BIOCELL

Tech Science Press

Doi:10.32604/biocell.2025.072104

REVIEW

# The Therapeutic Potential of iNKT Cells in the Treatment of Ovarian Cancer

**Anna Pawłowska-Łachut*, Dorota Suszczyk and Iwona Wertel**

Independent Laboratory of Cancer Diagnostics and Immunology, Medical University of Lublin, Chodźki 1, Lublin, 20-093, Poland

*Corresponding Author: Anna Pawłowska-Łachut. Email: anna.pawlowska-lachut@umlub.edu.pl

Received: 19 August 2025; Accepted: 24 November 2025; Published: 21 April 2026

**ABSTRACT:** Ovarian cancer (OC) remains the most lethal gynecological malignancy, and it is characterized by high heterogeneity, early metastatic dissemination, and frequent recurrence within 12–18 months after primary therapy. Despite progress in clinical management and drug development, the mortality rate remains high, and the biological drivers of OC aggressiveness are not fully understood. A major contributor to therapeutic resistance and disease progression is the ovarian tumor microenvironment (TME), which supports tumor growth and immune evasion. Its complexity poses significant challenges to the development of effective therapies. Current treatments, especially in advanced or recurrent stages, have limited efficacy. While immune checkpoint inhibitors (ICIs), such as anti-programmed death receptor (PD-1) and anti-programmed death ligand-1 (PD-L1) monoclonal antibodies, have revolutionized the treatment landscape of several solid tumors, their effectiveness in OC remains modest due to the non-inflamed, immunosuppressive nature of the disease. This highlights the need for alternative, more robust immunotherapeutic strategies. Invariant natural killer T (iNKT) cells engineered with chimeric antigen receptors (CARs) or T cell receptors (TCRs) are emerging as powerful candidates for next-generation adoptive cell therapies. This study aims to evaluate the therapeutic potential of iNKT cells in OC and to discuss their capacity to overcome immune resistance within the TME as a promising approach for next-generation immunotherapy. These dual-specific effector cells combine innate and adaptive properties, offering advantages such as human leukocyte antigen (HLA)-independent tumor recognition, natural tumor site homing, and the ability to modulate immunosuppressive TME. Preclinical studies have demonstrated their potential to overcome immune resistance and enhance antitumor responses in solid tumors, including OC. Altogether, iNKT-based therapies represent a promising and versatile platform to address the urgent need for more effective treatments for ovarian cancer.

**KEYWORDS:** iNKT cells; adoptive cell therapy; ovarian cancer; tumor microenvironment

## 1 Ovarian Cancer as a Silent Killer of Women

Ovarian cancer (OC) is considered to be the third most common and the most fatal malignancy of the female reproductive tract. According to data published by the World Health Organization (WHO) in 2022, as many as 324,603 women were diagnosed with the disease, and 206,956 died as a result of OC [1]. The prognosis for the next twenty years is pessimistic. According to WHO predictions, the estimated number of new OC cases in 2045 will total 476,893 and 645,373 deaths [1]. The estimated numbers of new cases and deaths associated with OC are presented in Fig. 1.

At the early stages, the disease is asymptomatic or has vague symptoms, thus OC is mostly diagnosed at advanced stages (approximately 70% of cases). Nonspecific symptoms include, among others, dyspeptic manifestations, changes in bowel movement and urinary frequency, abdominal pain, and bloating. Most patients with OC report having experienced above mentioned symptoms before diagnosis, but due to

their commonness, healthcare providers often do not investigate their underlying cause. Therefore, OC often remains undiagnosed until advanced stages (III and IV according to the International Federation of Gynecology and Obstetrics [FIGO]) [2,3].

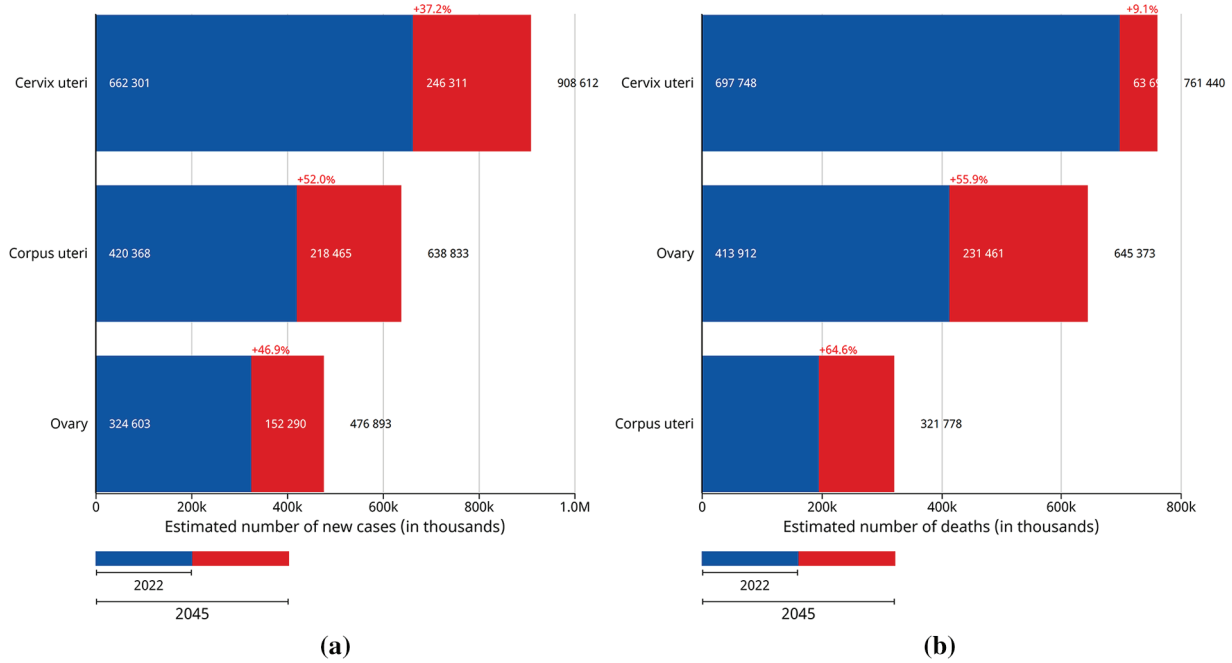


**Figure 1:** The estimated numbers of new cases (**a**) and deaths (**b**) associated with ovarian cancer (OC) [1]

The prognosis for women with OC is worrisome, and the five-year survival rate is only 47% [4]. Despite that the rate for patients with early disease (I FIGO stage) is 90%, it drops precipitously to 25% when metastases occur [2]. Standard management of OC involves cytoreductive surgery followed by platinum-based chemotherapy. Although over 80% of patients initially achieve remission and subsequent treatments are often ineffective due to drug resistance. Platinum-resistant disease remains particularly difficult to treat, with limited therapeutic options and poor survival outcomes. Therefore, developing new strategies to overcome resistance and improve patient prognosis has become a major research priority. Advances in molecular profiling have deepened our understanding of OC biology, leading to the emergence of targeted therapies designed to enhance efficacy and reduce toxicity by selectively interfering with key pathways driving tumor growth and progression [5].

The advances in treatment of OC include combining the standard therapy, i.e., platinum and taxane-based chemotherapy and primary debulking surgery, with biological drugs [6,7]. Targeted therapies aim to inhibit tumor growth by interfering with specific molecular or metabolic pathways and represent a major focus of OC research. Key approaches include antiangiogenic agents that block tumor blood vessel formation, thereby restricting tumor expansion, though resistance remains a challenge. Poly(ADP-ribose) polymerase inhibitor (PARPi) exploits defects in DNA repair, particularly in breast cancer susceptibility gene (BRCA) 1/2-mutated or homologous recombination-deficient tumors by inducing synthetic lethality. Agents like olaparib, niraparib, and rucaparib have shown strong clinical benefits, especially in recurrent OC, and continue to be a major research focus [5]. In 2014, the Food and Drug Administration (FDA) approved PARPi, i.e., olaparib, followed by the approval of vascular endothelial growth factor inhibitor (VEGFi) bevacizumab in OC treatment. Moreover, in 2020, the FDA approved their combination in the treatment of OC patients with

BRCA mutations [8]. To date, the diagnosis of the disease mainly relies on laparoscopy, measurement of CA-125 concentration in serum, and diagnostic imaging [2,9,10]. Selected drugs approved by the FDA for the treatment of OC are presented in Table 1.

**Table 1:** Selected FDA-approved drugs in the treatment of ovarian cancer (OC). Formulated based on: [11,12]

| Generic drug | Drug brand | Type | Indications |
|---|---|---|---|
| Olaparib | Lynparza | Poly (ADP-ribose) polymerase inhibitor (PARPi) | First-line maintenance of homologous recombination deficiency (HRD) positive advanced ovarian cancer (OC); First line/maintenance of recurrent or germline BRCA-mutated advanced OC |
| Niraparib | Zejula | PARPi | First line maintenance of advanced OC; Maintenance of recurrent OC |
| Rucaparib | Rubraca | PARPi | Deleterious BRCA-mutated recurrent OC |
| Bevacizumab | Avastin, Vegzelma, Mvasi, Zirabev, Alymsys | Vascular endothelial growth factor inhibitor | Recurrent OC (platinum-resistant/sensitive); FIGO III or IV after initial surgical resection |
| Cisplatin | N/A | Alkylating agent | Metastatic OC |
| Cyclophosphamide | N/A | Alkylating agent | Ovary adenocarcinoma |
| Carboplatin | N/A | Alkylating agent | Advanced (previously untreated) or recurrent OC |
| Thiotepa | N/A | Alkylating agent | Ovary adenocarcinoma |
| Paclitaxel | N/A | Antimicrotubule drug | Previously untreated/treated OC |
| Liposomal doxorubicin | Doxil | Cytotoxic drug | After failure of platinum-based chemotherapy (progressed or recurred) |
| Gemcitabine | N/A | Antimetabolite drug | In combination with carboplatin advanced OC (relapsed ≥6 months after platinum-based therapy) |
| Mirvetuximab; soravtansine-gynx | Elahere | Antibody-drug conjugated folate-receptor alpha | Folate receptor-alpha positive OC |

Note: Abbreviation: BRCA: Breast Cancer Susceptibility Gene; N/A: Not Applicable.

## 2 Characteristics of Ovarian Cancer Microenvironment

Interactions within the tumor microenvironment (TME) are potential targets for the development of novel and effective therapies for OC patients [13–15]. Signals from the TME, such as cytokines, microRNAs (miRNAs), chemokines, influence host immune cells, leading to alterations in their immunophenotype and consequently modulating their functions. The ovarian TME represents a strong and dynamic immunosuppressive niche consists of cellular (i.e., regulatory T cells (Tregs), invariant natural killer T (iNKT) cells, natural killer (NK) cells, natural killer T (NKT) cells, dendritic cells (DCs), myeloid-derived suppressive cells

(MDSCs), endothelial cells, cancer-associated fibroblasts (CAFs), tumor-associated macrophages (TAMs), adipocytes), as well as non-cellular components (cytokines, extracellular matrix (ECM), blood and lymph vessels) [16–19]. The interplay between these components, acidity, hypoxia, and pathological angiogenesis, and danger-associated molecular patterns (DAMPs) released via tumor cells, leads to recruitment of immunosuppressive subpopulations of host immune cells. To initiate an antitumor response, NK cells, B cells, cluster of differentiation 3 ($CD3^+$), $CD4^+$, $CD8^+$ tumor-infiltrating T cells (TILs), DCs, and macrophages are attracted. Nevertheless, OC cells may evade elimination via immunoediting even in a proinflammatory ecosystem [20]. It is well known that immune checkpoints (ICPs), including programmed cell death pathway, exert inhibitory impact on effector immune cells such as T cells and NK cells [15,21–24]. The interactions between iNKT cells and the ovarian tumor within the TME appear particularly intriguing. This subpopulation has been shown to engage in crosstalk with both the TME and cancer cells simultaneously, acting through diverse molecular mechanisms [16].

A unique characteristic feature of the OC microenvironment is its distinctly immunosuppressive nature, which classifies it as a "cold" tumor. "Hot" and "cold" tumors differ in their immune activity. While "hot" tumors exhibit strong immune infiltration, efficient antigen presentation, and active cytotoxic T cell responses, "cold" tumors are defined by weak immune cell infiltration, poor antigen presentation, and a suppressive immune milieu. In the case of OC, this "cold" phenotype is reflected by low levels of $CD8^+$ and activated $CD4^+$ T cells that promote peritoneal dissemination, and an increased presence of Tregs. Researchers have further distinguished two immunologically "cold" patterns in OC: ovarian lesions with scarce but dysfunctional cell infiltration dominated by Tregs, and omental lesions infiltrated mainly by non-tumor-specific bystander immune cells. Notably, while "hot" tumors tend to respond favorably to chemotherapy, patients with "cold" ovarian tumors show improved outcomes when treated with a combination of chemotherapy and dendritic cell-based vaccines, which enhance the tumor's immunogenic potential [20,25]. What is more, in the ovarian TME, extensive hypoxia not only stimulates the secretion of pro-angiogenic factors but also promotes the accumulation of Tregs. These cells play multiple immunosuppressive roles, including limiting lymphocyte infiltration, impairing the maturation of antigen-presenting cells (APCs) necessary for T cell activation, and supporting the development of tumor-associated macrophages (TAMs) [20].

Thus, the objective of this study is to explore the therapeutic potential of iNKT cells in the treatment of ovarian cancer. Specifically, we aimed to assess how iNKT cell-based immunotherapies, including those utilizing chimeric antigen receptor (CAR) or T cell receptor (TCR) engineering, could overcome the immunosuppressive TME and enhance antitumor immune responses. By summarizing current knowledge, preclinical, and clinical findings, this work seeks to provide a comprehensive overview of the rationale and perspectives for developing iNKT cell-based strategies as a next-generation immunotherapy for OC.

## 3 Methodology

A comprehensive literature search was conducted to identify relevant studies on iNKT cells and their therapeutic potential in ovarian cancer. The search was performed in PubMed and Scopus, databases for articles published between 2000 and 2025, with a particular focus on studies from the last five years. The following keywords and their combinations were used: "ovarian cancer," "iNKT cells," "immunotherapy," "chimeric antigen receptor invariant natural killer T (CAR-iNKT)", "T cell receptor invariant natural killer T (TCR-iNKT)", and "tumor microenvironment."

The analysis included original experimental and clinical studies investigating iNKT cells or iNKT-based immunotherapies in cancer, as well as studies providing mechanistic or translational insights into the role of iNKT cells within the TME. Relevant review articles summarizing current knowledge and recent advances in

this field were also considered. We excluded publications not written in English, studies unrelated to cancer or lacking relevance to immunotherapy, and conference abstracts or papers without accessible full text.

## 4 Challenges in OC Treatment

In spite of increasing awareness of OC, including initiatives such as World Ovarian Cancer Day (observed on May 8) and implementation of novel drugs, the survival trends of OC patients have not altered significantly. This poor outcome results from an asymptomatic course of the disease, the lack of screening tools, as well as the absence of reliable markers to distinguish malignant from benign ovarian tumors [2,26]. It should be highlighted implementation of effective treatment for OC patients is challenging due to the high heterogeneity of the disease, including molecular, genetic, and immunological aspects [26]. Another problem in OC treatment is the relapsing nature of the disease [27]. Approximately 70% of patients with advanced stages of OC (III and IV FIGO) experience recurrence within 12–18 months after primary treatment, and the malignancy shows reduced sensitivity to platinum-based agents [28–32]. Therefore, there is an urgent need to develop new diagnostic strategies, identify biomarkers capable of distinguishing benign ovarian tumors from malignant ones, and finally, implement novel, targeted therapies that will be beneficial for OC patients regardless of the stage of the disease.

