cells with high yield and purity [105]. The feeder-free culture adopted a system of plate-bound DLL4 and VCAM-1 to induce T cell commitment from stem cells [124,125]. The generated allogeneic HSC-engineered iNKT cells displayed potent antitumor efficacy and antiviral capacity [105]. They could be further engineered with CARs to enhance their tumor targeting and gene-edited with CRISPR-Cas9 to ablate surface MHC molecules [79]. Another feeder-free differentiation culture system spanned from iPSC maintenance to T cell proliferation stages, enabling large-scale iPSC-derived T cell generation for cancer immunotherapy [126]. Further improvements on the feeder-free cultures will be necessary to achieve a more stable and efficient immune cell production.

Stem cell gene editing—The common premise of engineering immune cells for cancer immunotherapy is to grant immune cells the ability to specifically target tumor cells. TCRs and CARs are widely used in stem cell engineering to enhance immune cell specificity given their validation in mature PBMC T cells. (Figure 1C) [15]. Tumor antigen-specific TCRs are typically obtained from patient-derived, tumor-responsive T cell clones [15,127], humanized mouse models [128,129], or using phage display technology [130,131]. Unlike TCRs which require CD3 co-expression and are MHC-restricted, synthetic CARs function independently of MHC restriction and can be applied to other cells such as NK cells, macrophages and myeloid cells [45,114], enhancing tumor targeting capability [132]. In addition to directly engineering mature PBMC-derived or stem-cell-derived immune cells, retroviral or lentiviral vectors are used to stably introduce TCRs and CARs into stem cells (Figure 1D) [133]. These vectors represent a promising approach to generate long-lasting immune cells with defined antigen specificity [134,135].

Gene-editing technologies, including zinc finger nucleases (ZFNs), transcription activator-like effector nucleases (TALENs), and CRISPR-Cas9 have been utilized in cancer immunotherapy (Figure 1C) [36,38,136–139]. These technologies enable efficient gene knockout, site specific knock-in, and genome-wide screen in target cells, including immune cells and stem cells. Knock-out of TCR genes (e.g., *TRAC* and *TRBC*) avoids T cell-triggered GvHD, knock-out of MHC-related genes (e.g., *B2M* and *CIITA*) reduces host T cell-mediated allorejection, and knock-out of immune checkpoint genes (e.g., *PDCD1, LAG3, CTLA4*, and *DGKα*) improves immune cell antitumor efficacy (Figure 2) [38,137,140–150]. Knock-in of a CAR gene into the *TRAC* locus via CRISPR-Cas9 results in a uniform CAR expression on T cells, lack of endogenous TCR expression, and enhanced T cell potency [151,152]. In addition, CRISPR/Cas9-mediated genome-wide screening of immune and stem cells can help to identify gene targets for cell-based therapies [153–158]. Compared to engineering CAR/TCRs or gene editing in mature immune cells, gene editing in stem cells

could mean a reduction of required materials, such as lentivirus and CRISPR-Cas9/gRNA. It might also lead to higher gene editing efficiency [79,159].

Gene knock-outs or knock-ins that enhance the antitumor capacity of immune cells are being widely explored. For example, incorporating IL-15 into NK or iNKT cells improves their *in vivo* persistence and tumor killing [160–163]. Introducing HLA-E or HLA-G into MHC knock-out cells grants resistance to host NK cell-mediated allorejection [79,164,165]. Disrupting CD52 makes these cells resistant to lymphodepleting drugs such as alemtuzumab [26,166] (Figure 2).

Stem cell clinical manufacturing—Cell-based therapy has revolutionized cancer treatment. Unique features of cellular products, such as high variability, a long manufacturing process, low stability, complicated storage and transportation, insufficient characterization, and an unclear mechanism of action, make these cells different from other chemical drug or antibody products [167]. Therefore, a GMP-grade regulation is especially necessary for cell therapy products. In compliance with official standards, such as the United States Pharmacopoeia (USP) or the European Pharmacopoeia (EurPh), complete characterization of cellular products should be tested, including identity, yield, purity, viability and potency. In addition, tumorigenicity and biocompatibility testing should be performed, if necessary. Multiple aspects should be taken into consideration, such as cell origin (autologous versus allogeneic), safety, immunogenicity, *in vivo* efficacy, persistence, administration route, exposure duration, use of combination products and others. [168].

One concern of PSC-derived cell products is the presence of residual undifferentiated PSCs, which could develop into teratomas in the recipient patients [169]. Several preclinical studies have reported that once PSCs are differentiated into immune cells, few residual PSCs persist and form teratomas in animal studies. These results have been considered sufficient to demonstrate the safety of PSC-derived cellular products, and therefore these products have entered phase I clinical testing [170]. Another concern of PSC-derived cell products is immune matching and tolerance [169]. Different strategies are being pursued to resolve the issue of allorejection, and these are discussed below. Additionally, high costs and a time-consuming production process are other problems in manufacturing. Different from a vaccine or an antibody, generating human immune cells requires longer periods in a GMP environment, multiple culture and engineering steps, and specific sets of reagents and final formulations. This issue is further complicated, as each patient and condition might require a different number of cells for treatment [171–173]. Nevertheless, the rapid development of PSC-related knowledge and technology will provide more and less expensive alternatives, bypassing the current limitations in iPSC protocols.

The Engineered Stem Cell Product: Challenges and Optimizations

Safety—In addition to traditional CAR-T cell adverse events, such as CRS and neurotoxicity [174], stem-cell-derived therapies have other safety concerns that must be addressed [175]. The self-renewal property of stem cells, in particular pluripotent stem cells, raises concerns about tumorigenicity [176]. It is also possible that undifferentiated and/or immature cells are retained in the final cell product. Even a few residual PSCs could result

in teratoma formation [177]. In addition, tumorigenic mutations can arise during *in vitro* culture, which is prone to cause genetic alterations, such as chromosomal abnormality, copy number variation, and single nucleotide mutations [178]. Allogeneic stem-cell-based products are subject to several *in vitro* and *in vivo* tumorigenicity-associated tests [179]. Karyotyping is traditionally used to monitor chromosomal abnormalities [180], such as chromosomal deletion, duplication, or rearrangement. Clones with such alterations are discarded. Next generation sequencing can identify single nucleotide and other genomic alterations. The ultimate effect of these smaller genetic events on the final cell product remains controversial and further research will indicate whether these changes should preclude utilizing specific cell products [177]. Flow cytometry and quantitative RT-PCR/droplet digital PCR are also employed to detect stem cells in the final product [181]. In products that rely on the use of iPSCs, reprogramming factors are at risk to cause tumorigenesis [182]. The Yamanaka factors, especially c-Myc, are often overexpressed and known as driver mutations of human cancers. Although current practice utilizes Sendai virus transduction of reprogramming factors for episomal expression [183], care must be taken to ensure that the reprogramming factors are not integrated as transgenes prior to clinical development [181,183,184]. Lastly, suicide switches can be included in stem-cell-derived therapies to eliminate the cells in case of tumor formation [185].

Although iPSC technology may ultimately unlock the door to autologous cell therapies of any cell type, the cumbersome and costly process of iPSC reprogramming and differentiation currently prohibits widespread application of individualized iPSC precision medicine. Thus, stem-cell-derived therapies will likely be allogeneic. An important safety concern for allogeneic cell therapies, particularly T cell-based therapies, is GvHD [186]. Our understanding of GvHD stems largely from the longstanding use of allogeneic HSCTs to treat hematological diseases [187]. It remains a major cause of patient morbidity and mortality [188–190]. Therefore, conventional αβ T cell-based allogeneic therapies require genetic engineering to remove or alter the endogenous TCR. TCR KO is a common approach for creating universal allogeneic T cell therapies (such as UCART). Other methods, such as gene insertion into the T cell *TRAC* locus, can be used to eliminate the risk of GvHD [139,151]. Importantly, several alternative immune cell populations obviate the need for genetic manipulation to avoid GvHD. Germline-encoded NK receptors and the TCRs on innate T cells do not respond to peptide-MHC mismatches [56]. A growing number of clinical trials validate the safety of these allogenic cell types for cancer immunotherapy [191]. Further support for the safety of innate and innate-like immune cells comes from extensive research identifying cellular components of allo-HSCT grafts that reduce the risk and severity of GvHD without diminishing normal immunological functions, including NKand iNKT cells [192,193].

Immunogenicity—Immune rejection is a substantial hurdle to the successful application of allogeneic cell therapies. Immunosuppressants are traditionally used to prevent allograft rejection, for instance after organ transplantations. However, severe side effects, including infections, can result from continued immunosuppression [194,195]. When transplanted into immune-privileged tissues, such as ocular or neurological tissues, allogeneic stem-cell-derived cells persist for years [177]. For non-immune-privileged areas, and to avoid continuous

immunosuppression, two strategies can help to prevent immune rejection in the patient: HLA haplotype banks that store human stem cells of different haplotypes and the engineering of hypoimmunogenic immune cell products.

HLA haplotyping is performed prior to solid organ transplantation and widely used in allo-HSCT, with millions of donors registered in worldwide bone marrow banks [196]. With over 26,000 known HLA alleles reported for humans [197], creating stem cell biobanks that cover thousands of unique haplotypes is not feasible. However, being able to select specific HLA pairs can maximize population coverage with a minimal number of HLA haplotypes [198]. The key advantage of a haplobanking strategy is that it does not require genetic engineering. Some of the limitations are the need for unique banks for different ethnicities, a benefit for only part of the population, and incomplete immune tolerance, even with HLA-matching [177].

Notably, cells can be engineered to evade the immune system. Genetic deletion of beta 2-microglobulin (B2M) and class II major histocompatibility complex transactivator (CIITA) prevent the expression of MHC Class I and II molecules [199]. Cells with genetic B2M and CIITA knockouts resist $CD8^+$ and $CD4^+$ host T cell allorejection, respectively [199]. Cells lacking MHC Class I expression are subject to NK “non-self” elimination, and animal and clinical studies have reported a role for monocytes and macrophages in graft rejection [200]. Moreover, HLA Class I and II knockouts were paired with CD47 overexpression in iPSCs [201]. CD47 is the canonical “don’t eat me” signal, and effectively reduces NK and macrophage-mediated elimination. In fully immunocompetent preclinical mouse allogeneic recipients, endothelial cells, smooth muscle cells, and cardiomyocytes derived from mouse hypoimmunogenic iPSCs were well tolerated. Human $B2M^{-/-}CIITA^{-/-}$ CD47 iPSC did not incite any detectable cellular IFN-γ response or antibody response in NSG-SGM3 mice and showed long-term engraftment, whereas non-CD47 engineered hiPSCs were rejected. Alternative hypoimmunogenic cell therapies, in which iPSCs lack B2M, CIITA, and NK activating ligand CD155, and express HLA-E were also tested [202]. T cells differentiated from the hypoimmunogenic iPSC lines showed longer survival than unmodified iPSC-derived T cells in the presence of allogeneic immunity, and importantly, were more resistant to NK killing than HLA-edited iPSC-derived T cells. Hypoimmunogenic iPSC-derived T cells engineered to express CAR displayed potent antitumor efficacy in mouse models of CD20-expressing leukemia or lymphoma. These studies indicate that hypoimmunogenic stem-cell-derived therapies have the potential to produce off-the-shelf therapies that exert therapeutic benefits within reasonable dosing regimens.

Antitumor efficacy—The increasing body of clinical experience confirming the safety of cell-based cancer therapies encourages a focus on enhancing antitumor efficacy. Although these efficacy improvements are primarily developed for mature cells, progress in stem cell research will allow concurrent investigation of such innovations in stem-cell-derived products.

A critical barrier to the success of engineered cell therapies is tumor recognition. In hematological malignancies, ubiquitous lineage-specific markers have engendered the striking success of CAR-T cells targeting CD19 or BCMA [203]. Importantly, the elimination of all B cells, cancerous and healthy, can be treated with immunoglobin infusions, providing

an advantage over strategies that eliminate cells expressing solid-tumor-associated antigens. Other hematological markers, such as CD20, CD22, CD30, CD33, and CD7, are being investigated as CAR-T cell targets [204]. Although CD19 and BCMA CAR-T cells achieve response rates upwards of 90% for certain cancers, relapse is common [141]. One possible explanation for relapse after CAR19 CAR T cell therapy is modulation of the CD19 antigen on cancerous cells. For instance, genetic modification leading to partial or complete downregulation of the CD19 receptor, or truncation of the protein, which then prevents binding by CD19 CAR-T cells. A lineage switching that leads to the development of a CD19-negative phenotype can also result in relapse [205].

Identifying tumor-specific antigens for solid tumors is more challenging. Solid tumors are highly heterogenous and tumor-overexpressed antigens are often present on non-disposable healthy tissue. The first wave of CAR-T cells for solid tumors have targeted HER2, EGFRvIII, mesothelin, CAIX, PSMA, and GPC3 [206]. A fatal case of HER2-targeting CAR-T cells, potentially due to CAR-T cells attacking lung epithelial cells expressing low levels of HER2, tempered solid tumor CAR-T cell enthusiasm, but many clinical studies have reported tolerable safety profiles [207] and HER2 has since been targeted safely [208]. Although antigen escape and antigen heterogeneity are two critical immunotherapy evasion mechanisms of cancer cells, several cell engineering strategies are being implemented to create therapies that target cancer cells via multiple antigens/pathways [209]. Stem-cell-derived products are uniquely positioned to provide improved tumor recognition given their genetic pliability. Importantly, research into efficacy optimization, such as CAR design and the overexpression of immunomodulatory proteins, was first performed using mature immune cells. As such, in most cases, engineered stem-cell-derived products would incorporate enhancements originally designed for mature cells. Considerations that are particularly relevant for stem cell engineering remain to be fully elucidated. Among them are, for instance, the optimal time frame to introduce the CAR and other molecules (i.e., pre or post-differentiation), the influence of genetic alterations on stem cell differentiation, and how to design stem-cell-optimized CARs.

One way to prevent antigen escape is to target multiple tumor antigens. Dual targeting CARs have been reported and clinical trials, predominantly for liquid cancers using CD19xCD22 or CD19xCD20 CARs, are currently ongoing [25,210,211]. Importantly, the evaluation of costimulatory domains and the orientation of antigen recognition modalities is necessary to produce optimized dual-targeting synthetic constructs [212,213]. An example is the synthetic Notch (synNotch) receptor-engineered cell therapy for the treatment of cancer [214,215]. synNotch receptors induce transcriptional activation after recognizing user-specified antigens. They can be used in a highly modular fashion to customize cytokine secretion profiles, differentiation, and local delivery of non-native therapeutic payloads, such as antibodies. SynNotch cellular programming has managed effective and controlled tumor cell killing by targeting antigens that are homogeneous but not fully tumor-specific in glioblastoma [216].

The antigen sensitivity of engineered receptors can influence the control of tumor cells expressing low levels of antigen. Native TCRs initiate T cell activation after recognizing only a few peptide-MHC complexes, whereas CARs require a higher antigen load [217].

HLA-independent T (HIT) cell receptors have been specifically developed to target tumors with low antigen density [218]. In this study, the authors edited the heavy and light chains of a traditional CAR scFv into the TRAC locus in human peripheral blood T cells in place of TCRα and β variable chains, reconfiguring the TCR, which maintained its natural CD3 engagement, to the CAR scFv target. HIT receptors consistently afforded superior antigen sensitivity and tumor recognition than CD28-based CARs, the most sensitive CAR design to date. Whether the HIT receptor is applicable to stem-cell-derived T cells remains to be demonstrated.

ADCC is mediated by the Fc receptor CD16a on immune cells and is the key effector mechanism of therapeutic monoclonal antibodies [219]. Natural CD16a undergoes activation-induced surface cleavage and different CD16a alleles have a range of antibody binding affinities, which can limit the therapeutic benefit of CD16a expression [220,221]. Researchers incorporated high affinity, non-cleavable CD16a expression into PSC–derived NK cells and observed enhanced antitumor activity in combination with antibody therapy [96].

In addition to genetically engineered tumor cell recognition, several immune cell-intrinsic tumor-targeting mechanisms can be harnessed for anticancer therapy. Tumor antigen-specific T cells can be reprogrammed into iPSCs, although TCR-mediated tumor targeting of the differentiated final cell therapies will be HLA-restricted [222]. The natural TCRs of iNKT, MAIT, and γδ T cells endow these T cell subpopulations with innate tumor cytotoxicity [223]. Adoptive transfer of engineered and non-engineered iNKT and γδ T cells for the treatment of cancer are ongoing [161,224]. NK cells and macrophages also possess inherent antitumor activity and are promising cell types for adoptive cancer treatment [223].

