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NK Cell Technology for Tumour Treatment: Advances, Advantages, Challenges, and Clinical Prospects

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Abstract. The natural cytotoxic properties of natural killer (NK) cells together with their suitability for genetic modification have attracted increasing interest in tumour immunotherapy. This review evaluates how NK cell-based therapies have developed, with particular attention to chimeric antigen receptor (CAR)-NK technology, therapeutic benefits, current limitations and clinical progress. NK cell-based therapies offer potential benefits including allogeneic "off-the-shelf" use, a favourable safety profile and multiple mechanisms of tumour recognition. Progress in receptor engineering, ex vivo expansion and renewable cell platforms has also improved the prospects for producing NK cell therapies on a broader scale. Early clinical studies have also reported encouraging responses in haematological malignancies, while studies in other tumours have demonstrated the feasibility of other targets and delivery approaches. The effectiveness of these therapies can nevertheless be reduced when the transferred cells do not persist, fail to enter tumour tissue efficiently or lose function after exposure to the tumour environment. Clinical investigations have shown that these therapies can be delivered to patients, although the amount of evidence and the responses observed vary considerably across tumour types. Overall, NK cell technology shows considerable potential for cancer treatment, but further development is required to achieve better clinical responses.

Keywords: natural killer cells, CAR-NK cells, tumour immunotherapy, adoptive cell therapy, tumour microenvironment

1. Introduction

Cancer remains a major global health burden and is responsible for approximately 9.7-10 million deaths each year. Around 20 million new cases occurred globally in 2022, and this figure is expected to rise by approximately 77%, reaching about 35 million cases per year by 2050 [1]. Tumours are highly heterogeneous among patients due to different tissue origins, random genetic mutations and responses to treatment, all of which make treatments particularly challenging. Therefore, there is an increasing need for novel therapeutic strategies that can improve treatment efficacy while reducing treatment related side effects.

A wide range of therapeutic methods has been used clinically as standard treatment, including chemotherapy, radiotherapy, targeted therapy, immunotherapy and surgery. More recently, therapeutic strategies that direct immune activity against malignant cells have become an important

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part of cancer treatment. One such strategy is adoptive cell therapy, in which immune cells with anti-tumour activity are used to target malignant cells directly [1]. Clinical advances have been reported with both immune checkpoint blockade and cell-based approaches such as chimeric antigen receptor (CAR) T-cell (CAR-T) therapy. However, each approach also has limitations. Chemotherapy and radiotherapy may damage healthy tissues and lead to serious side effects and complications, and are also less capable of treating metastatic tumours. Tumours can also acquire drug resistance due to their heterogeneity. CAR-T treatment has been particularly effective against some haematological malignancies, whereas comparable success has been more difficult to achieve in other tumours. The challenges include poor infiltration into solid tumour tissues and an immunosuppressive tumour microenvironment. Treatment can also produce clinically important toxicities, particularly cytokine release syndrome and neurological complications.

Natural killer (NK) cells provide an alternative platform for adoptive immunotherapy because tumour cell killing is part of their normal role in innate immunity [1]. Their activity does not require prior antigen sensitisation or major histocompatibility complex (MHC)-dependent recognition as T cells do. Genetic engineering can add CAR-mediated targeting to this existing activity, which means that CAR-NK cells may respond through both the introduced receptor and their endogenous NK cell receptors. These properties, together with the possibility of using donor-derived cells and the relatively favourable toxicity profile reported for NK cell products, have encouraged the development of NK cell therapies as an alternative to CAR-T therapy.

Development of NK cell products has also involved evaluating different starting materials. Peripheral blood, cord blood and induced pluripotent stem cells (iPSCs) offer distinct options for producing therapeutic NK cells and may differ in their suitability for consistent manufacturing [1]. Both experimental studies and early clinical investigations have subsequently extended NK cell therapy across several tumour settings. However, challenges including limited persistence, poor tumour infiltration and suppression within the tumour microenvironment still restrict the clinical effectiveness of NK cell therapy.

