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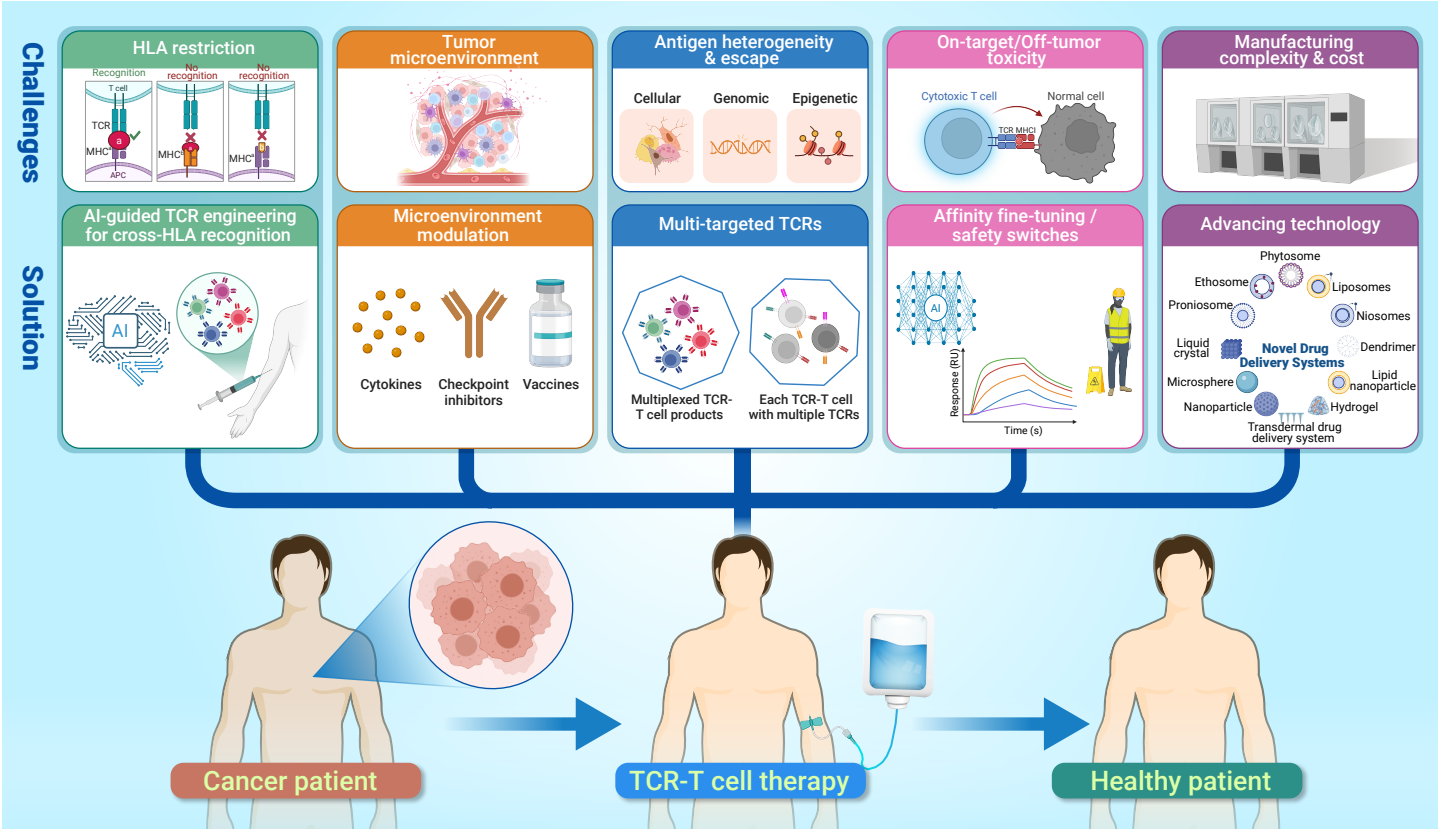
GRAPHICAL ABSTRACT



PUBLIC SUMMARY

- TCR-T (T-cell receptor-engineered T) therapy targets hidden internal cancer proteins as a breakthrough.
- The recent FDA approval of afami-cel marks a major milestone for advancing this personalized immunotherapy.
- Researchers leverage AI (Artificial Intelligence) and gene editing to overcome complex biological hurdles.
- New strategies aim to improve T-cell fitness and persistence within the suppressive tumor microenvironment.
- Combining TCR-T with other drugs could revolutionize care for patients with resistant solid malignancies.

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A prospective for TCR-T therapy in solid tumors: Pushing the boundaries of immunotherapy

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T-cell receptor-engineered T (TCR-T) cell therapy stands out as an innovative and promising approach in cancer immunotherapy, particularly for the treatment of solid tumors. This therapy differs from the well-established chimeric antigen receptor (CAR)-T therapy, which has revolutionized the treatment of hematologic malignancies. TCR-T therapy holds a unique edge in its capacity to recognize and target intracellular antigens, thereby broadening the spectrum of potential tumor-specific antigens that can be harnessed for therapeutic purposes. The recent FDA approval of afami-cel for synovial sarcoma marks a significant milestone in the journey of TCR-T therapy. This approval underscores the clinical potential of TCR-T in targeting solid tumors and opens up new avenues for therapeutic interventions. However, the broader clinical translation of TCR-T therapy is constrained by several critical barriers: the inherent requirement for HLA matching, the potent immunosuppressive tumor microenvironment, antigen heterogeneity leading to immune escape, and the risk of fatal on-target/off-tumor toxicities. In this review, we delineate the distinctive mechanistic advantages of TCR-T cells and provide a comprehensive analysis of the biological and technical hurdles encountered in solid tumor treatment. We synthesize findings from recent clinical trials and evaluate emerging optimization strategies, including TCR affinity fine-tuning and combinatorial approaches to modulate the immune landscape. Addressing these challenges is essential to provide a roadmap for enhancing the efficacy and safety of TCR-T therapies, ultimately facilitating their widespread adoption to improve survival outcomes for patients with refractory solid malignancies.

CURRENT LANDSCAPE AND UNIQUE ADVANTAGE

Cancer poses a significant global public health threat, marked by persistently elevated morbidity and mortality rates.[1] Recent advancements in medical research have led to substantial improvements in cancer treatment, resulting in enhanced patient survival rates. Solid tumors, which represent approximately 90% of adult cancers and originate primarily from epithelial tissues, remain a major focus of research efforts.[1] While conventional treatments such as surgery, radiation therapy, and chemotherapy can be effective for localized tumors, achieving complete eradication of residual cancer cells remains challenging.[2] This is partly attributed to the presence of micrometastases at the time of diagnosis in many patients, as well as treatment-induced immune system suppression and organ dysfunction.[2]

The immune system plays a pivotal role in the recognition and elimination of cancer.[3] Advances in understanding the tumor microenvironment and immune evasion mechanisms have fueled the development of innovative therapeutic strategies, notably adoptive cell immunotherapy.[4] This innovative treatment strategy involves *ex vivo* modification of a patient's own T cells, which are then reinfused to enhance their anti-tumor capabilities.[5] Adoptive cell immunotherapy, with chimeric antigen receptor (CAR)-T and T cell receptor (TCR)-T cell therapy as its flagship treatments, has emerged as a pivotal therapeutic modality in the ongoing fight against cancer.[6-8]

TCR-T therapy involves the genetic engineering of a patient's own T cells with a high-affinity, tumor-specific TCR gene. This process enables the modified T cells to specifically identify and eliminate tumor cells expressing a particular antigen.[9] Unlike CAR-T therapy, which employs a CAR molecule derived from B-cell receptors (BCRs) to directly recognize cell surface antigens in an MHC-independent manner, TCR-T therapy harnesses the natural TCR-MHC (major histocompatibility complex) interaction (Table 1).[9] While CARs are limited to surface-bound proteins that BCRs can natively bind, the TCR-T cells recognize endogenous intracellular antigens, such as mutated proteins, viral antigens, and cancer-testis antigens, which are presented on the cell surface by MHC molecules (Table 1).[9,10] By leveraging this unique recognition mechanism, TCR-T therapy can target the vast majority of intracellular proteins involved in tumor development, thereby significantly broadening the spectrum of druggable antigens and offering new therapeutic possibilities for cancer treatment.

CAR-T therapy has achieved remarkable success in hematologic malignancies, demonstrating complete remission rates of up to 80-90% in relapsed/refractory B-cell malignancies, but faces challenges in solid tumors.[7] This is mainly attributed to the lack of specific surface antigens on solid tumor cells, which frequently leads to "on-target, off-tumor" toxicities in vital organs, and the difficulty for CAR-T cells to penetrate and survive in the complex tumor microenvironment.[11] Specifically, the physical density of the extracellular matrix (ECM) acts as a "molecular fortress" to impede T-cell infiltration. Once inside the tumor core, these CAR-T cells are further neutralized by the immunosuppressive tumor microenvironment (TME), characterized by hypoxia, acidic pH, and inhibitory signaling from PD-L1 or TGF-β.[11]

In contrast, TCR-T therapy demonstrates several unique advantages in solid tumors. First, TCR-T therapy offers a broader range of targets compared to CAR-T therapy by targeting peptide fragments of intracellular proteins presented on the cell surface by MHC molecules (Table 1). Nearly 90% of a tumor cell's proteome is intracellular,[12] allowing TCR-T therapy to target a much wider variety of antigens, including those derived from mutations in key driver genes such as KRAS and p53, which are closely associated with tumor development.[13-15] Secondly, TCR-T therapy leverages the highly specific TCR-MHC recognition process and enhances it by using TCRs with even higher affinity and specificity (Table 1). This ensures precise targeting and minimizes off-target effects. Research has shown that TCR-T cells require significantly fewer peptide-MHC (pMHC) complexes for target antigen recognition compared to CARs, often as low as 1-50 pMHCs.[16] This inherent sensitivity grants TCR-T cells superior recognition and killing capabilities in solid tumors with high antigen heterogeneity or low antigen density. Last but not least, TCR-T cells utilize natural T-cell signaling pathways for their activation (Table 1). This precise orchestration of signaling events distinguishes them from CAR-T cells (Table 1). While CARs utilize a simplified, antibody-derived recognition domain linked to CD3ζ, TCR-T cells employ the complete, multi-subunit TCR-αβ/CD3 complex. The binding of a TCR to a pMHC initiates a precise cascade of signaling events, resulting in the formation of a highly organized immunological synapse.[17] This endogenous configuration allows for the detection of low-abundance intracellular antigens, facilitating high-sensitivity signal amplification and a more physiological cytokine profile compared to the MHC-independent, high-threshold activation characteristic of CAR-T therapy.[18] This physiological signaling potentially leads to more potent and sustained anti-tumor responses, with improved T-cell persistence and reduced "on-target, off-tumor" toxicity compared to some CAR-T constructs.

The foundational proof-of-concept of TCR-T therapy was established in the late 1980s, when researchers first demonstrated that the transfer of cloned α and β T-cell receptor chains could successfully redirect the antigen

Table 1. A comparison of CAR-T and TCR-T.

