

An Innovative Approach to Immuno Engineering: Nano Vaccines for Cancer Treatment

*Dr Samar Vihal

School of Pharmacy, GNA University, Phagwara-Hoshiarpur Rd, Sri Hargobindgarh, Dhak
Khati, Punjab, 144401, India

Corresponding email: Samarvihhal07@gmail.com

ABSTRACT: Nano vaccines represent a novel class of vaccines that utilize nanoparticles as carriers and immune-stimulating agents that have shown promise in both disease prevention and treatment, particularly in cancer. Their growing appeal in oncology stems from several key benefits, such as enhanced trafficking to lymph nodes, efficient antigen encapsulation and presentation, and the ability to elicit long-lasting anti-tumor immune responses. By employing nanoparticles to provide platforms for the co-delivery of adjuvant and antigen, nano vaccines offer an intriguing answer to these issues. A wide range of nanoparticle platforms have been engineered to deliver antigens and adjuvants specifically to lymphoid tissues and antigen-presenting cells (APCs). Nano vaccines enable precision immune system alteration to generate potent and durable anticancer responses by utilizing nanoscale properties including surface chemistry, changeable size, and targeted administration. This paper provides a comprehensive overview of recent advancements in cancer therapy using nano vaccines. Strategies for tumor antigen loading, lymph node trafficking, dendritic cell targeting, and immune checkpoint synergy the way that nano vaccines overcome the current limitations on immunotherapy are addressed. The clinical potential, regulatory barriers, and translational challenges of cancer immunotherapy based on nano vaccines are discussed in this review. This study aims to bring materials science and immuno-oncology closer together by offering insights into the design principles of next-generation cancer nano vaccines.

KEYWORDS: Cancer immunotherapy; Nano vaccines; Tumor antigens; Translational nanomedicine.

INTRODUCTION

Vaccines are among the most important medical breakthroughs, designed to trigger the immune system by introducing specific antigens. This approach has successfully protected people from deadly illnesses such as smallpox, measles, and whooping cough. Certain highly aggressive and treatment-resistant cancers, known for their high recurrence and metastasis rates, continue to pose significant challenges. Their poor treatment outcomes and substantial costs contribute to growing concern and highlight the urgent need for innovative cancer therapies (1). As a result, researchers have increasingly turned their attention to cancer immunotherapy, now recognized as the fourth primary approach to cancer treatment. Within this field, cancer nano vaccines have appeared as a promising approach, using nanocarriers to transport vaccine components into the body. These systems have shown strong anti-tumor immune responses and have significantly advanced efforts in cancer prevention and therapy (2). The identification of tumor-specific antigens, combined with the development of tailored neoantigen vaccines and immune inhibitors checkpoints capable of reversing tumor-mediated immune exhaustion, has renewed interest in cancer vaccines as a viable strategy in cancer immunotherapy. Cancer

nano vaccines function as active immunotherapeutic agents that utilize exosomes, tumor cells, proteins, peptides, or nucleic acid sequences carrying tumor-specific antigens (TSAs) or tumor-associated antigens (TAAs) to stimulate a targeted immune response aimed at suppressing tumor progression. Numerous challenges remain in the clinical development of cancer nano vaccines, such as the tumor microenvironment's strong immunosuppressive effects, the decreased expression of major histocompatibility complex (MHC) class I (MHC-I) on cancer cells, the incapacity to activate antitumor immunity, and the ineffective activation of antigen-presenting cells (APCs). Nanotechnology breakthroughs have produced nano vaccines with enhanced modulation capabilities in addition to the ability to transcend the limitations of conventional vaccinations (3).

Nanoparticles offer several advantages as vaccine delivery platforms. First, they can promote the release of tumor antigens, enhancing immune activation. Additionally, they allow for the co-delivery of antigens and adjuvants, which facilitates efficient activation of antigen-presenting cells (APCs) while minimizing the risk of immune tolerance associated with uptake by immature APCs. Second, nanoparticles enable more precise spatial and temporal

control of vaccine distribution. By tailoring their physicochemical characteristics such as size, surface charge, and composition their accumulation in lymph nodes can be significantly improved (4). Beyond their role as delivery vehicles, nanoparticles can also stimulate various immune responses and, in some cases, function as adjuvants themselves. In this context, self-adjuvating nano vaccines refer to nanomaterials that possess inherent immunostimulatory properties, potentially eliminating the need for additional adjuvants or reducing their required dose. This review highlights the underlying mechanisms nano vaccines in cancer treatment, examines their status in clinical research, and discusses their benefits and current challenges in advancing cancer immunotherapy.

COMPONENTS OF NANO VACCINES

As an evolution of traditional vaccine platforms, nano vaccines harness nanotechnology to revolutionize immunization approaches. They utilize engineered nanocarriers (NCs) to deliver antigens, adjuvants, or other immune-modulating molecules. These nanocarriers can be precisely designed with specific sizes, shapes, surface characteristics, and material compositions to improve antigen presentation, enhance immunogenicity, and achieve targeted delivery (5). Typically, nano vaccines consist of three key

components: the antigen, the adjuvant, and the nanocarrier itself. Each element contributes critically to the immune process initiating immune recognition of tumor-specific antigens, boosting immune activation, and sustaining the antitumor response collectively acting to strengthen the overall immune defense against cancer (6). The components of cancer nano vaccines are illustrated in Figure 1.

Antigens are a central component of vaccines, responsible for activating the immune system to identify and respond to specific targets. Broadly, cancer antigens are categorized into two types: tumor-associated antigens (TAAs) and tumor-specific antigens (TSAs) (7). Tumor-associated antigens (TAAs) are proteins that can be found on both normal and cancerous cells, though they tend to be present in tumor cells as high amount, making them viable targets for immunotherapy. Despite not being exclusive to cancer, their elevated presence in tumors enables immune recognition. Examples include MART-1, expressed in both melanocytes and melanoma cells; HER2/neu, frequently overexpressed in certain breast cancers; alpha-fetoprotein (AFP) in liver cancer; and carcinoembryonic antigen (CEA) in colorectal cancer. However, because TAAs are also present on healthy cells, they can

sometimes induce immune tolerance or trigger autoimmunity. In contrast, tumor-specific antigens (TSAs) result from somatic mutations unique to cancer cells, making them highly specific targets for immunotherapy. Nano vaccines incorporating TSAs can offer greater precision by targeting these mutation-derived antigens, which reflect the distinct molecular profile of each tumor. Common examples of TSAs include mutated forms of TP53, KRAS, and BRAF genes. Nonetheless, the significant genetic variability among tumors and patients

presents challenges for broad application. This necessitates individualized approaches involving genomic sequencing and computational analysis, which add complexity and increase the cost of vaccine development as illustrated in Table 1. The main distinctions between TAAs and TSAs in terms of their origin, expression, immunogenicity, and clinical usefulness are shown in this table, along with the benefits and drawbacks of each for use in tailored and targeted cancer immunotherapy (8).

Table 1. Comparing tumor-specific antigens (TSAs) and tumor-associated antigens (TAAs) in relation to the development of cancer nano vaccines

Feature	Tumor-Associated Antigens (TAAs)	Tumor-Specific Antigens (TSAs)
Origin	Present on both healthy and cancerous cells	Arise from somatic mutations unique to cancer cells
Expression Pattern	Overexpressed in tumor tissues but also found in normal cells	Exclusively expressed in tumor cells
Examples	MART-1 (melanoma), HER2/neu (breast cancer), AFP (hepatocellular carcinoma), CEA (colorectal cancer)	Mutated TP53, KRAS, BRAF
Immune Response	May result in immune tolerance or off-target effects due to expression in normal tissue	Highly specific immune activation with reduced risk of autoimmunity

Personalization	Broadly applicable across patients	Requires individualized tumor profiling and mutation identification
Advantages	Easier to identify and validate; applicable to larger patient populations	High specificity and precision; minimizes damage to normal tissues
Challenges	Risk of autoimmunity and immune evasion; less specific targeting	High tumor heterogeneity; complex identification; increased cost and technical demand
Use in Nano vaccines	Used to stimulate general antitumor immunity	Enables personalized nano vaccine strategies tailored to patient-specific tumor mutations

Adjuvants represent a critical component of nano vaccine formulations, playing a crucial role in enhancing immune response to antigens. These can be cytokines or toll-like receptor (TLR) agonists, or nanocarrier-based systems like liposomes and polymeric nanoparticles. By stimulating antigen-presenting cells (APCs) and inducing localized inflammation, adjuvants help initiate and amplify immune activation. In nano vaccine platforms, they not only improve antigen presentation but also support the maturation of immune cells, thereby significantly boosting overall vaccine effectiveness (9).