The following sections examine the biological basis and development of NK cell therapy and then consider its therapeutic advantages and the barriers that restrict its activity. Current clinical applications are then evaluated alongside the main directions for future development.

2. Biological basis and development of NK cell-based tumour therapy

2.1. NK cell recognition and anti-tumour activity

Whether an NK cell attacks a target depends on the combined signals generated by its surface receptors. Recognition of MHC-I by inhibitory receptors such as killer cell immunoglobulin-like receptors (KIRs) and natural killer group 2 member A (NKG2A) normally restrains cytotoxic activity, whereas receptors including natural killer group 2 member D (NKG2D), natural killer cell p30-related protein (NKp30), natural killer cell p44-related protein (NKp44) and natural killer cell p46-related protein (NKp46) can respond to stress-related changes on abnormal cells [1]. Loss of MHC-I on a tumour cell can therefore weaken inhibitory input and favour NK cell activation through the missing-self mechanism. Once this balance shifts towards activation, several routes can contribute to target cell elimination. Perforin and granzymes released from cytotoxic granules can initiate apoptosis, while Fas ligand and tumour necrosis factor-related apoptosis-inducing ligand (TRAIL) provide an additional death receptor route. Antibody-bound targets may instead be recognised through cluster of differentiation 16 (CD16), a process that produces antibody-dependent cellular cytotoxicity (ADCC). Activated NK cells can also release interferon gamma (IFN-γ) and

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tumour necrosis factor (TNF), extending their effect beyond direct target-cell killing [1]. Although these mechanisms allow NK cells to recognise tumour cells without antigen-specific receptors, natural NK cell activity alone may be insufficient to achieve sustained tumour control.

2.2. Development of CAR-NK cells

Natural NK cell recognition does not always generate enough activity to control tumour growth, particularly when malignant cells reduce activating signals or create conditions that suppress NK cell function. CAR modification adds a predefined antigen recognition route to address this limitation. In a typical CAR, target binding occurs through an extracellular recognition region, which is linked through hinge and membrane-spanning components to intracellular domains responsible for activating the cell. This engineered system operates alongside rather than instead of the native NK cell machinery. CAR-NK cells can therefore respond to the selected antigen through the CAR while retaining endogenous receptor mediated cytotoxicity.

This concept has subsequently entered clinical testing. Marin et al. administered interleukin-15 (IL-15)-expressing, cluster of differentiation 19 (CD19)-directed allogeneic CAR-NK cells to patients with CD19-positive B-cell malignancies [2]. A different clinical application was examined by Burger et al., who delivered human epidermal growth factor receptor 2 (HER2)-directed CAR-NK cells intracranially in recurrent glioblastoma [3].

2.3. Cell sources and preparation for CAR-NK therapy

Several types of starting material have been used to generate CAR-NK products: donor peripheral blood, cord blood, iPSCs and established NK cell lines. The choice of starting material can affect cell expansion, functional activity, consistency and clinical application, which makes cell source selection an important part of CAR-NK manufacturing.

Peripheral blood supplies mature cytotoxic NK cells directly, although the number collected is usually too small for treatment without substantial expansion in culture. Culture protocols therefore commonly use cytokine stimulation, including interleukin-2 (IL-2), IL-15 or interleukin-21 (IL-21), sometimes in combination with feeder cells to increase cell yield while supporting NK cell function. However, effective manufacturing requires a balance between cell quantity and functional quality. In the study by Zhang et al., exposing the cells first to IL-21 and subsequently to IL-15 produced a large increase in cell numbers without losing the required functional characteristics [4]. Variation between donors may still affect the consistency of peripheral-blood-derived products.

Cord blood represents another donor-derived starting material and is compatible with both ex vivo growth and genetic modification. Its feasibility as a CAR-NK source has been demonstrated by Marin et al. In the research, the therapeutic product was generated from cord-blood NK cells and administered to patients with B-cell malignancies [2]. Their study also showed that characteristics of the original cord-blood unit were associated with CAR-NK expansion and treatment response, which suggested that starting material can influence the final product. iPSCs differ from these donor sources because the starting population can be renewed and propagated before being directed towards an NK cell lineage [5].