Feature	TCR-T	CAR-T
Receptor Origin	Native or affinity-enhanced TCR	Single-chain variable fragment (scFv) from BCR
Recognition Type	Recognizes peptide-MHC complex	Direct binding (like an antibody)
Target Scope	Surface and Intracellular (MHC-restricted)	Surface only (MHC-independent)
Antigen Sensitivity	Very high (1–10 pMHC molecules)	Lower (typically requires ~1,000+ molecules)
Binding affinity (K_D)	Typically high (1 nM–1 μM)	Typically lower (1–100 μM)
Complex Structure	Octameric: TCR-αβ heterodimer + CD3 (γε, δε, ζζ)	Monomeric/Dimeric: scFv + Hinge + TM + Signaling domains
ITAM Count	10 ITAMs per complex (from the CD3 chains)	3 ITAMs (typically from one CD3ζ chain, though variants exist)
Synapse Formation	Highly organized (cSMAC/pSMAC)	Non-classical/less spatially organized immunological synapses
Co-stimulation	Provided in *trans* (e.g., via CD28 or 4-1BB on the APC)	Provided in *cis* (built into the CAR endodomain)
Transduction Cascade	"Orchestrated" cascade	"Direct" cascade
Triggering Events	pMHC binding to TCR/CD3 complex	Direct scFv-antigen binding to cell surface antigen
Proximal Signaling	require Lck phosphorylation and ZAP70 recruitment	CD3ζ and co-stimulatory domains are phosphorylated
The Signalosome	ZAP-70 phosphorylates LAT and SLP-76, forming a scaffold that recruits PLCγ	Often bypasses the requirement for a formal LAT-based signalosome
Outcome of Signaling Transduction	A regulated, rhythmic calcium flux that favors T cell memory formation and persistence	Rapid, intense "spikes" of signaling. This leads to faster tumor killing but also increases the risk of T cell exhaustion and cytokine release syndrome
Clinical efficacy	Demonstrated efficacy in selected solid tumors	High efficacy in hematological malignancies
Treatment-related toxicities	Generally lower incidence of systemic toxicities	Higher incidence of cytokine release syndrome and neurotoxicity
FDA approval	First FDA-approved TCR-engineered T cell therapy, afami-cel (TECELRA®), for synovial carcinoma (HLA-A*02:01/MAGE-A4) in 2024	Six FDA-approved products since 2017, primarily for B-cell malignancies

specificity of recipient T cells.[19] In 1988, Steven Rosenberg at NIH pioneered tumor-infiltrating lymphocytes (TILs) therapy by extracting, expanding, and reinfusing tumor-specific T cells in treating melanoma patients.[20] A melanoma antigen called MAGE-A1, which is recognized by T cells, was identified from a melanoma patient in 1991.[21] Three years later, TILs isolated from melanoma patients recognized the melanocyte differentiation proteins MART-1 and gp100 had been discovered.[22,23] These studies demonstrated the existence of tumor-reactive T cells that recognize non-mutated shared antigens. While early clinical efforts in the 2000s, most notably those targeting the MART-1 antigen in melanoma, confirmed the feasibility of TCR-T therapy.[24] It also underscored significant challenges regarding "on-target, off-tumor" toxicities and the relatively low affinity of natural TCRs against self-antigens.[24] In the last decade, more robust responses have been observed in clinical trials of an affinity-enhanced TCR targeting NY-ESO-1.[25-27] These early lessons paved the way for the modern era of TCR-T therapy, characterized by sophisticated affinity maturation, the use of high-throughput screening for neoantigen-specific TCRs, and the development of strategies to prevent TCR mispairing, thereby enhancing both the safety and potency of the platform.

NAVAGATING THE CHALLENGES

Despite its promise, TCR-T therapy for solid tumors faces several significant challenges that require ongoing research and innovative solutions (Figure 1).

Human leukocyte antigen (HLA) restriction

It is a significant limitation in TCR-T therapy as it confines applicability to patients with matching HLA haplotypes. Each TCR recognizes a specific peptide presented by a particular HLA allele, necessitating personalized approaches and severely limiting broad applicability.[10] Currently, most clinical trials focus on common HLA types such as HLA-A*02:01, which limits the eligible patient population and prevents TCR-T therapy from achieving the widespread applicability of "off-the-shelf" CAR-T therapies.[10] This limitation is evident in clinical practice, with trials for specific TCR-T therapies, such as human papillomavirus 16 (HPV16) E6 antigen-specific TCR-T therapy, exclusively enrolling HLA-A*02:01-positive patients.[28] However, the distribution of this allele varies considerably among different ethnic groups. Its allele frequency is approximately 27% in individuals of European descent, but only 11.9% in African Americans and 6.5% in Asian populations. This significantly impacts treatment accessibility for patients of different ethnicities.[29] Therefore, expanding HLA coverage or developing "universal" TCRs has become a critical research direction.

Tumor microenvironment (TME) suppression

The TME poses a significant challenge in TCR-T cell therapy by suppressing the immune system and preventing infused T cells from effectively targeting and destroying tumor cells. The TME is a complex network composed of malignant cells, stromal fibroblasts, distorted blood vessels, and a dense ECM that work together to create a hostile environment for T cells.[30] It can prevent infused T cells from reaching, infiltrating, and effectively destroying tumor cells. When T cells enter this hostile environment, they can become exhausted and dysfunctional, which leads to poor treatment outcomes.[30] Firstly, the TME is filled with immunosuppressive cells that actively inhibit T cells. These include tumor-associated macrophages (TAMs), myeloid-derived suppressor cells (MDSCs), and regulatory T cells (Tregs).[30] TAMs and MDSCs usually produce reactive oxygen species (ROS) and arginase, which deplete essential amino acids required for T cell activation. Additionally, Tregs outcompete effector T cells for IL-2, a vital growth factor. These cells also secrete molecules like IL-10 and TGF-β that inhibit T cell activation and can lead to T cell exhaustion or death.[30,31] Furthermore, chronic antigen stimulation and inhibitory signals from cytokines and immune check-

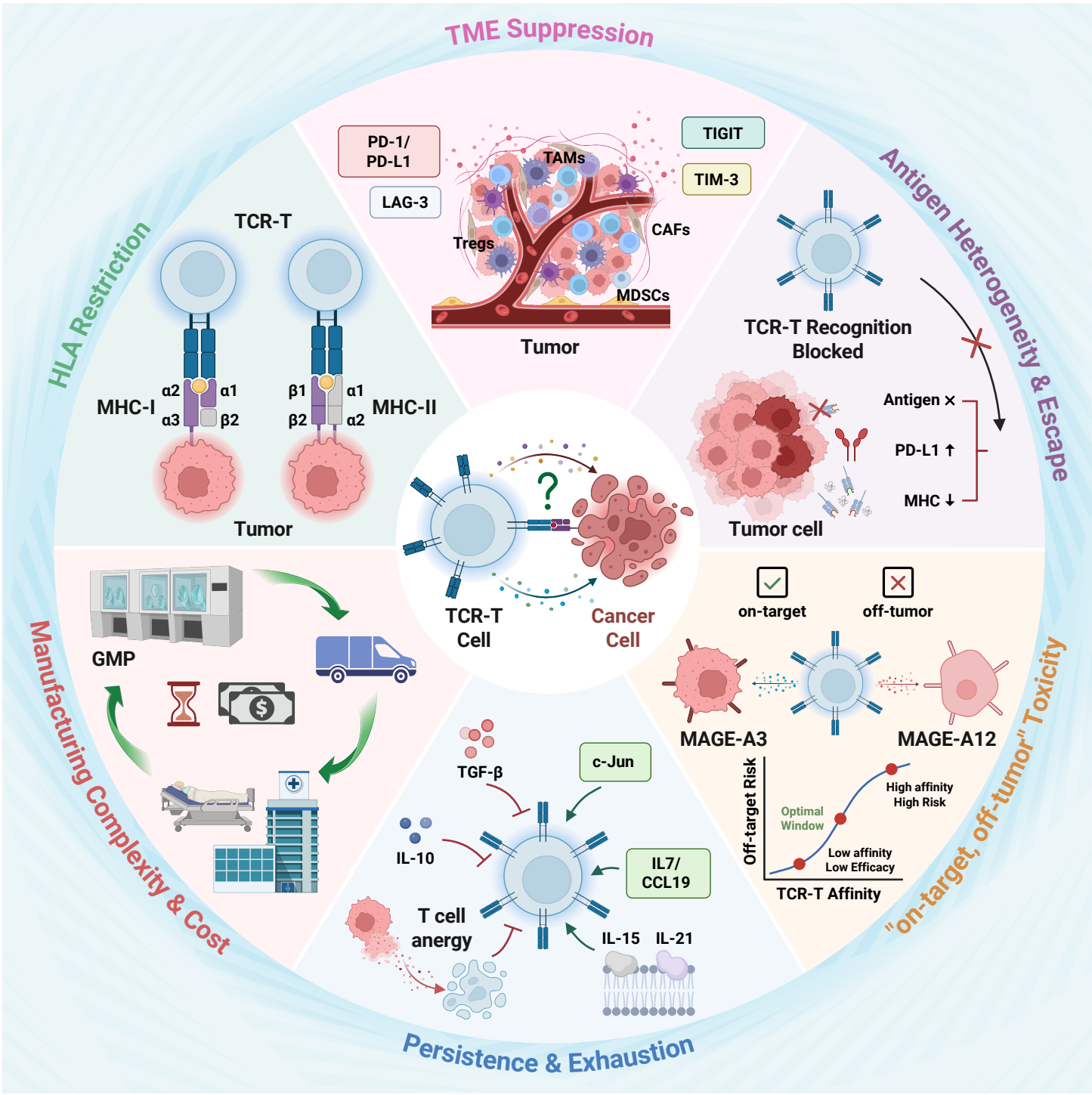

Figure 1. Roadblocks in TCR-T cell therapy This schematic summarizes the main obstacles limiting TCR-T efficacy and translation. These include: (1) HLA restriction, confining applicability to specific HLA types; (2) Tumor microenvironment suppression, impairing T cell infiltration and function; (3) Antigen heterogeneity and escape, enabling tumor recurrence; (4) On-target/off-tumor toxicity, from cross-reactivity with normal tissues; (5) T-cell persistence and exhaustion, reducing long-term activity; and (6) Manufacturing complexity and cost, limiting scalability. Together, these barriers underscore the need for innovative strategies to enhance TCR-T therapy in solid tumors. Image created with Biorender.com with permission.

points contribute to T cell exhaustion, characterized by the overexpression of inhibitory receptors such as PD-1, LAG-3, TIM-3, and TIGIT.[32] To overcome these challenges, combining TCR-T cell therapy with immune checkpoint blockade (ICB) might be a promising strategy to release these brakes on T cell activity and enhance their anti-tumor response.