The third essential element of nano vaccines is the use of nanocarriers (NCs),

which function as nanocarrier delivery systems for encapsulating antigens as well as adjuvants. These carriers protect the payload from premature degradation, enable sustained controlled release, and enhance targeted delivery to specific cells or tissues. Constructed from materials such as lipids, polymers, or inorganic substances, NCs offer distinct advantages in terms of biocompatibility, structural stability, and tunability. Their design can be optimized with surface modifications to target immune cell receptors or concocted to respond to specific stimuli within the microenvironment of tumor (TME), allowing for on-demand release. As multifunctional platforms, NCs provide precise control over the kinetics of antigen

and adjuvant release, ultimately contributing to improved therapeutic outcomes of nano vaccine-based cancer treatments (10).

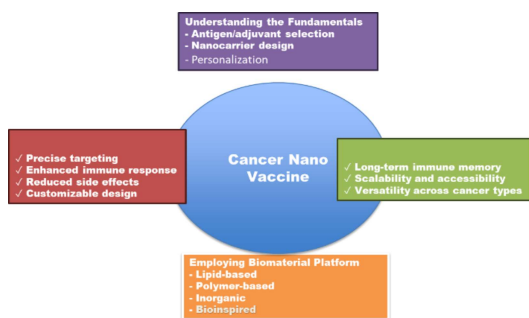


Figure 1. Components of cancer nano vaccines.

DECIPHERING THE IMMUNOMODULATORY MECHANISMS OF NANO VACCINES IN CANCER DISSEMINATION PATHWAYS

Cancer metastasis is a complex process that typically involves local invasion, entry of cancer cells into the circulation (intravasation), exit into distant tissues (extravasation), formation of micro metastases, and subsequent proliferation at secondary sites. Broadly, tumor progression can be divided into three key phases: growth of the primary tumor; dissemination via circulating tumor cells (CTCs); and metastatic colonization, which

includes the development of pre-metastatic niches and the establishment of disseminated tumor cells at distant organs. CTCs spread mainly through two pathways blood circulation and lymphatic vessels eventually reaching the bloodstream. Notably, cancer cells often migrate to nearby lymph nodes via lymphatic routes, where they may evade immune detection and support the development of secondary tumor sites. The metastatic cascade is further complicated by tumor-induced local and systemic immune suppression. This immunosuppressive environment is largely shaped by components such as regulatory T cells (Tregs), myeloid-derived suppressor cells, tumor-associated macrophages, and neutrophils, all of which help tumors escape immune surveillance as illustrated in Figure 2.

Nano vaccines represent a promising approach to modulate signaling pathways involved in metastasis. They have been shown to inhibit pro-metastatic signals, including hypoxia-inducible factors (HIFs), transforming growth factor- β (TGF- β), and the Wnt/ β -catenin axis, while activating immune-stimulatory pathways like cGAS STING and SHP-1/JAK2/STAT3. These signaling modulations can influence key factors such as epithelial–mesenchymal transition (EMT), cancer cell stemness, hypoxic tumor microenvironments, and

immune activation ultimately affecting the metastatic potential of tumors both *In vitro* and *In vivo*. Overall, disrupting metastatic cascade such as those involving hypoxia-inducible factors (HIFs) or transforming growth factor- β (TGF- β) can interfere with successful metastasis. This understanding has driven extensive research and clinical trials focused on enhancing immune responses to prevent and combat tumor spread (11).

The cancer vaccine response involves a series of five key steps: targeted antigen delivery to lymph nodes (LNs), co-delivery of antigen and adjuvant, precise antigen recognition, internalization and release of the antigen, and finally, antigen cross-presentation to activate T-cell responses. A variety of immune cells are involved throughout this process, including macrophages, dendritic cells (DCs), B cells, natural killer (NK) cells, as well as CD4⁺ and CD8⁺ T cells. Tumor vaccines initiate immune activation by directing both antigens and adjuvants to antigen-presenting cells (APCs). APCs particularly macrophages and dendritic cells serve as critical mediators of both innate and adaptive immunity. Due to their central role in modulating immune activation and tolerance, DCs are pivotal in the design of nano vaccines and are often considered the cornerstone of immune regulation. Upon

immunosuppressive elements or associated signaling pathways at any stage of the

administration of a nano vaccine, APCs, especially DCs, are the primary cells responsible for capturing its components. The adjuvants stimulate these cells, which then present the processed antigens on their surfaces using major histocompatibility complex class I and II (MHC I/II) molecules. Once activated, these APCs migrate to secondary lymphoid tissues like the spleen and lymph nodes, where they undergo maturation and initiate adaptive immune responses. This leads to the activation of T and B lymphocytes. CD8⁺ T cells, when activated via MHC-bound peptides, can directly eliminate tumor cells. In contrast, B cells require support from CD4⁺ T cells to become fully functional, producing antibodies that contribute to tumor cell destruction through antibody-mediated mechanisms. Cancer vaccines also promote the differentiation of naïve CD8⁺ T cells into cytotoxic T lymphocytes (CTLs), and naïve CD4⁺ T cells into T helper type 1 (Th1) or type 2 (Th2) subsets. Th1 cells release cytokines such as interleukin-2 (IL-2) and interferon-gamma (IFN- γ) to stimulate CTL activity and enhance cellular immune responses. Th2 cells, on the other hand, support B cell activation through CD40–CD40L signaling. Altogether, these mechanisms

illustrate how the immunosuppressive tumor microenvironment (TME) plays a major role in facilitating metastasis.

metastasis and reprogram the immune landscape toward tumor elimination (12).

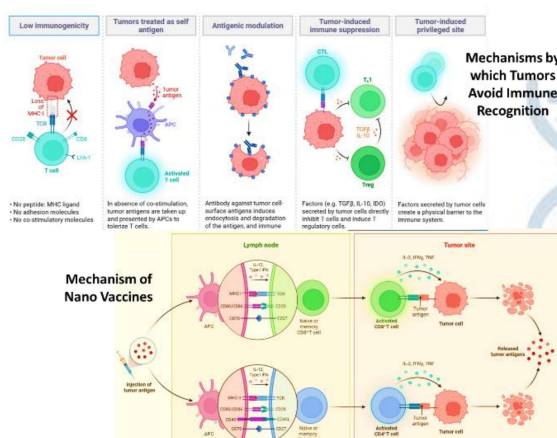


Figure 2. Mechanisms of tumor immune evasion and activation by cancer vaccines. (Top) Tumors escape immune recognition through mechanisms such as low immunogenicity, antigenic modulation, immune suppression, and physical exclusion from immune cells. (Bottom) Cancer vaccines enhance antigen presentation by dendritic cells (APCs) in lymph nodes, leading to activation of CD8⁺ and CD4⁺ T cells, which migrate to tumor sites and mediate tumor cell killing via cytokine secretion and direct cytotoxicity.

RECENT WORK DONE ON CANCER NANO VACCINES

Tumor cell-derived nano vaccines utilize components sourced from tumor cells,

Targeting this immunosuppressive environment through cancer vaccines may represent a promising strategy to counteract

offering the significant advantage of tumor-associated antigens. Despite this benefit, their therapeutic effectiveness and clinical translation are often hindered by complex fabrication processes, the safety concerns associated with external adjuvants, and suboptimal targeting of lymph nodes (LNs) and uptake by dendritic cells (DCs). In this work, scientist addressed these limitations by introducing the endoplasmic reticulum (ER) stress-inducing agent α -mangostin (α M) into tumor cells using poly(d,l-lactide-co-glycolide) (PLGA) nanoparticles. This approach led to the generation of self-assembled, tumor cell-derived nano vaccines (α M-Exos) via the natural exocytosis of nanoparticles through tumor-derived exosomes (TEXs). These α M-Exos not only displayed a wide range of tumor antigens but also carried elevated levels of 70 kDa heat shock proteins (Hsp70s), which were upregulated as a result of ER stress. Hsp70s served dual roles as intrinsic adjuvants and enhancers of LN accumulation and DC uptake. Upon subcutaneous administration, α M-Exos showed efficient LN migration and rapid DC internalization, enabling synchronized delivery of tumor antigens and adjuvants. This led to potent activation of antitumor

immunity and the establishment of durable immune memory. These findings highlight a robust, self-assembled tumor cell-derived nano vaccine platform, offering a multifunctional and promising strategy for effective cancer immunotherapy (13).