Implementation of ICPs inhibitors (ICIs) appears to be a breakthrough approach in OC treatment. Nonetheless, the response rate for programmed death receptor-1 (PD-1)/programmed death ligand-1 (PD-L1) inhibitors is low to moderate and totals 6%–15% [26]. ICIs restore T cell-mediated antitumor activity by blocking PD-1/PD-L1 signaling, but their efficacy as monotherapy is modest, prompting investigation into combination strategies [5]. Pembrolizumab, an anti-PD-1 monoclonal antibody (mAbs), blocks the PD-1/PD-L1 interaction, thereby sustaining T cell activity and promoting tumor cell apoptosis. Owing to its proven antitumor efficacy in cancers such as melanoma [33], lung [34], and renal carcinoma [35], it has been explored in OC. Early clinical trials with PD-1 inhibitors like nivolumab and pembrolizumab demonstrated modest response rates, with some complete responses observed in PD-L1 positive patients [5,36]. Notably, higher PD-L1 expression correlated with better outcomes. Nevertheless, the overall therapeutic benefit of pembrolizumab in OC remains limited. This reduced efficacy is attributed to the immunosuppressive TME, where dysfunctional immune cell infiltration impairs T cell activity. Moreover, PD-L1 expression alone does not reliably predict clinical response. Some PD-L1 positive patients fail to respond, while certain PD-L1 negative cases benefit, indicating that additional biomarkers and combination strategies are needed to enhance immunotherapy outcomes in OC [5].

Although OC cells have been shown to express other ICPs, such as cytotoxic T lymphocyte associated protein 4 (CTLA-4), and higher levels of tumor-infiltrating lymphocytes correlate with improved patient survival, clinical trials of ICPs in OC have so far produced limited efficacy. To date, no ICIs have been approved by the FDA for the treatment of OC [37]. Among the factors contributing to resistance to ICIs in OC are a network of miRNAs that have an impact on ICPs pathways, heterogeneity of the disease, low to moderate density of TILs, as well as cellular and non-cellular interactions in ovarian TME [22,38–41]. What is more, OC is marked by reduced tumor mutation burden and low microsatellite instability [42,43]. It is well known that immunotherapies based on ICIs are mostly beneficial in tumors that exhibit a high level of both these indicators [44]. Our previous work provides a detailed discussion of the mechanisms underlying resistance to immunotherapy in the treatment of ovarian cancer [45].

Another challenge in the implementation of ICIs in the treatment of OC is hyper- and pseudo-progression. Hyperprogression is a phenomenon characterized by accelerated tumor growth following immunotherapy and is considered a potential adverse effect of this treatment. It should be stressed that patients who experience hyperprogressive disease (HPD) show worse survival [46–49]. Boland et al. [50] in

their retrospective study showed that 51.6% ($n$ = 46) OC patients from the cohort ($n$ = 89) experienced HPD after ICIs treatment. Radiographic and clinical disease progression led to termination of treatment within ≤12 weeks [50]. Although several factors are considered potential risk factors for HPD, including age >65 years [51], female gender [15], multiple metastases sites [52,53], the current evidence remains inconclusive. To date, no reliable clinical predictors have been identified to predict HPD.

Another crucial phenomenon following ICIs treatment is pseudoprogression, which initially appears as a disease progression but, in contrast to HPD, it is followed by a subsequent positive objective response. Pseudoprogression results in the emergence of new lesions and an apparent increase in tumor burden via T cell recruitment to the tumor as a result of ICIs implementation. Thus, tumor size appears increased due to the presence of infiltrating T cells, rather than an actual increase in malignant cell burden [54]. Nevertheless, in case of pseudoprogression, the treatment should be continued [54]. The causes of pseudoprogression are still poorly understood. The phenomenon was described in several malignancies, including melanoma [55–57], non-small cell lung cancer (NSCLC), renal carcinoma [58], as well as OC [59,60]. Currently, pseudoprogression is identified retrospectively using imaging data and may lead to premature termination of effective therapy [61]. Thus, it is necessary to develop biomarkers capable of distinguishing HPD from pseudoprogression to guide clinical decisions regarding treatment continuation.

Given the complexity of OC treatment, largely due to its heterogeneous and immunosuppressive TME, iNKT cells offer a promising immunotherapeutic approach capable of overcoming these challenges through their unique ability to target tumors and modulate the immune landscape.

## 5 Characterization and Role of iNKT Cells

iNKT cells are effector cells and constitute approximately 0.01%–1% of T cells. They are considered a subpopulation of innate-like, non-conventional T cells that have common receptors with NK cells. The term iNKT cells is used because an invariant TCR is expressed by the majority of NKT cells [62,63]. The subset is marked by CD1d, a molecule related to major histocompatibility complex (MHC) class I, that presents antigens such as self-lipids, glycolipids, and microbial ones. Thus, iNKT cells and microorganisms are in persistent crosstalk. Moreover, iNKT cells may be activated via cytokines, i.e., IL-12, as well as through toll-like receptor (TLR), and they are predominant immune system cells that maintain homeostasis in mucosal tissues, e.g., intestine, lungs, and induce antimicrobial immune response [64–66]. iNKT cells take part in immune response targeted against infections by activating other immune system cells through interferon γ (IFN-γ), and through direct cytotoxicity [64–66]. iNKT cells play a dual and powerful role in antitumor immunity. Activated iNKT cells play a crucial role in antitumor immunity, as they are capable of eliminating cancer cells in a CD1d-dependent manner. They recognize glycolipid antigens presented by CD1d molecules on tumor cells, triggering direct cytotoxic responses. This ability represents a key functional feature of iNKT cells and holds significant potential for advancing cancer immunotherapy strategies [67,68]. They not only exert direct cytotoxic effects against tumor cells but also modulate the TME to enhance immune responses. Upon activation, iNKT cells release large amounts of Th1 and Th2 cytokines, which help regulate the activity of dendritic cells, NK cells, and cytotoxic T cells. They can deplete TAMs and MDSCs, thereby restoring immune surveillance and promoting effective infiltration and function of effector immune cells [69,70]. iNKT cells also promote the activation and maturation of DCs, enhancing their ability to present antigens and stimulate T cells. Through this interaction, iNKT cells bridge innate and adaptive immunity, ultimately strengthening tumor-specific T cell responses [71].

This population secretes cytokines, i.e., granulocyte-macrophage colony-stimulating factor (GM-CSF), tumor necrosis factor α (TNF-α), IFN-γ, macrophage inflammatory proteins 1α (MIP-1α) and 1β (MIP-1β), regulated on activation normal T cell expressed and secreted (RANTES),

eotaxin, transforming growth factor β (TGF-β), and interleukins (ILs): 2, 4, 5, 6, 10, 13, 17, and 21. Moreover, they express surface markers such as NK1.1, CD44, and CD69 [63,72], and they can eliminate recognized cells via Fas ligand (FasL), granzyme B, and perforin [63]. Through these combined mechanisms, iNKT cells strengthen both innate and adaptive antitumor immunity, making them an important target in the development of novel immunotherapeutic strategies.

Taking into account that iNKT cells coexpress TCR as well as NK receptors and cytokine receptors, their mode of action is far more rapid in response to cytokines or TCR signals via releasing cytokines and enhancing the adaptive immune response enhancement. iNKT cells are localized in peripheral tissues, where they often reside long term and contribute to diverse immune functions. Depending on the tissue environment and the immunological context, they can play either beneficial or detrimental roles, i.e., from maintaining tissue homeostasis to responding to infections and participating in tumor surveillance [64,73,74].

## 6 Role of iNKT in OC and Other Malignancies

iNKT cells hold significant potential in OC therapy due to their unique immunological properties. Unlike conventional T cells, iNKT cells can rapidly produce large amounts of both pro-inflammatory and regulatory cytokines upon activation, enabling them to influence various components of the TME. This capacity for immune modulation makes them especially valuable in OC, where the TME is often highly immunosuppressive [75,76]. iNKT cells are rapidly activated at the onset of immune responses through signals delivered by CD1d-restricted semi-invariant TCRs as well as cytokine receptors, including IL-12R and IL-18R. Once activated, iNKT cells perform both indirect and direct effector functions. Indirectly, they regulate and enhance the activity of other immune cells, while directly, they can exert cytotoxic effects on target cells. Following activation, iNKT cells promptly release large amounts of cytokines such as IFN-γ and IL-4, which drive the activation and maturation of antigen presenting cells (APCs), NK cells, and cytotoxic T cells. In addition to these, iNKT cells secrete a diverse set of other cytokines and mediators, including IL-2, IL-5, IL-6, IL-10, IL-13, IL-17, and IL-22, as well as chemokines like chemokine (C-C motif) ligand 3 (CCL3), CCL4, and CCL5, which further shape and amplify immune responses. Through CD40-CD40L interactions, iNKT cells also promote the activation and functional maturation of APCs. In terms of direct cytotoxicity, activated iNKT cells utilize granzyme and perforin release to induce apoptosis in target cells. Moreover, they express Fas ligand and TNF-related apoptosis-inducing ligand (TRAIL), which can initiate cell death via the death receptor pathway. Collectively, these mechanisms allow iNKT cells to serve as potent modulators of both innate and adaptive immunity [77].

The activation of iNKT cells results in a rapid and robust production of IFNγ, which plays a predominant role in the activation of NK cells. Then, NK cells are capable of targeting and eliminating tumor cells lacking MHC class I molecules. Following this early innate response, a cascade of adaptive immune activation occurs, including the expansion of antigen-specific $CD8^+$ and $CD4^+$ T cells that mediate cytotoxic responses against MHC-positive tumor cells and contribute to the formation of long-lasting anti-tumor immune memory. This cell population is also recruited via chemotactic signals derived from TME and infiltrates primary and metastatic sites in various types of solid tumors, where it promotes the maturation of DCs, further enhancing both innate and adaptive immunity [78–80].

iNKT cells reprogram immunosuppressive myeloid populations within the TME through CD1d-dependent recognition of lipid antigens. Regarding MDSCs, iNKT cells drive their differentiation into functional DCs, leading to activation of $CD8^+$ and $CD4^+$ T cells as well as NK cytotoxicity. They also reduce the number and suppressive activity of $CD1d^+$ MDSCs, promoting their maturation into antigen-presenting cells. In the case of TAMs, iNKT cells selectively eliminate pro-tumoral M2-like macrophages while supporting the survival of anti-tumoral M1-like macrophages. This occurs through coordinated CD1d, CD40,

and Fas signaling, i.e., CD40L-CD40 interaction protects M1 cells from Fas-mediated apoptosis, whereas M2 cells remain sensitive to Fas-induced death. Moreover, iNKT cells induce IL-12 production by DCs via CD1d- and CD40-dependent interactions, reinforcing a Th1-type immune response with enhanced IFN-γ secretion and cytotoxic T cell activation. Overall, iNKT cells remodel the TME from an immunosuppressive to a proinflammatory, tumoricidal state [69,78,81]. The models of iNKT cell activation and their remodeling of the TME are presented in Fig. 2.

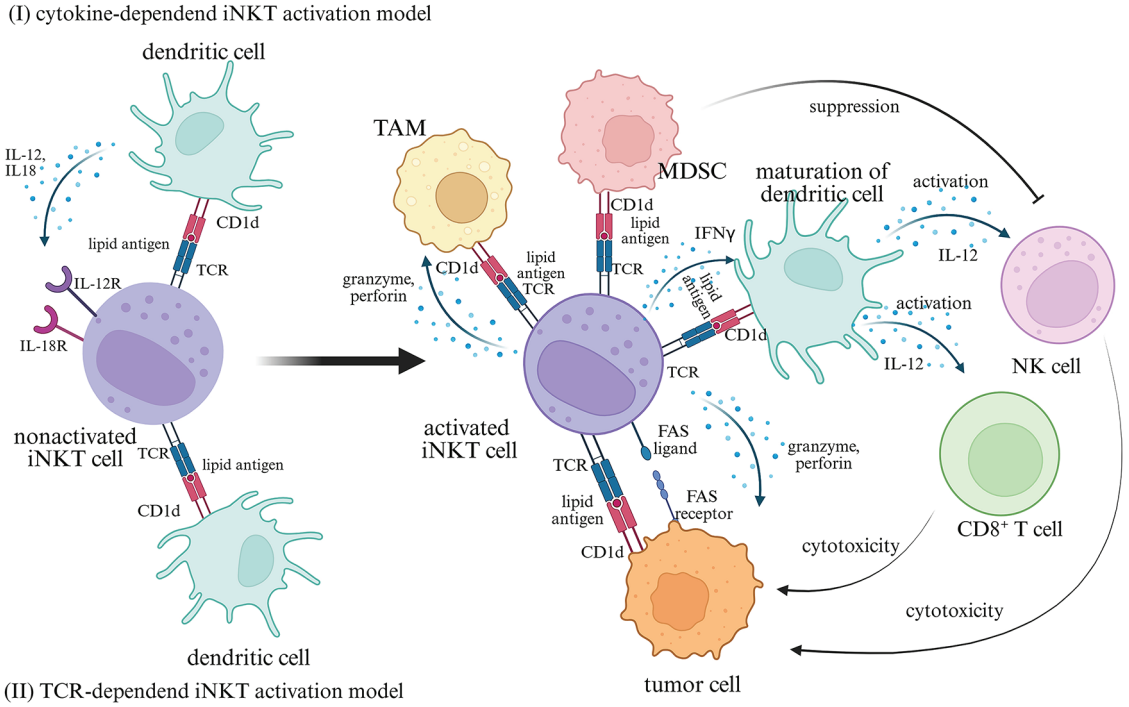


**Figure 2:** The models of iNKT cell activation and their remodeling of the TME. IL: interleukin; CD: cluster of differentiation; TCR: T cell receptor; iNKT: invariant natural killer T cell; TAM: tumor-associated macrophage; MDSC: myeloid-derived suppressor cell; IFN: interferon; NK: natural killer cell. Created in https://BioRender.com

Although iNKT cells have traditionally been considered to mediate anti-tumor responses through perforin-dependent cytotoxicity, this mechanism has been challenged. Inhibition of perforin activity using concanamycin A did not abolish the cytotoxic effect of iNKT cells, suggesting alternative pathways are involved. Moreover, iNKT cell-mediated killing does not require CD1d expression on tumor cells, indicating that their invariant TCR is not essential for target cell recognition and lysis [62].

Moreover, iNKT cells are not restricted by classical human leukocyte antigen (HLA) presentation, allowing broader applicability across patients without requiring HLA matching. They can also be engineered with CARs or TCRs to target tumor-associated antigens (TAAs), combining innate-like rapid responses with tumor specificity. Importantly, their ability to activate other immune cells, such as DCs and NK cells, may help overcome immune evasion mechanisms commonly observed in ovarian tumors [82].

Data available in the literature underscore the therapeutic promise of iNKT cell-based therapy, which has demonstrated the ability to target both OC cells and the TME via diverse molecular pathways [16]. Winkler et al. [63] found in their study a negative correlation between the concentration of CA125 in the serum of OC patients and a decreased number of NKT cells within tumor tissue. This phenomenon may be

related to the inflammatory process. The ovarian patients' recurrence-free survival and overall survival (OS) may be related to iNKT cells frequency and their location [83].

Li et al. [16] found that CD1d is a potential biomarker and therapeutic target within ovarian TME. CD1d molecule is mainly expressed on immunosuppressive cells, i.e., MDSCs and TAMs, regardless of platinum sensitivity and advancement of the disease. Interestingly, iNKT cells may target TAMs and MDSCs with CD1d expression without influencing effector cells such as NK cells, T cells, and B cells. The cytotoxicity of iNKT cells against healthy monocytes is reduced because CD1d expression on these monocytes is significantly lower compared to that on cells within ovarian TME [16]. Importantly, the presence of CAR target antigens on recurrent OC cells may enhance iNKT cell function through CAR engineering. Overall, these findings support the potential of iNKT cells as innovative carriers in next-generation cell-based immunotherapies, offering a promising approach for treating recurrent OC [16,84,85].

Clinical trials have shown decreased frequencies of iNKT cells and their defective functions in various types of malignancies, which are associated with unfavorable OS in both hematological malignancies and solid tumors such as head and neck cancer (HNC), neuroblastoma, and prostate cancer [78,86–88]. Per contra, a high frequency of circulating or intratumoral iNKT is related to beneficial OS in hematological malignancies, neuroblastoma, and colorectal cancer [78,89–92].