The TME also presents metabolic challenges for T cells, as it is characterized by low oxygen and the accumulation of detrimental metabolites that are detrimental to T cell function. Specifically, high levels of lactate lower the pH, impairing T cell proliferation,[33] while adenosine binds to A2A receptors to inhibit TCR signaling.[34,35] Furthermore, the buildup of kynurenine, a product of tryptophan metabolism, directly promotes T cell apoptosis and the induction of a dysfunctional state.[36-38] In addition, the accumulation of molecules like cholesterol and nutrient deprivation can also lead to T cell exhaustion, limiting their persistence and effectiveness.[39,40]

Beyond cellular suppression, solid tumors present physical barriers. Cancer-associated fibroblasts (CAFs) deposit large amounts of collagen, forming a dense extracellular matrix that acts as a physical barrier, preventing T cells from penetrating the core of the tumor. Additionally, CAFs release signaling

molecules that keep T cells restricted to the outer edges of the tumor, creating a "cold tumor" microenvironment with low T cell presence.[41] Notably, even after successful transendothelial migration into the target tissue, T cells can be excluded from the tumor nests and sequestered within the stroma by specific secreted molecules up-regulated in the TME.[42,43] Moreover, the disorganized and dysfunctional blood vessels within the tumor also impede the migration of T cells from the bloodstream into the tumor tissue.[44] For instance, the endothelial-cell-surface protein CD93 has been identified as a key regulator of vascular dysfunction, and its pathway can be targeted to normalize vessels and enhance T-cell infiltration.[45] Therefore, improving TCR-T cell therapy requires a multi-pronged approach to address these barriers. Future strategies will involve engineering T cells to resist exhaustion, breaking down physical barriers in the TME, and targeting the metabolic vulnerabilities of the tumor to create a more favorable environment for T cells. By overcoming these challenges, researchers aim to significantly improve the effectiveness of TCR-T cell therapy in treating solid tumors.

Antigen heterogeneity and escape

Tumors are complex and diverse, with some cancer cells altering or reducing the targeted antigen expression, which allows them to evade the immune response and potentially reappear. Even if TCR-T cell therapy effectively destroys tumor cells expressing a specific antigen, there may still be tumor subgroups that lack this antigen, leading to tumor relapse. For example, antigens like gp100 and NY-ESO-1 show substantial inter-tumor and intra-tumor heterogeneity in melanoma patients, severely affecting the stability and predictability of antigen targets.[46] In light of this, the most optimal target for TCR-T cell therapy would be an antigen universally expressed across all cancer cells while maintaining high tumor-specificity. Such antigens must be minimally expressed or absent in normal tissues, thereby widening the therapeutic window and minimizing potential "on-target, off-tumor" toxicities. This dual focus on coverage and specificity ensures the therapy can effectively eradicate the tumor mass while preventing recurrence and ensuring patient safety.[10]

Tumor cells have developed various ways to evade recognition and destruction by TCR-T cells, which poses a significant challenge in cancer treatment. One common mechanism is the loss or reduction of specific antigens that TCR-T cells are programmed to target. This allows a subset of cancer cells to go undetected by TCR-T cell therapy, enabling them to survive and potentially lead to disease relapse.[47-49] Another strategy employed by tumor cells is to interfere with the presentation of antigens on their surfaces through MHC molecules, which are crucial for TCR-T cell activation. This can be achieved by downregulating MHC molecule expression or altering the pathways responsible for antigen processing and presentation, such as TAP and β2-macroglobulin.[50] Additionally, tumor cells can undergo a transformation known as epithelial-mesenchymal transition, resulting in changes to their physical and molecular properties. This transformation often leads to a decrease in target antigens and MHC-I molecules, while boosting the presence of immunosuppressive molecules like PD-L1.[51] These altered cancer cells with a stem cell-like phenotype become resilient and are a frequent source of tumor recurrence.[52] To counteract antigen escape, researchers are developing new strategies. Instead of targeting a single antigen, TCR-T cell therapies are designed to recognize multiple antigens simultaneously. This diversified targeting strategy reduces the risk of all targeted antigens disappearing simultaneously, making it more challenging for cancer cells to evade detection. Complementary to the use of multiple targets, targeting shared neoantigens, such as $KRAS^{G12V}$ or $KRAS^{G12D}$ mutations, is a promising strategy because these "trunk" mutations are vital for tumor growth, making them less likely to be lost and therefore a more consistent and effective therapeutic target.[53] By developing innovative strategies that address antigen escape mechanisms, researchers are striving to enhance the efficacy and durability of TCR-T cell therapies in treating cancer.

"On-target/off-tumor" toxicity

While TCR-T cells can recognize a wider range of antigens presented by HLA molecules, this also increases the risk of inadvertently targeting similar antigens on healthy tissues, leading to potentially fatal off-target toxicity. In clinical trials targeting antigens such as MAGE-A3, MART-1, MAGE-A12, and CEA, fatal toxicities have been observed when TCR-T cells cross-reacted with related antigens present in normal tissues. For example, in a trial targeting MAGE-A3, TCR-T cells recognizing a similar peptide in the heart protein TITIN led to fatal cardiac toxicity.[54] In another trial, neurotoxicity occurred when TCR-T cells recognized low levels of MAGE-A12 in the brain tissue of patients.[55] Additionally, MART-1, a common target in melanoma therapy, has been linked to multi-organ failure and neurological damage in a TCR-T clinical trial because it is also present in normal melanocytes in the skin, eyes, and ears.[56] Furthermore, targeting antigens like CEA, which is also expressed in the healthy colon lining, can lead to severe diarrhea.[57] These cases show that even when a target antigen is primarily associated with a tumor, its presence in healthy tissues, even at trace levels, can lead to severe consequences. Boosting the TCR's affinity to enhance anti-tumor effects must be done cautiously, as it can make the T cells less able to distinguish between cancer cells and healthy ones. To ensure the safety of TCR-T therapy, researchers must thoroughly evaluate the potential for cross-reactivity with healthy tissues before selecting target antigens. By prioritizing tumor specificity and minimizing expression in normal tissues, the risk of on-target/off-tumor toxicity can be significantly reduced, making TCR-T cell therapy a safer and more effective treatment option for cancer patients.

T-cell persistence and exhaustion

Maintaining long-term persistence and preventing T-cell exhaustion in the harsh TME are crucial for durable responses. In the immunosuppressive environment of a solid tumor, TCR-T cells can undergo exhaustion, characterized by a decrease in proliferative capacity, weakened cytotoxicity, and the expression of inhibitory receptors such as PD-1 and TIM-3.[58] Clinical evidence shows that while TCR-T cells can initially expand and fight tumors, their numbers often drop significantly within weeks or months.[59,60] This lack of long-term persistence is a key factor limiting the therapy's effectiveness. T-cell exhaustion is driven by a multifactorial process involving sustained tumor antigen stimulation, chronic inhibitory receptor signaling, and metabolic stress within the TME. Specifically, prolonged exposure to tumor antigens drives T-cells towards a terminal dysfunctional state, while the presence of immune-suppressing elements like PD-L1 and TGF-β, combined with nutrient deprivation and hypoxia, further hastens T-cell dysfunction.[61-63] Improving the *in vivo* persistence and anti-exhaustion properties of TCR-T cells is crucial for enhancing their effectiveness and efficacy in cancer treatment.

Manufacturing complexity and cost

The manufacturing process for TCR-T cell therapy used in treating solid tumors is a significant obstacle to its widespread adoption due to its complexity and high costs.[64] The current TCR-T therapy cell products rely on autologous T cells, meaning they're made from the patient's own T cells, for individualized preparation. The entire workflow, from T cell collection and activation to gene modification, enrichment, and expansion under good manufacturing practice (GMP) conditions, is highly complex and time-consuming, often taking several weeks to complete. This not only demands highly specialized facilities and skilled personnel but also substantially increases production costs.[65,66] Since each therapy is personalized to the individual patient, mass production is challenging, leading to further difficulties in scaling up production. Furthermore, for patients with compromised immune systems, the quality of their T cells may be subpar, resulting in a less effective final product.[66] These challenges highlight the need for streamlining the manufacturing process and finding ways to reduce costs to make TCR-T cell therapy more accessible to a broader population of patients in need.

CLINICAL VALIDATION AND EMERGING EFFICACY

The FDA has granted accelerated approval for TECELRA (afamitresgene autoleucel), the first TCR-T cell product targeting MAGE-A4, for treating adult patients with unresectable or metastatic synovial sarcoma after previous chemotherapy. The number of TCR-T cell products in clinical trials has been steadily increasing (Figure 2A), with a focus on targeting highly immunogenic antigens such as cancer germline antigens (42.86%) and onco-viral proteins (11.8%) (Figure 2B). This approach offers significant benefits as these antigens are highly immunogenic and have lower risks of off-target effects. Additionally, most TCRs in these trials are restricted by the prevalent HLA-A*02

allele, expanding the range of eligible patient populations (Figure 2C). This trend highlights the potential of TCR-T cell therapy in treating various types of cancer, especially those with limited treatment options. The approval of TECELRA represents a significant milestone in the advancement of personalized immunotherapy for cancer patients.

Despite advances in TCR therapy with two products reaching Phase 3 trials (Table S1), the current efficacy in stabilizing disease progression remains moderate (Table 2), particularly for patients with advanced or refractory cancers. Collective clinical data (Table 2) underscore three pivotal translational challenges that must be addressed to improve patient outcomes. First, the safety profile presents a dual challenge. Approximately 13.3% (4/30) of analyzed trials targeted tissue differentiation antigens, such as CEA, gp100, and MART-1. While these trials demonstrated proof-of-concept with objective response rates (ORR) ranging from 18.8% to 33.3%, they were significantly hindered by on-target, off-tumor toxicities. Due to the expression of these antigens in healthy tissues such as skin, eyes, and colon, severe autoimmune adverse events were frequently reported, including severe colitis in CEA-targeted trials[57] and high incidences of cutaneous toxicity, uveitis, and hearing loss in gp100 [67] and MART-1 trials.[67,68] The majority of trials (21/30) focused on cancer germline antigens, which currently represent the most extensively studied and promising target class. NY-ESO-1 has shown exceptional and durable efficacy across various solid tumors, particularly in synovial sarcoma (ORR >50%) and multiple myeloma (ORR up to 80% in NCT01352286).[27,69-76] Its safety profile is generally manageable, with adverse events primarily consisting of cytokine release syndrome (CRS) and hematologic toxicities associated with lymphodepletion. While the MAGE-A family has been widely targeted, outcomes have been mixed. Notably, MAGE-A3-targeted trials (NCT01350401 and NCT01273181) resulted in fatal outcomes due to TCR cross-reactivity with similar peptides in the heart and brain. This underscores the critical necessity of rigorous preclinical safety assessments. A smaller subset of trials (5/30) explored onco-viral proteins, such as HBV-related antigens and HPV E6/E7. These targets displayed superior safety profiles because their expression is strictly restricted to virus-infected malignant cells.[28,48] Clinically, remarkable tumor regression was observed in HPV-associated cancers, a 50% ORR for HPV16 E7 in NCT02858310. In HBV-related hepatocellular carcinoma (HCC), stable disease (SD) was the primary outcome, demonstrating the potential for controlling tumor progression.[77-79] Recent trials have also highlighted the potential of novel targets. Alpha-fetoprotein (AFP), predominantly expressed in HCC and other aggressive malignancies like cholangiocarcinoma, has achieved an SD rate of 57.1%, offering value in disease stabilization.[80] PRAME, another cancer germline antigen with minimal expression in healthy tissues, has emerged as a highly promising target. Recent trials (NCT03686124) reported impressive ORRs (48.1% ~ 61.5%).[81] Similar to NY-ESO-1, the safety profile of PRAME-targeted therapies is characterized by manageable CRS and hematologic toxicities, with no reports of fatal off-target events to date. Beyond immediate safety concerns, the long-term clinical utility of TCR-T therapy is hindered by poor response durability and restrictive patient selection criteria. Clinical data indicate that while TCR-T cells often exhibit robust initial expansion leading to impressive tumor regression, their persistence frequently declines within weeks or months, inevitably leading to disease progression. Furthermore, treatment eligibility is currently dominated by HLA restriction, with over 60% of clinical trials exclusively targeting HLA-A*02 positive individuals (Figure 2). This heavy reliance creates a significant bottleneck in recruitment and limits equitable treatment access across diverse ethnic populations. To overcome these hurdles, next-generation strategies must prioritize molecular reprogramming to enhance T-cell fitness against the suppressive tumor microenvironment and explore HLA supertypes to broaden patient eligibility and ensure global accessibility. As researchers continue to develop TCR therapies, addressing these challenges and improving overall patient outcomes remain paramount.