Tumor tissues and cells serve as valuable sources of antigens for developing cancer vaccines. However, incorporating antigens into vaccines and ensuring their efficient delivery to antigen-presenting cells (APCs) remains a significant hurdle, primarily due to their poor solubility. To address this issue, this study employs 8 M urea to solubilize the water-insoluble components of tumor tissues and cells. These solubilized materials are then reassembled into nanoscale structures, forming nano vaccines derived from the entire contents of micrometre-sized tumor cells. To enhance immune activation, a diverse set of antigens both water-soluble and water-insoluble are integrated into and onto the nano vaccine structures. This dual encapsulation strategy significantly improves uptake by APCs and elicits stronger therapeutic responses in models of melanoma and breast cancer, compared to nano vaccines containing only soluble components. Moreover, combining the nano vaccines with anti-PD-1 antibodies and metformin further boosts their efficacy. In prophylactic models, the nano vaccines demonstrated complete

protection against lung cancer and a 70% prevention rate in melanoma-bearing mice. Immunological analyses reveal that the vaccines stimulate tumor-specific T cells and activate both innate and adaptive immune responses within the tumor microenvironment. In summary, this research introduces a broadly applicable approach for creating nano vaccines from whole tumor cells, offering a promising strategy for cancer immunotherapy and prevention (14).

The induction of tumor-specific T cells plays a vital role in effective cancer immunotherapy. However, one of the main challenges is coordinating the timely and spatially precise activation of antigen cross-presentation alongside innate immune stimulation in antigen-presenting cells (APCs). In this study, Luo et al., present a streamlined nano vaccine composed of a straightforward physical blend of a tumor antigen and a synthetic polymer nanoparticle known as PC7A NP. This formulation elicits a robust cytotoxic T-cell response with minimal systemic cytokine production. PC7A NP facilitates efficient cytosolic delivery of tumor antigens to APCs located in draining lymph nodes, thereby enhancing antigen presentation on the cell surface. Concurrently, it activates genes stimulated by type I interferons. This immune activation relies on the stimulator

of interferon genes (STING) pathway but is independent of the Toll-like receptor and MAVS (mitochondrial antiviral-signaling) pathways. In preclinical models of melanoma, colorectal cancer, and HPV E6/E7-related tumors, the nano vaccine significantly suppressed tumor growth. When combined with anti-PD-1 checkpoint blockade, the treatment demonstrated strong synergy, achieving 100% survival over 60 days in the TC-1 tumor model. Upon rechallenge with TC-1 cells, previously cured animals exhibited complete resistance to tumor growth, indicating the establishment of durable antitumor immune memory. Overall, this STING-activating nano vaccine presents a straightforward, effective, and low-toxicity approach for enhancing antitumor immune responses, offering considerable promise for future cancer immunotherapy applications (15).

Harnessing the innate immune system through nano vaccines offers a powerful approach to eliciting tumor-specific immune responses, making it a promising avenue in cancer immunotherapy. However, conventional vaccine strategies often require the co-delivery of antigens and adjuvants, a process that can be technically challenging and difficult to scale for widespread use.

To overcome these limitations, researcher developed a tumor-targeted nano vaccine platform that incorporates adjuvant activity directly into the delivery system. This was achieved using branched polyguanidine (PolyGu)-based nano vaccines, created by chemically modifying polyethylenimine (PEI) with various guanidine groups. This modification repurposes the inherent cytotoxicity of PEI into a beneficial innate immune-stimulating effect. Among these, the nano vaccine constructed with poly(phenyl biguanidine) (Poly-PBG) demonstrated strong immunostimulatory effects. It effectively activated dendritic cells and promoted their maturation through both TLR4 and NLRP3 signaling pathways, resulting in enhanced immune responses in vivo. The PolyGu-based nano vaccines significantly suppressed tumor progression and improved survival in mouse models. Additionally, this platform shows great potential for enhancing mRNA-based nano vaccine designs, opening opportunities for developing personalized cancer immunotherapies. Overall, this study presents a novel self-adjuvating nano vaccine strategy, moving the field closer to practical and scalable cancer immunotherapy solutions (16).

Although the simultaneous delivery of DNA, RNA, and peptide therapeutics holds significant promise for cancer theranostic,

effective platforms for such co-delivery remain limited. In this study, researcher introduce a novel nano vaccine system composed of self-assembled intertwining DNA-RNA nano capsules (iDR-NCs), designed to co-deliver immunostimulatory DNA CpG motifs, short hairpin RNA (shRNA), and tumor-specific peptide neoantigens directly to antigen-presenting cells (APCs) within lymph nodes. The fabrication of these nano capsules involved several steps: (1) tandem production of CpG DNA and shRNA using rolling circle replication and transcription simultaneously, (2) assembly of these products into DNA-RNA microflower structures, (3) compression of the microflowers into nanoscale iDR-NCs using PEGylated cationic polypeptides, and (4) encapsulation of peptide neoantigens within the nano capsules. The co-delivered CpG and shRNA function synergistically to enhance APC activation and prolong antigen presentation. When loaded with neoantigens, the iDR-NCs substantially increased the frequency of neoantigen-specific CD8⁺ T cells—by approximately eightfold compared to CpG alone promoted T cell memory formation and significantly suppressed the growth of neoantigen-expressing colorectal tumors. In summary, iDR-NCs represent a promising platform for the triple delivery of DNA, RNA, and peptide components, offering a powerful

and synergistic approach for cancer immunotherapy. In this work, scientist developed a novel class of nucleic acid-based nanomaterials, termed intertwining DNA-RNA nano capsules (iDR-NCs), designed through a one-pot reaction system that combines DNA and RNA synthesis. These nano capsules were subsequently used to construct immunotherapeutic nano vaccines by incorporating tumor-specific neoantigens. To achieve this, we optimized rolling circle replication (RCR) and rolling circle transcription (RCT) to occur simultaneously in a single solution, enabling the concurrent production of tandem DNA and RNA strands. These strands then self-assembled into DNA-RNA microflower structures (MFs). Unlike prior strategies that used RCR or RCT separately for fabricating nucleic acid nanostructures, this represents the first demonstration of a hybrid DNA-RNA nanostructure formed through the integration of both processes in a single system. This approach allows for the combination of DNA and RNA-based immune modulators within one nanomedicine. Zhu et al., designed these MFs to include immunostimulatory CpG DNA and Stat3-targeting short hairpin RNA (shRNA), aiming to co-activate TLR9 and STAT3 pathways within antigen-presenting cells (APCs) to promote a strong immune response. For enhanced lymph

node targeting and safety, we employed a biocompatible PEG-grafted polypeptide (PPT-g-PEG) to condense the MFs into nanoscale capsules (NCs). While polyethylenimine (PEI) is also known to compact similar nanostructures, its high cytotoxicity makes it less favourable for clinical use. The PPT-g-PEG copolymer not only improves biocompatibility but also facilitates the encapsulation of tumor neoantigens via hydrophobic interactions, taking advantage of the typically hydrophobic nature of these peptides. Additionally, hydrophilic neoantigens could be incorporated through chemical conjugation to the polymer. Importantly, this polymer is designed to shed its PEG coating in the acidic endolysosomal environment of APCs, thereby enhancing the proton sponge effect of the polypeptide backbone and improving endosomal escape and cytosolic delivery of the therapeutic payloads. Advanced imaging techniques and flow cytometry confirmed that iDR-NCs carrying tumor neoantigens successfully accumulated in lymph nodes and were internalized by APCs. In vitro assays demonstrated that the CpG and Stat3 shRNA components synergistically stimulated APC activation.