Critical bottlenecks in the widespread clinical application of innate and innate-like immune cells are their scarcity, fecundity, and genetic pliability. Starting at the stem cell state mitigates these issues by exploiting stem cell engineering techniques and expansion potential. For example, a single cord blood donation can be expanded to generate upwards of ten thousand doses of HSC-derived iNKT (HSC-iNKT) cells that retain iNKT TCR functionality and tumor targeting, and further CAR engineering results in superior antitumor efficacy [79]. In 2019, Zeng et al. used an “NK cell-promoting” protocol to differentiate γδ T-iPSCs, which produced “γδ natural killer T cells” that were cytotoxic to a broad spectrum of cancers through γδ TCR and NK killing mechanisms [225]. CAR-engineered, PSC-derived macrophages also exhibit potent cancer cytotoxicity *in vitro* and *in vivo* [114]. Therefore, utilizing stem cells to produce unique cell populations can potentially expand the armamentarium for cancer treatment and may be instrumental in combatting tumor antigen heterogeneity and escape mechanisms.

Persistence—Their enhanced ability to expand and persist might endow less differentiated T cells with better antitumor effects compared to fully differentiated effector T cells [226,227]. Increasing the persistence of innate and innate-like immune cells and their stem-cell-derived counterparts may be especially important given their traditionally short lifespans. Several stem cell engineering strategies have been developed to improve long-term durability of these cells.

Autonomous cytokine secretion is one way to boost the persistence of engineered cell products. Transducing CAR-T cells to express IL-15, IL-12, IL-2, IL-18, and other cytokines promotes *in vitro* and *in vivo* survival [228]. IL-15 signaling is especially important for the maintenance of NK and innate-like T cells [229], and has been incorporated into NK, iNKT, and γδ T cell-based therapies [161,230,231]. IL-15 signaling can be potentiated by incorporating IL-15 secretion, membrane-bound IL-15 expression, and/or genetic modifications targeting the IL-15 pathway [232–234]. Genetic knockout of cytokine-inducible SH2-containing protein (CIS; encoded by the gene CISH), a negative regulator of IL-15 signaling, in iPSC-NK cells increased IL-15-mediated JAK-STAT signaling activity. $CISH^{-/-}$ iPSC-NK cells displayed improved *in vivo* persistence and inhibition of tumor progression in a leukemia xenograft model, which coincided with CISH KO-mediated metabolic fitness advantages [96]. Other transcription factors, such c-Jun and BATF [235,236], protect CAR-T cells from exhaustion and enhance persistence. Further research will be needed to assess the feasibility of these methods in stem cell products. Modifications to improve cell persistence and survival will have to be optimized for each stem cell product and effects on differentiation must be assessed. Thus far, IL-15 signaling modifications have not been reported to negatively influence differentiation, production, or phenotype of stem-cell-derived NK and iNKT cells.

Heterogeneity—Each PSC line differs from others in gene profiling, epigenetic status and differentiation propensity [177,237–239]. A comparison of the differentiation potential of 17 human ESC lines found that some lines exhibited > 100-fold differences in the propensity to differentiate into specific lineages [238]. Others reported distinct differentiation capacities of multiple PSC lines and indicated that PSC lines with lower differentiation potential exhibit an abnormal epigenetic status and are prone to teratoma formation [237,240]. The heterogenicity of PSC lines may limit the broad application of PSC-derived immune cell therapy. Researchers have developed approaches to convert primed into naïve PSCs to eliminate PSC heterogeneity. A combination of five kinase inhibitors was reported (i.e., inhibitors of MEK, GSK3, BRAF, ROCK, and SRC) that induces naïve ESCs [241]. Equally, short-term expression of NANOG and KLF2 reset the human pluripotent state, and the naive state of PSC cells is maintained in the presence of a protein kinase C (PKC) inhibitor [242]. Although these methods are promising, the potential loss of genetic integrity and imprinting in naïve PSCs needs to be carefully considered to avoid problems [243,244].

Therapeutic Platforms and Clinical Trials

iPSC-derived NK cells—The use of iPSC-derived NK cells is receiving increased interest. iPSCs are a renewable cell source that can be expanded indefinitely to produce homogenous NK cells, addressing the manufacturing and supply chain bottlenecks associated with primary NK cells. Preclinically, iPSC-NK cells have shown powerful antitumor functions against a variety of cancers in xenograft models [86,91,96]. Hermanson et al. showed that the antitumor activity of iPSC-NK cells against MA148 and A1847 ovarian tumor cells was as effective as primary NK cells [88]. In a representative study, non-KIR expressing NK cells derived from donor peripheral blood-iPSCs had greater cytotoxicity against ovarian cancer SKOV3, colorectal cancer SW480 and HCT-8, breast cancer MCF7, and head and neck cancer SCC-25 cells compared to primary NK cells [68]. Engineered

iPSC-NK cells that express CARs targeting CD19, CD33 or GPC3 demonstrated improved antitumor efficacies against $CD19^+$, $CD33^+$ or $GPC3^+$ tumor cells [93,245,246]. Further, the antigen-specific NK cell signaling and anti-tumor activity of iPSC-NK cells could be enhanced by utilizing a CAR containing the transmembrane domain of NKG2D [86]. iPSC-NK cells with a deletion of the IL-15 signaling regulatory protein CISH demonstrated an improved metabolic profile, increased expansion and persistence, and enhanced cytotoxicity in human AML xenograft tumor models [96].

Phase I clinical trials are underway for universal off-the-shelf iPSC-NK cell products, including several of Fate Therapeutics' iPSC-NK cell therapies (Clinicaltrials.gov Identifier NCT03841110, NCT04023071, NCT05182073, NCT04245722). Fate Therapeutics showed the safety and tolerability of allogeneic iPSC-NK cells in liquid and solid cancer patients[247], and has advanced to clinical investigations with engineered iPSC-NK cell products. iPSC-NK cells engineered with CD19-targeting CAR, high-affinity, non-cleavable CD16 Fc receptor, and IL-15/IL-15 receptor fusion promoting cytokine-autonomous persistence iPSC NK cell therapy, showed promising results for treating B-cell lymphoma [248], with 13 of 19 patients achieving an OR with a single dose of the cell therapy. In multiple myeloma patients, Fate Therapeutics' BCMA-targeting cell product resulted in 10 of 14 patients achieving objective responses. These trials provide clinical support for the high tolerability of allogeneic iPSC-NK cell therapies and show signs of antitumor efficacy.

Besides Fate Therapeutics, several companies are developing next-generation iPSC-NK cells for cancer treatment. Shoreline Biosciences generate CISH knock-out iPSC-NK cell products with increased durability and activity for use in hematologic and solid tumor contexts [96]. Century Therapeutics develop iPSC-NK cells with multiple targets such as CD19, CD19xCD79b, CD133xEGFR and Nectin-4 to treat B cell malignancies, glioblastoma, acute myeloid leukemia, and other solid tumors [249]. Overall, by being easily engineered, cultivated on a large scale, and adapted to diverse cancers with high safety, iPSC-NK cells have become a viable alternative to conventional CAR-T cells for cancer immunotherapy [29,98,114,250–252]. Nevertheless, *in vivo* persistence and viability of iPSC-NK cells, as well as their efficiency in conjunction with other immune checkpoint inhibitors, are still unclear and must be elucidated before iPSC-NK cell therapies can be widely used in the clinic.

iPSC-derived immune cells—In 2013, two Japanese research groups generated rejuvenated iPSC-derived, antigen-specific T cells [65,66]. Human HIV-1 or MART-1-specific $CD8^+$ T cells were reprogrammed to pluripotency by transducing retroviral vectors encoding OCT3/4, SOX2, KLF4, and c-MYC. The T-iPSCs were then redifferentiated into $CD8^+$ T cells, which displayed the same antigen-specific killing activity and TCR rearrangement pattern as the original $CD8^+$ T cell clone from the patient [64–66]. A safeguard system, inducible caspase-9, was introduced into the iPSCs to ameliorate the tumorigenic potential of undifferentiated iPSCs [253]. Using a similar technology, Wakao et al. reprogrammed human MAIT cells into iPSCs and redifferentiated the iPSCs to MAIT cells with antimycobacterial activity [67]. In the same year, Themeli et al. combined T-iPSC and CAR technologies to develop CD19 CAR-T to treat B cell malignancies [76]. These CD19 CAR-T cells displayed a phenotype closely resembling that of γδ T cells [76]. Following the T-iPSC

technology, several groups generated rejuvenated iNKT [63], NK cells [68], and dendritic cells [112] from reprogrammed iPSCs. Due to the unlimited availability of iPSCs, these technologies provide a valuable source of off-the-shelf, allogeneic cell products. Further research also confirmed the translational relevance of iPSC-based strategies. In 2018, Minagawa et al. utilized CRISPR/Cas9 to delete *RAG2* in T-iPSCs and prevent an unwanted TCR rearrangement, and then transduced the T-iPSCs with an antigen-specific TCR to endow T cells with tumor targeting capacity [254]. The resulting TCR-stabilized, regenerated cytotoxic T cells displayed an effective antitumor ability in xenograft cancer models [254]. In 2021, the same group reported a clinically applicable and scalable technology to regenerate T cells from iPSCs derived from an antigen-specific cytotoxic T cell clone, or from TCR-transduced iPSCs, as starting materials [126]. A feeder-free, serum-free differentiation culture protocol was also developed to achieve an efficient iPSC differentiation procedure that can be adapted towards clinical application [126]. Overall, these are promising approaches to generate large numbers of tumor-targeting immune cells for the study of T cell differentiation and potential clinical application.

iPSC-derived CAR-T cells—iPSC-derived CAR-T cells have the potential to be an infinite source of phenotypically defined, expandable, and functional CAR-T cells for off-the-shelf cancer therapy [255]. However, compared to iPSC-NK cells, the generation of iPSC-derived CAR-T cells has been challenging and typically requires a preexisting TCR which directs *in vitro* T cell differentiation [62,65,66,81,254]. In addition, T cell differentiation requires notch ligand engagement and can be impaired by CAR expression [76]. Recently, highly functional CAR T cells were generated through iPSC reprogramming from $CD62L^+$ naive and memory T cells, followed by CD19-CAR engineering and MS5-DLL4 stromal cell-dependent 3D-organoid system differentiation [256]. The primary $CD62L^+$ T cells have superior persistence and therapeutic potential in CAR-T cell treatment, and the pre-existing TCR induces T cell differentiation in a directed manner [227]. The resulting iPSC-derived CD19 CAR-T cells demonstrated conventional αβ T cell phenotypes, homogeneous TCR repertoire, and strong antitumor reactivity [256]. Another iPSC-derived CAR-T cell product was generated by combining histone methyltransferase EZH1 repression, stromal-free T cell differentiation from iPSCs, and CAR engineering [257]. Repression of EZH1 promotes *in vitro* differentiation and maturation of T cells derived from iPSCs. The mature iPSC-T cells are similar to peripheral blood αβ T cells in phenotype and functionality [257]. In addition, CAR-engineered iPSC-T cells showed enhanced cytokine production, potent cytotoxicity, and superior persistence in preclinical mouse models [257]. Overall, these iPSC-derived CAR-T cell platforms lay the groundwork for future efforts targeted at creating an infinite number of potent, allogeneic “off-the-shelf” CAR-T cells.

HSC-derived iNKT cells—iNKT cells are another potentially promising cell population for cancer immunotherapy. However, the low frequency and high variability of iNKT cells in humans limit their clinical applications [258]. To overcome these challenges, an HSC-iNKT cell platform was developed (Figure 3A). The first generation was based on autologous HSC genetically engineered to express the iNKT TCR that were transferred back into the patient, potentially providing therapeutic levels of iNKT cells for a lifetime (Figure 3B) [106]. However, this approach is expensive and difficult to deliver to all patients in need [37].

Subsequent development of the HSC-iNKT cell platform has centered on producing off-the-shelf HSC-iNKT cells and it was recently shown that HSC-engineering followed by *in vitro* differentiation resulted in allogeneic HSC-iNKT cells with high yield and purity (Figure 3C and 3D) [79,259]. Allogeneic HSC-iNKT cells have innate cancer-killing capacity and low risk of GvHD and, to enhance their therapeutic potential, these cells can be engineered to express CARs and undergo ablations of HLA Class I and II [79]. Furthermore, HSC-iNKT cells can modulate the tumor microenvironment, as they can effectively target and eliminate tumor-associated macrophages and other immunosuppressive cells [105,260,261].

CONCLUSION

The remarkable success of CAR-T cells in treating hematological malignancies has ignited the field of cell therapy. In 2012, at the age of 6 years old, Emily Whitehead was the first patient to receive CAR-T cells for acute lymphoblastic leukemia and recently celebrated ten years of cancer free survival. By all evidence, she is cured [262]. Our goal is to make Emily's story a reality for all cancer patients. However, CAR-T cell therapies are limited by their autologous nature, suboptimal long-term efficacy for many patients, and a lack of potency in solid tumors. Off-the-shelf cell therapies that overcome tumor antigen plasticity, heterogeneity and the immunosuppressive tumor microenvironment can advance the current cell therapy paradigm. Work to date suggests that many immune cell subtypes, such as innate-like T cells, NK cells, and macrophages, might be useful in cancer therapy. The benefits of applying unconventional T and innate immune cells are twofold: they have intrinsic antitumor capabilities and pose little risk of causing GvHD. Unfortunately, isolation, gene engineering, and expansion of mature immune cells still represent several bottlenecks in the development of these therapies. Maintaining the desired cell phenotype during expansion, generating sufficient cell numbers for multiple doses, and performing extensive genetic manipulation are all remaining challenges.

Herein lies the allure of stem cell engineering, which promises scalable, easily modifiable and homogenous cell products that are optimized for safety and efficacy. As of yet, hypoimmunogenic, persistent, and potent stem-cell-derived therapies are under development. Despite this promise, several outstanding questions still need to be addressed. It was recently shown the iPSC-derived CAR-T cells exhibit reduced CAR expression compared to PBMC-derived CAR-T cells [256]. In addition, previous work indicated αβ iPSC-derived CAR-T cells are closely related to γδ T cells based on gene expression analysis [76]. It is noteworthy that multiple stem cell engineering and differentiation protocols have been documented, which have the potential to yield considerable variability in the resultant cell products, even when such products are classified as being of the same cellular subtype. We also do not know whether cells created *in vitro* are comparable to their naturally occurring counterparts. Stem-cell-derived products might be more affected by changes in gene regulation, such as epigenetic silencing. Importantly, most of the engineering and optimization strategies applied to stem cells were developed in mature T cells. Are there any modifications, such as different CAR designs or cytokine secretion patterns, that could enhance specific features of stem-cell-derived products? It will also be important to assess whether stem cell engineering influences the differentiation potential, identity and function of the final product. CARs, for example, can cause tonic signaling, which could impact stem

cell differentiation. Leveraging cutting-edge technologies, such as multi-omics, CRISPR screens, and automated culture systems, will enable researchers to better understand the underlying mechanisms of engineered stem cell functionality and perform high throughput screens to optimize the potential of stem cell therapies [263,264].

Given the complexity of diseases such as cancer, combination therapies will likely be necessary to achieve durable therapeutic benefits [265]. Engineered stem cell products, with their off-the-shelf nature, hold promise as a component of treatment regimens that include established cancer treatments, such as surgery, radiation, chemotherapy, and targeted therapies, as well as emerging cancer immunotherapies, such as immune checkpoint inhibitors, oncolytic viruses, cancer vaccines, and adoptive cellular therapies [15,57]. The versatility of engineered stem cell products and their ability to be tailored and scaled for specific cancer types make them a promising addition to the oncology arsenal, with the potential to enhance treatment outcomes and improve patients' quality of life. As the field continues to evolve, it is crucial that we maintain a cautious and meticulous approach to ensure the safe and effective translation of stem-cell-based therapies to clinical settings.