FUTURE DIRECTIONS AND PROMISING STRATEGIES

Addressing HLA restriction

TCR-T cell therapy is limited by HLA restrictions, where T cells need to identify a specific target peptide carried by a corresponding HLA allele. This limitation significantly narrows down the number of patients who can benefit from this therapy. Overcoming these restrictions will be crucial in expanding the reach and effectiveness of TCR-T cell therapy (Figure 3).

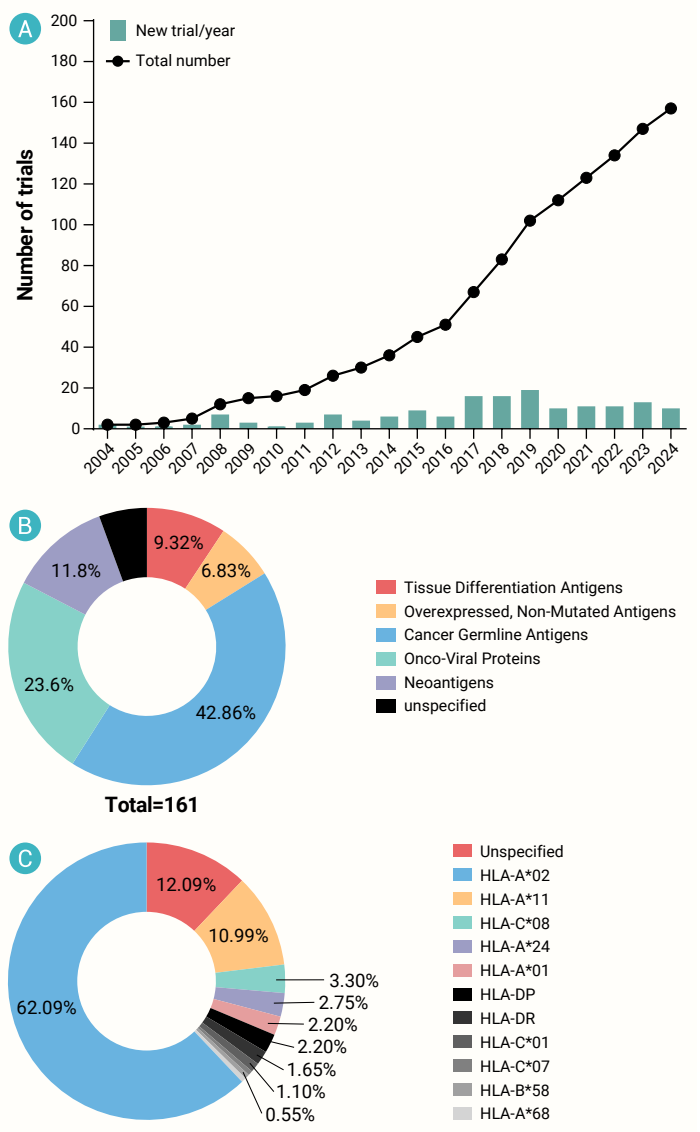

Figure 2. An analysis of trends in TCR-T trials registered on clinicaltrials.gov (A) Trends in annual new TCR-T trials and growth in cumulative registrations. (B) Frequency of different antigen types across 161 tests. (C) Distribution of HLA restriction types in 161 TCR-T clinical trials. Cutoff date of November 1, 2025.

Researchers have been exploring innovative approaches to overcoming the narrow restrictions posed by the diversity of HLA alleles. One of the most promising concepts that has emerged is the idea of HLA supertypes. By identifying structural and peptide-binding similarities among certain class I HLA molecules, researchers can group different HLA alleles into a single supertype. This grouping allows for the identification of HLA molecules that are capable of binding and presenting the same antigenic epitopes.[82] The similarity among members of an HLA supertype arises from the striking resemblance at key positions within the peptide-binding groove of the HLA molecules. For example, HLA alleles such as HLA–A*02:01, A*02:03, A*02:06, and A*02:10 share structural features that allow them to bind and present the identical NY–ESO–1 antigen peptide (SLLMWITQC).[83] This concept has paved the way for the development of "promiscuous" TCR-T products that have the potential to treat a broader patient cohort. An excellent example of the application of the HLA supertype concept is the FDA-approved TCR-T product, TECELRA. This product is engineered to recognize the MAGE–A4 peptide when presented by multiple HLA alleles such as HLA–A*02:01, HLA–A*02:02, HLA–A*02:03, and HLA–A*02:06.[84,85] Utilizing TCRs that recognize epitopes presented by an HLA supertype significantly expands the treatable population for TCR-T therapies, going beyond the limitations of single-allele match-

Table 2. Summary of results from TCR-T cell trials for solid tumors.

Trial Number	Phase	Antigen Class	Target	HLA Restriction	Disease	Response Rate	Adverse Events	Date	PMID
NCT00923806	Phase 1/2	Tissue Differentiation Antigens	CEA	HLA-A*02:01	Colorectal cancer	1 of 3, 33.3% PR.	Severe colitis	2011 Mar	57
NCT00509496	Phase 2	Tissue Differentiation Antigens	gp100	HLA-A*02:01	Metastatic melanoma	3 of 16, 18.8% (1 CR; 2 PR)	AEs: Skin toxicity (93.8%), Uveitis (25%), Hearingloss (31.3%)	2009 Jul	67
NCT00509288	Phase 2	Tissue Differentiation Antigens	MART-1	HLA-A*02:01	Metastatic melanoma	6 of 20, 30.0% PR	AEs: Skin toxicity (70%), Uveitis (60%), Hearing loss (50%), Erythematous skin rash (15%), Acute respiratory distress (10%).	2009 Jul	67
NCT00910650	Phase 2	Tissue Differentiation Antigens	MART-1	HLA-A*02:01	Metastatic melanoma	6 of 13, 46.2% SD	Adverse Events (AEs): Transient Tumor Regression (69.2%), Skin Rash (23.1%), Acute Respiratory Distress (15.4%), Cytokine Release (15.4%), Delayed Pancytopenia (7.7%).	2014 May	68
NCT03899415	Phase 1	Onco-Viral Proteins	HBsAg	HLA-A*02:01, HLA-C*08:01	Recurrent HBV-related HCC post-liver transplantation	1 of 8, 12.5% PR; 3 of 8, 37.5% SD	AEs: Grade 3 increase of liver enzymes (12.5%), Grade 1 AEs with mild elevation of liver enzymes during the first treatment cycle (12.5%).	2021 Dec	79
NCT02719782	Phase 1	Onco-Viral Proteins	HBsAg HBcAg	HLA-B*58:01/HBsAg, HLA-C*08:01/HBsAg, HLA-A*02:01/HBsAg, HLA-A*11:01/HBcAg,	Recurrent HBV-related HCC post-liver transplantation	0 of 6, 0% ORR (0 PR, 0 SD)	AEs: Pyrexia (50%, all Grade 1)	2023 Aug	82
NCT04677088	Phase 1	Onco-Viral Proteins	HBsAg HBeAg	HLA class I	Recurrent HBV-related HCC post-liver transplantation	4 of 6, 66.7% SD	AEs: grade 1 to 2 leukopenia (33.3%), grade 1/2 elevations in creatine kinase (50%).	2025 Sep	77
NCT02280811	Phase 1/2	Onco-Viral Proteins	HPV16E6	HLA-A*02:01	Metastatic HPV16-positive epithelial cancers	2 of 12, 16.7% PR	Adverse Events (AEs): Lymphopenia (100%); Neutropenia (100%); Thrombocytopenia (100%); Anemia (84.6%); Febrile neutropenia (38.5%); Infection (30.8%); Diarrhea (7.7%); Hyperbilirubinemia (7.7%); Rash (7.7%); Syncope (7.7%); Bronchial obstruction (7.7%); Hemorrhage (pulmonary) (7.7%); Hypoxia (7.7%); Pleural effusion (7.7%); Prolonged intubation (7.7%)	2019 Oct	28
NCT02858310	Phase 1/2	Onco-Viral Proteins	HPV16E7	HLA-A*02:01	HPV-16-associated epithelial cancers	6 of 12, 50% PR; 4 of 12, 33.3% SD	AEs: Neutropenia (100%); Leukopenia (100%); Anemia (100%); Febrile neutropenia (67%); Thrombocytopenia (67%); Hypophosphatemia (50%); Hyponatemia (33%); Pulmonary edema (25%); Hypoxia (25%); Increased aspartate aminotransferase (25%).	2021 Mar	48

Table 2. *(Continued)*

Trial Number	Phase	Antigen Class	Target	HLA Restriction	Disease	Response Rate	Adverse Events	Date	PMID
NCT01343043	Phase 1	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01 and HLA-A*02:06	Synovial sarcoma	1 of 12, 8.3% CR; 5 of 12, 41.7% PR	AEs: Lymphopenia (100%); Leukopenia (92%); Neutropenia (83%); Anemia (83%); Hypophosphatemia (75%); Thrombocytopenia (67%); Cytokine release syndrome (42%); Febrile neutropenia (17%).	2018 Aug	76
NCT02070406 NCT01697527	Phase 1	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01	Advanced sarcoma and metastatic melanoma.	1 of 6, 16.7% CR; 3 of 6, 50.0% PR in the ESO cohort; 2 of 4, 50.0% PR in the INY cohort.	Adverse Events (AEs): Bone marrow failure (10%, attributed to conditioning chemotherapy); Cytokine storm (20%, attributed to IL-2); Transaminitis (10%, attributed to IL-2 and ipilimumab); Ventricular tachycardia (10%, attributed to IL-2); Respiratory distress requiring intubation (10%, attributed to IL-2).	2019 Apr	75
NCT02366546	Phase 1	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01 and HLA-A*02:06	Recurrent solid tumors expressing the NY-ESO-1 antigen	3 of 9, 33.3% PR	AEs: Cytokine release syndrome (33%); Fever (33%)	2022 Jun	74
NCT04318964	Phase 1	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01	Soft tissue sarcomas (STSs)	5 of 12, 41.7% PR; median PFS was 7.2 months	AEs (≥grade 3): lymphopenia (100%), neutropenia (92%), leukopenia (83%), and anemia (33%), which were primarily attributable to the precondition of lymphodepletion. TEAE(grade 3–4): fever (8%), thrombocytopenia (8%), hypokalemia (8%), increased alanine aminotransferase (8%), proteinuria (8%), hypertriglyceridemia (8%), grade 2 cytokine release syndrome (16.7%).	2023 Aug 1	73
NCT03240861	Phase 1	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01	Advanced solid tumors	1 of 3, 33.3% PR; 1 of 3, 33.3% SD	AEs: Grade 4 cytopenias (100%, attributed to conditioning chemotherapy); Grade 3 hypotension (33%, possibly related to TCR T-cell product).	2025 Jul	72
NCT00670748	Phase 2	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01	Metastatic synovial cell sarcoma and metastatic melanoma	1 of 18, 5.6% CR; 10 of 18, 55.6% PR in synovial cell sarcoma patients; 4 of 20, 20.0% CR; 7 of 20, 35.0% PR in melanoma patients	Adverse Events (AEs): IL-2–related transient toxicities (100%); Febrile neutropenia (1 patient with E. coli bacteremia leading to septic shock and death).	2015 Mar	27