Among established NK cell lines, NK-92 has been used extensively as a platform for CAR modification. Because NK-92 cells can proliferate continuously, the clinical product must be irradiated before administration. This safety measure also restricts how long the cells remain after treatment. Burger et al. used this irradiated CAR-NK-92 platform to target HER2 in recurrent glioblastoma [3]. Overall, no single source provides every desirable feature. Starting material is

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therefore not simply a manufacturing choice; it also helps determine the biological characteristics of the therapeutic product.

3. Advantages of CAR-NK cell therapy

Several features of the NK cell platform are relevant to its translation into treatment. Donor-derived products may allow cells to be prepared in advance, and the safety characteristics of NK cells make this allogeneic strategy more practical. CAR modification adds defined antigen targeting without removing endogenous NK cell recognition, while renewable sources may offer greater control over manufacturing. These features are related because product availability, safety, biological activity and manufacturing consistency together determine how practical NK cell therapy may become in clinical practice.

3.1. Allogeneic and off-the-shelf potential

A key practical difference between NK cell-based therapies and conventional autologous CAR-T approaches is how the starting cells can be obtained. CAR-T products are commonly produced separately from each patient's own cells, so treatment depends on completing an individual manufacturing process. In contrast, NK cells may be collected from donor or renewable sources and prepared for use in more than one recipient. This creates the possibility of producing batches in advance rather than beginning manufacturing only after an individual patient requires treatment [5].

The importance of this approach extends beyond treatment availability. A single allogeneic source may potentially be used to produce multiple therapeutic doses, which reduces the need to repeat the manufacturing process for each patient. This could support larger scale production and potentially reduce treatment costs compared with fully individualised therapy [6]. The different NK cell sources described in section 2.4 also provide flexibility in selecting starting materials suitable for large scale production.

However, the term "off-the-shelf " does not mean that NK cell production is already simple or inexpensive. Products prepared in advance still require reliable expansion, storage and quality control, while cryo-preservation can negatively affect NK cell viability and function [5]. Therefore, the current advantage of NK cells is better considered as their potential for being more scalable and readily available before clinical treatment, rather than just an established solution to the cost and complexity of adoptive cell therapy.

3.2. Safety profile

The feasibility of using cells from another donor also depends strongly on safety. Donor T cells may respond to mismatched recipient antigens through their T cell receptors and subsequently damage recipient tissues, producing graft-versus-host disease (GvHD). NK cell activation is governed differently: the response reflects the combined input from activating and inhibitory receptors rather than antigen recognition through a T cell receptor. Consequently, allogeneic NK cell treatment has a substantially lower association with GvHD [5].

The toxicity pattern observed with CAR-NK treatment also differs from that commonly associated with CAR-T cells. Severe cytokine release syndrome (CRS) and neurological toxicity are major concerns after CAR-T therapy, whereas these events have generally been less severe in studies of CAR-NK cells. Differences in the cytokines released by activated NK and T cells have been proposed as one explanation, although the mechanism remains uncertain [6]. Clinical observations

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are consistent with this safety profile. Marin et al. reported only limited CRS and observed neither neurotoxicity nor GvHD following allogeneic cord-blood CAR-NK treatment [2]. This provides patient-level evidence for the safety characteristics proposed based on NK cell biology.

3.3. Duality of anti-tumour activity and manufacturing consistency

CAR engineering provides antigen-specific recognition while NK cells retain their endogenous receptors and natural cytotoxic mechanisms. As a result, CAR-NK cells are not completely dependent on the CAR for tumour recognition and may continue to respond through their natural activating receptors or by recognising antibody-coated targets through CD16 [5].

This second route of recognition becomes especially relevant when antigen expression differs between tumour cells. CAR-mediated activity depends on expression of the selected target antigen, but antigen expression may vary between tumour cells or decrease during treatment. Retaining CAR-independent NK-cell activity therefore provides an additional route for recognising abnormal cells and reducing full dependence on a single tumour-associated antigen [6]. However, this should be considered an additional protective mechanism rather than a complete solution to antigen escape, as tumours can evade immune recognition through several pathways.