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REFERENCES

1. Wilson JM (2009). A history lesson for stem cells. Science (80-.). 324, 727–728. 10.1126/science.1174935.
2. Singh AK, and McGuirk JP (2016). Allogeneic Stem Cell Transplantation: A Historical and Scientific Overview. Cancer Res. 76, 6445–6451. 10.1158/0008-5472.CAN-16-1311. [PubMed: 27784742]
3. Dunn GP, Old LJ, and Schreiber RD (2004). The Immunobiology of Cancer Immunosurveillance and Immunoediting. Immunity 21, 137–148. 10.1016/j.immuni.2004.07.017. [PubMed: 15308095]
4. Rosenberg SA, Yang JC, Sherry RM, Kammula US, Hughes MS, Phan GQ, Citrin DE, Restifo NP, Robbins PF, Wunderlich JR, et al. (2011). Durable complete responses in heavily pretreated patients with metastatic melanoma using T-cell transfer immunotherapy. Clin. Cancer Res 17, 4550–4557. 10.1158/1078-0432.CCR-11-0116. [PubMed: 21498393]
5. Radvanyi LG (2015). Tumor-Infiltrating Lymphocyte Therapy: Addressing Prevailing Questions. Cancer J. 21.
6. June CH (2007). Adoptive T cell therapy for cancer in the clinic. J. Clin. Invest 117, 1466–1476. 10.1172/JCI32446. [PubMed: 17549249]
7. Kansagra A, Farnia S, and Majhail N (2020). Expanding Access to Chimeric Antigen Receptor T-Cell Therapies: Challenges and Opportunities. Am. Soc. Clin. Oncol. Educ. B, e27–e34. 10.1200/EDBK_279151.
8. Mirzaei HR, Rodriguez A, Shepphird J, Brown CE, and Badie B (2017). Chimeric Antigen Receptors T Cell Therapy in Solid Tumor: Challenges and Clinical Applications . Front. Immunol 8.

9. Roddie C, O'Reilly M, Dias Alves Pinto J, Vispute K, and Lowdell M (2019). Manufacturing chimeric antigen receptor T cells: issues and challenges. Cytotherapy 21, 327–340. 10.1016/j.jcyt.2018.11.009. [PubMed: 30685216]
10. Srivastava S, and Riddell SR (2018). Chimeric Antigen Receptor T Cell Therapy: Challenges to Bench-to-Bedside Efficacy. J. Immunol 200, 459 LP–468. 10.4049/jimmunol.1701155. [PubMed: 29311388]
11. Xia A-L, Wang X-C, Lu Y-J, Lu X-J, and Sun B (2017). Chimeric-antigen receptor T (CAR-T) cell therapy for solid tumors: challenges and opportunities. Oncotarget 8, 90521–90531. 10.18632/oncotarget.19361. [PubMed: 29163850]
12. Lim WA, and June CH (2017). The Principles of Engineering Immune Cells to Treat Cancer. Cell 168, 724–740. 10.1016/j.cell.2017.01.016. [PubMed: 28187291]
13. Mikkilineni L, and Kochenderfer JN (2017). Chimeric antigen receptor T-cell therapies for multiple myeloma. Blood 130, 2594–2602. 10.1182/blood-2017-06-793869. [PubMed: 28928126]
14. Ramos CA, Heslop HE, and Brenner MK (2016). CAR-T Cell Therapy for Lymphoma. Annu. Rev. Med 67, 165–183. 10.1146/annurev-med-051914-021702. [PubMed: 26332003]
15. Themeli M, Rivière I, and Sadelain M (2015). New cell sources for T cell engineering and adoptive immunotherapy. Cell Stem Cell 16, 357–366. 10.1016/j.stem.2015.03.011. [PubMed: 25842976]
16. Ruella M, Xu J, Barrett DM, Fraietta JA, Reich TJ, Ambrose DE, Klichinsky M, Shestova O, Patel PR, Kulikovskaya I, et al. (2018). Induction of resistance to chimeric antigen receptor T cell therapy by transduction of a single leukemic B cell. Nat. Med 24, 1499–1503. 10.1038/s41591-018-0201-9. [PubMed: 30275568]
17. Good CR, Aznar MA, Kuramitsu S, Samareh P, Agarwal S, Donahue G, Ishiyama K, Wellhausen N, Rennels AK, Ma Y, et al. (2021). An NK-like CAR T cell transition in CAR T cell dysfunction. Cell 184, 6081–6100.e26. 10.1016/j.cell.2021.11.016. [PubMed: 34861191]
18. Deng Q, Han G, Puebla-Osorio N, Ma MCJ, Strati P, Chasen B, Dai E, Dang M, Jain N, Yang H, et al. (2020). Characteristics of anti-CD19 CAR T cell infusion products associated with efficacy and toxicity in patients with large B cell lymphomas. Nat. Med 26, 1878–1887. 10.1038/s41591-020-1061-7. [PubMed: 33020644]
19. Arcangeli S, Bove C, Mezzanotte C, Camisa B, Falcone L, Manfredi F, Bezzecchi E, El Khoury R, Norata R, Sanvito F, et al. (2022). CAR T-cell manufacturing from naive/stem memory T-lymphocytes enhances antitumor responses while curtailing cytokine release syndrome. J. Clin. Invest 10.1172/JCI150807.
20. Fraietta JA, Lacey SF, Orlando EJ, Pruteanu-Malinici I, Gohil M, Lundh S, Boesteanu AC, Wang Y, O'Connor RS, Hwang W-T, et al. (2018). Determinants of response and resistance to CD19 chimeric antigen receptor (CAR) T cell therapy of chronic lymphocytic leukemia. Nat. Med 24, 563–571. 10.1038/s41591-018-0010-1. [PubMed: 29713085]
21. Fraietta JA, Nobles CL, Sammons MA, Lundh S, Carty SA, Reich TJ, Cogdill AP, Morrissette JJD, DeNizio JE, Reddy S, et al. (2018). Disruption of TET2 promotes the therapeutic efficacy of CD19-targeted T cells. Nature 558, 307–312. 10.1038/S41586-018-0178-z. [PubMed: 29849141]
22. Majzner RG, and Mackall CL (2018). Tumor Antigen Escape from CAR T-cell Therapy. Cancer Discov. 8, 1219–1226. 10.1158/2159-8290.CD-18-0442. [PubMed: 30135176]
23. Sabatino M, Hu J, Sommariva M, Gautam S, Fellowes V, Hocker JD, Dougherty S, Qin H, Klebanoff CA, Fry TJ, et al. (2016). Generation of clinical-grade CD19-specific CAR-modified CD8+ memory stem cells for the treatment of human B-cell malignancies. Blood 128, 519–528. 10.1182/blood-2015-11-683847. [PubMed: 27226436]
24. Turtle CJ, Hanafi L-A, Berger C, Gooley TA, Cherian S, Hudecek M, Sommermeyer D, Melville K, Pender B, Budiarto TM, et al. (2016). CD19 CAR-T cells of defined CD4+:CD8+ composition in adult B cell ALL patients. J. Clin. Invest 126, 2123–2138. 10.1172/JCI85309. [PubMed: 27111235]
25. Shah NN, Zhu F, Taylor C, Schneider D, Krueger W, Worden A, Yim S, Fenske TS, Hamadani M, Johnson B, et al. (2018). A Phase 1 Study with Point-of-Care Manufacturing of Dual Targeted, Tandem Anti-CD19, Anti-CD20 Chimeric Antigen Receptor Modified T (CAR-T) Cells for Relapsed, Refractory, Non-Hodgkin Lymphoma. Blood 132, 4193. 10.1182/blood-2018-99-110194.

26. Benjamin R, Graham C, Yallop D, Jozwik A, Ciocarlie O, Jain N, Jabbour EJ, Maus MXV, Frigault M, Boissel N, et al. (2018). Preliminary Data on Safety, Cellular Kinetics and Anti-Leukemic Activity of UCART19, an Allogeneic Anti-CD19 CAR T-Cell Product, in a Pool of Adult and Pediatric Patients with High-Risk CD19+ Relapsed/Refractory B-Cell Acute Lymphoblastic Leukemia. Blood 132, 896–896. 10.1182/blood-2018-99-111356.
27. Murthy H, Iqbal M, Chavez JC, and Kharfan-Dabaja MA (2019). Cytokine Release Syndrome: Current Perspectives. ImmunoTargets Ther. 8, 43–52. 10.2147/ITT.S202015. [PubMed: 31754614]
28. Labanieh L, Majzner RG, and Mackall CL (2018). Programming CAR-T cells to kill cancer. Nat. Biomed. Eng 2, 377–391. 10.1038/s41551-018-0235-9. [PubMed: 31011197]
29. Liu E, Marin D, Banerjee P, Macapinlac HA, Thompson P, Basar R, Nassif Kerbauy L, Overman B, Thall P, Kaplan M, et al. (2020). Use of CAR-Transduced Natural Killer Cells in CD19-Positive Lymphoid Tumors. N. Engl. J. Med 382, 545–553. 10.1056/nejmoa1910607. [PubMed: 32023374]
30. Petersen CT, Hassan M, Morris AB, Jeffery J, Lee K, Jagirdar N, Staton AD, Raikar SS, Spencer HT, Sulchek T, et al. (2018). Improving T-cell expansion and function for adoptive T-cell therapy using ex vivo treatment with PI3Kd inhibitors and VIP antagonists. Blood Adv. 2, 210–223. 10.1182/bloodadvances.2017011254. [PubMed: 29386194]
31. Majzner RG, Ramakrishna S, Yeom KW, Patel S, Chinnasamy H, Schultz LM, Richards RM, Jiang L, Barsan V, Mancusi R, et al. (2022). GD2-CAR T cell therapy for H3K27M-mutated diffuse midline gliomas. Nature 603, 934–941. 10.1038/s41586-022-04489-4. [PubMed: 35130560]
32. Mount CW, and Gonzalez Castro LN (2022). Advances in Chimeric Antigen Receptor (CAR) T-Cell Therapies for the Treatment of Primary Brain Tumors. Antibodies 11. 10.3390/antib11020031.
33. Qi C, Gong J, Li J, Liu D, Qin Y, Ge S, Zhang M, Peng Z, Zhou J, Cao Y, et al. (2022). Claudin18.2-specific CAR T cells in gastrointestinal cancers: phase 1 trial interim results. Nat. Med 28, 1189–1198. 10.1038/s41591-022-01800-8. [PubMed: 35534566]
34. Depil S, Duchateau P, Grupp SA, Mufti G, and Poirot L (2020). 'Off-the-shelf' allogeneic CAR T cells: development and challenges. Nat. Rev. Drug Discov 19, 185–199. 10.1038/S41573-019-0051-2. [PubMed: 31900462]
35. Ruella M, and Kenderian SS (2017). Next-Generation Chimeric Antigen Receptor T-Cell Therapy: Going off the Shelf. BioDrugs 31, 473–481. 10.1007/s40259-017-0247-0. [PubMed: 29143249]
36. Sommer C, Boldajipour B, Kuo TC, Bentley T, Sutton J, Chen A, Geng T, Dong H, Galetto R, Valton J, et al. (2019). Preclinical Evaluation of Allogeneic CAR T Cells Targeting BCMA for the Treatment of Multiple Myeloma. Mol. Ther 27, 1126–1138. 10.1016/j.ymthe.2019.04.001. [PubMed: 31005597]
37. Perez C, Gruber I, and Arber C (2020). Off-the-Shelf Allogeneic T Cell Therapies for Cancer: Opportunities and Challenges Using Naturally Occurring "Universal" Donor T Cells. Front. Immunol 11, 583716. 10.3389/fimmu.2020.583716. [PubMed: 33262761]
38. Ren J, Liu X, Fang C, Jiang S, June CH, and Zhao Y (2017). Multiplex genome editing to generate universal CAR T cells resistant to PD1 inhibition. Clin. Cancer Res 23, 2255–2266. 10.1158/1078-0432.CCR-16-1300. [PubMed: 27815355]
39. Chaidos A, Patterson S, Szydlo R, Chaudhry MS, Dazzi F, Kanfer E, McDonald D, Marin D, Milojkovic D, Pavlu J, et al. (2012). Graft invariant natural killer T-cell dose predicts risk of acute graft-versus-host disease in allogeneic hematopoietic stem cell transplantation. Blood 119, 5030–5036. 10.1182/blood-2011-11-389304. [PubMed: 22371885]
40. Liu E, Tong Y, Dotti G, Shaim H, Savoldo B, Mukherjee M, Orange J, Wan X, Lu X, Reynolds A, et al. (2018). Cord blood NK cells engineered to express IL-15 and a CD19-targeted CAR show long-term persistence and potent antitumor activity. Leukemia 32, 520–531. 10.1038/leu.2017.226. [PubMed: 28725044]
41. Tugues S, Amorim A, Spath S, Martin-Blondel G, Schreiner B, De Feo D, Lutz M, Guscetti F, Apostolova P, Haftmann C, et al. (2018). Graft-versus-host disease, but not graft-versus-leukemia immunity, is mediated by GM-CSF-licensed myeloid cells. Sci. Transl. Med 10. 10.1126/scitranslmed.aat8410.

42. Delfanti G, Dellabona P, Casorati G, and Fedeli M (2022). Adoptive Immunotherapy With Engineered iNKT Cells to Target Cancer Cells and the Suppressive Microenvironment. Front. Med 9.
43. Bae E-A, Seo H, Kim I-K, Jeon I, and Kang C-Y (2019). Roles of NKT cells in cancer immunotherapy. Arch. Pharm. Res 42, 543–548. 10.1007/s12272-019-01139-8. [PubMed: 30859410]
44. Godfrey DI, Le Nours J, Andrews DM, Uldrich AP, and Rossjohn J (2018). Unconventional T Cell Targets for Cancer Immunotherapy. Immunity 48, 453–473. 10.1016/j.immuni.2018.03.009. [PubMed: 29562195]
45. Zhang C, Hu Y, and Shi C (2020). Targeting Natural Killer Cells for Tumor Immunotherapy. Front. Immunol 11. 10.3389/fimmu.2020.00060.
46. Li Y-R, Zhou K, Wilson M, Kramer A, Zhu Y, Dawson N, and Yang L (2022). Mucosal-associated invariant T cells for cancer immunotherapy. Mol. Ther 10.1016/j.ymthe.2022.11.019.
47. Cortés-Selva D, Dasgupta B, Singh S, and Grewal IS (2021). Innate and Innate-Like Cells: The Future of Chimeric Antigen Receptor (CAR) Cell Therapy. Trends Pharmacol. Sci 42, 45–59. 10.1016/j.tips.2020.11.004. [PubMed: 33250273]
48. Bulcha JT, Wang Y, Ma H, Tai PWL, and Gao G (2021). Viral vector platforms within the gene therapy landscape. Signal Transduct. Target. Ther 6, 53. 10.1038/S41392-021-00487-6. [PubMed: 33558455]
49. Ellis GI, Sheppard NC, and Riley JL (2021). Genetic engineering of T cells for immunotherapy. Nat. Rev. Genet 22, 427–447. 10.1038/s41576-021-00329-9. [PubMed: 33603158]
50. Weber EW, Maus MV, and Mackall CL (2020). The Emerging Landscape of Immune Cell Therapies. Cell 181, 46–62. 10.1016/j.cell.2020.03.001. [PubMed: 32243795]
51. Cascalho M, and Platt JL (2005). Basic mechanisms of humoral rejection. Pediatr. Transplant 9, 9–16. 10.1111/j.1399-3046.2004.00231.x. [PubMed: 15667605]
52. Montgomery RA, Cozzi E, West LJ, and Warren DS (2011). Humoral immunity and antibody-mediated rejection in solid organ transplantation. Semin. Immunol 23, 224–234. 10.1016/j.smim.2011.08.021. [PubMed: 21958960]
53. Liu E, Marin D, Banerjee P, Macapinlac HA, Thompson P, Basar R, Nassif Kerbauy L, Overman B, Thall P, Kaplan M, et al. (2020). Use of CAR-Transduced Natural Killer Cells in CD19-Positive Lymphoid Tumors. N. Engl. J. Med 382, 545–553. 10.1056/NEJMoa1910607. [PubMed: 32023374]
54. Figueiredo C, and Blasczyk R (2015). A future with less HLA: potential clinical applications of HLA-universal cells. Tissue Antigens 85, 443–449. 10.1111/tan.12564. [PubMed: 25864470]
55. Gornalusse GG, Hirata RK, Funk SE, Riolobos L, Lopes VS, Manske G, Prunkard D, Colunga AG, Hanafi L-A, Clegg DO, et al. (2017). HLA-E-expressing pluripotent stem cells escape allogeneic responses and lysis by NK cells. Nat. Biotechnol 35, 765–772. 10.1038/nbt.3860. [PubMed: 28504668]
56. Li Y-R, Dunn ZS, Zhou Y, Lee D, and Yang L (2021). Development of Stem Cell-Derived Immune Cells for Off-the-Shelf Cancer Immunotherapies. Cells 10. 10.3390/cells10123497.
57. Li Y-R, Zhou Y, Kramer A, and Yang L (2021). Engineering stem cells for cancer immunotherapy. Trends in cancer. 10.1016/j.trecan.2021.08.004.
58. Zhu H, Lai Y-S, Li Y, Blum RH, and Kaufman DS (2018). Concise Review: Human Pluripotent Stem Cells to Produce Cell-Based Cancer Immunotherapy. Stem Cells 36, 134–145. 10.1002/stem.2754. [PubMed: 29235195]
59. Vodyanik MA, Bork JA, Thomson JA, and Slukvin II (2005). Human embryonic stem cell-derived CD34+ cells: Efficient production in the coculture with OP9 stromal cells and analysis of lymphohematopoietic potential. Blood 105, 617–626. 10.1182/blood-2004-04-1649. [PubMed: 15374881]
60. Kaufman DS, Hanson ET, Lewis RL, Auerbach R, and Thomson JA (2001). Hematopoietic colony-forming cells derived from human embryonic stem cells. Proc. Natl. Acad. Sci. U. S. A 98, 10716–10721. 10.1073/pnas.191362598. [PubMed: 11535826]
61. Ng ES, Davis R, Stanley EG, and Elefanty AG (2008). A protocol describing the use of a recombinant protein-based, animal product-free medium (APEL) for human embryonic stem cell

differentiation as spin embryoid bodies. Nat. Protoc 3, 768–776. 10.1038/nprot.2008.42. [PubMed: 18451785]