Table 2. *(Continued)*

Trial Number	Phase	Antigen Class	Target	HLA Restriction	Disease	Response Rate	Adverse Events	Date	PMID
NCT02992743	Phase 2	Cancer Germline Antigens	NY-ESO-1	HLA-A*02:01, HLA-A*02:05, HLA-A*02:06	Advanced or metastatic myxoid/round cell liposarcoma (MRCLS)	Cohort 1 (Reduced-Dose LDR): 2 of 10, 20% PR; 8 of 10, 80% SD. Cohort 2 (Standard-Dose LDR): 4 of 10, 40% PR; 5 of 10, 50% SD.	Cohort 1: Lymphopenia (100%); Leukopenia (90%); Neutropenia (90%); Anemia (70%); Thrombocytopenia (80%); Hypophosphatemia (60%); Febrile neutropenia (20%); Cytokine release syndrome (60%). Cohort 2: Leukopenia (100%); Cytokine release syndrome (100%); Anemia (90%); Thrombocytopenia (80%); Hypophosphatemia (80%); Febrile neutropenia (40%); Rash/maculopapular rash (30%); Hyponatremia (20%)	2025 May	71
NCT03168438	Phase 1	Cancer Germline Antigens	NY-ESO-1/LAGE-1a	HLA-A*02:01, HLA-A*02:05, HLA-A*02:06	Relapsed/refractory multiple myeloma	1 of 6, 12.5% CR; 2 of 6, 37.5% PR	AEs: Anemia, leukopenia, and thrombocytopenia (100%); Lymphopenia and neutropenia (83%); grade 1/2 cytokine release syndrome (50%).	2023 Apr 11	70
NCT01352286	Phase 2	Cancer Germline Antigens	NY-ESO-1/LAGE-1	HLA-A*02:01	Relapsed, refractory, or high-risk multiple myeloma	20 of 25, 80.0% OR at Day 42; 19 of 25, 76.0% OR at Day 100; 11 of 25, 44.0% OR at 1 year.	Adverse Events (AEs): Thrombocytopenia (88%); Diarrhea (96%); Febrile neutropenia (68%); Anemia (56%); Leukopenia (56%); Neutropenia (44%); Hypophosphatemia (24%); Hypocalcemia (20%); Graft-versus-host disease (24%); Hypokalemia (12%); Rash (12%); Stomatitis (12%); Hyponatremia (8%); Hypoxia (8%).	2019 Jul	69
UMIN000002395	Phase 1	Cancer Germline Antigens	MAGE-A4	HLA-A*24:02	Recurrent or metastatic MAGE-A4-expressing esophageal squamous cell carcinoma	0 of 10, 0%	Skin reaction at injection site (40.0%)	2015 May	161
NCT04044859	Phase 1	Cancer Germline Antigens	MAGE-A4	HLA-A*02	Advanced cancers expressing the MAGE-A4 antigen	2 of 5, 40.0% PR	No DLTs, SAEs, GvHD, CRS, or neurotoxicity reported.	2020 Nov	162
NCT03132922	Phase 1	Cancer Germline Antigens	MAGE-A4	HLA-A*02	2 (5.3%) esophageal, 1 (2.6%) gastric, 3 (7.9%) head and neck, 1 (2.6%) melanoma, 2 (5.3%) NSCLC, 9 (23.7%) ovarian, 2 (5.3%) urothelial, 2 (5.3%) MRCLS, and 16 (42.1%) SS	9 of 38, 24% PR; 19 of 38, 50% SD	38 (100%) TEAEs (≥grade 3): Grade ≥3 decreased counts for lymphocytes (97%), neutrophils (87%), platelets (42%), anemia (63%), Grade ≥ 3 febrile neutropenia (32%), and pancytopenia (11%), prolonged neutropenia (24%).	2023 Jan	137,163

Table 2. *(Continued)*

Trial Number	Phase	Antigen Class	Target	HLA Restriction	Disease	Response Rate	Adverse Events	Date	PMID
NCT04044768	Phase 2	Cancer Germline Antigens	MAGE-A4	HLA-A*02	Advanced synovial sarcoma or myxoid round cell liposarcoma	ORR: 37% (19/52); Synovial Sarcoma ORR: 39% (17/44); Myxoid Round Cell Liposarcoma ORR: 25% (2/8). All responses were PR.	Most Common Grade ≥3 AEs: Cytopenias (Lymphopenia 96%, Neutropenia 85%, Leukopenia 81%). Cytokine Release Syndrome (CRS): 71% (37/52), mostly Grade 1-2, one Grade 3 event. Neurotoxicity: One Grade 1 immune effector cell-associated neurotoxicity syndrome (ICANS) case. No treatment-related deaths.	2024 Apr	164
NCT01350401	Phase 1/2	Cancer Germline Antigens	MAGE-A3	HLA-A*01	Metastatic melanoma and multiple myeloma	N/A	Off-target toxicity in cardiac tissue leading to 2 patients' deaths	2013 Aug	54
NCT02111850	Phase 1/2	Cancer Germline Antigens	MAGE-A3	HLA-DPB1*04:01	Metastatic or locally advanced refractory/recurrent cancers	1 of 17, 5.9% CR; 3 of 17, 17.6% PR	Adverse Events (AEs): Fever >39.0°C (58.8%); Elevated ALT/AST (11.8% grade 3–4); Elevated creatinine (11.8% grade 3–4); Hypoxia (5.9% grade 4); Dyspnea (5.9% grade 4); Rash (5.9% grade 2); Atrial fibrillation (5.9% grade 3); Renal failure (5.9% grade 3); Confusion (5.9% grade 3)	2017 Oct	165
NCT01273181	Phase 1/2	Cancer Germline Antigens	MAGE-A3/A9/A12	HLA-A*02:01	Metastatic melanoma, Synovial cell sarcoma, Esophageal cancer.	2 of 9, 22.2% CR; 3 of 9, 33.3% PR.	AEs: Neurologic toxicity (33.3%): Mental status changes, seizures, coma (2 fatal); Transient ischemic attack (11.1%), Hypotension (33.3%), Pulmonary edema (11.1%)	2013 Feb	55
NCT02592577	Phase 1	Cancer Germline Antigens	MAGE-A10	HLA-A*02	Advanced non-small cell lung cancer (NSCLC), including adenocarcinoma and squamous cell carcinoma.	1 of 11, 9.1% PR; 5 of 11, 45.5% SD	AEs: Lymphopenia (100%); Leukopenia (91%); Neutropenia (82%); Anemia (82%); Thrombocytopenia (45%); Hyponatremia (45%); Nausea (64%); Pyrexia (55%); Constipation (45%); Peripheral edema (45%); Chills (36%); Decreased appetite (36%); Fatigue (36%); Pneumonia (36%)	2022 Jan	166
NCT02989064	Phase 1	Cancer Germline Antigens	MAGE-A10	HLA-A*02	Advanced head and neck squamous cell carcinoma (HNSCC), melanoma, or urothelial carcinoma	4 of 10, 40% SD	AEs: Leukopenia (100%); Lymphopenia (100%); Neutropenia (100%); Anemia (90%); Thrombocytopenia (60%); Hyponatremia (60%); Fatigue (70%); Pyrexia (60%); Rash (50%); Decreased appetite (50%); Hypophosphatemia (40%); Constipation (40%); Hypotension (40%)	2022 Mar 18	138

Table 2. *(Continued)*

Trial Number	Phase	Antigen Class	Target	HLA Restriction	Disease	Response Rate	Adverse Events	Date	PMID
NCT04639245 NCT05430555	Phase 1/2	Cancer Germline Antigens	MAGE-A1	HLA-A*02:01	MAGEA1-positive, advanced and/or metastatic solid tumor	11 of 16, 68.8% SD	15 (93.8%) AEs (≥grade 3): cytopenias (neutropenia (13/16), lymphopenia (12/16), anemia (8/16), thrombocytopenia (8/16), and leukopenia (4/16) related to lymphodepletion. TEAEs (≥20%): nausea (43.8%), diarrhea (31.3%), fatigue (31.3%), fever (31.3%), rash (31.3%), and chills (25%).	2024 Jul 22	167
NCT03132792	Phase 1	Cancer Germline Antigens	Alpha-fetoprotein (AFP)	HLA-A*02:01	20 of 21 Advanced Hepatocellular Carcinoma, 1 of 21 Gastric Hepatoid Carcinoma	1 of 21, 4.8% CR; 1 of 21, 4.8% PR; 12 of 21, 57.1% SD	AEs: Neutropenia/neutrophil count decreased (52.4%); Leukopenia/white blood cell decreased (38.1%); Anemia/red blood cell decreased (28.6%); Cytokine release syndrome (28.6%, grades 1-2: n=5; grade 4: n=1); Increased aspartate aminotransferase (28.6%); Pyrexia (28.6%).	2025 Aug	168
NCT03686124	Phase 1	Cancer Germline Antigens	PRAME	HLA-A*02	PRAME recurrent and/or refractory solid tumors, including melanoma and sarcoma	phase 1a, u/cORR was 48.1% (13/27), cORR equaled 18.5% (5/27); phase 1b, u/cORR was 61.5% (8/13), cORR of 54.5% (6/11)	TEAEs (≥grade 3): lymphodepletion neutropenia (82.9%), lymphopenia (61%), leukopenia (56.1%). 92.7% cytokine release syndrome (CRS): grade 1 (46.3%), grade 2 (41.5%), grade 3 (4.9%).	2025 Jul	81

ing. Furthermore, researchers are exploring structure-guided TCR engineering to broaden the HLA recognition spectrum. For instance, *Karuppiah et al.* developed a TCR capable of recognizing the PRAME peptide antigen (ELFSYLIEK) presented by both HLA-A*03:01 and HLA-A*11:01, while maintaining similar affinity and functional activity.[86] This achievement of cross-HLA allele recognition provides a feasible approach to expanding the target population for TCR-T therapies. In addition to these advancements, *Foy et al.* have proposed a non-viral CRISPR technology platform that allows for the precise insertion of individualized neoantigen-TCRs into the endogenous TCR locus of T cells.[60] This strategy eliminates the dependence of traditional TCR-T cell therapy on specific HLA types, laying a strong technical foundation for developing personalized TCR-T cells for patients with diverse HLA types and showcasing the broad potential applications of this technology.