A further advantage is the possibility of developing more standardised NK-cell products. Primary donor-derived cells can vary between individuals, whereas renewable cell sources might be able to provide a more consistent starting population of cells for manufacturing. iPSCs are particularly relevant because they can be expanded and genetically modified before differentiation into NK cells. This could allow defined engineered cell populations to be produced repeatedly from the same starting material. Importantly, the feasibility of this approach has progressed into clinical development. There is a study demonstrating that an iPSC-derived genetically modified NK-cell product could be manufactured and administered clinically [7].

Overall, the advantages of NK and CAR-NK cells work together to support their clinical use rather than act as separate features. The allogeneic nature of NK and CAR-NK cells would be much less useful without a favourable safety profile, because donor-derived products must be suitable for use across different patients. Renewable cell sources may further strengthen this approach by improving manufacturing consistency and supporting larger scale production. The combination of CAR-mediated targeting and natural NK cell recognition reduces complete dependence on a single tumour antigen, which may be particularly relevant in heterogeneous tumours. Together, these features give NK cell therapy a different development pathway from highly individualised cell therapies such as autologous CAR-T treatment. However, these advantages can only translate into lasting clinical benefit if the therapeutic cells can survive, reach tumour tissue and remain functional.

4. Challenges limiting CAR-NK cell therapy

Despite the advantages discussed above, NK and CAR-NK cells still face several biological barriers after manufacturing. These limitations occur at different stages of treatment and are closely connected. Therapeutic cells must first survive for long enough to maintain anti-tumour activity, then migrate towards the tumour and enter the tumour tissue. Even after reaching the tumour site, they must remain functional under conditions such as hypoxia, nutrient competition, acidity and immunosuppressive signalling. Failure at any of these stages can reduce the overall therapeutic effect. This section therefore examines the main barriers affecting persistence, tumour homing and infiltration, and NK cell function within the tumour microenvironment (Table 1).

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4.1. Limited persistence and functional fitness

One limitation becomes apparent after treatment, which is that transferred NK and CAR-NK cells may remain in the patient for only a limited period. A sufficient number of functional cells must remain in vivo to maintain continuous anti-tumour activity. Maintaining these cells after infusion requires continued survival signals, with IL-15 having an especially important role. Limited persistence may therefore reduce the duration of tumour control and restrict the long-term effectiveness of treatment.

Persistence is not determined only by cell survival, but also by whether the remaining cells retain sufficient functional fitness. Li et al. found that interactions with metabolically active tumour cells changed the metabolic state of CAR-NK cells and were accompanied by weaker anti-tumour function [8]. These cells showed loss of metabolic function, including impaired mitochondrial function, and their accumulation was later associated with tumour resistance following CAR-NK treatment [8]. This suggests that CAR-NK cells may remain present but progressively lose the metabolic function required for effective tumour killing. The same study found that IL-15 engineering could improve metabolic fitness and anti-tumour activity, further emphasising the importance of survival and metabolic signalling in maintaining CAR-NK function [8].

4.2. Poor tumour homing and infiltration

Even functionally active NK cells cannot efficiently eliminate tumour cells unless they reach the tumour site. NK cell trafficking is regulated by interactions between chemokines produced within tissues and corresponding receptors expressed on NK cells. Core chemokine pathways include the C-X-C motif chemokine receptor 3 (CXCR3)–C-X-C motif chemokine ligand 9 (CXCL9) / CXCL 10/ CXCL 11axis, the C-C chemokine receptor type 5 (CCR5)–C-C motif chemokine ligand 5 (CCL5) axis, and the C-X3-C motif chemokine receptor 1 (CX3CR1)–C-X3-C motif chemokine ligand 1 (CX3CL1) axis. However, tumour derived chemokine profiles do not always correspond to the receptors expressed by therapeutic NK cells. The receptor profile may also change during culture and expansion, which means that manufacturing itself can modify how efficiently the resulting NK cells migrate [9].