62. Kennedy M, Awong G, Sturgeon CM, Ditadi A, LaMotte-Mohs R, Zúñiga-Pflücker JC, and Keller G (2012). T lymphocyte potential marks the emergence of definitive hematopoietic progenitors in human pluripotent stem cell differentiation cultures. Cell Rep. 2, 1722–1735. 10.1016/j.celrep.2012.11.003. [PubMed: 23219550]
63. Kitayama S, Zhang R, Liu TY, Ueda N, Iriguchi S, Yasui Y, Kawai Y, Tatsumi M, Hirai N, Mizoro Y, et al. (2016). Cellular Adjuvant Properties, Direct Cytotoxicity of Re-differentiated Vα24 Invariant NKT-like Cells from Human Induced Pluripotent Stem Cells. Stem Cell Reports 6, 213–227. 10.1016/j.stemcr.2016.01.005. [PubMed: 26862702]
64. Nishimura T, and Nakauchi H (2019). Generation of antigen-specific T cells from human induced pluripotent stem cells. Methods Mol. Biol 1899, 25–40. 10.1007/978-1-4939-8938-6_3. [PubMed: 30649763]
65. Nishimura T, Kaneko S, Kawana-Tachikawa A, Tajima Y, Goto H, Zhu D, Nakayama-Hosoya K, Iriguchi S, Uemura Y, Shimizu T, et al. (2013). Generation of rejuvenated antigen-specific T cells by reprogramming to pluripotency and redifferentiation. Cell Stem Cell 12, 114–126. 10.1016/j.stem.2012.11.002. [PubMed: 23290140]
66. Vizcardo R, Masuda K, Yamada D, Ikawa T, Shimizu K, Fujii S-I, Koseki H, and Kawamoto H (2013). Regeneration of human tumor antigen-specific T cells from iPSCs derived from mature CD8(+) T cells. Cell Stem Cell 12, 31–36. 10.1016/j.stem.2012.12.006. [PubMed: 23290135]
67. Wakao H, Yoshikiyo K, Koshimizu U, Furukawa T, Enomoto K, Matsunaga T, Tanaka T, Yasutomi Y, Yamada T, Minakami H, et al. (2013). Expansion of functional human mucosal-associated invariant T cells via reprogramming to pluripotency and redifferentiation. Cell Stem Cell 12, 546–558. 10.1016/j.stem.2013.03.001. [PubMed: 23523177]
68. Zeng J, Tang SY, Toh LL, and Wang S (2017). Generation of "Off-the-Shelf" Natural Killer Cells from Peripheral Blood Cell-Derived Induced Pluripotent Stem Cells. Stem Cell Reports 9, 1796–1812. 10.1016/j.stemcr.2017.10.020. [PubMed: 29173894]
69. Brentjens RJ, Rivière I, Park JH, Davila ML, Wang X, Stefanski J, Taylor C, Yeh R, Bartido S, Borquez-Ojeda O, et al. (2011). Safety and persistence of adoptively transferred autologous CD19-targeted T cells in patients with relapsed or chemotherapy refractory B-cell leukemias. Blood 118, 4817–4828. 10.1182/blood-2011-04-348540. [PubMed: 21849486]
70. Brentjens RJ, Davila ML, Riviere I, Park J, Wang X, Cowell LG, Bartido S, Stefanski J, Taylor C, Olszewska M, et al. (2013). CD19-targeted T cells rapidly induce molecular remissions in adults with chemotherapy-refractory acute lymphoblastic leukemia. Sci. Transl. Med 5. 10.1126/scitranslmed.3005930.
71. Davila ML, Riviere I, Wang X, Bartido S, Park J, Curran K, Chung SS, Stefanski J, Borquez-Ojeda O, Olszewska M, et al. (2014). Efficacy and toxicity management of 19-28z CAR T cell therapy in B cell acute lymphoblastic leukemia. Sci. Transl. Med 6, 224ra25. 10.1126/scitranslmed.3008226.
72. Kochenderfer JN, Dudley ME, Feldman SA, Wilson WH, Spaner DE, Maric I, Stetler-Stevenson M, Phan GQ, Hughes MS, Sherry RM, et al. (2012). B-cell depletion and remissions of malignancy along with cytokine-associated toxicity in a clinical trial of anti-CD19 chimeric-antigen-receptor-transduced T cells. Blood 119, 2709–2720. 10.1182/blood-2011-10-384388. [PubMed: 22160384]
73. Robbins PF, Morgan RA, Feldman SA, Yang JC, Sherry RM, Dudley ME, Wunderlich JR, Nahvi AV, Helman LJ, Mackall CL, et al. (2011). Tumor regression in patients with metastatic synovial cell sarcoma and melanoma using genetically engineered lymphocytes reactive with NY-ESO-1. J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol 29, 917–924. 10.1200/JCO.2010.32.2537.
74. Rapoport AP, Stadtmauer EA, Binder-Scholl GK, Goloubeva O, Vogl DT, Lacey SF, Badros AZ, Garfall A, Weiss B, Finklestein J, et al. (2015). NY-ESO-1-specific TCR-engineered T cells mediate sustained antigen-specific antitumor effects in myeloma. Nat. Med 21, 914–921. 10.1038/nm.3910. [PubMed: 26193344]
75. Zhang Y, Liu Z, Wei W, and Li Y (2022). TCR engineered T cells for solid tumor immunotherapy. Exp. Hematol. Oncol 11, 38. 10.1186/s40164-022-00291-0. [PubMed: 35725570]

76. Themeli M, Kloss CC, Ciriello G, Fedorov VD, Perna F, Gonen M, and Sadelain M (2013). Generation of tumor-targeted human T lymphocytes from induced pluripotent stem cells for cancer therapy. Nat. Biotechnol 31, 928–933. 10.1038/nbt.2678. [PubMed: 23934177]
77. Abramson JS, Palomba ML, Gordon LI, Lunning MA, Wang M, Arnason J, Mehta A, Purev E, Maloney DG, Andreadis C, et al. (2020). Lisocabtagene maraleucel for patients with relapsed or refractory large B-cell lymphomas (TRANSCEND NHL 001): a multicentre seamless design study. Lancet (London, England) 396, 839–852. 10.1016/S0140-6736(20)31366-0. [PubMed: 32888407]
78. Kharfan-Dabaja MA, Yassine F, Alhaj Moustafa M, Iqbal M, and Murthy H (2021). Lisocabtagene maraleucel in relapsed or refractory diffuse large B cell lymphoma: What is the evidence? Hematol. Oncol. Stem Cell Ther 10.1016/j.hemonc.2021.09.004.
79. Li Y-R, Zhou Y, Kim YJ, Zhu Y, Ma F, Yu J, Wang Y-C, Chen X, Li Z, Zeng S, et al. (2021). Development of allogeneic HSC-engineered iNKT cells for off-the-shelf cancer immunotherapy. Cell reports. Med 2, 100449. 10.1016/j.xcrm.2021.100449.
80. Seet CS, He C, Bethune MT, Li S, Chick B, Gschweng EH, Zhu Y, Kim K, Kohn DB, Baltimore D, et al. (2017). Generation of mature T cells from human hematopoietic stem and progenitor cells in artificial thymic organoids. Nat. Methods 14, 521–530. 10.1038/nmeth.4237. [PubMed: 28369043]
81. Montel-Hagen A, Seet CS, Li S, Chick B, Zhu Y, Chang P, Tsai S, Sun V, Lopez S, Chen HC, et al. (2019). Organoid-Induced Differentiation of Conventional T Cells from Human Pluripotent Stem Cells. Cell Stem Cell 24, 376–389.e8. 10.1016/j.stem.2018.12.011. [PubMed: 30661959]
82. Smith MJ, Webber BR, Mohtashami M, Stefanski HE, Zúñiga-Pflücker JC, and Blazar BR (2015). In Vitro T-Cell Generation From Adult, Embryonic, and Induced Pluripotent Stem Cells: Many Roads to One Destination. Stem Cells 33, 3174–3180. 10.1002/stem.2115. [PubMed: 26227158]
83. Robey E, Chang D, Itano A, Cado D, Alexander H, Lans D, Weinmaster G, and Salmon P (1996). An activated form of Notch influences the choice between CD4 and CD8 T cell lineages. Cell 87, 483–492. 10.1016/s0092-8674(00)81368-9. [PubMed: 8898201]
84. Robey E (1999). Regulation of T cell fate by Notch. Annu. Rev. Immunol 17, 283–295. 10.1146/annurev.immunol.17.1.283. [PubMed: 10358760]
85. Yasutomo K, Doyle C, Miele L, Fuchs C, and Germain RN (2000). The duration of antigen receptor signalling determines CD4+ versus CD8+ T-cell lineage fate. Nature 404, 506–510. 10.1038/35006664. [PubMed: 10761920]
86. Li Y, Hermanson DL, Moriarity BS, and Kaufman DS (2018). Human iPSC-Derived Natural Killer Cells Engineered with Chimeric Antigen Receptors Enhance Anti-tumor Activity. Cell Stem Cell 23, 181–192.e5. 10.1016/j.stem.2018.06.002. [PubMed: 30082067]
87. Goldenson BH, Zhu H, Wang YM, Heragu N, Bernareggi D, Ruiz-Cisneros A, Bahena A, Ask EH, Hoel HJ, Malmberg K-J, et al. (2020). Umbilical Cord Blood and iPSC-Derived Natural Killer Cells Demonstrate Key Differences in Cytotoxic Activity and KIR Profiles. Front. Immunol 11, 561553. 10.3389/fimmu.2020.561553. [PubMed: 33178188]
88. Hermanson DL, Bendzick L, Pribyl L, McCullar V, Vogel RI, Miller JS, Geller MA, and Kaufman DS (2016). Induced Pluripotent Stem Cell-Derived Natural Killer Cells for Treatment of Ovarian Cancer. Stem Cells 34, 93–101. 10.1002/stem.2230. [PubMed: 26503833]
89. Rezvani K, Rouce R, Liu E, and Shpall E (2017). Engineering Natural Killer Cells for Cancer Immunotherapy. Mol. Ther 25, 1769–1781. 10.1016/j.ymthe.2017.06.012. [PubMed: 28668320]
90. Cany J, van der Waart AB, Spanholtz J, Tordoir M, Jansen JH, van der Voort R, Schaap NM, and Dolstra H (2015). Combined IL-15 and IL-12 drives the generation of CD34+-derived natural killer cells with superior maturation and alloreactivity potential following adoptive transfer. Oncoimmunology 4, 1–2. 10.1080/2162402X.2015.1017701.
91. Cichocki F, Bjordahl R, Gaidarova S, Mahmood S, Abujarour R, Wang H, Tuininga K, Felices M, Davis ZB, Bendzick L, et al. (2020). iPSC-derived NK cells maintain high cytotoxicity and enhance in vivo tumor control in concert with T cells and anti-PD-1 therapy. Sci. Transl. Med 12. 10.1126/scitranslmed.aaz5618.
92. Fang F, Xiao W, and Tian Z (2017). NK cell-based immunotherapy for cancer. Semin. Immunol 31, 37–54. 10.1016/j.smim.2017.07.009. [PubMed: 28838796]

93. Tang X, Yang L, Li Z, Nalin AP, Dai H, Xu T, Yin J, You F, Zhu M, Shen W, et al. (2018). First-in-man clinical trial of CAR NK-92 cells: safety test of CD33-CAR NK-92 cells in patients with relapsed and refractory acute myeloid leukemia. Am. J. Cancer Res 8, 1083–1089. [PubMed: 30034945]

94. Woll PS, Martin CH, Miller JS, and Kaufman DS (2005). Human Embryonic Stem Cell-Derived NK Cells Acquire Functional Receptors and Cytolytic Activity. J. Immunol 175, 5095–5103. 10.4049/jimmunol.175.8.5095. [PubMed: 16210613]

95. Zhu H, and Kaufman DS (2019). An Improved Method to Produce Clinical-Scale Natural Killer Cells from Human Pluripotent Stem Cells. Methods Mol. Biol 2048, 107–119. 10.1007/978-1-4939-9728-2_12. [PubMed: 31396935]

96. Zhu H, Blum RH, Bernareggi D, Ask EH, Wu Z, Hoel HJ, Meng Z, Wu C, Guan K-L, Malmberg K-J, et al. (2020). Metabolic Reprograming via Deletion of CISH in Human iPSC-Derived NK Cells Promotes In Vivo Persistence and Enhances Anti-tumor Activity. Cell Stem Cell 27, 224–237.e6. 10.1016/j.stem.2020.05.008. [PubMed: 32531207]

97. Abel AM, Yang C, Thakar MS, and Malarkannan S (2018). Natural killer cells: Development, maturation, and clinical utilization. Front. Immunol 9, 1–23. 10.3389/fimmu.2018.01869. [PubMed: 29403488]

98. Liu H, Wang S, Xin J, Wang J, Yao C, and Zhang Z (2019). Role of NKG2D and its ligands in cancer immunotherapy. Am. J. Cancer Res 9, 2064–2078. [PubMed: 31720075]

99. Ruggeri L, Mancusi A, Capanni M, Urbani E, Carotti A, Aloisi T, Stern M, Pende D, Perruccio K, Burchielli E, et al. (2007). Donor natural killer cell allorecognition of missing self in haploidentical hematopoietic transplantation for acute myeloid leukemia: challenging its predictive value. Blood 110, 433–440. 10.1182/blood-2006-07-038687. [PubMed: 17371948]

100. Hu W, Wang G, Huang D, Sui M, and Xu Y (2019). Cancer immunotherapy based on natural killer cells: Current progress and new opportunities. Front. Immunol 10, 1–16. 10.3389/fimmu.2019.01205. [PubMed: 30723466]

101. Mehta RS, and Rezvani K (2018). Chimeric antigen receptor expressing natural killer cells for the immunotherapy of cancer. Front. Immunol 9, 1–12. 10.3389/fimmu.2018.00283. [PubMed: 29403488]

102. Zhu H, Blum RH, Bjordahl R, Gaidarova S, Rogers P, Lee TT, Abujarour R, Bonello GB, Wu J, Tsai P-F, et al. (2020). Pluripotent stem cell-derived NK cells with high-affinity noncleavable CD16a mediate improved antitumor activity. Blood 135, 399–410. 10.1182/blood.2019000621. [PubMed: 31856277]

103. Alfarra H, Weir J, Grieve S, and Reiman T (2020). Targeting NK Cell Inhibitory Receptors for Precision Multiple Myeloma Immunotherapy . Front. Immunol 11.