Enhancing T-cell fitness and persistence

Improving T-cell function and longevity *in vivo* is a critical area of research, with conventional approaches often involving the use of cytokines. Now, more advanced strategies are focusing on targeted delivery, molecular reprogramming, and metabolic adjustment (Figure 3). These strategies aim to enhance T-cell fitness and persistence *in vivo*, ultimately improving the body's immune response.

Cytokines, such as interleukins, are essential for immune cell function and are commonly used to enhance T-cell activity and prevent exhaustion. Providing T cells with additional cytokine signals could help them remain functional. Studies have shown that co-expressing cytokines like IL-7, CCL19, IL-15, and IL-21 can significantly boost T-cell function and promote the development of memory T cells.[87,88] *Ex vivo* culture methods commonly involve supplementing T cells with IL-7 and IL-15 to promote the differentiation of memory stem T cells (T_{SCM}).[89] However, systemic cytokine administration carries a significant risk of dose-limiting toxicity during *in vivo* therapy. Researchers are therefore developing strategies for precise cytokine delivery to the TME. One promising approach is to engineer T cells to co-express cytokines like IL-15 and IL-21 along with their TCR/CAR directly on the cell membrane. This targeted delivery strategy enhances the tumor-killing capacity of T cells without causing systemic toxicity.[87] Similar strategies have been successfully employed in optimizing CAR-T cell therapy. For example, engineering CAR-T cells to secrete IL-7 can regulate T-cell differentiation, improve anti-tumor efficacy, and prolong *in vivo* survival.[90] Additionally, expressing an IL-10-Fc fusion protein in CAR-T cells can reprogram intratumoral T cells toward oxidative phosphorylation, boosting their expansion and effector function.[91] Overall, the precise delivery of cytokines to T cells shows great promise in enhancing their anti-tumor activity and overcoming exhaustion, offering new possibilities for improving cancer immunotherapy.

While systemic cytokine administration is limited by toxicity, recent breakthroughs in protein engineering have yielded IL-2 variants designed to selectively enhance effector T-cell function.[92] Traditional IL-2 therapy often inadvertently stimulates $CD25^+$ Tregs, which can suppress the anti-tumor response. To address this, "not-alpha" IL-2 variants have been engineered to lose affinity for the high-affinity IL-2Rα (CD25) while retaining binding to the IL-2Rβγ signaling complex.[93,94] thereby prioritizing the expansion of $CD8^+$ effector and NK cells over Tregs. Furthermore, the development of "orthogonal" IL-2 and receptor pairs represents a major leap in TCR-T/CAR-T opti-

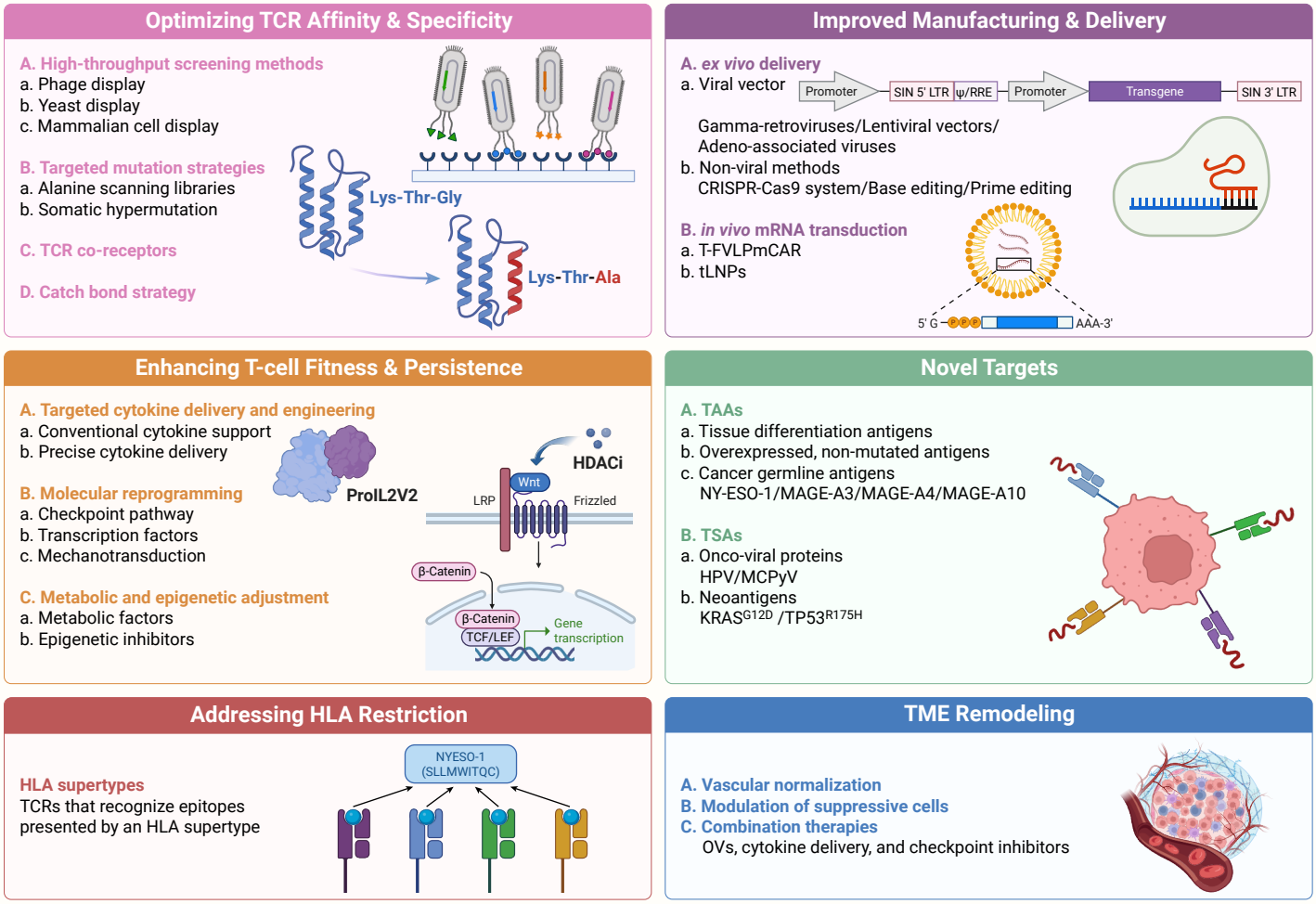

Figure 3. Emerging strategies to enhance the safety, efficacy, and durability of TCR-T cell therapy By fine-tuning TCR affinity and specificity, researchers can improve tumor targeting while reducing the risk of off-target effects. Additionally, efforts to improve T-cell fitness and persistence can ensure sustained anti-tumor activity. Addressing HLA restriction expands the potential pool of patients who can benefit from this therapy. Advancements in manufacturing and delivery methods enhance treatment efficiency and safety. Identifying new targets can increase treatment selectivity. Combining TCR-T therapy with oncolytic virotherapy helps transform the tumor microenvironment. Overall, these strategies offer a roadmap for advancing TCR-T therapy in solid tumors. Image created with Biorender.com with permission.

mization.[95,96] In this approach, both the IL-2 cytokine and its corresponding receptor IL-2Rα are mutated so they only bind to one another, but not to their wild-type counterparts. By transducing TCR-T cells with this orthogonal receptor, researchers can provide a selective fitness signal that only supports the engineered cells, effectively decoupling their survival from systemic cytokine levels and avoiding off-target immune activation.[96] Such strategies have shown significant potential in maintaining T-cell stemness and metabolic fitness within the hostile solid tumor microenvironment.

T-cell exhaustion is regulated by a complex network of transcription factors like TCF-1, NFAT, and TOX, leading to the upregulation of immune checkpoints like PD-1, CTLA-4, TIM-3, and LAG-3. These immune checkpoints inhibit T-cell activation when bound by ligands. Researchers are exploring various pathways to combat exhaustion, one being the inhibition of immune checkpoint expression or function.

One recent study developed an anti-TIM3-Pro-IL2 fusion protein, combining a TIM3-Fc fragment with a modified IL-2 (ProIL2V2) that remains active at low pH and has a higher affinity for IL-2Rβ. This fusion protein specifically targets dysfunctional T cells, effectively rejuvenating exhausted T cells.[97] Another approach is modulating transcription factors *via* gene editing. Overexpressing Tcf1 in T cells promotes the generation of T_{ex}-stem cells, reduces inhibitory receptor expression, and increases the secretion of various cytokines.[98] Similarly, c-Jun overexpression enhances *in vitro* and *in vivo* T-cell expansion, reduces terminal differentiation, and boosts anti-tumor efficacy.[99] Researchers are also exploring biomechanical regulation of exhaustion, such as the identification of Osr2 as a mechanosensitive transcription factor that exacerbates CD8+ T-cell exhaustion in TEM cells. By manipulating Osr2 signaling in engineered T cells, it may be possible to improve their therapeutic effectiveness against solid tumors.[100] This multi-faceted approach to combating T-cell exhaustion shows promise for enhancing the efficacy of immunotherapy in treating various diseases.

T-cell exhaustion is intrinsically linked to metabolic dysfunction, characterized by reduced glucose uptake, suppressed glycolysis, mitochondrial impairment, and cholesterol accumulation.[39,101,102] Modifying T cells' metabolism and enhancing the pool of initial T cells for therapy may unleash the full potential of TCR-T cell therapy and ensure long-lasting anti-tumor effects. For example, inhibiting isocitrate dehydrogenase 2 (IDH2) in T cells can activate acetyl-CoA-mediated epigenetic regulation, thereby fostering a memory-phenotype differentiation.[103] Another study found that succinate promotes mitochondrial adaptation and enhances T-cell vitality by driving BNIP3-mediated mitophagy. It also remodels the histone methylation landscape in CD8+ T cells, promoting the expression of stemness-associated genes.[104] Supplementing T cells with D-mannose can activate the Wnt signaling pathway *via* O-GlcNAc transferase. This, in turn, promotes Tcf7 expression and maintains epigenetic stemness, ultimately boosting anti-tumor activity.[105]

Epigenetic regulation has become a promising tool in boosting T-cell function through various inhibitors. Epigenetic intervention with a class I HDAC inhibitor (HDACi) in T cells can activate the Wnt/β-catenin signaling pathway, resulting in enhanced anti-tumor efficacy, improved memory formation, and increased resistance to exhaustion.[106] Inhibiting EZH1/2 in tumor cells makes them more immunogenic, which in turn enhances adoptive T-cell therapy by

improving T-cell activation, infiltration, and tumor-killing function.[107] Blocking BET proteins in T cells alleviates the exhausted state, leading to reduced inhibitory receptor expression, enhanced metabolic fitness, and improved proliferative capacity.[108]

Optimizing TCR affinity and specificity

Researchers are employing cutting-edge techniques such as high-throughput screening and rational design to enhance the affinity and specificity of TCRs for tumor antigens. The objective is to optimize their performance while reducing the chance of off-target effects. Emphasizing neoantigens, which are exclusive to an individual's tumor, plays a crucial role in diminishing adverse reactions. By fine-tuning TCRs, scientists aim to develop targeted therapies that effectively combat cancer while minimizing potential side effects for patients (Figure 3).