Although the *In vivo* synergy between CpG and shRNA has yet to be fully explored, our nano vaccines loaded with the Adpgk

neoantigen elicited robust neoantigen-specific CD8⁺ T cell responses and effectively suppressed tumor progression. Given the rarity of naturally occurring neoantigen-specific cytotoxic T lymphocytes (CTLs) in cancers such as colorectal cancer, enhancing these responses through exogenous delivery is a critical goal in cancer immunotherapy. Our results suggest that the combination of synergistic adjuvants and peptide neoantigens within iDR-NCs holds great promise for this purpose. Notably, our iDR-NC/Adpgk formulation induced potent antitumor T cell responses and showed therapeutic benefit in models resembling metastatic disease—a particularly challenging context in oncology. While complete tumor regression was not achieved with the nanovaccine alone, these findings support the potential of combining iDR-NCs with additional modalities such as immune checkpoint inhibitors, multi-epitope vaccines, cytokine therapies, or chemotherapy. For instance, increased PD-1 expression observed on CD8⁺ T cells post-vaccination suggests a potential for synergy with PD-1 blockade to overcome T cell exhaustion and further improve outcomes. Moreover, the modular nature of the iDR-NC platform allows for loading of multiple antigens, potentially broadening the T cell response and enhancing tumor eradication across heterogeneous cell

populations. In conclusion, this study introduces a versatile and generalizable strategy for constructing hybrid DNA-RNA nanocarriers capable of co-delivering nucleic acid adjuvants and peptide neoantigens. The iDR-NC platform, particularly when using CpG and Stat3 shRNA in combination with PPT-g-PEG condensation and neoantigen encapsulation, represents a promising avenue for developing personalized and combinatorial cancer immunotherapies (17).

Metastasis and tumor recurrence following surgical removal remain major challenges in effective cancer treatment. In this study, researcher presented a broadly applicable approach to develop personalized nano vaccines using a cationic fluoropolymer for post-operative cancer immunotherapy. These nano vaccines are created by simply combining the fluoropolymer with a model antigen, such as ovalbumin, resulting in nanoparticles that stimulate dendritic cell maturation through Toll-like receptor 4 (TLR4) signaling. They also enhance cytosolic delivery of the antigen, thereby promoting efficient cross-presentation. This nano vaccine formulation successfully suppressed the growth of established B16-OVA melanoma tumors. More significantly, when the fluoropolymer was mixed with cell membranes derived from a

patient's own resected primary tumor, and used in combination with immune checkpoint blockade therapy, it effectively reduced tumor recurrence and metastasis in two subcutaneous cancer models and an orthotopic breast cancer model. In the orthotopic setting, the treatment also generated durable immune memory capable of rejecting tumor rechallenge. Overall, this work introduces a straightforward and versatile platform for creating personalized nano vaccines that may help prevent cancer recurrence and metastatic spread after surgery. This study presents a novel approach for developing tumor nano vaccines using fluoroalkane-modified cationic polymers, termed F-PEIs. These polymers can spontaneously self-assemble into nanoparticles upon simple mixing with protein antigens or tumor cell membrane-derived antigens, resulting in uniformly sized formulations. Notably, nano vaccines based on these fluoropolymers demonstrated enhanced antitumor immune responses compared to traditional adjuvants like CpG and alum. This improvement is likely due to the nanoparticles' capability for cytosolic delivery of antigens, promoting efficient antigen cross-presentation, along with their capacity to activate dendritic cells by engaging pathways such as TLR4 signaling. The distinctive chemical nature of fluoroalkane chains exhibiting both hydrophobic and

lipophobic characteristics makes F-PEI a unique dual-function material, serving as both an antigen carrier and an immune-stimulating component. Additionally, the study found that immune responses initiated by these fluoropolymer-based nano vaccines could be significantly enhanced when combined with immune checkpoint blockade (ICB) therapy. Furthermore, leveraging the fluoropolymer's ability to encapsulate tumor cell membranes, personalized cancer vaccines were created by combining F13-PEI with membranes derived from surgically resected tumors. In preclinical models including two subcutaneous tumor models and one orthotopic tumor model with spontaneous metastasis these F13-PEI/Mem nano vaccines, administered alongside ICB therapy, effectively treated metastatic cancers that could not be fully addressed by surgery alone (18).

Chemotherapeutic agents can enhance the effectiveness of cancer immunotherapy by promoting the release of damage-associated molecular patterns (DAMPs) and inducing mutations that give rise to neoantigens, thereby increasing the immunogenic potential of tumors. However, their clinical utility is often restricted by dose-dependent toxicity and inherently low immunogenicity, which limits their ability to initiate a strong antitumor immune

response. To address this, scientist proposed that exposing cancer cells in vitro to extremely high concentrations of different chemotherapeutic drugs could artificially amplify the expression and diversity of both DAMPs and neoantigens, thus boosting their immunogenic profile. Based on this hypothesis, Scientist developed in vitro chemotherapy-induced (IVCI) nano vaccines using lysates derived from these treated cancer cells. These nano vaccines encapsulate a wide array of tumor-associated neoantigens and DAMPs, offering both broad antigenicity and intrinsic immunostimulatory capacity. In preclinical studies using CT26 tumor-bearing mice, the IVCI nano vaccines significantly enhanced immune activation, particularly when combined with immune checkpoint blockade therapy, leading to increased infiltration of CD4⁺ and CD8⁺ T cells within the tumor microenvironment. This strategy introduces a novel approach for artificially boosting tumor immunogenicity, offering a promising pathway for designing more effective nano vaccine-based chemoimmunotherapies (19).

Tumor vaccines have emerged as a major focus in cancer immunotherapy, aiming to harness the body's immune system by introducing tumor-associated antigens to trigger targeted immune responses against

malignant cells. In this study, a straightforward nano vaccine formulation was created to generate antigen-specific antitumor immunity. This nano vaccine employed polyethylenimine (PEI), a polycationic carrier, to co-deliver the model antigen ovalbumin (OVA) along with the immunostimulatory CpG oligonucleotide through electrostatic interactions. *In vitro* experiments demonstrated that the nano vaccine effectively stimulated DC maturation. When applied *In vivo*, the nano vaccine exhibited antitumor activity, which was further improved by co-administration with hyaluronidase (HAase). HAase increased the permeability of tumor tissues by degrading hyaluronic acid (HA) within the extracellular matrix (ECM), thereby enhancing the infiltration of vaccine-induced, tumor-specific T cells. The combined use of the PEI-based nano vaccine and HAase led to a pronounced therapeutic benefit, largely due to a greater accumulation of OVA-specific cytotoxic T lymphocytes within tumors. This approach offers a practical and effective strategy for enhancing antitumor immune responses in cancer immunotherapy (20).

Cancer can adapt mechanisms to escape immune surveillance, making immunotherapy a powerful strategy to re-engage the immune system in targeting tumors. Among various immune-

stimulating agents, unmethylated CpG-containing oligonucleotides are known for their ability to activate Toll-like receptor 9 (TLR9) within the endolysosomal compartments of antigen-presenting cells (APCs), making them attractive adjuvants for cancer treatment. Despite their promise, clinical use of synthetic CpG faces limitations including poor delivery efficiency to APCs, rapid degradation in the body, short half-life, and systemic side effects due to unintended distribution. To address these limitations, this study introduces DNA-inorganic hybrid nanovaccines (hNVs), constructed through the self-assembly of CpG concatemers and magnesium pyrophosphate (Mg_2PPI). This inorganic component provides structural stability, making the hNVs resistant to enzymatic breakdown and heat, key features for maintaining vaccine integrity during storage and administration. Once administered, fluorescently labelled hNVs were shown to accumulate efficiently within the endo lysosomes of APCs. There, the acidic environment dissolves Mg_2PPI , releasing CpG analog to initiate immune activation. The result was enhanced production of inflammatory cytokines and upregulation of co-stimulatory molecules in APCs. Compared to free CpG, the hNVs demonstrated extended retention at the tumor site and minimized adverse effects such as spleen enlargement. In a murine

melanoma model, just two doses of hNVs significantly suppressed tumor progression and outperformed free CpG treatment. These findings suggest that hNVs offer a robust and stable delivery system with strong immunotherapeutic potential for cancer treatment (21).