104. Fujii S. ichiro, Shimizu K, Okamoto Y, Kunii N, Nakayama T, Motohashi S, and Taniguchi M (2013). NKT cells as an ideal anti-tumor immunotherapeutic. Front. Immunol 4, 1–7. 10.3389/fimmu.2013.00409. [PubMed: 23355837]

105. Li YR, Dunn ZS, Jr GG, Carmona C, Zhou Y, Lee D, Yu J, Huang J, Kim JT, Arumugaswami V, et al. (2022). Development of off - the - shelf hematopoietic stem cell - engineered invariant natural killer T cells for COVID - 19 therapeutic intervention. Stem Cell Res. Ther, 1–15. 10.1186/s13287-022-02787-2. [PubMed: 34998430]

106. Zhu Y, Smith DJ, Zhou Y, Li YR, Yu J, Lee D, Wang YC, Di Biase S, Wang X, Hardoy C, et al. (2019). Development of Hematopoietic Stem Cell-Engineered Invariant Natural Killer T Cell Therapy for Cancer. Cell Stem Cell 25, 542–557.e9. 10.1016/j.stem.2019.08.004. [PubMed: 31495780]

107. Smith DJ, Liu S, Ji S, Li B, McLaughlin J, Cheng D, Witte ON, and Yang L (2015). Genetic engineering of hematopoietic stem cells to generate invariant natural killer T cells. Proc. Natl. Acad. Sci. U. S. A 112, 1523–1528. 10.1073/pnas.1424877112. [PubMed: 25605948]

108. Wakao H (2020). Reprogramming of MAIT Cells to Pluripotency and Redifferentiation. Methods Mol. Biol 2098, 237–257. 10.1007/978-1-0716-0207-2_16. [PubMed: 31792827]

109. Wakao H, and Fujita H (2013). Toward the realization of cell therapy: the advent of MAIT cells from iPSCs. Cell Cycle 12, 2341–2342. 10.4161/CC.25706. [PubMed: 23856578]

110. Zhou Y, Li Y-R, Zeng S, and Yang L (2021). Methods for Studying Mouse and Human Invariant Natural Killer T Cells. Methods Mol. Biol 2388, 35–57. 10.1007/978-1-0716-1775-5_4. [PubMed: 34524660]
111. Hansen M, Varga E, Aarts C, Wust T, Kuijpers T, von Lindern M, and van den Akker E (2018). Efficient production of erythroid, megakaryocytic and myeloid cells, using single cell-derived iPSC colony differentiation. Stem Cell Res. 29, 232–244. 10.1016/j.scr.2018.04.016. [PubMed: 29751281]
112. Horton C, Davies TJ, Lahiri P, Sachamitr P, and Fairchild PJ (2020). Induced pluripotent stem cells reprogrammed from primary dendritic cells provide an abundant source of immunostimulatory dendritic cells for use in immunotherapy. Stem Cells 38, 67–79. 10.1002/stem.3095. [PubMed: 31621975]
113. Sachamitr P, Leishman AJ, Davies TJ, and Fairchild PJ (2018). Directed Differentiation of Human Induced Pluripotent Stem Cells into Dendritic Cells Displaying Tolerogenic Properties and Resembling the CD141(+) Subset. Front. Immunol 8, 1935. 10.3389/fimmu.2017.01935. [PubMed: 29358940]
114. Zhang L, Tian L, Dai X, Yu H, Wang J, Lei A, Zhu M, Xu J, Zhao W, Zhu Y, et al. (2020). Pluripotent stem cell-derived CAR-macrophage cells with antigen-dependent anti-cancer cell functions. J. Hematol. Oncol 13, 153. 10.1186/s13045-020-00983-2. [PubMed: 33176869]
115. Kitchen SG, Bennett M, Galić Z, Kim J, Xu Q, Young A, Lieberman A, Joseph A, Goldstein H, Ng H, et al. (2009). Engineering antigen-specific T cells from genetically modified human hematopoietic stem cells in immunodeficient mice. PLoS One 4, 4–12. 10.1371/journal.pone.0008208.
116. Lan P, Tonomura N, Shimizu A, Wang S, and Yang YG (2006). Reconstitution of a functional human immune system in immunodeficient mice through combined human fetal thymus/liver and CD34+ cell transplantation. Blood 108, 487–492. 10.1182/blood-2005-11-4388. [PubMed: 16410443]
117. Melkus MW, Estes JD, Padgett-Thomas A, Gatlin J, Denton PW, Othieno FA, Wege AK, Haase AT, and Garcia JV (2006). Humanized mice mount specific adaptive and innate immune responses to EBV and TSST-1. Nat. Med 12, 1316–1322. 10.1038/nm1431. [PubMed: 17057712]
118. Kim S, Ahn SE, Lee JH, Lim D-S, Kim K-S, Chung H-M, and Lee S-H (2007). A Novel Culture Technique for Human Embryonic Stem Cells Using Porous Membranes. Stem Cells 25, 2601–2609. 10.1634/stemcells.2006-0814. [PubMed: 17628020]
119. Yu G, Kamano Y, Wang F, Okawa H, Yatani H, and Egusa H (2015). Feeder Cell Sources and Feeder-Free Methods for Human iPS Cell Culture BT - Interface Oral Health Science 2014. In, Sasaki K, Suzuki O, and Takahashi N, eds. (Springer Japan), pp. 145–159.
120. Lu Z, Zhu W, Yu Y, Jin D, Guan Y, Yao R, Zhang YA, Zhang Y, and Zhou Q (2010). Derivation and long-term culture of human parthenogenetic embryonic stem cells using human foreskin feeders. J. Assist. Reprod. Genet 27, 285–291. 10.1007/s10815-010-9408-5. [PubMed: 20393797]
121. Park Y, Kim JH, Lee SJ, Choi IY, Park SJ, Lee SR, Sung HJ, Yoo Y. Do, Geum DH, Choi CW, et al. (2011). Human feeder cells can support the undifferentiated growth of human and mouse embryonic stem cells using their own basic fibroblast growth factors. Stem Cells Dev. 20, 1901–1910. 10.1089/scd.2010.0496. [PubMed: 21231869]
122. Unger C, Gao S, Cohen M, Jaconi M, Bergstrom R, Holm F, Galan A, Sanchez E, Irion O, Dubuisson JB, et al. (2009). Immortalized human skin fibroblast feeder cells support growth and maintenance of both human embryonic and induced pluripotent stem cells. Hum. Reprod 24, 2567–2581. 10.1093/humrep/dep232. [PubMed: 19556288]
123. Pekkanen-Mattila M, Ojala M, Kerkelä E, Rajala K, Skottman H, and Aalto-Setälä K (2012). The effect of human and mouse fibroblast feeder cells on cardiac differentiation of human pluripotent stem cells. Stem Cells Int. 2012, 875059. 10.1155/2012/875059. [PubMed: 22315618]
124. Huijskens MJAJ, Walczak M, Koller N, Briedé JJ, Senden-Gijsbers BLMG, Schnijderberg MC, Bos GMJ, and Germeraad WTV (2014). Technical advance: ascorbic acid induces development of double-positive T cells from human hematopoietic stem cells in the absence of stromal cells. J. Leukoc. Biol 96, 1165–1175. 10.1189/jlb.1TA0214-121RR. [PubMed: 25157026]

125. Shukla S, Langley MA, Singh J, Edgar JM, Mohtashami M, Zúñiga-Pflöcker JC, and Zandstra PW (2017). Progenitor T-cell differentiation from hematopoietic stem cells using Delta-like-4 and VCAM-1. Nat. Methods 14, 531–538. 10.1038/nmeth.4258. [PubMed: 28394335]

126. Iriguchi S, Yasui Y, Kawai Y, Arima S, Kunitomo M, Sato T, Ueda T, Minagawa A, Mishima Y, Yanagawa N, et al. (2021). A clinically applicable and scalable method to regenerate T-cells from iPSCs for off-the-shelf T-cell immunotherapy. Nat. Commun 12, 430. 10.1038/S41467-020-20658-3. [PubMed: 33462228]

127. Johnson LA, Heemskerk B, Powell DJJ, Cohen CJ, Morgan RA, Dudley ME, Robbins PF, and Rosenberg SA (2006). Gene transfer of tumor-reactive TCR confers both high avidity and tumor reactivity to nonreactive peripheral blood mononuclear cells and tumor-infiltrating lymphocytes. J. Immunol 177, 6548–6559. 10.4049/jimmunol.177.9.6548. [PubMed: 17056587]

128. Cohen CJ, Zheng Z, Bray R, Zhao Y, Sherman LA, Rosenberg SA, and Morgan RA (2005). Recognition of fresh human tumor by human peripheral blood lymphocytes transduced with a bicistronic retroviral vector encoding a murine anti-p53 TCR. J. Immunol 175, 5799–5808. 10.4049/jimmunol.175.9.5799. [PubMed: 16237072]

129. Parkhurst MR, Joo J, Riley JP, Yu Z, Li Y, Robbins PF, and Rosenberg SA (2009). Characterization of genetically modified T-cell receptors that recognize the CEA:691-699 peptide in the context of HLA-A2.1 on human colorectal cancer cells. Clin. cancer Res. an Off. J. Am. Assoc. Cancer Res 15, 169–180. 10.1158/1078-0432.CCR-08-1638.

130. Li Y, Moysey R, Molloy PE, Vuidepot A-L, Mahon T, Baston E, Dunn S, Liddy N, Jacob J, Jakobsen BK, et al. (2005). Directed evolution of human T-cell receptors with picomolar affinities by phage display. Nat. Biotechnol 23, 349–354. 10.1038/nbt1070. [PubMed: 15723046]

131. Varela-Rohena A, Molloy PE, Dunn SM, Li Y, Suhoski MM, Carroll RG, Milicic A, Mahon T, Sutton DH, Laugel B, et al. (2008). Control of HIV-1 immune escape by CD8 T cells expressing enhanced T-cell receptor. Nat. Med 14, 1390–1395. 10.1038/nm.1779. [PubMed: 18997777]

132. Adair JE, Kubek SP, and Kiem HP (2017). Hematopoietic Stem Cell Approaches to Cancer. Hematol. Oncol. Clin. North Am 31, 897–912. 10.1016/j.hoc.2017.06.012. [PubMed: 28895855]

133. Milone MC, and O'Doherty U (2018). Clinical use of lentiviral vectors. Leukemia 32, 1529–1541. 10.1038/s41375-018-0106-0. [PubMed: 29654266]

134. Yang L, and Baltimore D (2005). Long-term in vivo provision of antigen-specific T cell immunity by programming hematopoietic stem cells. Proc. Natl. Acad. Sci. U. S. A 102, 4518–4523. 10.1073/pnas.0500600102. [PubMed: 15758071]

135. Yang L, Qin XF, Baltimore D, and Van Parijs L (2002). Generation of functional antigen-specific T cells in defined genetic backgrounds by retrovirus-mediated expression of TCR cDNAS in hematopoietic precursor cells. Proc. Natl. Acad. Sci. U. S. A 99, 6204–6209. 10.1073/pnas.092154599. [PubMed: 11983911]

136. Basar R, Daher M, and Rezvani K (2020). Next-generation cell therapies: the emerging role of CAR-NK cells. Blood Adv. 4, 5868–5876. 10.1182/bloodadvances.2020002547. [PubMed: 33232480]

137. Qasim W, Zhan H, Samarasinghe S, Adams S, Amrolia P, Stafford S, Butler K, Rivat C, Wright G, Somana K, et al. (2017). Molecular remission of infant B-ALL after infusion of universal TALEN gene-edited CAR T cells. Sci. Transl. Med 9, 1–9. 10.1126/scitranslmed.aaj2013.

138. Webber BR, Lonetree C-L, Kluesner MG, Johnson MJ, Pomeroy EJ, Diers MD, Lahr WS, Draper GM, Slipek NJ, Smeester BA, et al. (2019). Highly efficient multiplex human T cell engineering without double-strand breaks using Cas9 base editors. Nat. Commun 10, 5222. 10.1038/s41467-019-13007-6. [PubMed: 31745080]

139. Zhao J, Lin Q, Song Y, and Liu D (2018). Universal CARs, universal T cells, and universal CAR T cells. J. Hematol. Oncol 11, 132. 10.1186/s13045-018-0677-2. [PubMed: 30482221]

140. Torikai H, Reik A, Soldner F, Warren EH, Yuen C, Zhou Y, Crossland DL, Huls H, Littman N, Zhang Z, et al. (2013). Toward eliminating HLA class i expression to generate universal cells from allogeneic donors. Blood 122, 1341–1349. 10.1182/blood-2013-03-478255. [PubMed: 23741009]

141. Xu X, Sun Q, Liang X, Chen Z, Zhang X, Zhou X, Li M, Tu H, Liu Y, Tu S, et al. (2019). Mechanisms of Relapse After CD19 CAR T-Cell Therapy for Acute Lymphoblastic Leukemia and Its Prevention and Treatment Strategies . Front. Immunol 10.

142. Zhang Y, Zhang X, Cheng C, Mu W, Liu X, Li N, Wei X, Liu X, Xia C, and Wang H (2017). CRISPR-Cas9 mediated LAG-3 disruption in CAR-T cells. Front. Med 11, 554–562. 10.1007/s11684-017-0543-6. [PubMed: 28625015]

143. Beane JD, Lee G, Zheng Z, Mendel M, Abate-Daga D, Bharathan M, Black M, Gandhi N, Yu Z, Chandran S, et al. (2015). Clinical Scale Zinc Finger Nuclease-mediated Gene Editing of PD-1 in Tumor Infiltrating Lymphocytes for the Treatment of Metastatic Melanoma. Mol. Ther 23, 1380–1390. 10.1038/mt.2015.71. [PubMed: 25939491]

144. Jang Y, Choi J, Park N, Kang J, Kim M, Kim Y, and Ju JH (2019). Development of immunocompatible pluripotent stem cells via CRISPR-based human leukocyte antigen engineering. Exp. Mol. Med 51, 1–11. 10.1038/s12276-018-0190-2.

145. Jung I-Y, Kim Y-Y, Yu H-S, Lee M, Kim S, and Lee J (2018). CRISPR/Cas9-Mediated Knockout of DGK Improves Antitumor Activities of Human T Cells. Cancer Res. 78, 4692–4703. 10.1158/0008-5472.CAN-18-0030. [PubMed: 29967261]

146. Liu X, Zhang Y, Cheng C, Cheng AW, Zhang X, Li N, Xia C, Wei X, Liu X, and Wang H (2017). CRISPR-Cas9-mediated multiplex gene editing in CAR-T cells. Cell Res. 27, 154–157. 10.1038/cr.2016.142. [PubMed: 27910851]

147. Provasi E, Genovese P, Lombardo A, Magnani Z, Liu P-Q, Reik A, Chu V, Paschon DE, Zhang L, Kuball J, et al. (2012). Editing T cell specificity towards leukemia by zinc finger nucleases and lentiviral gene transfer. Nat. Med 18, 807–815. 10.1038/nm.2700. [PubMed: 22466705]

148. Rupp LJ, Schumann K, Roybal KT, Gate RE, Ye CJ, Lim WA, and Marson A (2017). CRISPR/Cas9-mediated PD-1 disruption enhances anti-Tumor efficacy of human chimeric antigen receptor T cells. Sci. Rep 7, 1–10. 10.1038/s41598-017-00462-8. [PubMed: 28127051]

149. Shi L, Meng T, Zhao Z, Han J, Zhang W, Gao F, and Cai J (2017). CRISPR knock out CTLA-4 enhances the anti-tumor activity of cytotoxic T lymphocytes. Gene 636, 36–41. 10.1016/j.gene.2017.09.010. [PubMed: 28888577]

150. Thongsin N, and Wattanapanitch M (2021). CRISPR/Cas9 Ribonucleoprotein Complex-Mediated Efficient B2M Knockout in Human Induced Pluripotent Stem Cells (iPSCs). Methods Mol. Biol 10.1007/7651_2021_352.

151. Eyquem J, Mansilla-Soto J, Giavridis T, Van Der Stegen SJC, Hamieh M, Cunanan KM, Odak A, Gönen M, and Sadelain M (2017). Targeting a CAR to the TRAC locus with CRISPR/Cas9 enhances tumour rejection. Nature 543, 113–117. 10.1038/nature21405. [PubMed: 28225754]

152. MacLeod DT, Antony J, Martin AJ, Moser RJ, Hekele A, Wetzel KJ, Brown AE, Triggiano MA, Hux JA, Pham CD, et al. (2017). Integration of a CD19 CAR into the TCR Alpha Chain Locus Streamlines Production of Allogeneic Gene-Edited CAR T Cells. Mol. Ther 25, 949–961. 10.1016/j.ymthe.2017.02.005. [PubMed: 28237835]

153. Joung J, Konermann S, Gootenberg JS, Abudayyeh OO, Platt RJ, Brigham MD, Sanjana NE, and Zhang F (2017). Genome-scale CRISPR-Cas9 knockout and transcriptional activation screening. Nat. Protoc 12, 828–863. 10.1038/nprot.2017.016. [PubMed: 28333914]

154. Joung J, Kirchgatterer PC, Singh A, Cho JH, Nety SP, Larson RC, Macrae RK, Deasy R, Tseng Y-Y, Maus MV, et al. (2022). CRISPR activation screen identifies BCL-2 proteins and B3GNT2 as drivers of cancer resistance to T cell-mediated cytotoxicity. Nat. Commun 13, 1–14. 10.1038/s41467-022-29205-8. [PubMed: 34983933]

155. Larson RC, Kann MC, Bailey SR, Haradhvala NJ, Llopis PM, Bouffard AA, Scarfó I, Leick MB, Grauwet K, Berger TR, et al. (2022). CAR T cell killing requires the IFNγR pathway in solid but not liquid tumours. Nature 604. 10.1038/s41586-022-04585-5.