These findings show that iNKT cells may utilize distinct mechanisms, different from those of conventional T cells or NK cells, to exert their anti-tumor effects [62,93] and are involved in the elimination of cancer cells by αGalCer-pulsed DCs (αGalCer/DCs), resulting in activation of host immune system cells [62,94]. Altogether, iNKT cell activation triggers a multifaceted anti-tumor response involving several immune cell types working in concert to eliminate malignant cells [62,94].

## 7 Immunotherapies Based on iNKT Cells

Various studies have confirmed that iNKT cells have a therapeutic potential in the treatment of solid cancers, including melanoma, HNC, and lung cancer [82,93,95–97]. iNKT cells represent a promising platform for the development of "off-the-shelf" effector cell products for adoptive cancer immunotherapy. However, to prevent immune-mediated rejection by the allogeneic host, it is necessary to modulate MHC expression on these cells. One strategy involves differentiating iNKT cells from human hematopoietic stem cells (HSCs) *in vitro*, which naturally express minimal levels of HLA class I and negligible HLA class II molecules. These stem cell-derived iNKT cells can then be further modified, for instance by introducing CARs, to create immune-evasive anti-tumor effectors. A challenge in using iNKT cells for adoptive therapy is their low abundance (around 0.01%) in peripheral blood (PB), and their frequency is even lower in patients with advanced malignancies. Nonetheless, iNKT cells are amenable to robust *ex vivo* expansion, and established protocols allow for their efficient activation and proliferation.

Chimeric antigen receptor invariant natural killer T (CAR-iNKT) cell therapy represents a promising advancement in cancer immunotherapy. iNKT cells are particularly well-suited for allogeneic "off-the-shelf" therapies due to their strong anti-tumor activity and minimal risk of triggering graft-vs.-host disease (GvHD). Since natural iNKT cells are scarce in PB, researchers have developed a method to generate these cells in large quantities by combining gene editing of HSCs with *in vitro* differentiation. The resulting allogeneic HSC-derived iNKT (AlloHSC-iNKT) cells closely mimic natural iNKT cells and attack tumors through multiple immune mechanisms while maintaining a favorable safety profile and low immunogenicity. Importantly, due to their limited dependence on HLA compatibility, iNKT cells from allogeneic donors may serve as a viable and scalable source for therapeutic use [78]. A schematic overview of CAR-iNKT cell therapy is shown in Fig. 3.

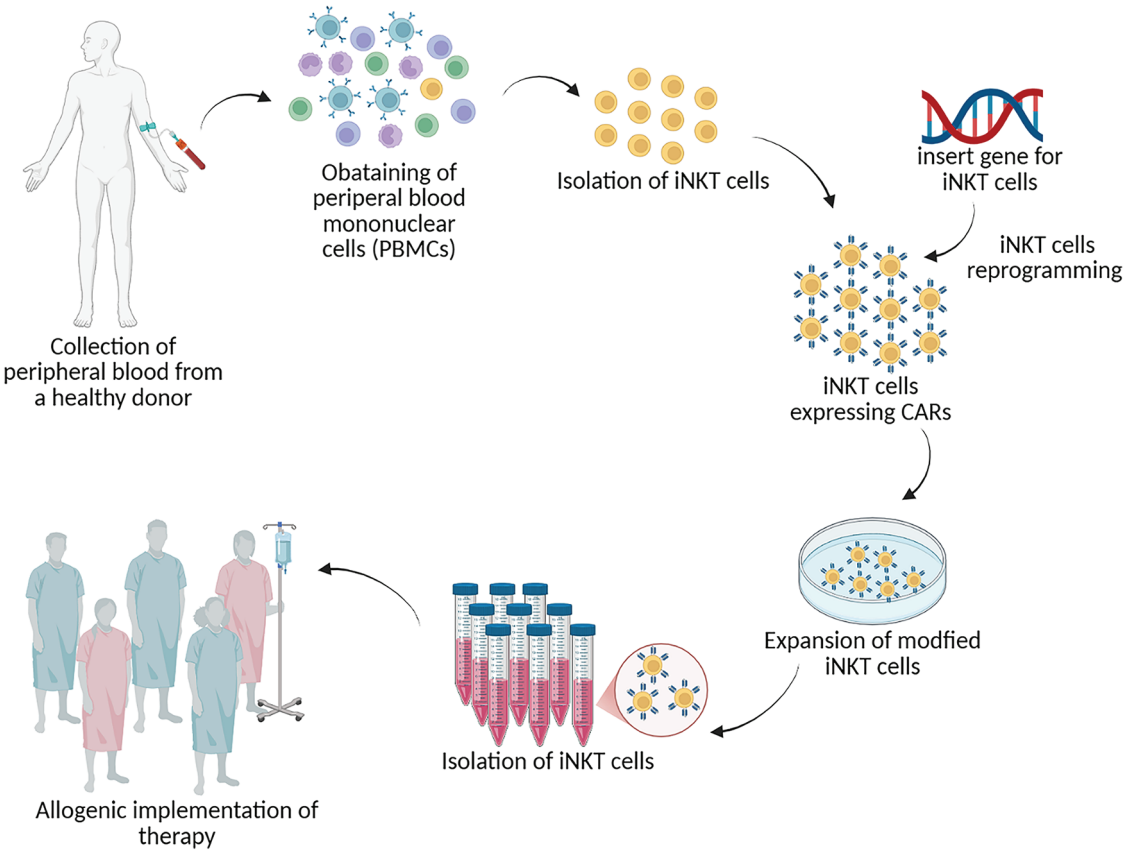


**Figure 3:** Schematic overview of the generation and implementation of CAR-iNKT cell therapy. PBMCs: peripheral blood mononuclear cells; CARs: chimeric antigen receptors. Created in https://BioRender.com

Preclinical findings support the therapeutic potential of CAR-iNKT cell products and provide a strong basis for their clinical translation [98]. Initially, therapies based on CAR primarily utilized conventional T cells (CAR-T cells therapy), which received FDA approval for the treatment of advanced lymphoma and acute lymphoblastic leukemia. CAR-T therapies have revolutionized blood cancer treatment but face challenges such as toxicity, high cost, and limited success in solid tumors. In contrast, iNKT cell therapies offer an "off-the-shelf", safer, and more scalable alternative, with natural resistance to graft-vs.-host disease and strong tumor-modulating abilities. Early studies show promise in both solid and hematologic cancers, suggesting iNKT cells could complement or enhance CAR-T therapies through immune modulation and combination strategies. CAR-T therapy's major strength, its powerful immune activation, is also its main limitation, often leading to severe toxicities such as cytokine release syndrome (CRS) and immune effector cell associated neurotoxicity syndrome (ICANS). These side effects can occur in a substantial proportion of patients, highlighting safety concerns with current CAR-T products. In contrast, iNKT cell therapies, whether engineered or natural, demonstrate a much safer toxicity profile. Their balanced cytokine secretion, limited *in vivo* expansion, and MHC-independent activity result in minimal CRS, negligible neurotoxicity, and no risk of graft-vs.-host disease, allowing for allogeneic use. While more clinical data are needed, iNKT therapies hold promise as safer, more accessible alternatives, particularly for patients who may not tolerate CAR-T therapy [99].

However, iNKT cells can also be used in this kind of treatment in both solid and hematological malignancies [100]. In spite of the very modest frequency of iNKT cells *in vivo*, this immune cell subset

eliminates tumor cells more efficiently than CAR-T cells. For instance, in murine model of lymphoma, the survival rate after implementing CAR-T cells was only 60%, whereas in the group treated with CAR19-iNKT cells it reached 90%. Interestingly, targeting both CD1d and CD19 simultaneously induces even deeper anticancer immune response and may reduce recurrence of the disease. This property is especially significant for ovarian cancer treatment. Moreover, CAR19-iNKT cells demonstrated the ability to penetrate the brain and effectively manage sizable tumor burdens and even in the presence of large tumors. These findings offer promising prospects for the development of novel therapeutic strategies for OC patients [101].

OC expresses multiple potential CAR targets, among which mesothelin, folate receptor alpha (FRα), mucin 16, and human epidermal growth factor receptor 2 (HER2) are the most extensively studied. These antigens show high expression on tumor cells with limited distribution in normal tissues, making them promising candidates for CAR therapies. Other emerging targets, such as claudin-6, protein tyrosine kinase 7 (PTK7), epithelial cell adhesion molecule (EPCAM), and Annexin A2 (ANXA2), are under preclinical evaluation and may broaden therapeutic options in the future [102]. Taking into account high heterogeneity of OC and its various subtypes, identifying target antigens for CAR therapies is challenging. Potential targets should be highly expressed on cancer cells and having none or marginal expression on host cells. All the described features meet the criteria of TAAs. A variety of notable TAAs have been studied in OC, each posing specific considerations and obstacles [103]. Dual-target CARs may offer a better strategy to reduce on-target off-tumor cytotoxicity. Interestingly, targeting FRα or mesothelin alone achieves approximately 48%–76% tumor elimination, while dual targeting increases tumor cell elimination to about 88% [104–106]. Early results indicate that these approaches are generally safe and technically feasible, though their clinical effectiveness remains limited. Further progress in CAR-T therapy for OC is hindered by factors such as tumor heterogeneity and inconsistent antigen expression, the presence of resistant and immune-evasive cancer stem cells, and the highly immunosuppressive peritoneal TME rich in TAMs and MDSCs [102]. Li et al. highlighted that a key advantage of CAR-NKT cells lies in their ability to reduce CRS by modulating macrophage activity, offering a safer alternative to conventional CAR-T cells, particularly in ovarian cancer where macrophage-driven CRS toxicity is pronounced [102]. Comparison of key features between CAR-T and CAR-iNKT cell therapies is presented in Table 2.

**Table 2:** Comparison of key features between chimeric antigen receptor-invariant natural killer T (CAR-iNKT) and CAR-T cell therapies

| Features | CAR-iNKT cells | CAR-T cells | Ref. |
|---|---|---|---|
| Mode of action | Tumor antigens are recognized via CAR and through preseting by cluster of differentiation 1d (CD1d) | Tumor antigens are recognized via CAR | [71,107] |
| Source | Allogenic (“off-the-shelf” therapy is possible) | Autologous | [71,108] |
| Autoimmunological reactivity | Lack of autoimmunology reactivity; there is a possibility of allogenic implementation | High reactivity, may lead to GvHD | [109–111] |
| Impact on TME | CAR-iNKT cells can actively modulate TME | Limited | [112,113] |
| Tumor infiltration | Tumors can be infiltrated by these cells | The infiltration of solid tumors is limited | [113,114] |

(Continued)

**Table 2 (continued)**

| Features | CAR-iNKT cells | CAR-T cells | Ref. |
|---|---|---|---|
| Clinical application | Solid tumor and hematological malignancies | Hematological malignancies mostly | [69,83,89,90] |
| AEs | Preliminary studies claimed that CAR-iNKT cells exhibit more mild AEs in comparison to CAR-T cells therapy | CAR-T CRES, CRS, ICANS | [115–117] |

Note: Abbreviation: TME: tumor microenvironment; CD: cluster of differentiation; AE: adverse event; CAR-iNKT: chimeric antigen receptor invariant natural killer T cell; CAR-T: chimeric antigen receptor T cell ; GvHD: graft-vs.-host disease; CRES: CAR-T cell-related encephalopathy syndrome; CRS: cytokine release syndrome; ICANS: immune effector cell–associated neurotoxicity syndrome.

iNKT cells act as natural adjuvants by releasing large amounts of IFN-γ, which enhances both innate and adaptive immune responses. Patients with higher levels of IFN-γ producing cells show better clinical outcomes, including significantly longer survival and reduced tumor progression. Beyond their therapeutic use as cells, iNKT cells can also serve as molecular adjuvants in cancer vaccines. When activated by lipid antigens such as αGalCer or its analogs, iNKT cells stimulate dendritic cells and boost $CD8^+$ T cells response, leading to stronger and more durable antitumor immunity. These CD1d-restricted vaccines have shown potential in enhancing immune protection not only against tumors but also against bacterial, viral, and parasitic infections [118,119].

A novel, scalable method for generating iNKT cells from cord blood offers a promising background for developing 'off-the-shelf' cancer immunotherapies. CAR-engineered iNKT cells have demonstrated potent anti-tumor activity against both hematologic malignancies and solid tumors, including OC. Their unique ability to function without inducing GvHD and to modulate the immunosuppressive TME makes them especially suitable for broader clinical application. This platform not only bypasses the need for patient-specific therapies but could significantly reduce production costs and improve treatment accessibility [120,121]. A detailed analysis of ovarian tumor cells and TME in samples from primary and recurrent OC revealed that hematopoietic stem cells-derived iNKT (HSC-iNKT) cells exert strong antitumor activity across diverse patient-derived tumor cells, regardless of tumor antigen or cancer stem cell marker expression. These findings highlight the therapeutic potential of HSC-iNKT cell therapy for OC, particularly in recurrent cases, by simultaneously eliminating tumor cells and reshaping the immunosuppressive microenvironment [85]. Currently, work is underway to implement CAR-iNKT cell therapy for ovarian cancer [122]. CAR-engineered iNKT cells have demonstrated the ability to selectively eliminate antigen-expressing tumor cell lines and patient-derived plasma cells *in vitro*, as well as suppress tumor growth in xenograft models *in vivo*, while preserving their native CD1d-restricted functionality [123–125].

Moreover, iNKT cells can be genetically modified to acquire an additional antigen specificities by introducing recombinant TCRs that target disease-relevant, particularly tumor-associated, antigens. For example, engineering iNKT cells with a human MHC class I-restricted TCR specific for a peptide from the 38-kDa protein of *Mycobacterium tuberculosis* enabled them to selectively eliminate monocyte-derived dendritic cells (Mo-DCs) loaded with the corresponding antigen [126]. Applying these strategies to TCR-engineered iNKT cells could enable the projecting of tumor-redirected effectors that not only target specific antigens but also retain the unique ability of iNKT cells to modulate the TME, thereby enhancing their overall anti-tumor efficacy [78]. Furthermore, PRAME-specific TCR-engineered iNKT cell therapy has

demonstrated promise in overcoming the challenges faced by conventional T cell approaches in treating solid tumors, including OC. Preclinical data suggest that these modified iNKT cells can selectively recognize and eliminate cancer cells [127–129]. Early clinical studies have demonstrated promising safety profiles and therapeutic benefits in both hematological malignancies and solid tumors, including OC [109].

The safety and toxicity profile of CAR-iNKT cell therapy remains one of its most critical limitations. Although iNKT cells naturally exhibit immunoregulatory and anti-inflammatory properties that may lower toxicity compared to conventional CAR-T cells, genetic modification and artificial activation can still provoke CRS or ICANS. These events arise from excessive cytokine secretion, such as IL-6, IFN-γ, and TNF-α, leading to systemic inflammation and potentially life-threatening complications. Another safety concern is the risk of off-target effects, as CAR-iNKT cells may recognize antigens expressed on normal tissues, resulting in unintended tissue damage. Moreover, the long term persistence and proliferation of engineered iNKT cells *in vivo* are not yet fully understood, raising questions about possible chronic immune activation or autoimmunity. To mitigate these risks, recent strategies include incorporating safety switch mechanisms (e.g., inducible caspase-9 systems) to allow rapid termination of CAR-iNKT activity in case of severe toxicity, as well as optimizing CAR design to balance efficacy and immune activation. Rigorous clinical monitoring protocols and early intervention strategies for CRS are also essential to ensure patient safety. While preclinical data suggest a more favorable toxicity profile compared to CAR-T cells, comprehensive clinical validation is still required to confirm the safety and tolerability of CAR-iNKT cell therapies in humans [71].