Recent research has increasingly recognized the gut microbiome as a key player in regulating cancer development and response to treatment. In particular, the microbial community residing in the gastrointestinal tract significantly contributes to the tumor microenvironment, especially in colorectal cancers. Emerging evidence highlights the potential of gut microbes particularly probiotics to boost the effectiveness of cancer immunotherapy. Bioactive molecules secreted by probiotics, such as lipopolysaccharide (LPS) and cyclic GMP-AMP (cGAMP), can engage pattern recognition receptors in the gut, triggering innate immune responses. This activation promotes the release of type I interferons, enhances dendritic cell (DC) recruitment and maturation, and subsequently stimulates both T cells and natural killer (NK) cells. As a result, probiotics may function as natural immune adjuvants that influence tumor progression and immune activation. Among various immunotherapy strategies, cancer nano vaccines especially those derived from

whole tumor components are effective in stimulating tumor antigen-specific T cell responses, which are crucial for targeting and eliminating cancer cells. However, the activation of these T cells typically requires days to weeks, presenting a window during which immune modulation by agents like probiotics may enhance the response. Studies have shown that strains such as *Lactobacillus rhamnosus* (LGG) and *Bifidobacterium longum* exert anti-inflammatory, anti-cancer, and anti-metastatic effects, in part by improving dendritic cell function and enhancing T cell-mediated immunity. Moreover, *B. longum* has been shown to reduce intestinal inflammation without impairing CTLA-4-based checkpoint blockade activity. Despite these promising features, directly applying probiotics, especially via oral delivery, presents several challenges. Biological barriers such as gastric acid can inactivate probiotics, while mechanical barriers like rapid gastrointestinal motility hinder their retention and colonization in the gut. To address these issues, surface modification techniques using materials like alginate, chitosan, polydopamine, and lipids have been explored to protect probiotics during transit. However, such protective coatings can sometimes compromise bacterial viability, reducing their therapeutic potential.

Therefore, improving probiotic delivery while preserving their activity and safety remains a critical hurdle in developing microbial-based cancer therapies. In this context, this study introduced a novel approach using a self-assembled biofilm-like coating to encapsulate probiotics, enhancing their stability, viability, and ability to withstand the harsh gastrointestinal environment. This formulation not only improves oral delivery and survival of the bacteria but also supports their effective colonization within the tumor microenvironment functioning as in-situ cancer vaccines. When combined with cancer nano vaccines prepared from tumor-derived components, the probiotic formulation significantly amplified both the preventive and therapeutic outcomes. This synergistic strategy boosts both innate and adaptive immune responses. Specifically, probiotics aid in the early stages of immune activation, facilitating more efficient T cell responses triggered by the nano vaccine. Importantly, this combined treatment approach also offers superior biocompatibility and safety compared to some clinical combination therapies currently in use.

Scientists enhanced the probiotic formulation by coating it with a lipid membrane, which improved its ability to colonize and survive in the gut. In mouse

models of colorectal cancer including both subcutaneous transplant models and chemically induced orthotopic colon cancer models this formulation demonstrated strong preventive and therapeutic effects on its own. When used in conjunction with cancer nano vaccines, the treatment showed markedly improved efficacy, whether administered orally or via intratumoral injection. In particular, the formulation was effective in preventing tumor development in chemically induced colon cancer models, where it reduced the number of tumors and amplified the vaccine's effect by increasing tumor-specific T cell populations. Additionally, the probiotic treatment altered the gut microenvironment, promoting the presence of goblet cells and upregulating protective proteins such as claudin and occludin, which may help fortify the intestinal barrier against tumor invasion. In summary, this study presents a promising probiotic-based formulation that can serve both as a standalone cancer preventive or therapeutic agent and as an immunological adjuvant to enhance the performance of cancer nanovaccines. The results demonstrate that this combined approach effectively mobilizes the immune system and improves treatment outcomes in colorectal cancer models (22).

Residual cancer stem-like cells (CSCs) often persist in the tissue surrounding a

surgically removed tumor, posing a significant threat for relapse and reducing the likelihood of a complete cure. At present, effective methods to monitor and eliminate these CSCs are lacking. To address this, we introduce a novel nano vaccine platform termed NICER, which utilizes a nanovesicle-based system designed to target both CSCs and bulk tumor cells simultaneously. This system incorporates CSC-specific antigens and an epigenetic nano-modulator within vesicles functionalized with a dendritic cell-targeting aptamer. The nanovesicles, sourced from tumors with high aldehyde dehydrogenase expression, inherently carry CSC-specific as well as tumor-associated antigens, enabling broad immunogenic coverage. Meanwhile, the encapsulated epigenetic modulator, directed at YTH N6-methyladenosine RNA binding protein 1, functions to suppress protease activity in dendritic cell lysosomes. This modulation enhances antigen cross-presentation through the MHC class I pathway, promoting a stronger immune response. In summary, the NICER vaccine offers a promising and comprehensive immunotherapy approach aimed at eliminating both CSCs and non-stem tumor cells, potentially preventing metastasis and improving long-term survival outcomes (23).

Cytomembrane-based cancer nano vaccines offer a promising strategy to trigger tumor-specific immune responses. However, many existing nano vaccine designs lack a thorough investigation into the mechanisms responsible for cancer prevention, treatment, and the formation of long-lasting immune memory, leaving their long-term therapeutic potential unclear. In this study, we developed a biomimetic nano vaccine platform, termed CCMP@R837, composed of poly(lactic-co-glycolic acid) (PLGA) nanoparticles encapsulating the immune-stimulating agent imiquimod (R837), and cloaked with cancer cell membranes (CCM) to serve as tumor-specific antigens. Notably, repeated immunization with CCMP@R837 in BALB/c mice established durable immune memory. Mice treated with this formulation showed reduced tumor progression and extended survival, with 75% surviving beyond 50 days post-tumor induction. This durable immunity was linked to increased infiltration of CD8⁺ cytotoxic T cells, reduced numbers of regulatory T cells in tumor tissues, and elevated levels of effector memory T cells in the spleen. Overall, CCMP@R837 represents a promising preventive cancer vaccine candidate with the potential for clinical application (24).

Senescent cancer cells possess strong immunogenic properties, which have been explored to trigger antitumor immune responses and support cancer treatment strategies. However, their direct use as live-cell vaccines is hindered by the immunosuppressive nature of the senescence-associated secretory phenotype (SASP) and their residual tumor-promoting activity. To address these challenges, this study presents a nano vaccine derived from senescent cancer cells that exhibits enhanced immunotherapeutic potential. The vaccine is constructed by coating an immunostimulatory nano adjuvant with membranes from senescent cancer cells, creating a biomimetic system that significantly outperforms live senescent cells in promoting dendritic cell (DC) uptake, improving targeting to lymph nodes, and boosting immune activation. Compared to nano vaccines based on tumor cells undergoing immunogenic cell death, the senescent cell-based formulation more effectively drives DC maturation, generates stronger antitumor immunity, and delivers improved therapeutic benefits in melanoma-bearing mice especially when used alongside α PD-1 therapy with reduced adverse effects. This work introduces a flexible strategy for harnessing the immunogenic benefits of senescent tumor cells while minimizing associated drawbacks, thereby advancing the

development of next-generation biomimetic nano vaccines for cancer immunotherapy (25).

Breast cancer remains the most frequently diagnosed cancer worldwide, with surgical removal being the primary treatment option. Advances in surgical techniques have shifted the focus toward minimally invasive and breast-conserving procedures. However, breast-conserving surgeries often carry a heightened risk of tumor recurrence, presenting a significant clinical challenge. To address this, the present study introduces a single-dose, injectable nano-in-gel vaccine (NIGel-Vax) designed for use after breast cancer surgery. This vaccine is formulated by combining protein antigens with a PEI-4BI_{mi}-Man adjuvant, which is then encapsulated within a hydrogel matrix composed of oxidized dextran (ODEX) and 4-arm PEG-ONH₂. When tumor lysate proteins from 4T1 cells were used as antigens, the NIGel-Vax achieved a 92% tumor inhibition rate and a 33% complete cure rate in a postoperative 4T1 mouse model. Moreover, when the tumor-associated antigen TROP2 protein was employed, the vaccine showed a 96% tumor suppression rate and a 50% cure rate in triple-negative breast cancer (TNBC) models. These findings highlight a promising strategy for improving

postoperative outcomes in breast cancer patients (26).