Reaching the tumour-associated vasculature is only the first step. NK cells must adhere to endothelial cells, leave the circulation and migrate through the extracellular matrix before reaching tumour cells. Abnormal tumour vasculature and dense stromal structures can restrict these processes. NK cell movement through tissues involves adhesion molecules and matrix-remodelling enzymes such as matrix metalloproteinases, which means that changes in these systems can further affect infiltration [9]. Therefore, increasing the cytotoxic capacity of NK cells alone may have limited therapeutic benefit if insufficient numbers of cells can physically reach tumour cells.

Table 1. Key barriers limiting NK and CAR-NK cell therapy: mechanisms and consequences

Barrier	Main Mechanism	Effect on NK Cell Therapy	Ref.
Limited persistence and functional fitness	Insufficient survival signalling and loss of metabolic fitness	Reduced persistence and anti-tumour activity	[8]

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Table 1. (continued)

Poor tumour homing and infiltration	Mismatch between tumour-derived chemokines and NK cell chemokine receptors; abnormal vasculature and stromal barriers	Reduced migration and entry of NK cells into tumour tissue	[9]
Hypoxia	Impaired induction of NKp30, NKp44, NKp46 and NKG2D under low-oxygen conditions	Weaker receptor-dependent tumour killing	[10]
Lactate and extracellular acidification	Intracellular acidification, mitochondrial dysfunction, increased mitochondrial ROS and reduced ATP production	Reduced NK-cell survival and increased apoptosis	[11]

4.3. Hypoxia and metabolic stress

NK cells that successfully enter tumour tissue face further suppression from the tumour microenvironment. One important feature is hypoxia, which commonly develops when rapid tumour growth exceeds the local oxygen supply. Balsamo et al. found that after cytokine stimulation, NK cells exposed to low oxygen showed weaker induction of several activating receptors, specifically NKp30, NKp44, NKp46 and NKG2D [10]. This was accompanied by reduced natural cytotoxicity against tumour targets [10]. Hypoxic adaptation was associated with increased hypoxia-inducible factor 1-alpha (HIF-1α), indicating that reduced oxygen availability can directly alter the functional phenotype of NK cells.

However, hypoxia does not suppress all NK cell killing mechanisms equally. A study has found that CD16 expression and antibody-dependent cellular cytotoxicity were relatively preserved under hypoxic conditions [10]. This distinction is important because it suggests that the tumour microenvironment can selectively impair particular NK cell functions rather than cause complete loss of cytotoxicity. The effect of hypoxia also occurs along with other forms of metabolic pressure. Tumour cells have high metabolic demands and compete with immune cells for nutrients, while CAR-NK cells can progressively lose metabolic fitness following interaction with tumour cells [8]. Consequently, even when NK cells successfully infiltrate a tumour, the local environment can reduce their ability to maintain effective cytotoxic activity.

4.4. Acidosis and immunosuppressive tumour microenvironment

In addition to hypoxia and nutrient competition, altered tumour metabolism creates another barrier through the accumulation of lactate and extracellular acidification. High rates of tumour glycolysis can increase the release of both lactate and protons into the surrounding environment. Using colorectal liver metastases, Harmon et al. linked these acidic metabolic conditions to impaired survival of resident NK cells [11]. When liver-resident NK cells were exposed to lactic acid, their intracellular pH fell and mitochondrial function deteriorated. This was accompanied by greater mitochondrial reactive oxygen species (ROS) and lower adenosine triphosphate (ATP) levels [11]. These ultimately promote NK cell apoptosis [11].

These metabolic effects occur together with immunological suppression. Suppression also arises from immune signalling within the tumour. Transforming growth factor beta (TGF-β) can inhibit

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NK cell function, while regulatory T cells (Tregs), myeloid-derived suppressor cells (MDSCs) and tumour-associated macrophages provide additional sources of inhibitory activity [9]. These mechanisms often act together, as hypoxia, nutrient competition, acidic conditions and inhibitory signalling can co-exist within the tumour microenvironment. As a result, improving only one aspect of NK cell function may be insufficient to restore anti-tumour activity.