156. Sanson KR, Hanna RE, Hegde M, Donovan KF, Strand C, Sullender ME, Vaimberg EW, Goodale A, Root DE, Piccioni F, et al. (2018). Optimized libraries for CRISPR-Cas9 genetic screens with multiple modalities. Nat. Commun 9, 1–15. 10.1038/S41467-018-07901-8. [PubMed: 29317637]

157. Shifrut E, Carnevale J, Tobin V, Roth TL, Woo JM, Bui CT, Li PJ, Diolaiti ME, Ashworth A, and Marson A (2018). Genome-wide CRISPR Screens in Primary Human T Cells Reveal

Key Regulators of Immune Function. Cell 175, 1958–1971.e15. 10.1016/j.cell.2018.10.024. [PubMed: 30449619]

158. Wang D, Prager BC, Gimple RC, Aguilar B, Alizadeh D, Tang H, Lv D, Starr R, Brito A, Wu Q, et al. (2021). Crispr screening of car t cells and cancer stem cells reveals critical dependencies for cell-based therapies. Cancer Discov. 11, 1192–1211. 10.1158/2159-8290.CD-20-1243. [PubMed: 33328215]
159. Zhang X, Cheng C, Sun W, and Wang H (2020). Engineering T Cells Using CRISPR/Cas9 for Cancer Therapy BT - RNA Interference and CRISPR Technologies: Technical Advances and New Therapeutic Opportunities. In, Sioud M, ed. (Springer US), pp. 419–433. 10.1007/978-1-0716-0290-4_23.
160. Fujisaki H, Kakuda H, Shimasaki N, Imai C, Ma J, Lockey T, Eldridge P, Leung WH, and Campana D (2009). Expansion of highly cytotoxic human natural killer cells for cancer cell therapy. Cancer Res. 69, 4010–4017. 10.1158/0008-5472.CAN-08-3712. [PubMed: 19383914]
161. Heczey A, Courtney AN, Montalbano A, Robinson S, Liu K, Li M, Ghatwai N, Dakhova O, Liu B, Raveh-Sadka T, et al. (2020). Anti-GD2 CAR-NKT cells in patients with relapsed or refractory neuroblastoma: an interim analysis. Nat. Med 26, 1686–1690. 10.1038/S41591-020-1074-2. [PubMed: 33046868]
162. Liu D, Song L, Wei J, Courtney AN, Gao X, Marinova E, Guo L, Heczey A, Asgharzadeh S, Kim E, et al. (2012). IL-15 protects NKT cells from inhibition by tumor-associated macrophages and enhances antimetastatic activity. J. Clin. Invest 122, 2221–2233. 10.1172/JCI59535. [PubMed: 22565311]
163. Xu X, Huang W, Heczey A, Liu D, Guo L, Wood M, Jin J, Courtney AN, Liu B, Di Pierro EJ, et al. (2019). NKT cells coexpressing a GD2-specific chimeric antigen receptor and IL15 show enhanced in vivo persistence and antitumor activity against neuroblastoma. Clin. Cancer Res 25, 7126–7138. 10.1158/1078-0432.CCR-19-0421. [PubMed: 31484667]
164. Hla A, Class H.L. a, Borrego BF, Ulbrecht M, Weiss EH, Coligan JE, and Brooks AG (1998). Protection from Natural Killer Cell – mediated Lysis. 187.
165. Lee N, Llano M, Carretero M, Akiko-lshitani, Navarro F, López-Botet M, and Geraghty DE (1998). HLA-E is a major ligand for the natural killer inhibitory receptor CD94/NKG2A. Proc. Natl. Acad. Sci. U. S. A 95, 5199–5204. 10.1073/pnas.95.9.5199. [PubMed: 9560253]
166. Benjamin R, Graham C, Yallop D, Jozwik A, Mirci-Danicar OC, Lucchini G, Pinner D, Jain N, Kantarjian H, Boissel N, et al. (2020). Genome-edited, donor-derived allogeneic anti-CD19 chimeric antigen receptor T cells in paediatric and adult B-cell acute lymphoblastic leukaemia: results of two phase 1 studies. Lancet (London, England) 396, 1885–1894. 10.1016/S0140-6736(20)32334-5. [PubMed: 33308471]
167. Bedford P, Jy J, Collins L, and Keizer S (2018). Considering Cell Therapy Product “Good Manufacturing Practice” Status. Front. Med 5, 118. 10.3389/fmed.2018.00118.
168. Giancola R, Bonfini T, and Iacone A (2012). Cell therapy: cGMP facilities and manufacturing. Muscles. Ligaments Tendons J. 2, 243–247. [PubMed: 23738304]
169. Carpenter MK, and Rao MS (2015). Concise review: making and using clinically compliant pluripotent stem cell lines. Stem Cells Transl. Med 4, 381–388. 10.5966/sctm.2014-0202. [PubMed: 25722426]
170. Carpenter MK, Frey-Vasconcells J, and Rao MS (2009). Developing safe therapies from human pluripotent stem cells. Nat. Biotechnol 27, 606–613. 10.1038/nbt0709-606. [PubMed: 19587662]
171. Panchision DM (2013). Meeting report: using stem cells for biological and therapeutics discovery in mental illness, April 2012. Stem Cells Transl. Med 2, 217–222. 10.5966/sctm.2012-0149. [PubMed: 23408104]
172. Kleitman N, Rao MS, and Owens DF (2013). Pluripotent stem cells in translation: a Food and Drug Administration-National Institutes of Health collaboration. Stem Cells Transl. Med 2, 483–487. 10.5966/sctm.2013-0042. [PubMed: 23757505]
173. Frey-Vasconcells J, Whittlesey KJ, Baum E, and Feigal EG (2012). Translation of stem cell research: points to consider in designing preclinical animal studies. Stem Cells Transl. Med 1, 353–358. 10.5966/sctm.2012-0018. [PubMed: 23197814]

174. Brudno JN, and Kochenderfer JN (2019). Recent advances in CAR T-cell toxicity: Mechanisms, manifestations and management. Blood Rev. 34, 45–55. 10.1016/j.blre.2018.11.002. [PubMed: 30528964]

175. Zhou Y, Li M, Zhou K, Brown J, Tsao T, Cen X, Husman T, Bajpai A, Dunn ZS, and Yang L (2022). Engineering Induced Pluripotent Stem Cells for Cancer Immunotherapy. Cancers 14. 10.3390/cancers14092266.

176. Furukawa Y, Hamano Y, Shirane S, Kinoshita S, Azusawa Y, Ando J, Nakauchi H, and Ando M (2022). Advances in Allogeneic Cancer Cell Therapy and Future Perspectives on “Off-the-Shelf” T Cell Therapy Using iPSC Technology and Gene Editing. Cells 11. 10.3390/cells11020269.

177. Yamanaka S (2020). Pluripotent Stem Cell-Based Cell Therapy-Promise and Challenges. Cell Stem Cell 27, 523–531. 10.1016/j.stem.2020.09.014. [PubMed: 33007237]

178. Ben-David U, Siranosian B, Ha G, Tang H, Oren Y, Hinohara K, Strathdee CA, Dempster J, Lyons NJ, Burns R, et al. (2018). Genetic and transcriptional evolution alters cancer cell line drug response. Nature 560, 325–330. 10.1038/s41586-018-0409-3. [PubMed: 30089904]

179. Sato Y, Bando H, Di Piazza M, Gowing G, Herberts C, Jackman S, Leoni G, Libertini S, MacLachlan T, McBlane JW, et al. (2019). Tumorigenicity assessment of cell therapy products: The need for global consensus and points to consider. Cytotherapy 21, 1095–1111. 10.1016/j.jcyt.2019.10.001. [PubMed: 31711733]

180. Peruzzi L, and Eroğlu HE (2013). Karyotype asymmetry: again, how to measure and what to measure? Comp. Cytogenet 7, 1–9. 10.3897/CompCytogen.v7i1.4431. [PubMed: 24260685]

181. Seki T, Yuasa S, Oda M, Egashira T, Yae K, Kusumoto D, Nakata H, Tohyama S, Hashimoto H, Kodaira M, et al. (2010). Generation of induced pluripotent stem cells from human terminally differentiated circulating T cells. Cell Stem Cell 7, 11–14. 10.1016/j.stem.2010.06.003. [PubMed: 20621043]

182. Okita K, Ichisaka T, and Yamanaka S (2007). Generation of germline-competent induced pluripotent stem cells. Nature 448, 313–317. 10.1038/nature05934. [PubMed: 17554338]

183. Li HO, Zhu YF, Asakawa M, Kuma H, Hirata T, Ueda Y, Lee YS, Fukumura M, Iida A, Kato A, et al. (2000). A cytoplasmic RNA vector derived from nontransmissible Sendai virus with efficient gene transfer and expression. J. Virol 74, 6564–6569. 10.1128/jvi.74.14.6564-6569.2000. [PubMed: 10864670]

184. Yu J, Hu K, Smuga-Otto K, Tian S, Stewart R, Slukvin II, and Thomson JA (2009). Human induced pluripotent stem cells free of vector and transgene sequences. Science 324, 797–801. 10.1126/science.1172482. [PubMed: 19325077]

185. Mashima H, Zhang R, Kobayashi T, Tsukamoto H, Liu T, Iwama T, Hagiya Y, Yamamoto M, Fukushima S, Okada S, et al. (2021). Improved safety of induced pluripotent stem cell-derived antigen-presenting cell-based cancer immunotherapy. Mol. Ther. - Methods Clin. Dev 21, 171–179. 10.1016/j.omtm.2021.03.002. [PubMed: 33816647]

186. Karantalis V, Schulman IH, Balkan W, and Hare JM (2015). Allogeneic cell therapy: a new paradigm in therapeutics. Circ. Res. 116, 12–15. 10.1161/CIRCRESAHA.114.305495. [PubMed: 25552688]

187. Appelbaum FR (2001). Haematopoietic cell transplantation as immunotherapy. Nature 411, 385–389. 10.1038/35077251. [PubMed: 11357147]

188. Hill GR, Betts BC, Tkachev V, Kean LS, and Blazar BR (2021). Current Concepts and Advances in Graft-Versus-Host Disease Immunology. Annu. Rev. Immunol 39, 19–49. 10.1146/annurev-immunol-102119-073227. [PubMed: 33428454]

189. Martinez-Cibrian N, Zeiser R, and Perez-Simon JA (2021). Graft-versus-host disease prophylaxis: Pathophysiology-based review on current approaches and future directions. Blood Rev. 48, 100792. 10.1016/j.blre.2020.100792. [PubMed: 33386151]

190. Penack O, Marchetti M, Ruutu T, Aljurf M, Bacigalupo A, Bonifazi F, Ciceri F, Cornelissen J, Malladi R, Duarte RF, et al. (2020). Prophylaxis and management of graft versus host disease after stem-cell transplantation for haematological malignancies: updated consensus recommendations of the European Society for Blood and Marrow Transplantation. Lancet. Haematol 7, e157–e167. 10.1016/S2352-3026(19)30256-X. [PubMed: 32004485]

191. Sabry M, and Lowdell MW (2020). Killers at the crossroads: The use of innate immune cells in adoptive cellular therapy of cancer. Stem Cells Transl. Med 9, 974–984. 10.1002/sctm.19-0423. [PubMed: 32416056]
192. Lan F, Zeng D, Higuchi M, Higgins JP, and Strober S (2003). Host conditioning with total lymphoid irradiation and antithymocyte globulin prevents graft-versus-host disease: the role of CD1-reactive natural killer T cells. Biol. blood marrow Transplant. J. Am. Soc. Blood Marrow Transplant 9, 355–363. 10.1016/s1083-8791(03)00108-3.
193. Yamasaki S, Henzan H, Ohno Y, Yamanaka T, Iino T, Itou Y, Kuroiwa M, Maeda M, Kawano N, Kinukawa N, et al. (2003). Influence of transplanted dose of CD56+ cells on development of graft-versus-host disease in patients receiving G-CSF-mobilized peripheral blood progenitor cells from HLA-identical sibling donors. Bone Marrow Transplant. 32, 505–510. 10.1038/sj.bmt.1704165. [PubMed: 12942097]
194. Fishman JA (2017). Infection in Organ Transplantation. Am. J. Transplant 17, 856–879. 10.1111/ajt.14208. [PubMed: 28117944]
195. Holt CD (2017). Overview of Immunosuppressive Therapy in Solid Organ Transplantation. Anesthesiol. Clin 35, 365–380. 10.1016/j.anclin.2017.04.001. [PubMed: 28784214]
196. van Rood JJ, and Oudshoorn M (2008). Eleven million donors in Bone Marrow Donors Worldwide! Time for reassessment? Bone Marrow Transplant. 41, 1–9. 10.1038/sj.bmt.1705866. [PubMed: 17982505]
197. Hurley CK (2021). Naming HLA diversity: A review of HLA nomenclature. Hum. Immunol 82, 457–465. 10.1016/j.humimm.2020.03.005. [PubMed: 32307125]
198. Taylor CJ, Bolton EM, Pocock S, Sharpies LD, Pedersen RA, and Bradley JA (2005). Banking on human embryonic stem cells: estimating the number of donor cell lines needed for HLA matching. Lancet (London, England) 366, 2019–2025. 10.1016/S0140-6736(05)67813-0. [PubMed: 16338451]
199. Lanza R, Russell DW, and Nagy A (2019). Engineering universal cells that evade immune detection. Nat. Rev. Immunol 19, 723–733. 10.1038/s41577-019-0200-1. [PubMed: 31417198]
200. Ordikhani F, Pothula V, Sanchez-Tarjuelo R, Jordan S, and Ochando J (2020). Macrophages in Organ Transplantation . Front. Immunol 11.
201. Deuse T, Hu X, Gravina A, Wang D, Tediashvili G, De C, Thayer WO, Wahl A, Garcia JV, Reichenspurner H, et al. (2019). Hypoimmunogenic derivatives of induced pluripotent stem cells evade immune rejection in fully immunocompetent allogeneic recipients. Nat. Biotechnol 37, 252–258. 10.1038/s41587-019-0016-3. [PubMed: 30778232]
202. Wang B, Iriguchi S, Waseda M, Ueda N, Ueda T, Xu H, Minagawa A, Ishikawa A, Yano H, Ishi T, et al. (2021). Generation of hypoimmunogenic T cells from genetically engineered allogeneic human induced pluripotent stem cells. Nat. Biomed. Eng 5, 429–440. 10.1038/s41551-021-00730-z. [PubMed: 34002062]
203. Holstein SA, and Lunning MA (2020). CAR T-Cell Therapy in Hematologic Malignancies: A Voyage in Progress. Clin. Pharmacol. Ther 107, 112–122. 10.1002/cpt.1674. [PubMed: 31622496]
204. Han D, Xu Z, Zhuang Y, Ye Z, and Qian Q (2021). Current Progress in CAR-T Cell Therapy for Hematological Malignancies. J. Cancer 12, 326–334. 10.7150/jca.48976. [PubMed: 33391429]
205. Shah NN, Maatman T, Hari P, and Johnson B (2019). Multi Targeted CAR-T Cell Therapies for B-Cell Malignancies . Front. Oncol 9.
206. Newick K, O'Brien S, Moon E, and Albelda SM (2017). CAR T Cell Therapy for Solid Tumors. Annu. Rev. Med 68, 139–152. 10.1146/annurev-med-062315-120245. [PubMed: 27860544]
207. Liu X, Zhang N, and Shi H (2017). Driving better and safer HER2-specific CARs for cancer therapy. Oncotarget 8, 62730–62741. 10.18632/oncotarget.17528. [PubMed: 28977984]
208. Navai SA, Derenzo C, Joseph S, Sanber K, Byrd T, Zhang H, Mata M, Gerken C, Shree A, Mathew PR, et al. (2019). Abstract LB-147: Administration of HER2-CAR T cells after lymphodepletion safely improves T cell expansion and induces clinical responses in patients with advanced sarcomas. Cancer Res. 79, LB-147-LB-147. 10.1158/1538-7445.AM2019-LB-147.