Several early-phase clinical trials have evaluated iNKT cell-based therapies in cancer patients using different approaches, including α-GalCer administration, α-GalCer-pulsed $CD1d^+$ APCs, and adoptive transfer of *ex vivo* expanded iNKT cells [130–132]. These studies consistently demonstrated a favorable safety profile, with no dose-limiting toxicities reported even at high cell doses (up to $1 \times 10^{10}/m^2$). While increases in iNKT cell counts and IFN-γ production were observed in many patients, clinical efficacy was generally limited, ranging from stable disease to occasional partial responses. Notably, a phase II trial in hepatocellular carcinoma showed that combining *ex vivo* expanded iNKT cells with transarterial embolization (TAE) improved both progression-free survival (PFS) and OS compared to TAE alone, with several complete responses observed. Ongoing trials are currently exploring autologous and allogeneic "off-the-shelf" iNKT cell therapies and nanoparticle-based iNKT activators (IMM60) in combination with other immunotherapeutic agents to enhance antitumor efficacy while maintaining safety [133–135].

iNKT cell therapies represent a groundbreaking advancement in cellular immunotherapy. This approach combines the targeted precision of CARs with the distinctive immunoregulatory abilities of iNKT cells. Their strong capacity to proliferate without exhaustion, lack of alloreactivity, enhanced ability to infiltrate tumors, potent cytotoxic effects, and their capability to remodel the TME are highly desirable in development of targeted therapies. Such features help overcome many of the limitations associated with traditional CAR-T and other CAR-engineered cell therapies [109]. Altogether, iNKT cells represent a promising platform for developing innovative immunotherapies aimed at reshaping the immune landscape in solid tumors, including OC, and improving clinical outcomes.

## 8 Future Perspectives of OC Treatment

Ovarian TME plays an important role in tumor initiation, progression, and metastasis. It is well known that the formation of peritoneal metastases contributes to high mortality and recurrence risk. Thus TME offers a wealth of therapeutic targets [136]. Moreover, it also contributes to the suppression of antitumor immune response inducted via ICPs [17,137]. Immune checkpoints represent promising therapeutic targets in emerging OC treatment. The response of OC patients to immunotherapy based on programmed cell death pathway (PD-1/PD-L1) inhibitors is modest. Alas, the most of clinical trials have been conducted

in patients with recurrent disease who have already received multiple lines of therapy. Therefore, to overcome the immunosuppressive ovarian TME, it is necessary to investigate their efficiency in first-line treatment [138,139]. The investigation of cellular crosstalk within the OC microenvironment using artificial TME models appears particularly valuable, given its dynamic nature, complexity, and differences observed between primary and recurrent tumors [25,140,141].

Following the discussion on iNKT cell based therapies, it is also important to consider NK cell based approaches, which share certain similarities but differ in key mechanisms and clinical applications. NK cells exert antitumor activity in OC through antibody-dependent cellular cytotoxicity, perforin and granzyme release, and cytokine secretion (IFN-γ, TNF-α). They can also induce apoptosis via Fas/FasL and TRAIL/TRAILR pathways. Clinical trials show that NK cell based immunotherapy is generally safe and may stabilize disease, though its overall efficacy remains limited. The TME and prior treatments can impair NK cell function, highlighting the need for combination or modulatory strategies. CAR-NK cell therapy, an emerging approach, enhances NK cell specificity through genetic modification with CARs targeting antigens such as HER2, epidermal growth factor receptor (EGFR), or mesothelin. Compared with CAR-T cells, CAR-NKs offer lower toxicity, reduced GvHD risk, and "off-the-shelf" potential. Preclinical data indicate promising antitumor effects, but further optimization, including overcoming TME suppression and expanding clinical validation, is essential for broader application in OC [142–144]. In contrast, iNKT cell therapy bridges innate and adaptive immunity. iNKT cells recognize glycolipid antigens presented by CD1d, enabling both direct cytotoxic effects and potent activation of NK, dendritic, and $CD8^+$ T cells. They remodel the TME by reducing immunosuppressive cells (MDSCs, TAMs) and enhancing antigen presentation. However, their scarcity and functional exhaustion in the OC microenvironment limit clinical application.

In summary, NK therapies offer strong, immediate cytotoxicity and safety advantages, while iNKT therapies provide broader immunomodulation and TME reprogramming. Combining or sequentially using both approaches could enhance overall therapeutic efficacy in OC. The promising approach in OC treatment is a combined therapy targeting other ICPs such as the T cell immunoglobulin and ITIM domain/CD155/DNAX accessory molecule-1 (TIGIT/CD155/DNAM-1) axis. Preclinical studies in a murine model of colorectal cancer showed that the dual blockade of PD-1/PD-L1 and TIGIT led to remission in the entire studied group vs. only partial tumor regression observed with the blockade of a single pathway. The approach stimulates the effector activity of T cells and NK cells, and redirects the immune system activity against the tumor. The understanding of the synergistic action of the TIGIT and PD-1/PD-L1 blockade is, however, still limited. Considering the positive impact of this combined therapy in malignancies, including lung and colorectal cancer, it appears to be a promising approach in OC treatment [145–149]. The rationale and clinical implementation of dual blockade in the treatment of patients with ovarian cancer were thoroughly outlined in our previous work [26]. Moreover, another dual immune checkpoint blockade targeting PD-1 and CTLA-4 has shown promising efficacy in advanced and OC, outperforming single agent immunotherapy. It improves response durability and surgical outcomes but exhibits subtype-dependent variability. Despite safety concerns and limited first-line data, ongoing trials aim to optimize combinations and identify predictive biomarkers, positioning dual blockade as a key emerging strategy in OC immunotherapy [37,150].

Bispecific antibodies (BsAbs) represent an emerging immunotherapeutic strategy in OC. By simultaneously binding two distinct targets, BsAbs enhance immune cell activation and tumor recognition. Preclinical studies have shown that BsAbs targeting TIGIT/PD-L1 improve antitumor efficacy compared with conventional anti-PD-L1mAbs. Another promising class, T cell redirecting bispecifics (TCBs), engage both T cells and TAAs such as mucin-16, mucin-1, AXL, and LYPD1, promoting T cell mediated cytotoxicity. Overall, BsAbs show significant potential to enhance immune responses in OC, though further studies are required to

optimize safety and clinical efficacy [151–154]. BsAbs offer precise, engineered immune redirection, whereas iNKT therapies provide broader immune modulation and TME remodeling.

To the date, few biological drugs targeted TME have been approved by the FDA, including VEGFi and PARPi [17]. The combination this kind of treatment with ICIs is also beneficial because it leads to sensitization of tumor to ICIs [138,139]. It should be highlighted that a major impediment in implement effective therapeutic approach for OC patients is absence of a congeneric target signature for OC. That immuneprofile may offer insights into potential biomarkers to monitor the disease progression in real time [16]. Selected interventional clinical trials investigating immunotherapy in ovarian cancer are summarized in Table 3.

**Table 3:** Selected interventional clinical trials investigating immunotherapy in OC

| Study Title | NCT Number | Acronym | Phase | Interventions | Sponsor |
|---|---|---|---|---|---|
| Chemotherapy Combined With Propranolol Hydrochloride as Neoadjuvant Therapy for Advanced High-grade Serous OC | NCT07125391 | N/A | 2 | Drug: Cohort A | Bai-Rong Xia |
| Low-Dose Radiation-Stereotactic body radiotherapy-Cadonilimab for Advanced Gastric, Colorectal and OC With Peritoneal Metastases | NCT06940921 | N/A | 1/2 | Radiation: Low-Dose Radiation + Stereotactic body radiotherapy<br>Drug: Cadonilimab | Zhang Tao |
| A Clinical Study on Fasudil Hydrochloride for Treatment of Gene-Specific OC | NCT06890858 | N/A | 2 | Drug: Fasudil Hydrochloride | Zhejiang Provincial People's Hospital |
| Vaccine Therapy Plus Pembrolizumab in Treating Advanced Ovarian, Fallopian Tube, or Primary Peritoneal Cavity Cancer | NCT05920798 | FRAPPE | 1/2 | Procedure: Biopsy<br>Procedure: Biospecimen Collection<br>Procedure: Computed Tomography | Mayo Clinic |
| Abemaciclib and Letrozole in Patients With Estrogen Receptor-positive Rare OC | NCT05872204 | ALEPRO | 2 | Drug: Abemaciclib<br>Drug: Letrozole | Universitaire Ziekenhuizen KU Leuven |
| A Study to Evaluate the Safety and Therapeutic Activity of GI-102 As a Single Agent and in Combination with Conventional Anti-cancer Drugs, Pembrolizumab or Trastuzumab Deruxtecan in Patients with Advanced Solid Tumors (KEYNOTE-G08) | NCT05824975 | N/A | 1/2 | Drug: GI-102 subcutaneous<br>Drug: GI-102<br>Drug: doxorubicin | GI Innovation, Inc. |
| NEOadjuvant Dendritic Cell Vaccination for OC | NCT05773859 | NEODOC | 1/2 | Biological: XP-DC vaccinations | Radboud University Medical Center |
| A Safety, Tolerability and Efficacy Study of NC410 Plus Pembrolizumab in Participants with Advanced Unresectable or Metastatic Solid Tumors | NCT05572684 | N/A | 1/2 | Drug: NC410<br>Drug: pembrolizumab | NextCure, Inc. |
| Safety and Efficacy of Anti-CD47, ALX148 in Combination with Liposomal Doxorubicin and Pembrolizumab in Recurrent Platinum-resistant OC | NCT05467670 | N/A | 2 | Drug: Pembrolizumab<br>Drug: ALX148<br>Drug: Doxorubicin | Alexander B Olawaiye, MD |

(Continued)

**Table 3 (continued)**

| Study Title | NCT Number | Acronym | Phase | Interventions | Sponsor |
|---|---|---|---|---|---|
| AK104 Combined With Chemotherapy as Neoadjuvant Treatment for Advanced OC | NCT05430906 | N/A | 2 | Drug: AK104—Chemotherapy | Hunan Cancer Hospital |
| Efficacy & Safety of Olvi-Vec and Platinum-doublet + Bevacizumab Compared to Physician's Choice of Chemotherapy and Bevacizumab in Platinum-Resistant/Refractory OC (OnPrime, GOG-3076) | NCT05281471 | N/A | 3 | Biological: olvimulogene nanivacirepvec<br>Drug: Platinum chemotherapy: carboplatin (preferred) or cisplatin<br>Drug: Non-platinum chemotherapy: Physician's Choice of gemcitabine, taxane (paclitaxel, docetaxel or nab-paclitaxel) or pegylated liposomal doxorubicin | Genelux Corporation |
| Pembrolizumab Combined With Bevacizumab With or Without Agonist Anti-cluster of differentiation (CD) 40 CDX-1140 for the Treatment of Patients With Recurrent OC | NCT05231122 | N/A | 2 | Biological: Anti-cluster of CD40 Agonist Monoclonal Antibody CDX-1140<br>Biological: Bevacizumab<br>Biological: Pembrolizumab | Roswell Park Cancer Institute |
| T-regulatory Cell Depletion with E7777 Combined with Pembrolizumab in Recurrent or Metastatic Solid Tumors | NCT05200559 | N/A | 1/2 | Drug: Pembrolizumab<br>Drug: E7777 | Alexander B Olawaiye, MD |
| Phase I/II Study of Autologous T Cells to Express T-Cell Receptors in Subjects With Solid Tumors | NCT05194735 | N/A | 1/2 | Biological: Neoantigen specific T cell expressing engineered T cell receptor (TCR-T cell) drug product<br>Biological: Aldesleukin (interleukine 2) | Alaunos Therapeutics |
| Pembrolizumab and Lenvatinib for the Treatment of Serous Ovarian Cancer Patients | NCT05114421 | N/A | 2 | Drug: Lenvatinib<br>Biological: Pembrolizumab | M.D. Anderson Cancer Center |
| Testing Nivolumab With or Without Ipilimumab in Deficient Mismatch Repair System (dMMR) Recurrent Endometrial Carcinoma | NCT05112601 | N/A | 2 | Procedure: Biospecimen Collection<br>Procedure: Computed Tomography<br>Biological: Ipilimumab | National Cancer Institute |
| A Beta-only IL-2 ImmunoTherapY Study | NCT05086692 | ABILITY-1 | 1/2 | Drug: MDNA11<br>Drug: Pembrolizumab (KEYTRUDA®) | Medicenna Therapeutics, Inc. |

(Continued)

**Table 3 (continued)**

| Study Title | NCT Number | Acronym | Phase | Interventions | Sponsor |
|---|---|---|---|---|---|
| Oregovomab in Combination With Bevacizumab Plus Chemo in breast cancer susceptibility gene (BRCA) Wild Type Platinum Sensitive Recurrent OC | NCT04938583 | N/A | 1/2 | Biological: Oregovomab<br>Drug: Bevacizumab<br>Drug: Paclitaxel<br>1 more | CanariaBio Inc. |
| APL-2 and Pembrolizumab vs. APL-2, Pembrolizumab, and Bevacizumab vs. Bevacizumab Alone for the Treatment of Recurrent Ovarian, Fallopian Tube, or Primary Peritoneal Cancer and Malignant Effusion | NCT04919629 | N/A | 2 | Biological: Bevacizumab<br>Procedure: Biopsy<br>Procedure: Biospecimen Collection | Roswell Park Cancer Institute |
| Immunotherapy Platform Study in Platinum Resistant High Grade Serous OC | NCT04918186 | IPROC | 2 | Drug: Durvalumab<br>Drug: BA3011<br>Drug: BA3021 | Canadian Cancer Trials Group |
| A Study of Maintenance DCVAC/OvCa After First-line Chemotherapy Added Standard of Care | NCT04834544 | N/A | 2 | Combination Product: DCVAC/OvCa<br>Combination Product: Placebo | Peking University Third Hospital |
| Efficacy of Tislelizumab and Spartalizumab Across Multiple Cancer Types in Patients with programmed death receptor 1 (PD-1)-high MRNA Expressing Tumors | NCT04802876 | ACROPOLI | 2 | Drug: Spartalizumab<br>Drug: Tislelizumab | SOLTI Breast Cancer Research Group |
| OSE2101 Alone or in Combination With Pembrolizumab vs. best supportive care in Patients With Platinum-sensitive Recurrent OC | NCT04713514 | TEDOVA | 2 | Drug: OSE2101<br>Drug: Pembrolizumab 25 mg/mL [Keytruda] | ARCAGY/ GINECO GROUP |
| Addition of Pembrolizumab to the Standard of Care Chemotherapy in Patients With small cell carcinoma of the ovary, hypercalcemic type | NCT04602377 | PembroSCCOHT | 2 | Drug: Pembrolizumab 25 mg/mL [Keytruda] | ARCAGY/ GINECO GROUP |
| Oregovomab Plus Chemo in Newly Diagnosed Patients With Advanced Epithelial OC Following Optimal Debulking Surgery | NCT04498117 | FLORA-5 | 3 | Biological: Oregovomab<br>Drug: Paclitaxel<br>Drug: Carboplatin | CanariaBio Inc. |
| Pembrolizumab and Carboplatin for the Treatment of Recurrent Ovarian, Fallopian Tube, or Primary Peritoneal Cancer | NCT04387227 | N/A | 2 | Drug: Carboplatin<br>Biological: Pembrolizumab<br>Procedure: Computed Tomography | University of Washington |
| ATr Inhibitor in Combination With Olaparib/Durvalumab (MEDI4736) in Gynaecological Cancers With AT-rich interactive domain-containing protein 1A (ARID1A) Loss or no Loss | NCT04065269 | ATARI | 2 | Drug: Ceralasertib<br>Drug: Olaparib<br>Drug: Durvalumab | Institute of Cancer Research, UK |

(Continued)