Taken together, these limitations are better understood as successive obstacles during treatment rather than as independent defects. Limited survival reduces the number of therapeutic cells available to reach the tumour, while inefficient homing and infiltration further decrease the number of cells that enter tumour tissue. Those that successfully reach the tumour must then maintain metabolic fitness and cytotoxic activity despite hypoxia, nutrient competition, acidity and immunosuppressive signals. These barriers can therefore accumulate, which means that reduced efficiency at one stage can make the limitations encountered at later stages even more important. This helps explain why NK cells with strong cytotoxic activity under controlled experimental conditions may show weaker therapeutic activity in vivo. It also suggests that improving a single feature of NK cells may be insufficient if other stages of the treatment process remain limited.

5. Clinical applications and ongoing trials

5.1. Clinical development of CAR-NK therapy

Clinical applications for CAR-NK therapy have developed most clearly in haematological malignancies. One reason is that malignant cells in these diseases are comparatively accessible, while antigens such as CD19 provide established therapeutic targets. Outside haematological disease, development is more difficult because variation between tumour cells, inefficient cell trafficking and local immunosuppression can all interfere with treatment. The phase 1/2 CD19 study therefore provides a useful example of the more advanced clinical evidence available in B cell disease. Of the 37 patients treated, 18 responded, which corresponded to a response rate of 48.6% at both the day 30 and day 100 assessments [2]. Patients who responded also showed stronger expansion and longer persistence of the infused CAR-NK cells [2]. Importantly, treatment outcome was also related to characteristics of the original cord-blood units. This suggests that efficacy is affected not only by CAR-mediated tumour recognition, but also by the biological properties of the therapeutic cell product. The findings therefore support the importance of cell expansion, persistence and source material selection in determining treatment outcome.

In comparison, clinical development in other tumours has been more limited and required different approaches to overcome barriers associated with tumour accessibility and the local tumour microenvironment. Burger et al. tested local intracranial delivery of an HER2-directed CAR-NK-92 product in a cohort of nine patients with recurrent glioblastoma [3]. Five patients achieved stable disease lasting between 7 and 37 weeks, although no objective responses were observed [3]. Rather than providing strong evidence of efficacy, this study is important because it demonstrates a different approach to CAR-NK delivery. Direct intracranial administration places the therapeutic cells at the tumour site and may reduce dependence on systemic trafficking, which is one of the most challenging factors discussed previously. The findings therefore indicate that placing the cells at the tumour site does not by itself resolve the other biological limitations affecting treatment.

Clinical exploration has also moved beyond the more established CAR targets. For example, Li et al. described the use of a roundabout guidance receptor 1 (Robo1)-directed CAR-NK product in one patient whose pancreatic ductal adenocarcinoma had metastasised to the liver [12]. Following treatment, disease control was reported for approximately five months [12]. This case expands the

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potential clinical application of CAR-NK therapy beyond commonly studied targets such as CD19 and HER2. However, evidence from a single patient cannot establish the efficacy of therapy and the outcomes should be interpreted more cautiously.

Taken together, these studies show different levels of maturity of current CAR-NK applications. Evidence in B cell malignancies has progressed to larger early phase studies with measurable response rates, whereas evidence in other tumours remains largely based on small early phase studies. Current evidence of clinical development supports the feasibility of CAR-NK therapy across different cases, but the outcomes of these studies also show that feasibility alone does not necessarily translate into consistent clinical efficacy. Clinical feasibility shows that these products can be manufactured with an acceptable safety profile for patients, whereas therapeutic efficacy also depends on whether enough active cells remain after infusion, arrive at the tumour site and continue to function there. Differences in target antigen expression and the tumour microenvironment may further contribute to the variable responses observed between patients and tumour types.