209. Marofi F, Motavalli R, Safonov VA, Thangavelu L, Yumashev AV, Alexander M, Shomali N, Chartrand MS, Pathak Y, Jarahian M, et al. (2021). CAR T cells in solid tumors: challenges and opportunities. Stem Cell Res. Ther 12, 81. 10.1186/s13287-020-02128-1. [PubMed: 33494834]

210. Cordoba S, Onuoha S, Thomas S, Pignataro DS, Hough R, Ghorashian S, Vora A, Bonney D, Veys P, Rao K, et al. (2021). CAR T cells with dual targeting of CD19 and CD22 in pediatric and young adult patients with relapsed or refractory B cell acute lymphoblastic leukemia: a phase 1 trial. Nat. Med 27, 1797–1805. 10.1038/s41591-021-01497-1. [PubMed: 34642489]

211. van der Schans JJ, van de Donk NWCJ, and Mutis T (2020). Dual Targeting to Overcome Current Challenges in Multiple Myeloma CAR T-Cell Treatment . Front. Oncol 10.

212. Hirabayashi K, Du H, Xu Y, Shou P, Zhou X, Fucá G, Landoni E, Sun C, Chen Y, Savoldo B, et al. (2021). Dual-targeting CAR-T cells with optimal co-stimulation and metabolic fitness enhance antitumor activity and prevent escape in solid tumors. Nat. Cancer 2, 904–918. 10.1038/s43018-021-00244-2. [PubMed: 34746799]

213. Zah E, Nam E, Bhuvan V, Tran U, Ji BY, Gosliner SB, Wang X, Brown CE, and Chen YY (2020). Systematically optimized BCMA/CS1 bispecific CAR-T cells robustly control heterogeneous multiple myeloma. Nat. Commun 11, 2283. 10.1038/S41467-020-16160-5. [PubMed: 32385241]

214. Roybal KT, Williams JZ, Morsut L, Rupp LJ, Kolinko I, Choe JH, Walker WJ, McNally KA, and Lim WA (2016). Engineering T Cells with Customized Therapeutic Response Programs Using Synthetic Notch Receptors. Cell 167, 419–432.e16. 10.1016/j.cell.2016.09.011. [PubMed: 27693353]

215. Srivastava S, Salter AI, Liggitt D, Yechan-Gunja S, Sarvothama M, Cooper K, Smythe KS, Dudakov JA, Pierce RH, Rader C, et al. (2019). Logic-Gated ROR1 Chimeric Antigen Receptor Expression Rescues T Cell-Mediated Toxicity to Normal Tissues and Enables Selective Tumor Targeting. Cancer Cell 35, 489–503.e8. 10.1016/j.ccell.2019.02.003. [PubMed: 30889382]

216. H. CJ, B. WP, S. SM, D. GR, W. LA, A. KN, M. DK, Wei Y, A. CD, Anna C, et al. (2021). SynNotch-CAR T cells overcome challenges of specificity, heterogeneity, and persistence in treating glioblastoma. Sci. Transl. Med 13, eabe7378. 10.1126/scitranslmed.abe7378. [PubMed: 33910979]

217. I. SA, Anusha R, J. KJ, G. IR, A. SS, Isabel L, L. TM, Vishaka M, Valentin V, Daniel S, et al. (2021). Comparative analysis of TCR and CAR signaling informs CAR designs with superior antigen sensitivity and in vivo function. Sci. Signal 14, eabe2606. 10.1126/scisignal.abe2606. [PubMed: 34429382]

218. Mansilla-Soto J, Eyquem J, Haubner S, Hamieh M, Feucht J, Paillon N, Zucchetti AE, Li Z, Sjöstrand M, Lindenbergh PL, et al. (2022). HLA-independent T cell receptors for targeting tumors with low antigen density. Nat. Med 28, 345–352. 10.1038/S41591-021-01621-1. [PubMed: 35027758]

219. Dixon KJ, Wu J, and Walcheck B (2021). Engineering Anti-Tumor Monoclonal Antibodies and Fc Receptors to Enhance ADCC by Human NK Cells. Cancers 13. 10.3390/cancers13020312.

220. Jing Y, Ni Z, Wu J, Higgins L, Markowski TW, Kaufman DS, and Walcheck B (2015). Identification of an ADAM17 cleavage region in human CD16 (FcγRIII) and the engineering of a non-cleavable version of the receptor in NK cells. PLoS One 10, e0121788. 10.1371/journal.pone.0121788. [PubMed: 25816339]

221. Koene HR, Kleijer M, Algra J, Roos D, von dem Borne AE, and de Haas M (1997). Fc gammaRIIIa-158V/F polymorphism influences the binding of IgG by natural killer cell Fc gammaRIIIa, independently of the Fc gammaRIIIa-48L/R/H phenotype. Blood 90, 1109–1114. [PubMed: 9242542]

222. Saito H, Okita K, Fusaki N, Sabel MS, Chang AE, and Ito F (2016). Reprogramming of Melanoma Tumor-Infiltrating Lymphocytes to Induced Pluripotent Stem Cells. Stem Cells Int. 2016, 8394960. 10.1155/2016/8394960. [PubMed: 27057178]

223. Woo S-R, Corrales L, and Gajewski TF (2015). Innate Immune Recognition of Cancer. Annu. Rev. Immunol 33, 445–474. 10.1146/annurev-immunol-032414-112043. [PubMed: 25622193]

224. Deng J, and Yin H (2022). Gamma delta (γδ) T cells in cancer immunotherapy; where it comes from, where it will go? Eur. J. Pharmacol 919, 174803. 10.1016/j.ejphar.2022.174803. [PubMed: 35131312]

225. Zeng J, Tang SY, and Wang S (2019). Derivation of mimetic γδ T cells endowed with cancer recognition receptors from reprogrammed γδ T cell. PLoS One 14, e0216815. [PubMed: 31071196]

226. Chan JD, Lai J, Slaney CY, Kallies A, Beavis PA, and Darcy PK (2021). Cellular networks controlling T cell persistence in adoptive cell therapy. Nat. Rev. Immunol 21, 769–784. 10.1038/s41577-021-00539-6. [PubMed: 33879873]

227. McLellan AD, and Ali Hosseini Rad SM (2019). Chimeric antigen receptor T cell persistence and memory cell formation. Immunol. Cell Biol 97, 664–674. 10.1111/imcb.12254. [PubMed: 31009109]

228. Yeku OO, and Brentjens RJ (2016). Armored CAR T-cells: utilizing cytokines and pro-inflammatory ligands to enhance CAR T-cell anti-tumour efficacy. Biochem. Soc. Trans 44, 412–418. 10.1042/BST20150291. [PubMed: 27068948]

229. Ohteki T (2002). Critical role for IL-15 in innate immunity. Curr. Mol. Med 2, 371–380. 10.2174/1566524023362519. [PubMed: 12108948]

230. Liu E, Tong Y, Dotti G, Shaim H, Savoldo B, Mukherjee M, Orange J, Wan X, Lu X, Reynolds A, et al. (2018). Cord blood NK cells engineered to express IL-15 and a CD19-targeted CAR show long-term persistence and potent antitumor activity. Leukemia 32, 520–531. 10.1038/leu.2017.226. [PubMed: 28725044]

231. Makkouk A, Yang XC, Barca T, Lucas A, Turkoz M, Wong JTS, Nishimoto KP, Brodey MM, Tabrizizad M, Gundurao SRY, et al. (2021). Off-the-shelf Vδ1 gamma delta T cells engineered with glypican-3 (GPC-3)-specific chimeric antigen receptor (CAR) and soluble IL-15 display robust antitumor efficacy against hepatocellular carcinoma. J. Immunother. cancer 9. 10.1136/jitc-2021-003441.

232. Allen AG, Pattali R, Izzo KM, Getgano JA, Wasko KM, Blaha LC, Zuris JA, Zhang K, Shearman MS, and Chang K-H (2021). A Bicistronic Vector Expressing CD16 and a Membrane Bound IL-15 Construct in iPSC Derived NK Cells Increased Cytotoxicity and Persistence. Blood 138, 4809. 10.1182/blood-2021-153258.

233. Christodoulou I, Koldobskiy M, Ho WJ, Marple A, Ravich WJ, Rahnama R, and Bonifant CL (2021). Engineered Interleukin-15 Autocrine Signaling Invigorates Anti-CD123CAR-NK Cells. Blood 138, 2806. 10.1182/blood-2021-146609.

234. Gerew A, Sexton S, Wasko KM, Shearman MS, Zhang K, Chang K-H, and Khan SQ (2021). Deletion of CISH and TGFβR2 in iPSC-Derived NK Cells Promotes High Cytotoxicity and Enhances In Vivo Tumor Killing. Blood 138, 2780. 10.1182/blood-2021-150731.

235. Lynn RC, Weber EW, Sotillo E, Gennert D, Xu P, Good Z, Anbunathan H, Lattin J, Jones R, Tieu V, et al. (2019). c-Jun overexpression in CAR T cells induces exhaustion resistance. Nature 576, 293–300. 10.1038/s41586-019-1805-z. [PubMed: 31802004]

236. Seo H, González-Avalos E, Zhang W, Ramchandani P, Yang C, Lio C-WJ, Rao A, and Hogan PG (2021). BATF and IRF4 cooperate to counter exhaustion in tumor-infiltrating CAR T cells. Nat. Immunol 22, 983–995. 10.1038/s41590-021-00964-8. [PubMed: 34282330]

237. Koyanagi-Aoi M, Ohnuki M, Takahashi K, Okita K, Noma H, Sawamura Y, Teramoto I, Narita M, Sato Y, Ichisaka T, et al. (2013). Differentiation-defective phenotypes revealed by large-scale analyses of human pluripotent stem cells. Proc. Natl. Acad. Sci. U. S. A 110, 20569–20574. 10.1073/pnas.1319061110. [PubMed: 24259714]

238. Osafune K, Caron L, Borowiak M, Martinez RJ, Fitz-Gerald CS, Sato Y, Cowan CA, Chien KR, and Melton DA (2008). Marked differences in differentiation propensity among human embryonic stem cell lines. Nat. Biotechnol 26, 313–315. 10.1038/nbt1383. [PubMed: 18278034]

239. Yamanaka S (2012). Induced pluripotent stem cells: past, present, and future. Cell Stem Cell 10, 678–684. 10.1016/j.stem.2012.05.005. [PubMed: 22704507]

240. Nishizawa M, Chonabayashi K, Nomura M, Tanaka A, Nakamura M, Inagaki A, Nishikawa M, Takei I, Oishi A, Tanabe K, et al. (2016). Epigenetic Variation between Human Induced Pluripotent Stem Cell Lines Is an Indicator of Differentiation Capacity. Cell Stem Cell 19, 341–354. 10.1016/j.stem.2016.06.019. [PubMed: 27476965]

241. Theunissen TW, Powell BE, Wang H, Mitalipova M, Faddah DA, Reddy J, Fan ZP, Maetzel D, Ganz K, Shi L, et al. (2014). Systematic identification of culture conditions for

induction and maintenance of naive human pluripotency. Cell Stem Cell 15, 471–487. 10.1016/j.stem.2014.07.002. [PubMed: 25090446]

242. Takashima Y, Guo G, Loos R, Nichols J, Ficz G, Krueger F, Oxley D, Santos F, Clarke J, Mansfield W, et al. (2014). Resetting transcription factor control circuitry toward ground-state pluripotency in human. Cell 158, 1254–1269. 10.1016/j.cell.2014.08.029. [PubMed: 25215486]
243. Di Stefano B, Ueda M, Sabri S, Brumbaugh J, Huebner AJ, Sahakyan A, Clement K, Clowers KJ, Erickson AR, Shioda K, et al. (2018). Reduced MEK inhibition preserves genomic stability in naive human embryonic stem cells. Nat. Methods 15, 732–740. 10.1038/s41592-018-0104-1. [PubMed: 30127506]
244. Theunissen TW, Friedli M, He Y, Planet E, O'Neil RC, Markoulaki S, Pontis J, Wang H, Iouranova A, Imbeault M, et al. (2016). Molecular Criteria for Defining the Naive Human Pluripotent State. Cell Stem Cell 19, 502–515. 10.1016/j.stem.2016.06.011. [PubMed: 27424783]
245. Imai C, Iwamoto S, and Campana D (2005). Genetic modification of primary natural killer cells overcomes inhibitory signals and induces specific killing of leukemic cells. Blood 106, 376–383. 10.1182/blood-2004-12-4797. [PubMed: 15755898]
246. Ueda T, Kumagai A, Iriguchi S, Yasui Y, Miyasaka T, Nakagoshi K, Nakane K, Saito K, Takahashi M, Sasaki A, et al. (2020). Non-clinical efficacy, safety and stable clinical cell processing of induced pluripotent stem cell-derived anti-glypican-3 chimeric antigen receptor-expressing natural killer/innate lymphoid cells. Cancer Sci. 111, 1478–1490. 10.1111/cas.14374. [PubMed: 32133731]
247. Hong D, Patel S, Patel M, Musni K, Anderson M, Cooley S, Valamehr B, and Chu W (2020). 380 Preliminary results of an ongoing phase I trial of FT500, a first-in-class, off-the-shelf, induced pluripotent stem cell (iPSC) derived natural killer (NK) cell therapy in advanced solid tumors. J. Immunother. Cancer 8, A231 LP–A232. 10.1136/jitc-2020-SITC2020.0380.
248. Cichocki F, Goodridge JP, Bjordahl R, Gaidarova S, Mahmood S, Abujarour R, Davis Z, Wang H, Tuininga K, Kodal B, et al. (2021). Off-the-Shelf, Multiplexed-Engineered iPSC-Derived NK Cells Mediate Potent Multi-Antigen Targeting of B-Cell Malignancies with Reduced Cytotoxicity Against Healthy B Cells. Blood 138, 407. 10.1182/blood-2021-148654.
249. Diem MC (2021). Engineered stem cells power expansive range of cancer therapies. Biopharma Deal., 4985.
250. Luevano M, Madrigal A, and Saudemont A (2012). Generation of natural killer cells from hematopoietic stem cells in vitro for immunotherapy. Cell. Mol. Immunol 9, 310–320. 10.1038/cmi.2012.17. [PubMed: 22705914]
251. Shah N, Li L, McCarty J, Kaur I, Yvon E, Shaim H, Muftuoglu M, Liu E, Orlowski RZ, Cooper L, et al. (2017). Phase I study of cord blood-derived natural killer cells combined with autologous stem cell transplantation in multiple myeloma. Br. J. Haematol 177, 457–466. 10.1111/bjh.14570. [PubMed: 28295190]
252. Vivier E, Ugolini S, Blaise D, Chabannon C, and Brossay L (2012). Targeting natural killer cells and natural killer T cells in cancer. Nat. Rev. Immunol 12, 239–252. 10.1038/nri3174. [PubMed: 22437937]
253. Ando M, Nishimura T, Yamazaki S, Yamaguchi T, Kawana-Tachikawa A, Hayama T, Nakauchi Y, Ando J, Ota Y, Takahashi S, et al. (2015). A Safeguard System for Induced Pluripotent Stem Cell-Derived Rejuvenated T Cell Therapy. Stem cell reports 5, 597–608. 10.1016/j.stemcr.2015.07.011. [PubMed: 26321144]
254. Minagawa A, Yoshikawa T, Yasukawa M, Hotta A, Kunitomo M, Iriguchi S, Takiguchi M, Kassai Y, Imai E, Yasui Y, et al. (2018). Enhancing T Cell Receptor Stability in Rejuvenated iPSC-Derived T Cells Improves Their Use in Cancer Immunotherapy. Cell Stem Cell 23, 850–858.e4. 10.1016/j.stem.2018.10.005. [PubMed: 30449714]
255. Sadeqi Nezhad M, Abdollahpour-Alitappeh M, Rezaei B, Yazdanifar M, and Seifalian AM (2021). Induced Pluripotent Stem Cells (iPSCs) Provide a Potentially Unlimited T Cell Source for CAR-T Cell Development and Off-the-Shelf Products. Pharm. Res 38, 931–945. 10.1007/s11095-021-03067-z. [PubMed: 34114161]
256. Wang Z, McWilliams-Koeppen HP, Reza H, Ostberg JR, Chen W, Wang X, Huynh C, Vyas V, Chang W-C, Starr R, et al. (2022). 3D-organoid culture supports differentiation of human

CAR+ iPSCs into highly functional CAR T cells. Cell Stem Cell 29, 515–527.e8. 10.1016/j.stem.2022.02.009. [PubMed: 35278370]