**Table 3 (continued)**

| Study Title | NCT Number | Acronym | Phase | Interventions | Sponsor |
|---|---|---|---|---|---|
| Testing the Addition of an Immunotherapy Drug, Tremelimumab, to the poly ADP-ribose polymerase (PARP) Inhibition Drug, Olaparib, for Recurrent Ovarian, Fallopian Tube, or Peritoneal Cancer | NCT04034927 | N/A | 2 | Procedure: Biospecimen Collection<br>Procedure: Computed Tomography<br>Procedure: Magnetic Resonance Imaging | National Cancer Institute |
| Study of an Immunotherapeutic, DPX-Survivac, in Combination With Low Dose Cyclophosphamide & Pembrolizumab, in Subjects With Selected Advanced & Recurrent Solid Tumors | NCT03836352 | N/A | 2 | Other: DPX-Survivac<br>Drug: Cyclophosphamide<br>Drug: Pembrolizumab | ImmunoVaccine Technologies, Inc. (IMV Inc.) |
| Trans-Artery/Intra-Tumor Infusion of Checkpoint Inhibitors Plus Chemodrug for Immunotherapy of Advanced Solid Tumors | NCT03755739 | N/A | 2/3 | Drug: Checkpoint inhibitor such as Pembrolizumab plus chemotherapy | Second Affiliated Hospital of Guangzhou Medical University |
| Systemic Immune Checkpoint Blockade and Intraperitoneal Chemo-Immunotherapy in Recurrent OC | NCT03734692 | N/A | 1/2 | Drug: Rintatolimod<br>Drug: Pembrolizumab<br>Drug: Cisplatin | Robert Edwards |
| ACTengine® IMA203/IMA203CD8 as Monotherapy or in Combination With Nivolumab in Recurrent and/or Refractory Solid Tumors | NCT03686124 | ACTengine | 1/2 | Biological: IMA203 Product<br>Biological: IMA203 product-flat dose<br>Biological: IMA203CD8 Product | Immatics US, Inc. |
| Administration of Autologous T-Cells Genetically Engineered to Express T-Cell Receptors Reactive Against Neoantigens in People With Metastatic Cancer | NCT03412877 | N/A | 2 | Drug: Cyclophosphamide<br>Drug: Fludarabine<br>Drug: Aldesleukin | National Cancer Institute |
| Immunotherapy With Neo-adjuvant Chemotherapy for OC | NCT03249142 | INeOV | 1/2 | Drug: ARM A Durvalumab/chemotherapy association<br>Drug: ARM B Durvalumab/Tremelimumab/chemotherapy association | ARCAGY/GINECO GROUP |
| P53MVA and Pembrolizumab in Treating Patients With Recurrent Ovarian, Primary Peritoneal, or Fallopian Tube Cancer | NCT03113487 | N/A | 2 | Biological: Modified Vaccinia Virus Ankara Vaccine Expressing p53<br>Biological: Pembrolizumab | City of Hope Medical Center |
| Pegylated Liposomal Doxorubicin Hydrochloride With Atezolizumab and/or Bevacizumab in Treating Patients With Recurrent Ovarian, Fallopian Tube, or Primary Peritoneal Cancer | NCT02839707 | N/A | 2/3 | Drug: Atezolizumab<br>Biological: Bevacizumab<br>Procedure: Computed Tomography | National Cancer Institute |

(Continued)

**Table 3 (continued)**

| Study Title | NCT Number | Acronym | Phase | Interventions | Sponsor |
|---|---|---|---|---|---|
| Nivolumab and Ipilimumab in Treating Patients With Rare Tumors | NCT02834013 | N/A | 2 | Procedure: Biospecimen Collection<br>Procedure: Computed Tomography<br>Procedure: Echocardiography Test | National Cancer Institute |
| Administering Peripheral Blood Lymphocytes Transduced With a CD70-Binding Chimeric Antigen Receptor to People With CD70 Expressing Cancers | NCT02830724 | N/A | 1/2 | Drug: Cyclophosphamide<br>Drug: Fludarabine<br>Drug: Aldesleukin | National Cancer Institute |
| Matched Paired Pharmacodynamics and Feasibility Study of Durvalumab in Combination With Chemotherapy in Frontline OC (N-Dur) | NCT02726997 | N/A | 1/2 | Drug: Carboplatin<br>Biological: Durvalumab<br>Other: Laboratory Biomarker Analysis | M.D. Anderson Cancer Center |
| Gene-Modified T Cells With or Without Decitabine in Treating Patients With Advanced Malignancies Expressing New York esophageal squamous cell carcinoma 1 (NY-ESO-1) | NCT02650986 | N/A | 1/2 | Drug: Cyclophosphamide<br>Drug: Decitabine<br>Other: Laboratory Biomarker Analysis | Roswell Park Cancer Institute |
| PARP-inhibition and cytotoxic T-lymphocyte associated protein 4 (CTLA-4) Blockade in BRCA-deficient OC | NCT02571725 | N/A | 1/2 | Drug: Olaparib<br>Drug: Tremelimumab | New Mexico Cancer Research Alliance |
| A Trial of Vigil for Participants with OC | NCT02346747 | VITAL | 2 | Biological: Vigil<br>Other: Placebo | Gradalis, Inc. |
| Immunotherapy Using Tumor Infiltrating Lymphocytes for Patients With Metastatic Cancer | NCT01174121 | N/A | 2 | Drug: Pembrolizumab (Keytruda)<br>Drug: Fludarabine<br>Drug: Cyclophosphamide | National Cancer Institute |
| Carboplatin, Paclitaxel and Gemcitabine Hydrochloride With or Without Bevacizumab After Surgery in Treating Patients With Recurrent Ovarian, Epithelial, Primary Peritoneal, or Fallopian Tube Cancer | NCT00565851 | N/A | 3 | Biological: Bevacizumab<br>Drug: Carboplatin<br>Drug: Docetaxel | National Cancer Institute |

Note: Abbreviation: OC: ovarian cancer; TCR-T cell: T cell expressing engineered T cell receptor; PD-1: programmed death receptor 1; ARID1A: AT-rich interactive domain-containing protein 1A; PARP: the poly ADP-ribose polymerase; NY-ESO-1: New York esophageal squamous cell carcinoma 1; CTLA-4: cytotoxic T-lymphocyte associated protein 4.

Combining iNKT-based therapy with PD-1/PD-L1 or CTLA-4 blockade offers a promising strategy to reverse T cell exhaustion in malignancies, including ovarian cancer. iNKT cells can both directly kill tumor cells and rapidly reshape the TME, providing costimulatory signals and cytokines that help restore activity of dysfunctional $CD8^+$ T cells. Adoptive transfer of iNKT cells alongside $PD\text{-}1^+$ $CD8^+$ T cells could therefore amplify antitumor activity by restoring effector functions in exhausted T cells while iNKT cells remodel suppressive niches [155,156]. Preclinical work [157] showed superior tumor control when iNKT cells

were given with tumor-specific T cells compared with either cell type alone, and early clinical studies using combined iNKT and PD-1$^{+}$ CD8$^{+}$ T cells have demonstrated feasibility and acceptable safety across solid tumors, with signals of clinical benefit in some patients [158]. Moreover, pairing iNKT therapy with ICIs such as anti-PD-1/PD-L1 or anti-CTLA-4 could provide complementary mechanisms. Implementation of ICIs leads to the release of inhibitory brakes on T cells while iNKT cells supply activation and microenvironmental reprogramming, making the combination a rational approach to overcome exhaustion and enhance durable responses in OC. Further controlled trials are needed to define optimal dosing, scheduling, and predictive biomarkers [37,151].

Another challenge is biomarker selection, which is crucial for identifying OC patients most likely to respond to ICIs. While individual biomarkers such as PD-L1 expression, HRD, microsatellite instability, tumor mutational burden, and specific gene mutations (e.g., ARID1A, STAT1, APOBEC3A) have been explored, none alone reliably predict response. Integrative approaches combining genomic, transcriptomic, and proteomic signatures along with immune cell infiltration patterns and chemokine expression (e.g., chemokine (C-X-C motif) ligand 9 (CXCL9), CXCL10, CXCL13) offer greater predictive accuracy. Developing composite biomarker panels and dynamic assessment models is essential for advancing personalized immunotherapy in OC [159,160].

To improve OC patients' outcomes, it is important to design combination therapies based on genomic data, molecular testing, and real-time changes in the TME. This can help identify useful biomarkers and allow for more personalized treatment, avoid HPD, and distinguish it from pseudoprogression. It is crucial to investigate their background to identify predictive factors and improve decision-making regarding the implementation of ICPs based immunotherapy or early termination of the treatment [17,45,48]. The determination of OC patients at risk of developing HPD is crucial to avoid the abrupt progression of malignancy [161]. Close cooperation between scientists, clinicians, and drug developers is essential to make further progress in this field, and not only to prolong OS but also to improve the quality of life of OC patients. Therefore establishment of molecular, genomic, and immune signatures of OC may result in the development of targeted therapies that could be beneficial for OC patients [2,45].

Therefore, harnessing NKT cells represents a potentially effective strategy in the treatment of OC. Optimizing the selection of α-GalCer-based agonists and developing methods to restore physiological levels of iNKT cells in OC patients may provide deeper insight into how to effectively integrate these cells into immunotherapeutic approaches.

## 9 Limitations

A key limitation of this review is the lack of ongoing clinical trials investigating the use of CAR-iNKT cells or other iNKT-based therapies specifically in patients with OC. The current evidence base is therefore derived primarily from preclinical experiments, fundamental immunology studies, or clinical trials conducted in other malignancies, particularly solid tumors with biological characteristics comparable to OC. While these studies provide valuable mechanistic insights and proof-of-concept data, their findings cannot yet be directly extrapolated to OC patients. Further clinical research is required to validate the therapeutic potential, safety, and efficacy of iNKT-based therapies in this context and to determine their role within the evolving landscape of OC immunotherapy.

A major limitation in advancing CAR-iNKT cell therapies lies in the extremely low abundance of iNKT cells in humans, typically representing only 0.01%–1% of peripheral blood T cells. This scarcity requires substantial *ex vivo* expansion to obtain sufficient cell numbers for clinical use. The problem is even more pronounced in autologous settings, where cancer progression and prior immunosuppressive treatments can markedly deplete endogenous iNKT pools and impair their effector functions. Although this presents a

significant bottleneck, optimized expansion protocols have been developed using α-GalCer–pulsed APCs or anti-CD3 stimulation, together with cytokine support such as IL-2, IL-7, IL-15, or IL-21, enabling the generation of clinically relevant iNKT products. Another key limitation is the functional and phenotypic heterogeneity of iNKT cells. Distinct subsets, particularly $CD4^+$ and $CD4^-$ iNKT cells, differ in cytokine profiles and cytotoxic potential. $CD4^+$ iNKT cells exhibit broad cytokine production (e.g., GM-CSF, TNF-α, IFN-γ, IL-4, IL-2), acting in an adjuvant-like manner to enhance antigen-specific T cell responses, whereas $CD4^-$ subsets display stronger cytolytic and Th1-biased activity. Moreover, memory-like $CD62L^+$ iNKT cells show superior persistence, proliferation, and antitumor efficacy. Clinical studies confirmed that higher frequencies of $CD62L^+$ CAR-iNKT cells in infusion products correlate with better *in vivo* expansion and therapeutic responses, highlighting the importance of subset composition for treatment success [109].

Although off-the-shelf iNKT cell-based therapy shows great potential for OC, several challenges remain. Safety concerns, particularly the risk of "on-target, off-tumor" toxicity, need to be minimized by selecting more specific targets such as claudin 6 or mucin 16. Further optimization of CAR design and large-scale production methods is required to ensure safety, efficacy, and feasibility. Ultimately, well-designed clinical trials are necessary to confirm the therapeutic value of allogeneic CAR-iNKT cells and other iNKT-based strategies in ovarian cancer treatment.

## 10 Conclusions

OC is marked by high heterogeneity, early metastases occurrence, and recurrence; thus, the mortality rate remains extremely high. Despite advances in medicine and clinical trials, the biological mechanisms behind the aggressiveness of OC are still not fully understood. The ovarian TME plays an important role in promoting tumor growth, spread, and resistance, which makes it a promising target for new treatments. However, the complexity of interactions between cancer cells, host immune system cells, soluble factors, miRNAs, and other noncellular components makes it difficult to develop effective new drugs. Taking into account the limited efficacy of available treatment in advanced stages of OC and in recurrent disease, targeted therapies are highly needed. Immunotherapies based on ICIs, such as anti-PD-1 or anti-PD-L1 monoclonal antibodies, have become game changers in the treatment of solid malignancies, including melanoma, renal cancer, and lung cancer. However, the response rate to this kind of treatment, especially in monotherapy, is limited because OC tumors are typically noniflamed. Therefore, combined therapies targeting multiple ICIs as well as other biological factors may be beneficial for OC patients.

CAR- and TCR-engineered iNKT cells emerge as promising candidates for next-generation dual-specific effector cell therapies. Their unique properties justify further research aimed at evaluating their anti-tumor potential in adoptive cell therapy settings, especially in comparison to conventional T cells. Notably, these iNKT-based approaches may offer significant advantages, such as eliminating the need for HLA matching and enabling targeted recognition of TAAs. Furthermore, their intrinsic capacity to modulate the TME highlights their therapeutic promise in reshaping immune responses in cancer patients. There is a critical need for more effective therapies for OC, particularly in light of frequent recurrence and resistance to platinum-based treatments. In summary, iNKT cells represent a promising alternative to conventional T cells in cancer immunotherapy. Their unique features, including CD1d-restricted recognition, natural migration to tumor sites, and capacity to modulate the immunosuppressive TME, make them well-suited for the projection of next-generation adoptive cell therapies targeting solid tumors, including ovarian cancer. However, further studies, including clinical trials that are currently lacking, are necessary to confirm the efficacy and safety of iNKT-based therapy. Most of the available data comes from preclinical studies, which limit the ability to fully assess its therapeutic potential.

**Acknowledgement:** None.

**Funding Statement:** This work was supported by the Medical University of Lublin, grant number PBmb2.

**Author Contributions:** The authors confirm contribution to the paper as follows: Conceptualization, Anna Pawłowska-Łachut; methodology, Anna Pawłowska-Łachut; software, Anna Pawłowska-Łachut, Iwona Wertel; formal analysis, Dorota Suszczyk; investigation, Anna Pawłowska-Łaachut, Iwona Wertel; resources, Anna Pawłowska-Łachut; writing—original draft preparation, Anna Pawłowska-Łachut; writing—review and editing, Anna Pawłowska-Łachut, Dorota Suszczyk; supervision, Iwona Wertel; project administration, Anna Pawłowska-Łachut; funding acquisition, Anna Pawłowska-Łachut, Iwona Wertel. All authors reviewed the results and approved the final version of the manuscript.