Tumor vaccines have generally shown limited clinical efficacy, largely due to challenges such as suboptimal dendritic cell (DC) targeting, poor cross-presentation of antigens, immunosuppressive signaling within DCs, and the presence of an inhibitory tumor microenvironment (TME). To overcome these obstacles, a novel approach involving nanovaccines with dual targeting of both draining lymph nodes (DLNs) and tumors was developed. This strategy utilized newly synthesized heterocyclic lipidoid and polyester materials to enable dual-targeted immunotherapy. Additionally, oligo hyaluronic acid (HA) and DMG-PEG2000-Mannose were incorporated to formulate nanovaccines capable of delivering STAT3 siRNA and model antigens. These nanocarriers were engineered for efficient cytoplasmic delivery and targeted accumulation in DLNs and tumor sites.

The dual-targeted nano vaccines significantly enhanced antigen presentation and DC maturation, activated the STING pathway, promoted pro-apoptotic activity, and elicited potent antitumor immune responses. They also mitigated the immunosuppressive nature of the TME by reducing inhibitory immune cell populations and reprogramming tumor-

associated neutrophils from the pro-tumorigenic N2 phenotype to the antitumor N1 phenotype. Among the four nanovaccine formulations tested, the MALO@HBNS constructed from heterocyclic lipidoid and polyester—demonstrated superior therapeutic performance. When combined with anti-PD-L1 therapy, MALO@HBNS exhibited a markedly enhanced anticancer effect. This study presents a promising dual-targeted nanovaccine platform with strong potential for integration into combination cancer immunotherapies involving immune checkpoint blockade (27).

CLINICAL TRIALS

The summary of the cancer nano vaccines in clinical trials are enlisted in Table 2. In Phase I clinical trials, researchers primarily focus on evaluating the safety and determining appropriate dosage levels of experimental nano vaccines. These early-stage studies are critical for identifying potential side effects and establishing the maximum tolerated dose. The main objective is to create a safe foundation for later trial phases, which will assess the vaccine's effectiveness and broader clinical benefits in larger patient groups. One ongoing Phase I trial (NCT05714748) is investigating the safety and preliminary efficacy of a lipid nanoparticle (LNP)-encapsulated mRNA vaccine designed to

treat Epstein–Barr virus (EBV)-positive advanced cancers. This study represents a key step in developing a novel therapeutic vaccine with independently held intellectual property. This clinical trial is recruiting participants and clinical trial number is NCT05714748 (28).

Another active Phase I study NCT05475106 is testing the safety and potential efficacy of customized neoantigen peptide vaccines combined with Leukine (Sargramostim) in patients with various cancers. Participants receive intradermal injections of personalized multi-peptide vaccines alongside Leukine, following a structured schedule over several weeks. The study is ethically approved and aligned with the principles outlined in the Declaration of Helsinki. This clinical trial is recruiting participants (29).

In addition, RNA lipid particle (RNA-LP) vaccines are being assessed in a Phase I trial (NCT04573140) targeting newly diagnosed glioblastoma (GBM) in adults. Standard treatments like surgery and chemoradiation are followed by personalized immunotherapy. Tumor samples are collected to extract RNA, which is then formulated into liposomal vaccines. Vaccination begins four weeks post-radiation, with patients receiving three biweekly doses, followed by monthly injections for up to 14 months. Clinical

monitoring, including MRI and health assessments, continues for up to two years, with more frequent evaluations during the first year (30).

Phase II trials shift the focus to evaluating the efficacy of nanovaccines in larger patient groups. These studies aim to determine how long immune responses last, assess the impact on disease progression, and further analyze safety profiles. They also explore how vaccines might work in conjunction with other therapies like chemotherapy, radiation, or other targeted treatments. A Phase I/II trial (NCT00952692) examined the safety and immune response generated by a therapeutic combination of the immune agent dHER2+AS15 ASCI with lapatinib in patients with metastatic breast cancer that overexpresses the HER2 protein and shows resistance to trastuzumab. The immune agent comprises a truncated HER2 protein (dHER2) and an adjuvant system (AS15), which includes monophosphoryl lipid A (MPL), CpG oligodeoxynucleotides (ODN), and QS21 components designed to stimulate the immune system. Administered intramuscularly every two weeks in cycles, the vaccine was given alongside daily oral doses of lapatinib, a dual inhibitor of HER1 and HER2 (31).

Another Phase II trial (NCT00828009) is evaluating the safety of combining the

BLP25 liposomal vaccine (tecemotide) with bevacizumab after chemoradiotherapy in patients with stage IIIA or IIIB non-small cell lung cancer (NSCLC). This approach investigates the potential synergy between immune activation through vaccination and anti-angiogenic therapy via bevacizumab, aiming to prolong survival and delay disease progression. The Phase II trial NCT00828009 (E6508) evaluating the combination of the BLP25 (tecemotide) liposomal vaccine with bevacizumab after chemoradiation in unresectable Stage IIIA–IIIB NSCLC found the regimen to be well tolerated, with no new safety concerns reported. While this pilot study focused primarily on safety and side effects, activity signals were observed, such as instances of stable disease, but no definitive conclusions on efficacy (e.g., progression-free or overall survival benefits) have been published to date (1).

A separate Phase II study (NCT04580771) is assessing the safety and tolerability of the nanoparticle-based HPV-16 E6/E7 multi-peptide vaccine PDS0101 administered alongside standard chemoradiation in patients with locally advanced cervical cancer. The vaccine is intended to trigger a targeted immune response against HPV16-positive tumor cells, potentially enhancing the overall

effectiveness of conventional treatment (33).

Phase III clinical studies are designed to confirm the treatment's effectiveness on a larger scale and to further evaluate its safety. These trials typically compare the investigational therapy against the current standard of care to provide strong clinical evidence supporting its use. They also allow for the observation of side effects across diverse patient populations. A current Phase II/III trial (NCT05141721) includes an initial Phase II segment

that investigates the immune response elicited by a prime/boost strategy using GRT-C901 and GRT-R902 vectors, in combination with immune checkpoint inhibitors and standard chemotherapy (fluoropyrimidine and bevacizumab). The subsequent Phase III segment aims to validate the efficacy of this combined treatment by comparing it with standard chemotherapy alone, using progression-free survival as the primary endpoint (34).

Two large-scale Phase III studies are evaluating the individualized neoantigen vaccine V940 (formerly mRNA-4157) in combination with pembrolizumab. The first (NCT05933577) focuses on preventing recurrence in patients with high-risk melanoma, comparing the vaccine-plus-pembrolizumab combination to

pembrolizumab alone. The clinical trial is active (35).

The second (NCT06077760) targets patients with resected Stage II or IIIA/IIIB NSCLC, using a double-blind design to assess whether the addition of V940 improves disease-free survival relative to

pembrolizumab plus placebo. Participants receive nine doses of V940 every three weeks and pembrolizumab every six weeks for up to one year or until disease recurrence or toxicity becomes unacceptable. This clinical trial is recruiting participants (36).

Table 2. Summary of Cancer Nano vaccines in Clinical Trials (Phases I–II

Trial ID	Vaccine Type / Target	Indication	Treatment Description	Phase	Ref
NCT05714748	LNP-encapsulated mRNA vaccine	EBV-positive advanced malignancies	Investigates mRNA vaccine encapsulated in lipid nanoparticles for treating EBV-associated cancers.	Phase I	28
NCT05475106	Personalized neoantigen peptide + Leukine	Solid tumors with neoantigens	Intradermal administration of personalized multipeptide vaccine with Leukine (Sargramostim).	Phase I	29
NCT04573140	RNA-lipid particle vaccine	Newly diagnosed glioblastoma (GBM)	Post-surgery and chemoradiation, patients receive RNA-loaded liposomal vaccines over 14 months.	Phase I	30

NCT00952692	dHER2+AS15 ASCI + Lapatinib	HER2- positive metastatic breast cancer	Truncated HER2 protein with adjuvants (MPL, CpG ODN, QS21) plus oral lapatinib for trastuzumab- resistant tumors.	Phase I/II	31
NCT00828009	BLP25 liposomal vaccine (tecemotide) + Bevacizumab	Stage IIIA/IIIB NSCLC	Post- chemoradiation treatment with vaccine and anti-angiogenic antibody (bevacizumab).	Phase II	32
NCT04580771	Liposomal HPV-16 E6/E7 peptide vaccine (PDS0101)	Locally advanced cervical cancer	Vaccine combined with standard chemoradiation to enhance immune response against HPV16+ tumor cells.	Phase II	33
NCT05141721	GRT-C901 + GRT-R902 + CPI + chemo	Advanced cancer	Heterologous prime/boost strategy with checkpoint inhibitors and chemotherapy vs. chemotherapy alone.	Phase II/III	34
NCT05933577	V940 (mRNA- 4157) + Pembrolizumab	High-risk melanoma	Individualized mRNA vaccine plus anti-PD-1 antibody to prevent	Phase III	35

			recurrence post-surgery.		
NCT06077760	V940 (mRNA-4157) + Pembrolizumab	Stage II/IIIA/IIIB NSCLC (adjuvant)	Randomized, double-blind trial comparing V940 + pembrolizumab vs. placebo + pembrolizumab following tumor resection.	Phase III	36