Various high-throughput screening methods have been developed to enhance the ability of TCRs to recognize tumors by isolating high-affinity, tumor-specific TCRs. One such method is phage display, which generates massive libraries for screening with up to 10^11 variants.[109] However, since it uses a bacterial system, the TCRs might not fold correctly, potentially leading to false positives. Additionally, phage display cannot directly compare the binding strength of different TCRs.[110] On the other hand, yeast display is a more suitable alternative for mammalian proteins as it accurately handles folding and modifications, resulting in a higher-quality library. By combining yeast display with flow cytometry, researchers can analyze fluorescence intensity and directly compare the affinity of each TCR variant.[111] Another method, mammalian cell display, involves expressing TCR libraries on TCR-negative T cells, providing the advantage of a complete TCR signaling system. This allows for direct screening by measuring T-cell activation. However, it's limited by the low efficiency of retroviral transduction, restricting library sizes to around 10^4 compared to the 10^7 to 10^10 libraries possible with phage and yeast display.[112] Each of these screening techniques offers unique benefits and challenges in the quest to identify high-affinity, tumor-specific TCRs for therapeutic purposes.

Beyond high-throughput screening, researchers use targeted mutation strategies to improve TCR function. One such approach involves using alanine scanning libraries to systematically alter amino acid residues in the CDR3 α and β regions of the TCR to alanine. By identifying key amino acid residues that influence TCR affinity, researchers can refine TCR mutation schemes for improved efficacy.[113,114] For example, researchers introduced random mutations into a $KRAS^{G12D}$-targeting TCR and, through multiple rounds of phage display, increased its affinity by nearly a hundredfold.[115] In addition to this random mutation technique, researchers can leverage the natural evolution process of B cells through somatic hypermutation for *in vivo* antibody affinity optimization. This process involves the AID enzyme introducing random point mutations across the TCR genes to generate a diverse pool of mutants, enabling the selection of high-affinity variants.[116,117] TCR co-receptors are also crucial for recognition and activation. For instance, if the CD8 binding site on MHC-I is mutated, TCR activation is directly inhibited.[118] Similarly, the CD4 co-receptor can significantly boost antigen recognition for MHC-II-restricted TCRs.[119] By understanding and optimizing these key components, researchers can effectively modulate TCR function and improve its therapeutic potential.

The pursuit of high affinity in TCRs has long been a common goal, but recent research has shown that it can come with drawbacks. Super high-affinity TCRs can cause dangerous cross-reactivity with healthy tissues and may even lead to T-cell exhaustion.[120] Some studies have found that lower-affinity TCRs can actually maintain their function better over time.[121] In a striking example, reducing TCR signal strength by deleting CD8 with CRISPR/Cas9 was shown to improve a TCR's anti-tumor ability.[122] This has led to new thinking. Instead of simply seeking high affinity, innovative techniques should be explored to improve the TCR's anti-tumor activity without dangerously increasing its affinity. A research group at Stanford developed a strategy to find mutants that form strong "catch bonds" with low-affinity pMHCs, resulting in robust T-cell activation. This approach led to a TCR that was more effective against tumors while having reduced cross-reactivity to the heart antigen TITIN, offering a fresh perspective on the relationship between TCR affinity and function.[123] Furthermore, during preclinical development, researchers should use more sophisticated screening methods to identify potential cross-reactive targets before a therapy enters clinical trials.[124,125]

Recent advances in AI-based approaches have revolutionized this space, moving beyond simple sequence alignment to deep learning architectures.[126,127] Models such as TCR-transformer or CNN-based encoders can now capture the complex biochemical grammar of the TCR-pMHC interface, allowing for the high-throughput prediction of novel TCRs and their specificities.[127-129] These AI-driven platforms assist in identifying potential off-target antigens by scanning the entire human proteome for structural similarities to the target peptide,[129] thereby mitigating the risk of clinical-grade cross-reactivity. By integrating these computational tools with experimental validation, the timeline for discovering safe, high-efficacy TCRs can be significantly shortened.[127] It also shifts the paradigm from simple predictive screening to the high-specificity *de novo* design of molecules. Groundbreaking research by Householder *et al.* demonstrated that deep learning tools, such as RFdiffusion and ProteinMPNN, can be harnessed to design novel α-helical TCR mimics from scratch. By employing "peptide-centric" engineering to enhance specificity and utilizing AI-driven in silico screening for precise off-target prediction, they have significantly streamlined safety assessment processes.[130] Building upon these foundations, Johansen *et al.* further optimized the design pipeline by introducing in silico cross-panning and creatively integrating Molecular Dynamics simulations to evaluate complex stability and reduce false-positive rates. Using this approach, they successfully engineered functional mini-binders for a specific neoantigen relying solely on AI-predicted pMHC structures, highlighting the immense potential of this methodology for personalized medicine.[131] Subsequently, Liu *et al.* showcased the broad utility of these methods by generating specific binders for 11 distinct pMHC targets. They introduced an efficient "Partial Diffusion" strategy to repurpose "privileged scaffolds". Rather than initiating a complete redesign, this strategy utilizes diffusion models to perform localized optimizations on validated protein frameworks, adapting them to new targets and substantially accelerating the development timeline.[132] Collectively, these advancements represent an increasingly mature framework for the rational, high-throughput design of potent and highly specific pMHC-targeting molecules. By significantly condensing experimental workflows, these AI-driven strategies provide a robust foundation for the advancement of personalized T-cell immunotherapies.

Novel targets

The ongoing search for new and precise tumor antigens is essential for the advancement of TCR-T cell therapy. Researchers are now prioritizing antigens with a strong focus on tumor specificity and little to no presence in normal tissues to ensure safety (Figure 3). TCRs are capable of identifying various antigens, categorized into tumor-associated antigens (TAAs) and tumor-specific antigens (TSAs) (Table 3). When considering which antigens to target, TSAs prove to be the superior choice over TAAs. This is because TSAs are solely present in cancerous cells, sparing healthy tissues from potential harm caused by T cell attacks.[133-135] By honing in on TSAs, the risk of off-target effects is significantly diminished, thereby increasing the effectiveness and safety of TCR-T cell therapy.

TAAs are proteins present on both cancer cells and some normal cells. These antigens are commonly elevated in tumors, aiding the immune system in recognizing and targeting cancerous cells. **Tissue differentiation antigens** are proteins found in specific tissue types that are often overexpressed or abnormally expressed in tumor cells. Some examples include MART-1,[67,68] gp100,[67] and CEA.[57] While TCRs targeting these antigens can have high affinity and have shown some clinical efficacy, their expression in normal tissues can lead to significant on-target/off-tumor toxicity.[57,67] It is essential to consider the potential risks associated with off-target effects when developing therapies targeting tissue differentiation antigens. Tumor cells often overproduce certain normal proteins, known as **overexpressed, non-mutated antigens**. This phenomenon is exemplified by Wilms' tumor 1 (WT1), a protein found in higher levels in various cancers. WT1 is a common marker for both hematologic and solid malignancies, serving as a potential target for cancer treatment and detection.[136] **Cancer germline antigens**, also called cancer-testis antigens, are proteins normally found in the testes and ovaries but

Table 3. Advantages and limitations of different antigen types in clinical applications.

	Classification	Example	Advantages & Disadvantages
TAAs	Tissue Differentiation Antigens	MART-1,[67] gp100,[67] CEA,[57]	High risk of off-target toxicity; naturally occurring TCRs may exhibit low affinity, necessitating TCR affinity modification may further mitigate off-target risks.[57,67,169]
	Overexpressed, Non-Mutated Antigens	WT1,[170]	Off-target risks; requiring sufficiently large differences from normal tissue expression levels.[171]
	Cancer Germline Antigens	NY-ESO-1,[73] MAGE-A3,[165] MAGE-A4,[137] MAGE-A10,[138]	Significantly reduced off-target effects on normal tissues; the same antigen may be expressed in multiple tumors; risk of antigen loss.[25,172,173]
	Onco-Viral Proteins	HPV16E6,[28] HPV16E7,[48] HBsAg, HBcAg,[77]	High immunogenicity with lower off-target toxicity, but limited applicability to virus-associated tumors only (approximately 8-10% of cancers).[174,175]
TSAs	Neoantigens	$KRAS^{G12D}$[145] $P53^{R175H}$[15]	High immunogenicity, low off-target toxicity, and risk of antigen loss.[176]
		Personalized neoantigens,[177]	Highly individualized design; prediction relies on whole-exome sequencing and HLA typing, resulting in high costs and prolonged timelines.[60,177]

aberrantly expressed by various tumors. One notable example is NY-ESO-1, which has displayed promising results in treating metastatic melanoma and synovial sarcoma.[27] The MAGE-A protein family, which includes MAGE-A3, MAGE-A4, and MAGE-A10, is a significant focus in TCR-T trials.[137-139] Currently, the only FDA-approved TCR-T product targets MAGE-A4 and has shown a 24% overall response rate in clinical trials.[137] Another potential target, PRAME, is also under investigation, showing a clinical response rate of 28.9% with no reported off-target toxicity.[81] These advancements in TCR therapy offer hope for improved treatments and outcomes for patients with various types of cancer.

TSAs are specific markers only present in tumor cells, making them ideal targets for T-cell immunotherapies. This unique characteristic allows for targeted treatment of cancer while minimizing harm to healthy cells. **Viral proteins** produced by oncoviruses like HPV can trigger cancer. The immune system sees these proteins as foreign invaders. The constant presence of HPV's oncoproteins E6 and E7 makes them promising targets for TCR-T cell therapies, especially for HPV-related cancers.[28,48] Clinical trials are also experimenting with TCR-T therapies for aggressive skin cancers linked to the Merkel cell polyomavirus (MCPyV).[140] These therapies offer hope in the fight against virus-induced cancers by targeting specific viral proteins that drive tumor growth. The potential of TCR-T therapy in combating these diseases shows promising outcomes in ongoing research. **Tumor-specific mutant antigens (Neoantigens)** arise from random mutations in the DNA of tumor cells. They are unique to the individual's tumor and are highly immunogenic.[141,142] These neoantigens are generally categorized into two types: private and public. The vast majority of neoantigens are "private", meaning they are unique to a single patient and require highly personalized therapy.[143,144] "Public" neoantigens (also called shared neoantigens) result from recurrent mutations found across multiple patients. A significant advantage of targeting public neoantigens, such as the aforementioned $KRAS^{G12D}$ and $TP53^{R175H}$, is their potential for broad clinical application.[15,145] Because these markers are frequently observed in specific cancer populations, a single validated TCR could be developed as an "off-the-shelf" therapeutic, making TCR-T therapy accessible to a much larger group of patients without the need for individual *de novo* antigen discovery. Apart from single-nucleotide variants (SNVs), tumors can also generate neoantigens through small insertions or deletions, known as indels. A recent study of 5,777 tumor samples across 19 cancer types found that kidney tumors had the highest proportion of indel mutations.[146] Interestingly, indels were shown to be a more abundant source of highly specific neoantigens compared to SNVs, generating a higher number of high-affinity neoantigens and mutation-specific binding antigens.[146,147] Furthermore, cancer cells can produce cryptic peptides from non-canonical translation of genomic regions that are normally not translated, such as untranslated regions (UTRs), long non-coding RNAs (lncRNAs), and alternative reading frames within protein-coding genes.[148] A compelling example of this phenomenon is the aberrant expression of endogenous retroviruses (ERVs). While typically silenced in healthy somatic tissues, ERVs can undergo upregulated transcription and translation in the context of cancer, serving as a potent source of tumor-specific cryptic antigens.[149] Researchers have already identified such cryptic antigens that can be recognized by T cells in pancreatic cancer.[148] By capitalizing on these distinct genetic characteristics of tumors, researchers aim to uncover more precise and effective neoantigens. Innovative methods, including deep learning algorithms, are being developed to predict and screen these antigens, including predicting TCR-antigen binding specificity.[150] This advancement holds promise in enhancing the effectiveness of cancer immunotherapy by targeting neoantigens more accurately.