**Availability of Data and Materials:** Not applicable.

**Ethics Approval:** Not applicable.

**Conflicts of Interest:** The authors declare no conflicts of interest to report regarding the present study.

## Abbreviations

| | |
|---|---|
| AE | Adverse effect |
| AlloHSC-iNKT | Allogeneic hematopoietic stem cell-derived invariant natural killer T |
| ANXA2 | Annexin A2 |
| APC | Antigen-presenting cell |
| ARID1A | AT-rich interactive domain-containing protein 1A |
| BRCA | Breast cancer susceptibility gene |
| BsAb | Bispecific antibody |
| CAF | Cancer-associated fibroblast |
| CAR-iNKT | Chimeric antigen receptor invariant natural killer T |
| CCL | Chemokine (C-C motif) ligand |
| CD | Cluster of differentation |
| CAR | Chimeric antigen receptor |
| CRES | Cell-related encephalopathy syndrome |
| CRS | Cytokine release syndrome |
| CTLA-4 | Cytotoxic T lymphocyte associated protein 4 |
| CXCL | Chemokine (C-X-C motif) ligand |
| DAMP | Danger-associated molecular pattern |
| DC | Dendritic cell |
| DNAM-1 | DNAX accessory molecule-1 |
| ECM | Cytokines, extracellular matrix |
| EGFR | Epidermal growth factor receptor |
| EPCAM | Epithelial cell adhesion molecule |
| FasL | Fas ligand |
| FRα | Folate receptor α |
| FIGO | International Federation of Gynecology and Obstetrics |
| FDA | The Food and Drug Administration |
| GM-CSF | Granulocyte-macrophage colony-stimulating factor |
| GvHD | Graft-vs.-host disease |
| HER2 | Human epidermal growth factor receptor 2 |
| HLA | Human leukocyte antigen |
| HNC | Head and neck cancer |

| | |
|---|---|
| HPD | Hyperprogressive disease |
| HRD | Homologous recombination deficiency |
| HSC | Hematopoietic stem cell |
| ICANS | Immune effector cell–associated neurotoxicity syndrome |
| ICI | Immune checkpoint inhibitor |
| ICP | Immune checkpoint |
| IFN-γ | IFN-γ |
| IL | Interleukin |
| iNKT | Invariant natural killer T |
| mAb | Monoclonal antibody |
| MDSC | Myeloid-derived suppressive cells |
| MHC | Major histocompatibility complex |
| MIP-1α | Macrophage inflammatory proteins α |
| MIP-1β | Macrophage inflammatory proteins β |
| miRNA | MicroRNA |
| Mo-DCs | Eliminate monocyte-derived dendritic cells |
| NK | Natural killer |
| NKT | Natural killer T |
| NSCLC | Non-small cells lung cancer |
| NY-ESO-1 | New York esophageal squamous cell carcinoma 1 |
| OC | Ovarian cancer |
| OS | Overall survival |
| PARP | Poly (ADP-ribose) polymerase |
| PARPi | Poly (ADP-ribose) polymerase inhibitor |
| PB | Peripheral blood |
| PBMC | Peripheral blood mononuclear cell |
| PD-1 | Programmed death receptor-1 |
| PD-L1 | Programmed death ligand-1 |
| PFS | Progression-free survival |
| PTK7 | Protein tyrosine kinase |
| RANTES | Regulated on activation normal T cell expressed and secreted |
| TAA | Tumor-associated antigen |
| TAE | Transarterial embolization |
| TAM | Tumor-associated macrophage |
| TCB | T cell redirecting bispecific |
| TCR | T cell receptors |
| TCR-iNKT | T cell receptor invariant natural killer T |
| TCR-T cell | T cell receptor T cell |
| TGF-β | Transforming growth factor β |
| TIGIT | The T-cell immunoglobulin and ITIM domain/CD155/DNAX |
| TIL | Tumor-infiltrating T cell |
| TLR | Toll-like receptor |
| TME | Tumor microenvironment |
| TNF-α | Tumor necrosis factor α |
| TRAIL | TNF-related apoptosis-inducing ligand |
| Treg | Regulatory T cells |
| VEGFi | Vascular endothelial growth factor inhibitor |
| WHO | World Health Organization |
| αGalCer/DCs | αGalCer-pulsed DCs |

## References


1. Cancer Today [Internet]. [cited 2025 Aug 2]. Available from: http://gco.iarc.fr/today/home.
2. Stewart C, Ralyea C, Lockwood S. Ovarian cancer: an integrated review. Semin Oncol Nurs. 2019;35(2):151–6.
3. Jayson GC, Kohn EC, Kitchener HC, Ledermann JA. Ovarian cancer. Lancet. 2014;384(9951):1376–88. doi:10.1016/s0140-6736(13)62146-7.
4. Lin H, Wang J, Wen X, Wen Q, Huang S, Mai Z, et al. A prognosis-predictive nomogram of ovarian cancer with two immune-related genes: CDC20B and PNPLA5. Oncol Lett. 2020;20(5):204.
5. Wang G, Yang H, Wang Y, Qin J. Ovarian cancer targeted therapy: current landscape and future challenges. Front Oncol. 2025;15:1535235. doi:10.3389/fonc.2025.1535235.
6. Huo X, Zhang X, Li S, Wang S, Sun H, Yang M. Identification of novel immunologic checkpoint gene prognostic markers for ovarian cancer. J Oncol. 2022;2022:8570882. doi:10.1155/2022/8570882.
7. Zhang Y, Cui Q, Xu M, Liu D, Yao S, Chen M. Current Advances in PD-1/PD-L1 Blockade in Recurrent Epithelial Ovarian Cancer. Front Immunol [Internet]. 2022 [cited 2025 Nov 4]. Available from: https://www.frontiersin.org/articles/10.3389/fimmu.2022.901772.
8. Commissioner O of the. U.S. Food and Drug Administration [Internet]. FDA; 2022 [cited 2022 Nov 4]. Available from: https://www.fda.gov/home.
9. Lheureux S, Braunstein M, Oza AM. Epithelial ovarian cancer: evolution of management in the era of precision medicine. CA A Cancer J Clin. 2019;69(4):280–304. doi:10.3322/caac.21559.
10. Rutten MJ, Leeflang MMG, Kenter GG, Mol BWJ, Buist M. Laparoscopy for diagnosing resectability of disease in patients with advanced ovarian cancer. Cochrane Database Syst Rev. 2014;20(5):CD009786. doi:10.1002/14651858.cd009786.
11. FDA-Approved Ovarian Cancer Treatments [Internet]. MPR; 2025 [cited 2025 Oct 16]. Available from: https://www.empr.com/charts/fda-approved-ovarian-cancer-treatments/.
12. Drugs@FDA: FDA-Approved Drugs [Internet]. [cited 2025 Oct 16]. Available from: https://www.accessdata.fda.gov/scripts/cder/daf/index.cfm.
13. Świderska J, Kozłowski M, Kwiatkowski S, Cymbaluk-Płoska A. Immunotherapy of ovarian cancer with particular emphasis on the PD-1/PDL-1 as target points. Cancers. 2021;13(23):6063. doi:10.3390/cancers13236063.
14. Chardin L, Leary A. Immunotherapy in ovarian cancer: thinking beyond PD-1/PD-L1. Front Oncol. 2021;11:795547. doi:10.3389/fonc.2021.795547.
15. Maiorano BA, Maiorano MFP, Lorusso D, Maiello E. Ovarian cancer in the era of immune checkpoint inhibitors: state of the art and future perspectives [Internet]. Medicine & Pharmacology; 2021 Aug [cited 2022 Mar 2]. Available from: https://www.preprints.org/manuscript/202108.0037/v1.
16. Li YR, Ochoa CJ, Zhu Y, Kramer A, Wilson M, Fang Y, et al. Profiling ovarian cancer tumor and microenvironment during disease progression for cell-based immunotherapy design. iScience. 2023;26(10):107952. doi:10.1016/j.isci.2023.107952.
17. Jiang Y, Wang C, Zhou S. Targeting tumor microenvironment in ovarian cancer: premise and promise. Biochim Et Biophys Acta (BBA)—Rev Cancer. 2020;1873(2):188361. doi:10.1016/j.bbcan.2020.188361.
18. Zhao Y, Shen M, Wu L, Yang H, Yao Y, Yang Q, et al. Stromal cells in the tumor microenvironment: accomplices of tumor progression? Cell Death Dis. 2023;14(9):587. doi:10.1038/s41419-023-06110-6.
19. El-Arabey AA, Alkhalil SS, Al-Shouli ST, Awadalla ME, Alhamdi HW, Almanaa TN, et al. Revisiting macrophages in ovarian cancer microenvironment: development, function and interaction. Med Oncol. 2023;40(5):142. doi:10.1007/s12032-023-01987-x.
20. Garlisi B, Lauks S, Aitken C, Ogilvie LM, Lockington C, Petrik D, et al. The complex tumor microenvironment in ovarian cancer: therapeutic challenges and opportunities. Curr Oncol. 2024;31(7):3826–44. doi:10.3390/curroncol31070283.
21. Ahmadi N, Kadi C, Benlamari M, Gaouzi Z, El Ouali EM, Bouziane A, et al. Efficacy of PD-1/PDL-1 inhibitors for ovarian cancer: a systematic review and network meta-analysis. Future Oncol. 2025;21(20):2637–47. doi:10.1080/14796694.2025.2530378.

22. Ghisoni E, Morotti M, Sarivalasis A, Grimm AJ, Kandalaft L, Laniti DD, et al. Immunotherapy for ovarian cancer: towards a tailored immunophenotype-based approach. Nat Rev Clin Oncol. 2024;21(11):801–17. doi:10.1038/s41571-024-00937-4.
23. Chen Y, Liu X, Hu Y, Xia L. Efficacy and safety of PD-1/PD-L1 immune checkpoint inhibitors in the treatment of recurrent ovarian cancer: a systematic review and meta-analysis. Medicine. 2024;103(18):e38019. doi:10.1097/md.0000000000038019.
24. Frentzas S, Austria Mislang AR, Lemech C, Nagrial A, Underhill C, Wang W, et al. Phase 1a dose escalation study of ivonescimab (AK112/SMT112), an anti-PD-1/VEGF-A bispecific antibody, in patients with advanced solid tumors. J Immunother Cancer. 2024;12(4):e008037. doi:10.1136/jitc-2023-008037.
25. Chen J, Yang L, Ma Y, Zhang Y. Recent advances in understanding the immune microenvironment in ovarian cancer. Front Immunol. 2024;15:1412328. doi:10.3389/fimmu.2024.1412328.
26. Pawłowska A, Skiba W, Suszczyk D, Kuryło W, Jakubowicz-Gil J, Paduch R, et al. The dual blockade of the TIGIT and PD-1/PD-L1 pathway as a new hope for ovarian cancer patients. Cancers. 2022;14(23):5757. doi:10.3390/cancers14235757.
27. Bonifácio VDB. Ovarian cancer biomarkers: moving forward in early detection. In: Serpa J, editor. Tumor microenvironment: the main driver of metabolic adaptation. Cham, Switzerland: Springer International Publishing; 2020. p. 355–63. doi:10.1007/978-3-030-34025-4_18.
28. Gęca K, Litwiński J, Ostrowski T, Świetlicka I, Polkowski WP, Skórzewska M. Exploring the survival determinants in recurrent ovarian cancer: the role of cytoreductive surgery and hyperthermic intraperitoneal chemotherapy. Cancers. 2024;16(11):2150. doi:10.3390/cancers16112150.
29. Kargo AS, Jensen PT, Lindemann K, Hjøllund NH, Liposits GI, Raaschou-Jensen N, et al. Association of patient-reported outcomes and ovarian cancer recurrence. Int J Gynecol Cancer. 2021;31(9):1248–59. doi:10.1136/ijgc-2021-002550.
30. Gupta M, Babic A, Beck AH, Terry K. TNF-α expression, risk factors, and inflammatory exposures in ovarian cancer: evidence for an inflammatory pathway of ovarian carcinogenesis? Hum Pathol. 2016;54:82–91. doi:10.1016/j.humpath.2016.03.006.
31. Nebgen DR, Lu KH, Bast RC. Novel approaches to ovarian cancer screening. Curr Oncol Rep. 2019;21(8):75. doi:10.1007/s11912-019-0816-0.
32. Chen K, Wang J, Yang M, Deng S, Sun L. Immunotherapy in recurrent ovarian cancer. Biomedicines. 2025;13(1):168. doi:10.3390/biomedicines13010168.
33. FDA Approves Pembrolizumab for Adjuvant Treatment of Stage IIB or IIC Melanoma|FDA [Internet]. [cited 2025 Oct 21]. Available from: https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-pembrolizumab-adjuvant-treatment-stage-iib-or-iic-melanoma.
34. FDA Approves Pembrolizumab as Adjuvant Treatment for Non-Small Cell Lung Cancer. FDA [Internet]. 2024 Aug 9 [cited 2025 Oct 21]. Available from: https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-pembrolizumab-adjuvant-treatment-non-small-cell-lung-cancer.
35. FDA Approves Pembrolizumab for Adjuvant Treatment of Renal Cell Carcinoma. FDA [Internet]. 2024 Aug 9 [cited 2025 Oct 21]. Available from: https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-pembrolizumab-adjuvant-treatment-renal-cell-carcinoma.
36. Matulonis UA, Shapira-Frommer R, Santin AD, Lisyanskaya AS, Pignata S, Vergote I, et al. Antitumor activity and safety of pembrolizumab in patients with advanced recurrent ovarian cancer: results from the phase II KEYNOTE-100 study. Ann Oncol. 2019;30(7):1080–7. doi:10.1093/annonc/mdz135.
37. Hsu CY, Muhammed TM, Uthirapathy S, Ahmad I, Ballal S, Kalia R, et al. CTLA-4 blockade in ovarian cancer immunotherapy: mechanisms and clinical strategies. Semin Oncol. 2025;52(4):152370. doi:10.1016/j.seminoncol.2025.152370.
38. Majidpoor J, Mortezaee K. The efficacy of PD-1/PD-L1 blockade in cold cancers and future perspectives. Clin Immunol. 2021;226(13):108707. doi:10.1016/j.clim.2021.108707.
39. Drakes ML, Czerlanis CM, Stiff PJ. Immune checkpoint blockade in gynecologic cancers: state of affairs. Cancers. 2020;12(11):3301. doi:10.3390/cancers12113301.

40. Agyemang AF, Lele S. The use of immunotherapy for treatment of gynecologic malignancies. In: Lele S, editor. Ovarian cancer. Brisbane, Australia: Exon Publications; 2022 [cited 2025 Aug 8]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK585994/
41. Mi JL, Xu M, Liu C, Wang RS. Interactions between tumor mutation burden and immune infiltration in ovarian cancer. Int J Clin Exp Pathol. 2020;13(10):2513–23.
42. Deshpande M, Romanski PA, Rosenwaks Z, Gerhardt J. Gynecological cancers caused by deficient mismatch repair and microsatellite instability. Cancers. 2020;12(11):3319. doi:10.3390/cancers12113319.
43. Evrard C, Alexandre J. Predictive and prognostic value of microsatellite instability in gynecologic cancer (endometrial and ovarian). Cancers. 2021;13(10):2434. doi:10.3390/cancers13102434.
44. Zhang L, Conejo-Garcia JR, Katsaros D, Gimotty PA, Massobrio M, Regnani G, et al. Intratumoral T cells, recurrence, and survival in epithelial ovarian cancer. N Engl J Med. 2003;348(3):203–13. doi:10.1056/nejmoa020177.
45. Pawłowska A, Rekowska A, Kuryło W, Pańczyszyn A, Kotarski J, Wertel I. Current understanding on why ovarian cancer is resistant to immune checkpoint inhibitors. Int J Mol Sci. 2023;24(13):10859. doi:10.3390/ijms241310859.
46. Saâda-Bouzid E, Defaucheux C, Karabajakian A, Coloma VP, Servois V, Paoletti X, et al. Hyperprogression during anti-PD-1/PD-L1 therapy in patients with recurrent and/or metastatic head and neck squamous cell carcinoma. Ann Oncol. 2017;28(7):1605–11. doi:10.1093/annonc/mdx178.
47. Lo Russo G, Moro M, Sommariva M, Cancila V, Boeri M, Centonze G, et al. Antibody-Fc/FcR interaction on macrophages as a mechanism for hyperprogressive disease in non–small cell lung cancer subsequent to PD-1/PD-L1 blockade. Clin Cancer Res. 2019;25(3):989–99. doi:10.1158/1078-0432.ccr-18-1390.
48. Frelaut M, Du Rusquec P, De Moura A, Le Tourneau C, Borcoman E. Pseudoprogression and hyperprogression as new forms of response to immunotherapy. BioDrugs. 2020;34(4):463–76. doi:10.1007/s40259-020-00425-y.
49. Arasanz H, Zuazo M, Bocanegra A, Gato M, Martínez-Aguillo M, Morilla I, et al. Early detection of hyperprogressive disease in non-small cell lung cancer by monitoring of systemic T cell dynamics. Cancers. 2020;12(2):344. doi:10.3390/cancers12020344.
50. Boland JL, Zhou Q, Martin M, Callahan MK, Konner J, O'Cearbhaill RE, et al. Early disease progression and treatment discontinuation in patients with advanced ovarian cancer receiving immune checkpoint blockade. Gynecol Oncol. 2019;152(2):251–8. doi:10.1016/j.ygyno.2018.11.025.
51. Champiat S, Dercle L, Ammari S, Massard C, Hollebecque A, Postel-Vinay S, et al. Hyperprogressive disease is a new pattern of progression in cancer patients treated by Anti-PD-1/PD-L1. Clin Cancer Res. 2017;23(8):1920–8. doi:10.1158/1078-0432.ccr-16-1741.
52. Kim CG, Kim KH, Pyo KH, Xin CF, Hong MH, Ahn BC, et al. Hyperprogressive disease during PD-1/PD-L1 blockade in patients with non-small-cell lung cancer. Ann Oncol. 2019;30(7):1104–13. doi:10.1093/annonc/mdz123.
53. Kanjanapan Y, Day D, Wang L, Al-Sawaihey H, Abbas E, Namini A, et al. Hyperprogressive disease in early-phase immunotherapy trials: clinical predictors and association with immune-related toxicities. Cancer. 2019;125(8):1341–9. doi:10.1002/cncr.31999.
54. Failing JJ, Dudek OA, Marin Acevedo JA, Chirila RM, Dong H, Markovic SN, et al. Biomarkers of hyperprogression and pseudoprogression with immune checkpoint inhibitor therapy. Future Oncol. 2019;15(22):2645–56. doi:10.2217/fon-2019-0183.
55. Di Giacomo AM, Danielli R, Guidoboni M, Calabrò L, Carlucci D, Miracco C, et al. Therapeutic efficacy of ipilimumab, an anti-CTLA-4 monoclonal antibody, in patients with metastatic melanoma unresponsive to prior systemic treatments: clinical and immunological evidence from three patient cases. Cancer Immunol Immunother. 2009;58(8):1297–306. doi:10.1007/s00262-008-0642-y.
56. Matos I, Martin-Liberal J, Hierro C, Ochoa De Olza M, Viaplana C, Costa M, et al. Incidence and clinical implications of a new definition of hyperprogression (HPD) with immune checkpoint inhibitors (ICIs) in patients treated in phase 1 (Ph1) trials. JCO. 2018;36(15 suppl):3032. doi:10.1200/jco.2018.36.15_suppl.3032.
57. Chiou VL, Burotto M. Pseudoprogression and immune-related response in solid tumors. J Clin Oncol. 2015;33(31):3541–3. doi:10.1200/jco.2015.61.6870.