CONCLUSION

Cancer nano vaccines are emerging as a transformative approach in immunotherapy, offering novel solutions to overcome the limitations of conventional cancer treatments. This review explores recent progress in the design and application of nano vaccines, emphasizing their capacity to provoke targeted immune responses against tumor-specific antigens and to strengthen the body's natural defense against cancer. Although substantial advancements have been made, key hurdles persist—such as improving targeted delivery systems, counteracting the immunosuppressive tumor microenvironment (TME), and conducting comprehensive clinical trials to confirm safety and efficacy. Looking forward, the integration of cutting-edge technologies including artificial intelligence, multiomics

approaches, and synthetic biology holds promise for the next generation of nano vaccine development. Combining nano vaccines with complementary therapies, such as immune checkpoint inhibitors or CAR-T cell therapy, may lead to synergistic strategies that significantly amplify antitumor immunity. Nevertheless, a focus on streamlined design remains essential to ensure scalability and cost-effectiveness. Promoting open-access data sharing and standardized protocols will aid in refining formulations, enhancing reproducibility, and enabling the use of AI-driven tools for optimization. While continued refinement of established platforms like lipid-based nanoparticles is important, exploring innovative nanomaterials will be equally critical for future breakthroughs. In clinical practice, employing patient stratification to guide recruitment may enhance trial success and

uncover new therapeutic possibilities for individuals who respond poorly to current nano vaccine strategies. Ultimately, advancing cancer nano vaccines from the laboratory to the clinic will depend on strong interdisciplinary collaboration among scientists, healthcare providers, industry stakeholders, and regulatory bodies.

FUNDING INFORMATION

The authors declare that no funds, grants, or other support were received during the preparation of this manuscript.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

1. Wang, Y., Liu, C., Fang, C., Peng, Q., Qin, W., Yan, X., & Zhang, K. (2024). Engineered Cancer Nanovaccines: A New Frontier in Cancer Therapy. *Nano-Micro Letters*, 17(1), 30. <https://doi.org/10.1007/s40820-024-01533-y>.
2. van der Bruggen, P., Traversari, C., Chomez, P., Lurquin, C., De Plaen, E., Van den Eynde, B., Knuth, A., & Boon, T. (1991). A Gene Encoding an Antigen Recognized by Cytolytic T Lymphocytes on a Human Melanoma. *Science*, 254(5038), 1643–1647. <https://doi.org/10.1126/science.1840703>.
3. Antonarelli, G., Corti, C., Tarantino, P., Ascione, L., Cortes, J., Romero, P., Mittendorf, E. A., Disis, M. L., & Curigliano, G. (2021). Therapeutic cancer vaccines revamping: Technology advancements and pitfalls. *Annals of Oncology*, 32(12), 1537–1551. <https://doi.org/10.1016/j.annonc.2021.08.2153>.
4. Liao, Z., Huang, J., Lo, P.-C., Lovell, J. F., Jin, H., & Yang, K. (2022). Self-adjuvating cancer nano vaccines. *Journal of Nanobiotechnology*, 20(1), 345. <https://doi.org/10.1186/s12951-022-01545-z>.
5. Fang, X., Lan, H., Jin, K., Gong, D., & Qian, J. (2022). Nanovaccines for Cancer Prevention and Immunotherapy: An Update Review. *Cancers*, 14(16), Article 16. <https://doi.org/10.3390/cancers14163842>.
6. Goldberg, M. S. (2019). Improving cancer immunotherapy through nanotechnology. *Nature Reviews Cancer*, 19(10), 587–602. <https://doi.org/10.1038/s41568-019-0186-9>.
7. Yao, M., Liu, X., Qian, Z., Fan, D., Sun, X., Zhong, L., & Wu, P. (2023). Research progress of nanovaccine in

- anti-tumor immunotherapy. *Frontiers in Oncology*, 13. <https://doi.org/10.3389/fonc.2023.1211262>.
8. Chen, I., Chen, M. Y., Goedegebuure, S. P., & Gillanders, W. E. (2021). Challenges targeting cancer neoantigens in 2021: A systematic literature review. *Expert Review of Vaccines*, 20(7), 827–837. <https://doi.org/10.1080/14760584.2021.1935248>
 9. Zaccariotto, G. de C., Bistaffa, M. J., Zapata, A. M. M., Rodero, C., Coelho, F., Quitiba, J. V. B., Lima, L., Sterman, R., Cardoso, V. M. de O., & Zucolotto, V. (2025). Cancer Nanovaccines: Mechanisms, Design Principles, and Clinical Translation. *ACS Nano*, 19(17), 16204–16223. <https://doi.org/10.1021/acsnano.4c15765>.
 10. Zhang, Y., Lin, S., Wang, X.-Y., & Zhu, G. (2019). Nanovaccines for cancer immunotherapy. *WIREs Nanomedicine and Nanobiotechnology*, 11(5), e1559. <https://doi.org/10.1002/wnan.1559>
 11. Luo, M., Wang, H., Wang, Z., Cai, H., Lu, Z., Li, Y., Du, M., Huang, G., Wang, C., Chen, X., Porembka, M. R., Lea, J., Frankel, A. E., Fu, Y.-X., Chen, Z. J., & Gao, J. (2017). A STING-activating nanovaccine for cancer immunotherapy. *Nature Nanotechnology*, 12(7), 648–654. <https://doi.org/10.1038/nnano.2017.52>
 12. Mohme, M., Riethdorf, S., & Pantel, K. (2017). Circulating and disseminated tumour cells. Mechanisms of immune surveillance and escape. *Nature Reviews Clinical Oncology*, 14(3), 155–167. <https://doi.org/10.1038/nrclinonc.2016.144>.
 13. Liang, K., Sun, Y., Xie, L., Liu, Y., You, Y., Xu, J., Ma, F., Huang, Y., Song, Q., Xiao, W., Huang, J., Gao, X., & Chen, J. (2024). Biologically Self-Assembled Tumor Cell-Derived Cancer Nanovaccines as an All-in-One Platform for Cancer Immunotherapy. *ACS Nano*, 18(8), 6702–6717. <https://doi.org/10.1021/acsnano.4c01050>
 14. Ma, L., Diao, L., Peng, Z., Jia, Y., Xie, H., Li, B., Ma, J., Zhang, M., Cheng, L., Ding, D., Zhang, X., Chen, H., Mo, F., Jiang, H., Xu, G., Meng, F., Zhong, Z., & Liu, M. (2021). Immunotherapy and Prevention of Cancer by Nanovaccines Loaded with Whole-Cell Components of Tumor Tissues or Cells. *Advanced Materials*, 33(43), 2104849. <https://doi.org/10.1002/adma.202104849>.
 15. Luo, M., Wang, H., Wang, Z., Cai, H., Lu, Z., Li, Y., Du, M., Huang, G., Wang, C., Chen, X., Porembka, M. R., Lea, J.,