5.2. Expanding NK cell therapeutic strategies

CAR engineering is only one route being used to enhance NK cell therapy. Another strategy is cytokine-induced memory-like (CIML) activation, in which prior cytokine exposure alters the subsequent anti-tumour response of NK cells. Shapiro et al. translated this concept into patients with advanced head and neck cancer by administering CIML NK cells alongside immunomodulatory treatment [13].

The combination treatment also produced opposing biological effects. Although ipilimumab promoted early expansion of the transferred NK cells, recipient cluster of differentiation 8 (CD8)+ T cells recovered more rapidly at the same time. This immune recovery was linked to earlier elimination of the human leukocyte antigen (HLA)-mismatched donor NK population [13]. These findings further emphasise that although combination treatments may improve NK cell activity, they may also reduce NK cell persistence.

Together, current clinical studies show that NK cell therapy can be developed through several different approaches. However, the evidence also indicates that clinical outcome depends on more than the cytotoxic capacity of the infused cells. Cell source characteristics, expansion, persistence, delivery and interactions with the recipient immune system can all influence therapeutic performance.

6. Future perspectives

Future development of NK cell therapy will therefore require greater attention to the overall performance of the therapeutic product rather than a simple increase in its cytotoxic activity. Effective NK cell therapies will need to combine reliable tumour recognition with sufficient persistence, metabolic fitness, tumour trafficking and resistance to the suppressive tumour microenvironment [1, 8]. At the same time, more standardised cell platforms may help reduce product variability and support larger scale manufacturing. The clinical development of iPSC-derived engineered NK cells provides one potential route towards more reproducible products [7].

Further engineering may also be used to address specific biological limitations of NK cells. Strategies that improve cytokine support, tumour homing or resistance to inhibitory signals may help therapeutic cells maintain their activity after administration. Gene editing approaches could provide another route for modifying these properties in a more controlled way. However, these

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modifications should be evaluated together rather than only according to their effect on cytotoxicity, as improvements in one function may not overcome limitations at other stages of treatment.

Most importantly, stronger clinical evidence is still required. Current evidence ranges from individual case reports to small phase I studies and larger early phase trials, which makes it difficult to compare the long-term effectiveness of different NK cell platforms directly. Future trials should test these products in larger cohorts, follow patients for longer periods and assess both therapeutic response and the behaviour of the transferred cells over time. Comparative studies will also become important as the field develops, especially for determining whether improvements observed in early-phase studies provide meaningful advantages over existing treatment approaches. Overall, NK-cell-based treatment has now moved beyond a primarily experimental concept and entered early clinical development, but its future success will depend on translating its favourable biological characteristics into long-lasting benefits for patients.

7. Conclusion

NK cell technology provides a promising approach to tumour treatment by combining natural anti-tumour activity with the potential for further engineering and allogeneic use. The field has developed from studies of basic NK cell tumour recognition to the clinical testing of genetically modified NK cell products. The ability of NK cells to combine CAR-mediated targeting with natural NK cell recognition provides more than one route for tumour cell elimination, while their relatively favourable safety profile supports the development of allogeneic products. The use of different cell sources including cord blood and iPSCs has also expanded the possibilities for producing more accessible products. Importantly, early clinical studies have moved these concepts beyond preclinical development and demonstrated clinical feasibility in patients. However, the evidence reviewed here shows that these advantages alone are not sufficient to ensure effective treatment. The therapeutic outcome depends on whether NK cells can remain functional, reach the tumour and maintain their activity within the tumour microenvironment. Experience from early patient studies indicates that both unmodified and CAR-engineered NK cell approaches can be delivered with manageable safety profiles, but consistent and durable responses have not yet been fully established across different types of tumour or different individuals. Nevertheless, the responses observed in B cell malignancies together with the clinical evaluation of alternative targets, cell sources and delivery approaches show that NK cell technology can be adapted to different therapeutic backgrounds. These developments provide a strong basis for continued optimisation rather than suggest a fundamental limitation of the therapeutic approach. The remaining question is therefore how consistently these biological advantages can be converted into meaningful benefit for patients. Progress in this area will shape how widely NK cell-based treatment can ultimately be used in cancer treatment.

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