58. Queirolo P, Spagnolo F. Atypical responses in patients with advanced melanoma, lung cancer, renal-cell carcinoma and other solid tumors treated with anti-PD-1 drugs: a systematic review. Cancer Treat Rev. 2017;59(23):71–8. doi:10.1016/j.ctrv.2017.07.002.
59. Passler M, Taube ET, Sehouli J, Pietzner K. Pseudo- or real progression? An ovarian cancer patient under nivolumab: a case report. World J Clin Oncol. 2019;10(7):247–55. doi:10.5306/wjco.v10.i7.247.
60. Li H, Zhou X, Zhang D, Wang G, Cheng X, Xu C, et al. Early onset immune-related adverse event to identify pseudo-progression in a patient with ovarian cancer treated with nivolumab: a case report and review of the literature. Front Med. 2020;7:366. doi:10.3389/fmed.2020.00366.
61. Ma Y, Wang Q, Dong Q, Zhan L, Zhang J. How to differentiate pseudoprogression from true progression in cancer patients treated with immunotherapy. Am J Cancer Res. 2019;9(8):1546–53.
62. Aoki T, Motohashi S, Koseki H. Regeneration of invariant natural killer T (iNKT) cells: application of iPSC technology for iNKT cell-targeted tumor immunotherapy. Inflamm Regen. 2023;43(1):27. doi:10.1186/s41232-023-00275-5.
63. Winkler I, Woś J, Bojarska-Junak A, Semczuk A, Rechberger T, Baranowski W, et al. An association of iNKT+/CD3+/CD161+ lymphocytes in ovarian cancer tissue with CA125 serum concentration. Immunobiology. 2020;225(6):152010. doi:10.1016/j.imbio.2020.152010.
64. Díaz-Basabe A, Strati F, Facciotti F. License to kill: when iNKT cells are granted the use of lethal cytotoxicity. Int J Mol Sci. 2020;21(11):3909. doi:10.3390/ijms21113909.
65. Hapil FZ, Wingender G. The interaction between invariant natural killer T cells and the mucosal microbiota. Immunology. 2018;155(2):164–75. doi:10.1111/imm.12958.
66. Kinjo Y, Takatsuka S, Kitano N, Kawakubo S, Abe M, Ueno K, et al. Functions of CD1d-restricted invariant natural killer T cells in antimicrobial immunity and potential applications for infection control. Front Immunol. 2018;9:1266. doi:10.3389/fimmu.2018.01266.
67. Tian C, Wang Y, Su M, Huang Y, Zhang Y, Dou J, et al. Motility and tumor infiltration are key aspects of invariant natural killer T cell anti-tumor function. Nat Commun. 2024;15(1):1213. doi:10.1038/s41467-024-45208-z.
68. Shyanti RK, Haque M, Singh R, Mishra M. Optimizing iNKT-driven immune responses against cancer by modulating CD1d in tumor and antigen presenting cells. Clin Immunol. 2024;269:110402. doi:10.1016/j.clim.2024.110402.
69. Liu Y, Wang G, Chai D, Dang Y, Zheng J, Li H. iNKT: a new avenue for CAR-based cancer immunotherapy. Transl Oncol. 2022;17:101342.
70. iNKT Cell Therapy in 2025: A Novel Paradigm in Cancer Immunotherapy—OncoDaily [Internet]. 2025 [cited 2025 Oct 20]. Available from: https://oncodaily.com/oncolibrary/inkt-cell-therapy.
71. Wang Z, Zhang G. CAR-iNKT cell therapy: mechanisms, advantages, and challenges. Curr Res Transl Med. 2025;73(1):103488. doi:10.1016/j.retram.2024.103488.
72. Tessmer MS, Fatima A, Paget C, Trottein F, Brossay L. NKT cell immune responses to viral infection. Expert Opin Ther Targets. 2009;13(2):153–62. doi:10.1517/14712590802653601.
73. Krijgsman D, Hokland M, Kuppen PJK. The role of natural killer T cells in cancer-a phenotypical and functional approach. Front Immunol. 2018;9:367. doi:10.3389/fimmu.2018.00367.
74. Crosby CM, Kronenberg M. Tissue-specific functions of invariant natural killer T cells. Nat Rev Immunol. 2018;18(9):559–74. doi:10.1038/s41577-018-0034-2.
75. Novak J, Lehuen A. Mechanism of regulation of autoimmunity by iNKT cells. Cytokine. 2011;53(3):263–70. doi:10.1016/j.cyto.2010.11.001.
76. Yamamura T, Sakuishi K, Illés Z, Miyake S. Understanding the behavior of invariant NKT cells in autoimmune diseases. J Neuroimmunol. 2007;191(1):8–15. doi:10.1016/j.jneuroim.2007.09.014.
77. Cui G, Abe S, Kato R, Ikuta K. Insights into the heterogeneity of iNKT cells: tissue-resident and circulating subsets shaped by local microenvironmental cues. Front Immunol. 2024;15:1349184. doi:10.3389/fimmu.2024.1349184.
78. Delfanti G, Dellabona P, Casorati G, Fedeli M. Adoptive immunotherapy with engineered iNKT cells to target cancer cells and the suppressive microenvironment. Front Med. 2022;9:897750. doi:10.3389/fmed.2022.897750.

79. Cortesi F, Delfanti G, Grilli A, Calcinotto A, Gorini F, Pucci F, et al. Bimodal CD40/Fas-dependent crosstalk between iNKT cells and tumor-associated macrophages impairs prostate cancer progression. Cell Rep. 2018;22(11):3006–20. doi:10.1016/j.celrep.2018.02.058.
80. Metelitsa LS. Anti-tumor potential of type-I NKT cells against CD1d-positive and CD1d-negative tumors in humans. Clin Immunol. 2011;140(2):119–29. doi:10.1016/j.clim.2010.10.005.
81. Lindau D, Gielen P, Kroesen M, Wesseling P, Adema GJ. The immunosuppressive tumour network: myeloid-derived suppressor cells, regulatory T cells and natural killer T cells. Immunology. 2013;138(2):105–15. doi:10.1111/imm.12036.
82. Look A, Burns D, Tews I, Roghanian A, Mansour S. Towards a better understanding of human iNKT cell subpopulations for improved clinical outcomes. Front Immunol. 2023;14:1176724. doi:10.3389/fimmu.2023.1176724.
83. Alhamawi RM, Aloufi N, Alamri AF, Altubayli FA, Alsairi RT, Alhamad RA, et al. Prognostic impact of invariant natural killer T cells in solid and hematological tumors; systematic review and meta-analysis. Cancer Biomark. 2024;41(2):155–64. doi:10.3233/cbm-240069.
84. Eakins J, O'conner L, David J, Bates M, Ibrahim O, Martin C, et al. EV270/#1221 Ex vivo upregulation of CD1d expression to enhance the therapeutic benefit of iNKT cells of ovarian cancer. Int J Gynecol Cancer. 2024;34:A224. doi:10.1136/ijgc-2024-igcs.389.
85. Li Y, Ochoa C, Yang L. 416 Profiling ovarian cancer tumor and microenvironment with disease progression for the design of immunotherapy strategies. J Immunother Cancer. 2023;11(Suppl):A465. doi:10.1136/jitc-2023-SITC2023.0416.
86. Molling JW, Langius JAE, Langendijk JA, Leemans CR, Bontkes HJ, Van Der Vliet HJJ, et al. Low levels of circulating invariant natural killer T cells predict poor clinical outcome in patients with head and neck squamous cell carcinoma. J Clin Oncol. 2007;25(7):862–8. doi:10.1200/jco.2006.08.5787.
87. Fujii SI, Shimizu K, Smith C, Bonifaz L, Steinman RM. Activation of natural killer T cells by alpha-galactosylceramide rapidly induces the full maturation of dendritic cells *in vivo* and thereby acts as an adjuvant for combined CD4 and CD8 T cell immunity to a coadministered protein. J Exp Med. 2003;198(2):267–79. doi:10.1084/jem.20030324.
88. Metelitsa LS, Wu HW, Wang H, Yang Y, Warsi Z, Asgharzadeh S, et al. Natural killer T cells infiltrate neuroblastomas expressing the chemokine CCL2. J Exp Med. 2004;199(9):1213–21. doi:10.1084/jem.20031462.
89. De Lalla C, Rinaldi A, Montagna D, Azzimonti L, Bernardo ME, Sangalli LM, et al. Invariant NKT cell reconstitution in pediatric leukemia patients given HLA-haploidentical stem cell transplantation defines distinct CD4+ and CD4-subset dynamics and correlates with remission state. J Immunol. 2011;186(7):4490–9. doi:10.4049/jimmunol.1003748.
90. Hishiki T, Mise N, Harada K, Ihara F, Takami M, Saito T, et al. Invariant natural killer T infiltration in neuroblastoma with favorable outcome. Pediatr Surg Int. 2018;34(2):195–201. doi:10.1007/s00383-017-4189-x.
91. Tachibana T, Onodera H, Tsuruyama T, Mori A, Nagayama S, Hiai H, et al. Increased intratumor Valpha24-positive natural killer T cells: a prognostic factor for primary colorectal carcinomas. Clin Cancer Res. 2005;11(20):7322–7. doi:10.1158/1078-0432.ccr-05-0877.
92. Shaulov A, Yue S, Wang R, Joyce RM, Balk SP, Kim HT, et al. Peripheral blood progenitor cell product contains Th1-biased noninvariant CD1d-reactive natural killer T cells: implications for posttransplant survival. Exp Hematol. 2008;36(4):464–72. doi:10.1016/j.exphem.2007.12.010.
93. Kawano T, Cui J, Koezuka Y, Toura I, Kaneko Y, Sato H, et al. Natural killer-like nonspecific tumor cell lysis mediated by specific ligand-activated Valpha14 NKT cells. Proc Natl Acad Sci U S A. 1998;95(10):5690–3. doi:10.1073/pnas.95.10.5690.
94. Fujii SI, Shimizu K, Okamoto Y, Kunii N, Nakayama T, Motohashi S, et al. NKT cells as an ideal anti-tumor immunotherapeutic. Front Immunol. 2013;4:409.
95. Takami M, Ihara F, Motohashi S. Clinical application of iNKT cell-mediated anti-tumor activity against lung cancer and head and neck cancer. Front Immunol. 2018;9:2021. doi:10.3389/fimmu.2018.02021.
96. Coupe N, Pinato DJJ, Fairchild JP, Pastore DRE, Innaimo S, Thompson D, et al. IMPORT-201 (IMP-MEL): a phase 1 first-in-human dose finding/randomized phase 2 study of a novel iNKT agonist IMM60 and pembrolizumab

for advanced melanoma and metastatic non-small cell lung cancer (NSCLC). JCO. 2023;41(16 suppl):2575. doi:10.1200/jco.2023.41.16_suppl.2575.
97. Cui G, Shimba A, Jin J, Ogawa T, Muramoto Y, Miyachi H, et al. A circulating subset of iNKT cells mediates antitumor and antiviral immunity. Sci Immunol. 2022;7(76):eabj8760. doi:10.1126/sciimmunol.abj8760.
98. Li YR, Zhou Y, Kim YJ, Zhu Y, Ma F, Yu J, et al. Development of allogeneic HSC-engineered iNKT cells for off-the-shelf cancer immunotherapy. Cell Rep Med. 2021;2(11):100449.
99. CAR T-Cell Therapy vs. iNKT Cell Therapy: The Future of Engineered Immune Cells in Cancer Treatment—OncoDaily [Internet]. 2025 [cited 2025 Oct 16]. Available from: https://oncodaily.com/oncolibrary/car-t-cell-therapy-vs-inkt-cell-therapy.
100. Zhou X, Wang Y, Dou Z, Delfanti G, Tsahouridis O, Pellegry CM, et al. CAR-redirected natural killer T cells demonstrate superior antitumor activity to CAR-T cells through multimodal CD1d-dependent mechanisms. Nat Cancer. 2024;5(11):1607–21. doi:10.1038/s43018-024-00830-0.
101. Fights Cancer More Effectively—Supercharged Natural Killer Cell (CAR-iNKT)—Creative Biolabs CAR-T Blog [Internet]. 2018 [cited 2025 Aug 4]. Available from: https://www.creative-biolabs.com/blog/car-t/fights-cancer-more-effectively-supercharged-natural-killer-cell-car-inkt/.
102. Li YR, Li Z, Zhu Y, Li M, Chen Y, Lee D, et al. Overcoming ovarian cancer resistance and evasion to CAR-T cell therapy by harnessing allogeneic CAR-NKT cells. Med. 2025;6(10):100804. doi:10.1016/j.medj.2025.100804.
103. Cutri-French C, Nasioudis D, George E, Tanyi JL. CAR-T cell therapy in ovarian cancer: where are we now? Diagnostics. 2024;14(8):819. doi:10.3390/diagnostics14080819.
104. Domínguez-Prieto V, Qian S, Villarejo-Campos P, Meliga C, González-Soares S, Guijo Castellano I, et al. Understanding CAR T cell therapy and its role in ovarian cancer and peritoneal carcinomatosis from ovarian cancer. Front Oncol. 2023;13:1104547. doi:10.3389/fonc.2023.1104547.
105. Liang Z, Dong J, Yang N, Li SD, Yang ZY, Huang R, et al. Tandem CAR-T cells targeting FOLR1 and MSLN enhance the antitumor effects in ovarian cancer. Int J Biol Sci. 2021;17(15):4365–76. doi:10.7150/ijbs.63181.
106. Terlikowska KM, Dobrzycka B, Terlikowski SJ. Chimeric antigen receptor design and efficacy in ovarian cancer treatment. Int J Mol Sci. 2021;22(7):3495. doi:10.3390/ijms22073495.
107. Mitra A, Barua A, Huang L, Ganguly S, Feng Q, He B. From bench to bedside: the history and progress of CAR T cell therapy. Front Immunol. 2023;14:1188049. doi:10.3389/fimmu.2023.1188049.
108. Dickinson MJ, Barba P, Jäger U, Shah NN, Blaise D, Briones J, et al. A novel autologous CAR-T therapy, YTB323, with preserved T-cell stemness shows enhanced CAR T-cell efficacy in preclinical and early clinical development. Cancer Discov. 2023;13(9):1982–97. doi:10.1158/2159-8290.cd-22-1276.
109. Niedzielska M, Chalmers A, Popis MC, Altman-Sharoni E, Addis S, Beulen R, et al. CAR-iNKT cells: redefining the frontiers of cellular immunotherapy. Front Immunol. 2025;16:1625426. doi:10.3389/fimmu.2025.1625426.
110. Sanber K, Savani B, Jain T. Graft-vs.-host disease risk after chimeric antigen receptor T-cell therapy: the diametric opposition of T cells. Br J Haematol. 2021;195(5):660–8. doi:10.1111/bjh.17544.
111. Farid KMN, Bug G, Schmitt A, Lang F, Schubert ML, Haberkorn U, et al. GVHD after CAR T-cell therapy post allogeneic hematopoietic cell transplantation—successfully treated by extracorporeal photopheresis. Front Immunol. 2024;15:1500177. doi:10.3389/fimmu.2024.1500177.
112. Zhang H, Nguyen J, Pham M, Smith A. iNKT cell modulation of tumor microenvironment (TME) and antitumor immunity: the role of antitumorigenic and protumorigenic iNKT cells. J Immunol. 2020;204(1 suppl):88–3. doi:10.4049/jimmunol.204.supp.88.3.
113. Tang L, Pan S, Wei X, Xu X, Wei Q. Arming CAR-T cells with cytokines and more: innovations in the fourth-generation CAR-T development. Mol Ther. 2023;31(11):3146–62. doi:10.1016/j.ymthe.2023.09.021.
114. Correa-Lara MVM, Lara-Vega I, Nájera-Martínez M, Domínguez-López ML, Reyes-Maldonado E, Vega-López A. Tumor-infiltrating iNKT cells activated through c-Kit/Sca-1 are induced by pentoxifylline, norcantharidin, and their mixtures for killing murine melanoma cells. Pharmaceuticals. 2023;16(10):1472. doi:10.3390/ph16101472.
115. Li YR, Zhu Y, Chen Y, Yang L. The clinical landscape of CAR-engineered unconventional T cells. Trends Cancer. 2025;11(6):520–39.