- Frankel, A. E., Fu, Y.-X., Chen, Z. J., & Gao, J. (2017). A STING-activating nanovaccine for cancer immunotherapy. *Nature Nanotechnology*, 12(7), 648–654. <https://doi.org/10.1038/nnano.2017.52>.
16. Zhang, X., Wang, K., Zhao, Z., Shan, X., Wang, Y., Feng, Z., Li, B., Luo, C., Chen, X., & Sun, J. (2024). Self-Adjuvanting Polyguanidine Nanovaccines for Cancer Immunotherapy. *ACS Nano*, 18(9), 7136–7147. <https://doi.org/10.1021/acsnano.3c11637>
17. Zhu, G., Mei, L., Vishwasrao, H. D., Jacobson, O., Wang, Z., Liu, Y., Yung, B. C., Fu, X., Jin, A., Niu, G., Wang, Q., Zhang, F., Shroff, H., & Chen, X. (2017). Intertwining DNA-RNA nano capsules loaded with tumor neoantigens as synergistic nano vaccines for cancer immunotherapy. *Nature Communications*, 8(1), 1482. <https://doi.org/10.1038/s41467-017-01386-7>
18. Xu, J., Lv, J., Zhuang, Q., Yang, Z., Cao, Z., Xu, L., Pei, P., Wang, C., Wu, H., Dong, Z., Chao, Y., Wang, C., Yang, K., Peng, R., Cheng, Y., & Liu, Z. (2020). A general strategy towards personalized nanovaccines based on fluoropolymers for post-surgical cancer immunotherapy. *Nature Nanotechnology*, 15(12), 1043–1052. <https://doi.org/10.1038/s41565-020-00781-4>.
19. Chen, G., Li, X., Li, R., Wu, K., Lei, Z., Dai, R., Roche, K., Wang, A. Z., & Min, Y. (2023). Chemotherapy-Induced Neoantigen Nanovaccines Enhance Checkpoint Blockade Cancer Immunotherapy. *ACS Nano*, 17(19), 18818–18831. <https://doi.org/10.1021/acsnano.3c03274>.
20. Guan, X., Chen, J., Hu, Y., Lin, L., Sun, P., Tian, H., & Chen, X. (2018). Highly enhanced cancer immunotherapy by combining nanovaccine with hyaluronidase. *Biomaterials*, 171, 198–206. <https://doi.org/10.1016/j.biomaterials.2018.04.039>.
21. Zhu, G., Liu, Y., Yang, X., Kim, Y.-H., Zhang, H., Jia, R., Liao, H.-S., Jin, A., Lin, J., Aronova, M., Leapman, R., Nie, Z., Niu, G., & Chen, X. (2016). DNA–inorganic hybrid nanovaccine for cancer immunotherapy. *Nanoscale*, 8(12), 6684–6692. <https://doi.org/10.1039/C5NR08821F>.
22. Xu, X., Zhang, M., Liu, X., Chai, M., Diao, L., Ma, L., Nie, S., Xu, M., Wang, Y., Mo, F., & Liu, M. (2023). Probiotics formulation and cancer nanovaccines show synergistic effect in immunotherapy and prevention of

- colon cancer. *iScience*, 26(7).
<https://doi.org/10.1016/j.isci.2023.107167>.
23. You, Q., Wu, G., Li, H., Liu, J., Cao, F., Ding, L., Liang, F., Zhou, B., Ma, L., Zhu, L., Wang, C., Yang, Y., & Chen, X. (2025). A nanovaccine targeting cancer stem cells and bulk cancer cells for postoperative cancer immunotherapy. *Nature Nanotechnology*, 1–14.
<https://doi.org/10.1038/s41565-025-01952-x>.
 24. Xiao, L., Huang, Y., Yang, Y., Miao, Z., Zhu, J., Zhong, M., Feng, C., Tang, W., Zhou, J., Wang, L., Zhao, X., & Wang, Z. (2021). *Biomimetic cytomembrane nanovaccines prevent breast cancer development in the long term*.
<https://doi.org/10.1039/D0NR08978H>.
 25. Yang, C., Chen, Y., Liu, J., Zhang, W., He, Y., Chen, F., Xie, X., Tang, J., Guan, S., Shao, D., Wang, Z., & Wang, L. (2024). Leveraging Senescent Cancer Cell Membrane to Potentiate Cancer Immunotherapy Through Biomimetic Nano vaccine. *Advanced Science*, 11(30), 2400630.
<https://doi.org/10.1002/advs.202400630>.
 26. Liu, T., Si, X., Liu, L., Ma, S., Huang, Z., Zhang, Y., Song, W., Zhang, Y., & Chen, X. (2024). Injectable Nano-in-Gel Vaccine for Spatial and Temporal Control of Vaccine Kinetics and Breast Cancer Postsurgical Therapy. *ACS Nano*, 18(4), 3087–3100.
<https://doi.org/10.1021/acsnano.3c08376>.
 27. Liu, Z., Liu, B., Feng, Y., Zhao, L., Wang, Q., He, H., Yin, T., Zhang, Y., Yang, L., Gou, J., & Tang, X. (2024). Dual-Targeted Self-Adjuvant Heterocyclic Lipidoid@Polyester Hybrid Nanovaccines for Boosting Cancer Immunotherapy. *ACS Nano*, 18(24), 15557–15575.
<https://doi.org/10.1021/acsnano.4c00392>.
 28. Peng, X. (2025). *A Phase I Study of mRNA Vaccine for Patients With EBV-positive Advanced Malignant Tumors* (Clinical Trial Registration No. NCT05714748). [clinicaltrials.gov. https://clinicaltrials.gov/study/NCT05714748](https://clinicaltrials.gov/study/NCT05714748)
 29. Instituto de Medicina Regenerativa. (2024). Pilot Study of Personalized Neoantigen Peptide Vaccines and Leukine for the Treatment of Neoplasms (Clinical Trial Registration No. NCT05475106). [clinicaltrials.gov. https://clinicaltrials.gov/study/NCT05475106](https://clinicaltrials.gov/study/NCT05475106).
 30. University of Florida. (2025). A Phase I/II Study of RNA-lipid Particle (RNA-LP) Vaccines for Newly Diagnosed Pediatric High-Grade Gliomas (pHGG) and Adult

- Glioblastoma (GBM) (Clinical Trial Registration No. NCT04573140). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT04573140).
<https://clinicaltrials.gov/study/NCT04573140>.
31. Morse, M. (2021). A Monocentric, Open-label Phase I/II Study to Assess dHER2+AS15 Cancer Immunotherapeutic Given in
32. ECOG-ACRIN Cancer Research Group. (2023). A Phase II Study of L-BLP25 and Bevacizumab in
- Unresectable Stage IIIA and IIIB Non-Squamous Non-Small Cell Lung Cancer After Definitive Chemoradiation (Clinical Trial Registration No. NCT00828009). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT00828009).
<https://clinicaltrials.gov/study/NCT00828009>.
33. M.D. Anderson Cancer Center. (2025). IMMUNOCERV: Evaluating the Safety of Chemoradiation Combined With PDS0101 Immunotherapy in Treating Locally Advanced Cervical Cancer (Clinical Trial Registration No. NCT04580771). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT04580771).
<https://clinicaltrials.gov/study/NCT04580771>.
34. Seattle Project Corporation. (2025). A Phase 2/3, Randomized, Open-Label Study of Maintenance GRT-C901/GRT-R902, A Neoantigen
- Combination With Lapatinib to Patients With ErbB2 Overexpressing Metastatic Breast Cancer Refractory to Trastuzumab (Clinical Trial Registration No. NCT00952692). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT00952692).
<https://clinicaltrials.gov/study/NCT00952692>.
- Vaccine, in Combination With Immune Checkpoint Blockade for Patients With Metastatic Colorectal Cancer (Clinical Trial Registration No. NCT05141721). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT05141721).
<https://clinicaltrials.gov/study/NCT05141721>.
35. Merck Sharp & Dohme LLC. (2025). A Phase 3, Randomized, Double-Blind, Placebo- and Active-Comparator-Controlled Clinical Study of Adjuvant V940 (mRNA-4157) Plus Pembrolizumab Versus Adjuvant Placebo Plus Pembrolizumab in Participants With High-Risk Stage II-IV Melanoma (INTerpath-001) (Clinical Trial Registration No. NCT05933577). [clinicaltrials.gov](https://clinicaltrials.gov/study/NCT05933577).
<https://clinicaltrials.gov/study/NCT05933577>.

36. Merck Sharp & Dohme LLC. (2025). A Phase 3, Randomized, Double-blind, Placebo- and Active-Comparator-Controlled Clinical Study of Adjuvant V940 (mRNA-4157) Plus Pembrolizumab Versus Adjuvant Placebo Plus Pembrolizumab in

Participants With Resected Stage II, IIIA, IIIB (N2) Non-small Cell Lung Cancer (INterpath-002) (Clinical Trial Registration No. NCT06077760). clinicaltrials.gov.

<https://clinicaltrials.gov/study/NCT06077760>



BTS International
Advanced
Pharmaceutical
Sciences Journal