Improved manufacturing and delivery

Enhancements in manufacturing and delivery are crucial for the improvement of adoptive T-cell therapies. Efficient gene delivery plays a vital role in engineering T cells for *in vivo* treatment (Figure 3). The field has progressed from basic viral vectors to more advanced non-viral methods and precision editing technologies.[151,152] These advancements are necessary for the optimization of T-cell therapies. By focusing on improved manufacturing processes and delivery systems, the potential for successful treatment outcomes can be significantly enhanced.

Initially, gamma-retroviruses were the primary choice for transducing T cells. However, they pose a significant safety risk because their integration sites often occur near gene promoters. If the inserted gene integrates near an oncogene or tumor suppressor gene, it can potentially transform normal cells into cancerous cells. This risk was tragically demonstrated in a patient undergoing gene therapy with a gamma-retrovirus, where insertional mutagenesis led to a case of clonal T cell acute lymphoblastic leukemia, highlighting the potential safety issues associated with these vectors.[153] Lentiviral vectors, on the other hand, present a more advantageous option compared to retroviral vectors. These vectors can accommodate longer gene segments, boast higher transduction efficiency, and provoke lower immunogenic responses. Nevertheless, the integration of lentiviral vectors still occurs randomly, thereby retaining the risk of insertional mutagenesis. Notably, there have been instances where gene insertion into the CBL gene led to the substantial expansion of T-cell clones.[154] Another safer alternative, adeno-associated viruses (AAVs), have emerged as viable contenders. AAVs possess the ability to infect both dividing and non-dividing cells while posing a lower risk of insertional mutagenesis than retroviral vectors. However, their packaging capacity is constrained to genes under 5 kb, and the gene integration rate of

Table 4. Challenges and Emerging Strategies in TCR-T Therapy.

Major Barrier	Description of Limitation	Emerging Strategies & Future Directions
HLA Restriction	Confines applicability to patients with matching HLA haplotypes.	Targeting HLA supertypes; structure-guided TCR engineering for cross-HLA recognition; and non-viral CRISPR platforms to eliminate specific HLA dependence.
Tumor Microenvironment	Immunosuppressive cells (TAMs, MDSCs, Tregs) and physical barriers (CAFs) inhibit T-cell infiltration and function.	Combination therapies with immune checkpoint blockade; engineering T cells to resist exhaustion; and using oncolytic viruses to remodel the TME.
Antigen Heterogeneity & Escape	Tumors may lack or lose targeted antigens, leading to immune evasion and relapse.	Developing multi-antigen targeting strategies; targeting vital driver mutations (e.g., KRAS, p53) that are less likely to be lost.
On-target/Off-tumor Toxicity	Cross-reactivity with similar antigens in healthy tissues can cause fatal damage.	Prioritizing highly specific TSAs; using AI-assisted algorithms for cross-reactivity prediction; and optimizing TCRs for "catch bonds" to improve sensitivity without increasing risk.
Persistence & Exhaustion	T-cell numbers drop over time due to chronic antigen stimulation and inhibitory signals.	Targeted cytokine delivery (e.g., IL-7, IL-15, IL-21); molecular reprogramming of transcription factors; and metabolic/epigenetic modulation.
Manufacturing Complexity & Cost	Personalized autologous production is time-consuming, expensive, and difficult to scale.	Advancing non-viral gene editing; developing *in vivo* mRNA delivery via LNPs or viral-like particles; and exploring allogeneic or iPSC-derived TCR-T cells.

AAVs remains relatively modest. An additional challenge arises from the high prevalence of pre-existing neutralizing antibodies to AAVs in the general population, with rates surpassing 30% in adults, severely impeding the effectiveness and accessibility of AAV-based therapies.[155]

The focus on developing non-viral methods for gene editing has intensified due to safety and efficiency concerns associated with viral vectors. One of the leading technologies in this field is the CRISPR-Cas9 system, which allows for precise gene editing while minimizing the risk of random insertion.[152] For instance, researchers have been able to utilize this system to simultaneously knock out endogenous TCR chains and introduce therapeutic TCR sequences downstream of the native TCR promoter in a single-step process,[60] improving transduction efficiency and enabling multiplex editing to enhance T-cell function or prevent exhaustion. However, traditional gene editing tools such as ZFNs, TALENs, and classic CRISPR-Cas9 rely on inducing DNA double-strand breaks (DSBs), which can lead to genotoxicity issues like chromosomal translocations and large deletions, especially when editing multiple genes. Efficiency, specificity, and controllability also remain suboptimal. In response to these challenges, the field has developed "CRISPR 2.0," which focuses on avoiding DSBs to enhance safety and precision in gene editing.[152] Newer techniques like base editing and prime editing have been introduced as part of CRISPR 2.0 to achieve single-base substitutions or small modifications without breaking the DNA double helix. Base editing involves fusing a catalytically impaired Cas9 with a deaminase to enable direct conversion of C → T or A → G bases, while prime editing uses a Cas9 nickase and a reverse transcriptase guided by a specialized pegRNA to "write in" new sequences at the target site.[152] These advancements significantly reduce the risk of genomic instability and offer new possibilities for safer, next-generation cell therapies.

A major challenge in traditional T-cell engineering is the high cost and complexity of *ex vivo* manufacturing processes that require lymphocyte depletion in patients. To make TCR-T therapy more accessible and affordable, researchers are exploring alternative strategies. The primary focus is on developing *in vivo* methods for direct T-cell modification, which would eliminate the need for complex *ex vivo* preparations and pre-conditioning chemotherapy, as well as prevent permanent insertion mutations.[156] One innovative approach involves using T-cell-specific fusogenic virus-like particles (T-FVLP$_{mCAR}$) that imitate the membrane fusion mechanism of HIV to efficiently and specifically produce CAR-T cells *in vivo*.[157] Another strategy uses targeted LNPs (tLNPs) to deliver mRNA directly to specific T cells *in vivo* via simple injection, generating CAR-T cells within the body.[158] Despite the potential of these methods, LNP-mediated mRNA transduction still faces challenges related to poor stability, low biocompatibility, degradation issues, and the risk of inducing systemic side effects *in vivo*.[159] Further research is necessary to confirm the long-term safety and efficacy of *in vivo* mRNA delivery methods for T-cell modification. These advancements hold promise for revolutionizing TCR-T therapy and making it more widely available to patients in need.

Allogeneic and iPSC-derived TCR-T cells present hopeful prospects as substitutes for autologous TCR-T therapy, potentially addressing its significant drawbacks. Although still in the early stages of development, these innovative methods show promise in resolving the limitations of current autologous treatments. Their potential to enhance TCR-T therapy effectiveness is a beacon of hope for future advancements in the field.

Combining TCR-T with oncolytic virotherapy

The integration of TCR-T therapy with other therapies presents a promising approach in the treatment of cancer. Along with the combination of cytokine therapy, immune checkpoint blockade, and epigenetic modulation, pairing TCR-T with oncolytic viruses (OVs) has shown potential in enhancing anti-cancer effects with strategies that "remodel" the TME. Rather than merely increasing the pool of available TAAs, OVs primarily benefit TCR-T therapy by several functions. Firstly, OV-mediated lysis triggers the release of pathogen-associated molecular patterns (PAMPs) and danger-associated molecular patterns (DAMPs), effectively turning "cold" tumors to "hot" tumors. Then, the resulting inflammatory response promotes the expression of adhesion molecules and chemokines like CXCL9 or CXCL10, which are critical for the recruitment and deep penetration of TCR-T cells into the tumor core. Furthermore, OVs can be engineered to express cytokines like IL-12 or IFN-γ that further counteract the suppressive nature of the TME, thereby sustaining the activity of infiltrated TCR-T cells.[160] This combined action of TCR-T and OVs works together to improve the effectiveness of T cell therapy in eliminating tumors, offering a new avenue in cancer treatment.

CONCLUSION AND FUTURE DIRECTIONS

TCR-T therapy is a groundbreaking frontier in solid tumor immunotherapy, offering hope and promise in the battle against cancer. Despite facing challenges (Figure 1 & Table 4), recent clinical successes, including FDA approvals and impressive results from trials targeting antigens like MAGE-A4, NY-ESO-1, and HPV-E7, demonstrate the potential of this innovative approach. By targeting a diverse range of tumor antigens and constantly improving T-cell engineering, tumor microenvironment modulation, and combination strategies, TCR-T therapy is poised to revolutionize treatment for patients with resistant solid malignancies. The future holds great potential for this therapy, with further breakthroughs on the horizon that will make it

a standard of care for a variety of solid tumors. This transformative therapy offers new hope for patients who have exhausted traditional treatment options, opening up a new era in cancer treatment that holds promise for improved outcomes and quality of life.

The advancement of TCR-T therapy for solid tumors holds immense promise for revolutionizing cancer treatment, but there are crucial areas that must be addressed to unlock its full potential (Figure 3 & Table 4). One of the primary challenges is overcoming HLA restriction, which can be achieved by developing versatile TCRs capable of targeting HLA supertypes, or creating personalized TCR-T products that are not dependent on HLA compatibility through innovative gene-editing technologies such as CRISPR 2.0. Furthermore, continuous effort is required for the discovery and validation of highly specific TSAs, such as neoantigens from indels or cryptic peptides, to enhance safety and reduce "on-target, off-tumor" toxicity. Additionally, efforts should be directed towards devising strategies to counter the immunosuppressive TME and prevent T-cell exhaustion. This may involve enhancing T cell persistence, metabolic fitness, and resistance to exhaustion signals through targeted cytokine delivery, molecular reprogramming, or epigenetic modulation. Moreover, the accessibility and affordability of TCR-T therapy need to be improved through the development of safer, non-viral, and potentially *in vivo* T-cell engineering and delivery methods. Simplifying the complex manufacturing process will be essential for the widespread adoption of this groundbreaking therapy. Finally, exploring innovative combination therapies, such as combining TCR-T with immune checkpoint inhibitors or oncolytic viruses, could further enhance T-cell efficacy and transform "cold" tumors into hotspots of immune activity. By addressing these critical areas, the potential of TCR-T therapy for solid tumors can be fully realized, bringing new hope to cancer patients worldwide.

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All authors contributed to and approved the final manuscript. Q.X. is the corresponding author, has full access to all the data and has final responsibility for the decision to submit for publication.

DECLARATION OF INTERESTS

The authors declare no competing interests.

DATA AND CODE AVAILABILITY

No data was used for the research described in the article.

SUPPLEMENTAL INFORMATION

It can be found online at https://doi.org/10.59717/j.xinn-med.2026.100212.

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