116. Ramos CA, Courtney AN, Lulla PD, Hill LC, Kamble RT, Carrum G, et al. Off-the-shelf CD19-specific CAR-NKT cells in patients with relapsed or refractory B-cell malignancies. Transplant Cell Ther. 2024;30(2 suppl):S41–2. doi:10.1016/j.jtct.2023.12.072.
117. Adkins S. CAR T-cell therapy: adverse events and management. J Adv Pract Oncol. 2019;10 Suppl 3:21–8.
118. Zhang D, Lu J. Global trends and frontiers in iNKT cells: a bibliometric and visualized analysis. Front Immunol. 2025;16:1618254. doi:10.3389/fimmu.2025.1618254.
119. Fujii S, Motohashi S, Shimizu K, Nakayama T, Yoshiga Y, Taniguchi M. Adjuvant activity mediated by iNKT cells. Semin Immunol. 2010;22(2):97–102. doi:10.1016/j.smim.2009.10.002.
120. Li YR, Zhou Y, Yu J, Kim YJ, Li M, Lee D, et al. Generation of allogeneic CAR-NKT cells from hematopoietic stem and progenitor cells using a clinically guided culture method. Nat Biotechnol. 2025;43(3):329–44. doi:10.1038/s41587-024-02226-y.
121. UCLA Scientists Develop Novel Technology that Positions 'Off-The-Shelf' Cancer Immunotherapy for the Clinic|UCLA Health [Internet]. [cited 2025 Jul 31]. Available from: https://www.uclahealth.org/news/release/ucla-scientists-develop-novel-technology-positions-shelf.
122. Combating Ovarian Cancer Using Stem Cell-Engineered Off-The-Shelf CAR-iNKT Cells—CIRM [Internet]. [cited 2025 Aug 4]. Available from: https://www.cirm.ca.gov/our-progress/awards/combating-ovarian-cancer-using-stem-cell-engineered-shelf-car-inkt-cells/.
123. Rotolo A, Caputo VS, Holubova M, Baxan N, Dubois O, Chaudhry MS, et al. Enhanced anti-lymphoma activity of CAR19-iNKT cells underpinned by dual CD19 and CD1d targeting. Cancer Cell. 2018;34(4):596–610.e11. doi:10.1016/j.ccell.2018.08.017.
124. Heczey A, Liu D, Tian G, Courtney AN, Wei J, Marinova E, et al. Invariant NKT cells with chimeric antigen receptor provide a novel platform for safe and effective cancer immunotherapy. Blood. 2014;124(18):2824–33. doi:10.1182/blood-2013-11-541235.
125. Poels R, Drent E, Lameris R, Katsarou A, Themeli M, Van Der Vliet HJ, et al. Preclinical Evaluation of invariant natural killer T cells modified with CD38 or BCMA chimeric antigen receptors for multiple myeloma. Int J Mol Sci. 2021;22(3):1096. doi:10.3390/ijms22031096.
126. Jiang ZM, Luo W, Wen Q, Liu SD, Hao PP, Zhou CY, et al. Development of genetically engineered iNKT cells expressing TCRs specific for the M. tuberculosis 38-kDa antigen. J Transl Med. 2015;13(1):141. doi:10.1186/s12967-015-0502-4.
127. De Santo C, Salio M, Masri SH, Lee LYH, Dong T, Speak AO, et al. Invariant NKT cells reduce the immunosuppressive activity of influenza A virus-induced myeloid-derived suppressor cells in mice and humans. J Clin Investig. 2008;118(12):4036–48. doi:10.1172/jci36264.
128. MiNK Therapeutics. MiNK therapeutics reports third quarter 2024 results and business update. New York, NY, USA: MiNK Therapeutics, Inc.; 2024.
129. Ko HJ, Lee JM, Kim YJ, Kim YS, Lee KA, Kang CY. Immunosuppressive myeloid-derived suppressor cells can be converted into immunogenic APCs with the help of activated NKT cells: an alternative cell-based antitumor vaccine. J Immunol. 2009;182(4):1818–28. doi:10.4049/jimmunol.0802430.
130. O'Neal J, Mavers M, Jayasinghe RG, DiPersio JF. Traversing the bench to bedside journey for iNKT cell therapies. Front Immunol. 2024;15:1436968. doi:10.3389/fimmu.2024.1436968.
131. Radboud University Medical Center. First-in-human Phase I Dose Escalation Study Assessing Safety, Tolerability and Preliminary Efficacy of Immunomodulatory Nanoparticles [Internet]. clinicaltrials.gov; 2025 Apr [cited 2025 Oct 23]. Report No.: NCT04751786. Available from: https://clinicaltrials.gov/study/NCT04751786.
132. Home—ClinicalTrials.gov [Internet]. [cited 2025 Aug 9]. Available from: https://clinicaltrials.gov/ct2/home.
133. Zhang M. Phase 1/2 Study of Natural Killer T Cell Infusion in Patients With Advanced Solid Tumor [Internet]. clinicaltrials.gov; 2022 Apr [cited 2025 Oct 23]. Report No.: NCT02562963. Available from: https://clinicaltrials.gov/study/NCT02562963.
134. Memorial Sloan Kettering Cancer Center. A Phase II Study of agenT-797 (Invariant Natural Killer T Cells), Botensilimab, a Novel Fc-enhanced CTLA-4 Inhibitor, Plus Balstilimab (Anti-PD-1) With Ramucirumab and Paclitaxel for Patients With Previously Treated, Advanced Esophageal, Gastric, or Gastro-esophageal Junction

Adenocarcinoma [Internet]. clinicaltrials.gov; 2025 Oct [cited 2025 Oct 23]. Report No.: NCT06251973. Available from: https://clinicaltrials.gov/study/NCT06251973.
135. Guo J, Bao X, Liu F, Guo J, Wu Y, Xiong F, et al. Efficacy of invariant natural killer T cell infusion plus transarterial embolization vs. transarterial embolization alone for hepatocellular carcinoma patients: a phase 2 randomized clinical trial. J Hepatocell Carcinoma. 2023;10:1379–88. doi:10.2147/jhc.s416933.
136. Sugarbaker PH. Prevention and treatment of peritoneal metastases: a comprehensive review. Indian J Surg Oncol. 2019;10(1):3–23. doi:10.1007/s13193-018-0856-1.
137. Quail DF, Joyce JA. Microenvironmental regulation of tumor progression and metastasis. Nat Med. 2013;19(11):1423–37. doi:10.1038/nm.3394.
138. Leary A, Tan D, Ledermann J. Immune checkpoint inhibitors in ovarian cancer: where do we stand? Ther Adv Med Oncol. 2021;13:17588359211039899.
139. Wei J, Li W, Zhang P, Guo F, Liu M. Current trends in sensitizing immune checkpoint inhibitors for cancer treatment. Mol Cancer. 2024;23(1):279. doi:10.1186/s12943-024-02179-5.
140. Dhawan U, Wang SM, Chu YH, Huang GS, Lin YR, Hung YC, et al. Nanochips of tantalum oxide nanodots as artificial-microenvironments for monitoring ovarian cancer progressiveness. Sci Rep. 2016;6(1):31998. doi:10.1038/srep31998.
141. Bu X, Mahoney KM, Freeman GJ. Learning from PD-1 resistance: new combination strategies. Trends Mol Med. 2016;22(6):448–51. doi:10.1016/j.molmed.2016.04.008.
142. Fan Z, Han D, Fan X, Zhao L. Ovarian cancer treatment and natural killer cell-based immunotherapy. Front Immunol. 2023;14:1308143. doi:10.3389/fimmu.2023.1308143.
143. Hou Y, Zhao X, Nie X. Enhancing the therapeutic efficacy of NK cells in the treatment of ovarian cancer (Review). Oncol Rep. 2024;51(3):50.
144. Ahmad GV, Nouri S, Mohammad gholian A, Abdollahi E, Ghorbaninezhad F, Tahmasebi S, et al. Breaking barriers: CAR-NK cell therapy breakthroughs in female-related cancers. Biomed Pharmacother. 2025;187(12):118071. doi:10.1016/j.biopha.2025.118071.
145. Genentech, Inc. A Phase II, Randomized, Blinded, Placebo-Controlled Study of Tiragolumab, An Anti-TIGIT Antibody, in Combination With Atezolizumab in Chemotherapy-Naïve Patients with Locally Advanced or Metastatic Non-Small Cell Lung Cancer [Internet]. clinicaltrials.gov; 2025 Jul [cited 2025 Aug 18]. Report No.: NCT03563716. Available from: https://clinicaltrials.gov/study/NCT03563716.
146. A Study of Tiragolumab in Combination With Atezolizumab in Chemotherapy-Naïve Patients with Locally Advanced or Metastatic Non-Small Cell Lung Cancer—Full Text View—ClinicalTrials.gov [Internet]. [cited 2025 Nov 6]. Available from: https://clinicaltrials.gov/ct2/show/NCT03563716.
147. McNulty R. FDA grants tiragolumab breakthrough therapy designation for PD-L1-high NSCLC. Evid Based Oncol. 2021;27:SP58–SP58.
148. Rodriguez-Abreu D, Johnson ML, Hussein MA, Cobo M, Patel AJ, Secen N. Primary analysis of a randomized, double-blind, phase II study of the anti-TIGIT antibody tiragolumab (tira) plus atezolizumab (atezo) vs. placebo plus atezo as first-line (1L) treatment in patients with PD-L1-selected NSCLC (CITYSCAPE). J Clin Oncol. 2020;38(15 suppl):9503. doi:10.1016/j.annonc.2021.10.217.
149. Cho BC, Abreu DR, Hussein M, Cobo M, Patel AJ, Secen N, et al. Tiragolumab plus atezolizumab vs. placebo plus atezolizumab as a first-line treatment for PD-L1-selected non-small-cell lung cancer (CITYSCAPE): primary and follow-up analyses of a randomised, double-blind, phase 2 study. Lancet Oncol. 2022;23(6):781–92. doi:10.1016/s1470-2045(22)00226-1.
150. Li D-M, Pei K-G, Yu X-Z, Qie M-R, Zhong L, Song L. Advances in PD-1 and CTLA-4 dual-target immunotherapy for ovarian cancer. Front Immunol. 2025;16:1686532. doi:10.3389/fimmu.2025.1686532.
151. Yeku OO, Rao TD, Laster I, Kononenko A, Purdon TJ, Wang P, et al. Bispecific T-cell engaging antibodies against MUC16 demonstrate efficacy against ovarian cancer in monotherapy and in combination With PD-1 and VEGF inhibition. Front Immunol. 2021;12:663379. doi:10.3389/fimmu.2021.663379.
152. Rotte A, Sahasranaman S, Budha N. Targeting TIGIT for immunotherapy of cancer: update on clinical development. Biomedicines. 2021;9(9):1277. doi:10.3390/biomedicines9091277.

153. Ma L, Gai J, Qiao P, Li Y, Li X, Zhu M, et al. A novel bispecific nanobody with PD-L1/TIGIT dual immune checkpoint blockade. Biochem Biophys Res Commun. 2020;531(2):144–51. doi:10.1016/j.bbrc.2020.07.072.
154. O'Brien E, Mayer CM, Arend RC. Exploring T-cell bispecific antibodies in gynecologic malignancy. Gynecol Oncol Rep. 2025;59(14):101772. doi:10.1016/j.gore.2025.101772.
155. Wang J, Cheng X, Jin Y, Xia B, Qin R, Zhang W, et al. Safety and clinical response to combined immunotherapy with autologous iNKT cells and PD-1$^{+}$CD8$^{+}$ T cells in patients failing first-line chemotherapy in stage IV pancreatic cancer. Cancer Res Commun. 2023;3(6):991–1003. doi:10.1158/2767-9764.crc-23-0137.
156. Hadfield MJ, Safran H, Purbhoo MA, Grossman JE, Buell JS, Carneiro BA. Overcoming resistance to programmed cell death protein 1 (PD-1) blockade with allogeneic invariant natural killer T-cells (iNKT). Oncogene. 2024;43(10):758–62. doi:10.1038/s41388-024-02948-y.
157. Delfanti G, Cortesi F, Perini A, Antonini G, Azzimonti L, de Lalla C, et al. TCR-engineered iNKT cells induce robust antitumor response by dual targeting cancer and suppressive myeloid cells. Sci Immunol. 2022;7(74):eabn6563. doi:10.1126/sciimmunol.abn6563.
158. Kim ST. Study to Evaluate the safety and efficacy of sunitinib, in subject with refractory solid tumors [Internet]. clinicaltrials.gov; 2022 Jun [cited 2025 Oct 24]. Report No.: NCT02691793. Available from: https://clinicaltrials.gov/study/NCT02691793.
159. Rong NJ, Liu Y, Fang K, Tan Y, Pan LP, Yan M, et al. Unraveling the potential biomarkers of immune checkpoint inhibitors in advanced ovarian cancer: a comprehensive review. Investig New Drugs. 2024;42(6):728–38. doi:10.1007/s10637-024-01478-4.
160. Xu Y, Zuo F, Wang H, Jing J, He X. The current landscape of predictive and prognostic biomarkers for immune checkpoint blockade in ovarian cancer. Front Immunol. 2022;13:1045957. doi:10.3389/fimmu.2022.1045957.
161. Yang C, Xia BR, Zhang ZC, Zhang YJ, Lou G, Jin WL. Immunotherapy for ovarian cancer: adjuvant, combination, and neoadjuvant. Front Immunol. 2020;11:2595. doi:10.3389/fimmu.2020.577869.