257. Jing R, Scarfo I, Najia MA, Lummertz da Rocha E, Han A, Sanborn M, Bingham T, Kubaczka C, Jha DK, Falchetti M, et al. (2022). EZH1 repression generates mature iPSC-derived CAR T cells with enhanced antitumor activity. Cell Stem Cell 29, 1181–1196.e6. 10.1016/j.stem.2022.06.014. [PubMed: 35931029]
258. Montoya CJ, Pollard D, Martinson J, Kumari K, Wasserfall C, Mulder CB, Rugeles MT, Atkinson MA, Landay AL, and Wilson SB (2007). Characterization of human invariant natural killer T subsets in health and disease using a novel invariant natural killer T cell-clonotypic monoclonal antibody, 6B11. Immunology 122, 1–14. 10.1111/j.1365-2567.2007.02647.x. [PubMed: 17662044]
259. Li Y-R, Zeng S, Dunn ZS, Zhou Y, Li Z, Yu J, Wang Y-C, Ku J, Cook N, Kramer A, et al. (2022). Off-the-shelf third-party HSC-engineered iNKT cells for ameliorating GvHD while preserving GvL effect in the treatment of blood cancers. iScience 25, 104859. 10.1016/j.isci.2022.104859. [PubMed: 36034226]
260. Li Y-R, Brown J, Yu Y, Lee D, Zhou K, Dunn ZS, Hon R, Wilson M, Kramer A, Zhu Y, et al. (2022). Targeting Immunosuppressive Tumor-Associated Macrophages Using Innate T Cells for Enhanced Antitumor Reactivity. Cancers 14. 10.3390/cancers14112749.
261. Metelitsa LS (2011). Anti-tumor potential of type-I NKT cells against CD1d-positive and CD1d-negative tumors in humans. Clin. Immunol 140, 119–129. 10.1016/j.clim.2010.10.005. [PubMed: 21095162]
262. Neff Newitt V (2022). The Incredible Story of Emily Whitehead & CAR T-Cell Therapy. Oncol. Times 44.
263. Johansson M, Ulfenborg B, Andersson CX, Heydarkhan-Hagvall S, Jeppsson A, Sartipy P, and Synnergren J (2022). Multi-Omics Characterization of a Human Stem Cell-Based Model of Cardiac Hypertrophy. Life (file///Users/yanruideli/Downloads/35487213.nbibBasel, Switzerland) 12. 10.3390/life12020293.
264. Brooks IR, Garrone CM, Kerins C, Kiar CS, Syntaka S, Xu JZ, Spagnoli FM, and Watt FM (2022). Functional genomics and the future of iPSCs in disease modeling. Stem cell reports 17, 1033–1047. 10.1016/j.stemcr.2022.03.019. [PubMed: 35487213]
265. Bayat Mokhtari R, Homayouni TS, Baluch N, Morgatskaya E, Kumar S, Das B, and Yeger H (2017). Combination therapy in combating cancer. Oncotarget 8, 38022–38043. 10.18632/oncotarget.16723. [PubMed: 28410237]

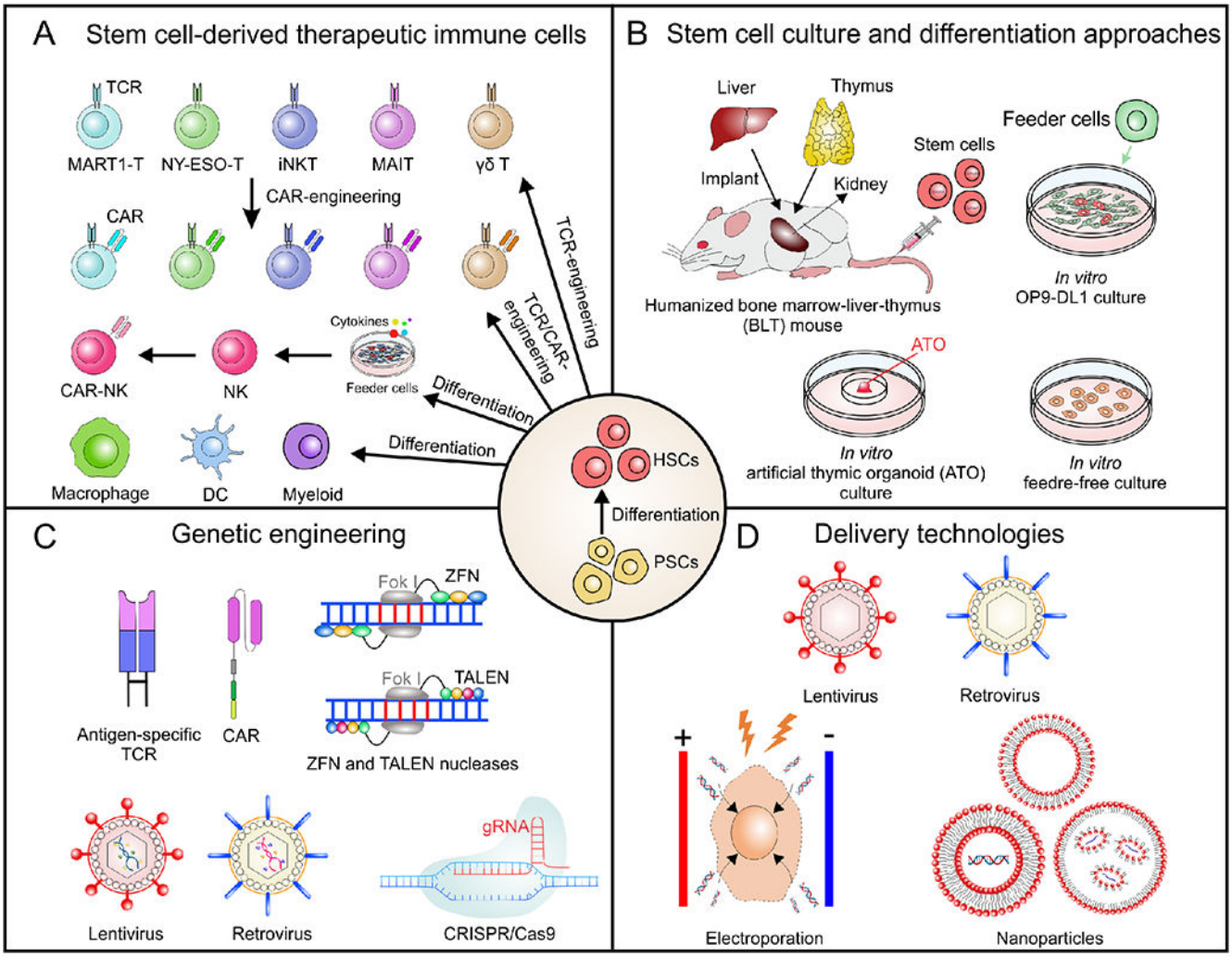

Figure 1. Stem cell engineering technologies, approaches, and therapeutic immune cells

(A) HSCs and PSCs can be engineered and differentiated into a variety of immune cells, such as conventional αβ T cells, innate T (i.e., iNKT, MAIT and γδ T) cells, NK cells, macrophages, dendritic cells, and myeloid cells. These immune cells could be further engineered with CARs to enhance their tumor targeting capacity.

(B) Various stem cell differentiation culture systems have been developed, such as a humanized Bone Marrow-Liver-Thymus (BLT) mouse model, *in vitro* feeder-dependent OP9-DL and artificial thymic organoid (ATO) cultures, and *in vitro* feeder-free cultures.

(C) Genetic engineering strategies have been explored in stem cells and immune cells for antitumor applications including CAR and TCR engineering, via gene editing using CRISPR/Cas9, designer nucleases like ZFN and TALEN, and viral vectors.

(D) In addition to lentiviral or retroviral transduction, delivery systems such as electroporation and nanoparticles achieve stable and efficient gene delivery to stem cells and their derivative immune cells.

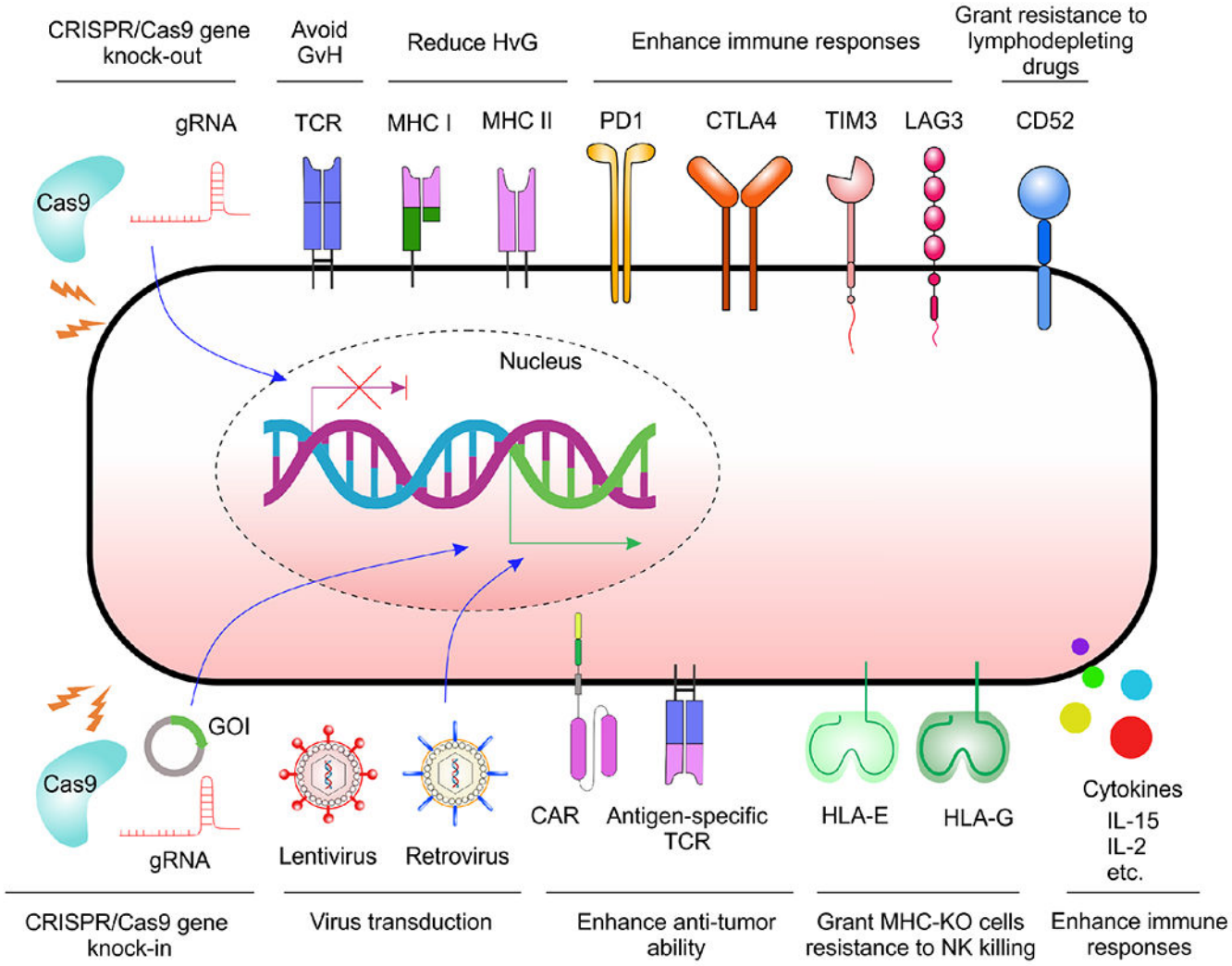

Figure 2. Applications of viral vector transduction and CRISPR/Cas9 gene-editing in stem-cell-based immune engineering

Lentiviral or retroviral-vector-mediated delivery of CARs, antigen-specific TCRs, and immune-enhanced genes (e.g., IL-2, IL-15) enhance the antitumor response of immune cells. CRISPR/Cas9 gene editing enables multiple gene knockouts to avoid graft-versus-host-disease (GvHD) (e.g., knockout of TCR), reduce allorejection (e.g., knockout of MHC I and MHC II), and enhance the immune response (e.g., knockout of immune checkpoint proteins such as PD-1, CTLA-4, TIM-3, and LAG-3). In addition, CRISPR/Cas9 allows the site-specific knock-in of genes of interest in target cells.

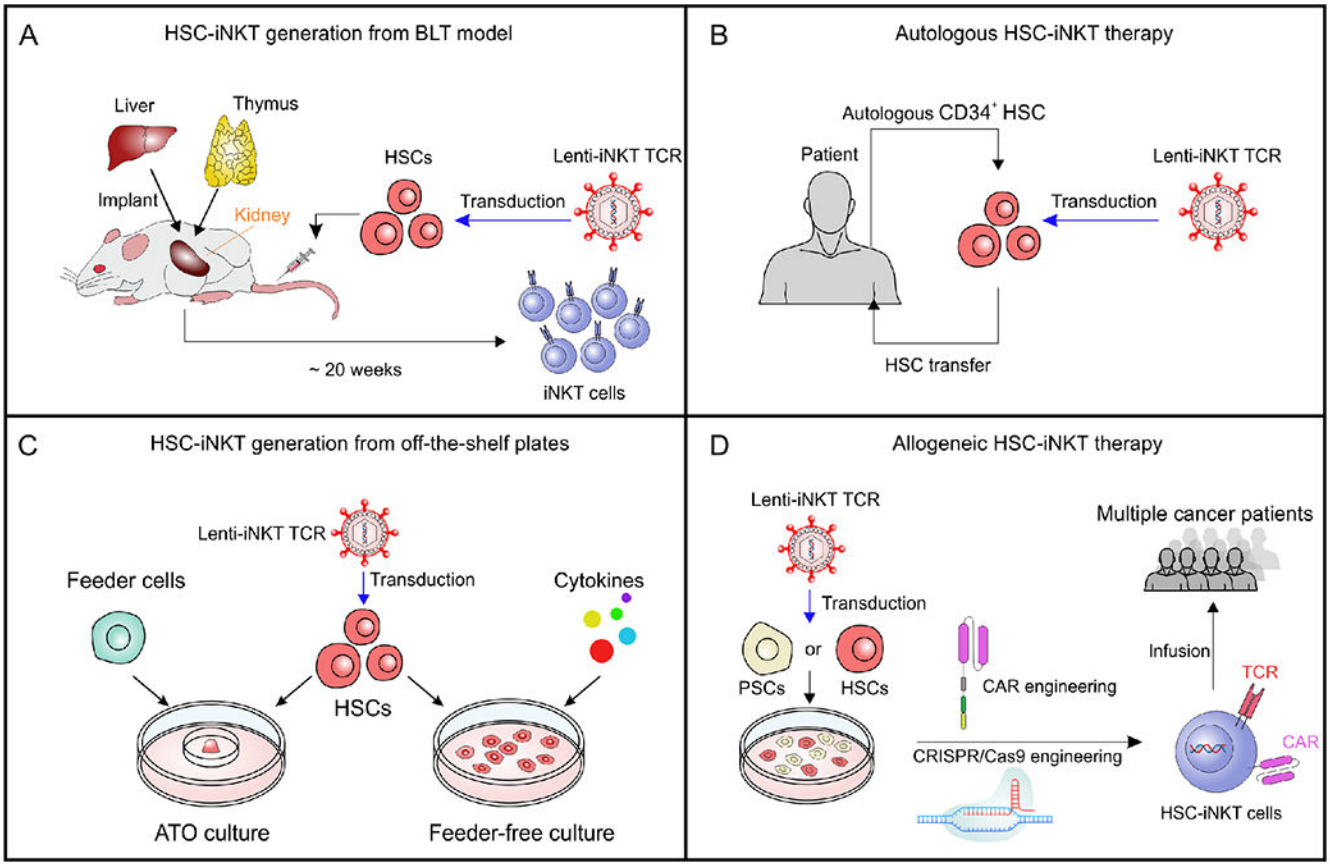

Figure 3. Development of HSC-engineered iNKT (HSC-iNKT) cell therapies for cancer

HSC-iNKT cells are presented as an immune cell example. The different stem cell differentiation and culture strategies could be easily applied to generate other TCR-engineered T cells, such as MAIT, γδ, and antigen-specific αβ T cells. The proposed autologous and allogenic cell therapy could also use other TCR-engineered T cells as cell carriers, depending on the tumor types.

(A) Generation of HSC-iNKT cells in a Bone Marrow-Liver-Thymus (BLT) humanized mouse model.

(B) Development of an autologous HSC-iNKT cell therapy for cancer.

(C) Generation of allogeneic HSC-iNKT cells in an ATO or a feeder-free culture.

(D) Development of an allogeneic HSC-iNKT cell therapy for cancer. CAR engineering and CRISPR/Cas9 gene editing could be incorporated into HSC-iNKT cells to enhance their immune response and safety profile.