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Nanotechnology-based strategies for vaccine development: accelerating innovation and delivery

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lipid nanoparticles; nanotechnology; polymer nanoparticles; vaccine development; virus-like particles

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ABSTRACT

The key role and impact of nanotechnology in vaccine development became particularly prominent following the outbreak of the coronavirus disease 2019 (COVID-19) pandemic in 2019. Especially in the process of designing and optimising COVID-19 vaccines, the application of nanomaterials significantly accelerated vaccine development and efficient delivery. In this review, we categorised and evaluated conventional vaccines, including attenuated live vaccines, inactivated vaccines, and subunit vaccines, highlighting their advantages and limitations. We summarised the development history, mechanisms, and latest technologies of vaccine adjuvants, emphasising their critical role in immune responses. Furthermore, we focused on the application of nanotechnology in the vaccine field, detailing the characteristics of nanoparticle vaccines, including virus-like particles, lipid-based carriers, inorganic nanoparticles, and polymer-based carriers. We emphasised their potential advantages in enhancing vaccine stability and immunogenicity, as well as their ability to deliver vaccines and present antigens through various routes. Despite facing challenges such as low drug loading efficiency, issues with long-term storage, high costs, and difficulties in large-scale production, nano-vaccines hold promise for the future. This review underscores the pivotal role and prospects of nanotechnology in vaccine development, offering new pathways and strategies to address current and future disease challenges.

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Introduction

Vaccination represents a significant milestone in human history, playing a vital role in disease prevention.[1] Since the introduction of the first vaccine over 300 years ago, vaccines have effectively mitigated serious health risks and societal challenges. Vaccination is an essential element of public health initiatives, particularly in economically disadvantaged regions. Nanovaccines have the potential to generate substantial economic benefits due to their simplified research and development process compared to conventional vaccines. This streamlined process allows for rapid production and deployment, cost advantages, and high efficacy. Additionally, nanovaccines, such as those for tumour prevention and infectious diseases like coronavirus disease 2019 (COVID-19), have the potential to significantly reduce social medical costs and contribute to overall economic growth. At the same time, as a new technology, nano vaccine has significant social value, which can drive the development of upstream and downstream related industries, the technological upgrading and transformation of corresponding enterprises, and the vigorous development of corresponding basic scientific research. As a pivotal achievement in the field of medicine, vaccines effectively stimulate and harness the body's immune response.[2] Vaccines elicit immune responses by mimicking the presence of pathogens, thereby enhancing the body's ability to mount a swift and robust defense upon encountering actual pathogens. Edward

Jenner, a British physician, achieved a significant milestone in vaccine development in 1798 by creating the cowpox virus vaccine.[3] This vaccine involved the isolation, inactivation, and injection of the disease-causing pathogen into humans, resulting in immunity against smallpox and establishing the basis for the development of conventional vaccines. Conventional vaccines commonly utilise weakened or killed pathogens, or their components, to activate the immune system and confer immunity against specific diseases.[4, 5] Nevertheless, there remain a multitude of infectious diseases for which existing vaccines demonstrate limited efficacy. This can be attributed to unintended consequences, restricted durability against a range of pathogen strains, and the possibility of allergic responses.[6] While conventional vaccines have improved antigen presentation through diverse delivery routes, there are instances where they may exhibit infectivity, particularly in the case of live attenuated vaccines, leading to the potential for virulence reversion and the induction of mild symptoms in vaccinated individuals.[7] Moreover, the use of protein-containing vaccines has been shown to potentially disturb immune homeostasis post-administration, leading to concerns regarding their stability and potential toxicity.[8, 9] The progression of nanotechnology has enabled the incorporation of nanoparticles with diverse characteristics into the medical field, including different compositions, sizes, shapes, and surface properties.[10, 11] Within the field of disease prevention, the incorporation of nanotechnology into the development of vaccines presents notable advantages, such as the ability of nanoparticles to act as a delivery system for various routes of administration and improve the efficacy of antigen presentation.[12-14] Furthermore, nanoparticles can serve as vaccine adjuvants to strengthen the immune response. Undoubtedly, nanotechnology is playing a pivotal role in progressing vaccine development to address unmet clinical requirements.[15] One notable advancement is the successful repurposing of lipid nanoparticles (LNPs), originally designed for drug delivery, into carriers for mRNA vaccines (Moderna and Pfizer/BioNTech COVID-19 vaccines).[16] These advancements highlight the promising potential for integrating nanotechnology into the development of vaccines.[17]

To ensure that this review encompasses the latest research findings and provides a comprehensive perspective, we employed a systematic literature review approach. First, we conducted a literature search in the PubMed database, a widely recognised resource in the biomedical field. The keywords used included "nanotechnology" and "vaccine", aimed at capturing all relevant studies on the application of nanotechnology in vaccine development. After the preliminary search, we screened the literature based on the following criteria: First, relevance; the literature must directly discuss or involve how nanotechnology is used in the design, optimisation, and delivery of vaccines. Second, timeliness; we prioritised recently published studies to reflect the latest advances in the field. Finally, we considered the quality of the literature; cited works had to come from peer-reviewed journals to ensure the reliability and scientific validity of the information. After screening, a total of 147 articles met our inclusion criteria, providing a solid theoretical foundation for this review. These articles cover not only traditional vaccine types but also explore the roles and potential advantages of various nanomaterials as carriers or adjuvants in vaccines. Furthermore, this review outlines various nanotechnology methods for vaccine development, including traditional approaches and the latest technologies. Special attention is given to the role of different nanomaterials in vaccines, highlighting their ability to improve vaccine stability, biodistribution, and cellular uptake. Additionally, we discuss the potential and prospects of nanoparticle vaccines (nanovaccines) (**Figure 1**).

Conventional Vaccines

Conventional vaccines are classified into attenuated live vaccines, inactivated vaccines, subunit vaccines, and other categories based on the various technologies utilised.[18] Recent progress in immunology has led to a transition from employing whole pathogens (e.g. inactivated or attenuated pathogens) to targeting specific components of the pathogen, such as proteins, polysaccharides, and nucleic acids. This shift includes genetic engineering alterations of viruses and bacteria, as well as the development of recombinant proteins and viral vector vaccines.[19] Each type of vaccine presents its own set of benefits and drawbacks.

Live vaccines are capable of inducing strong immune responses, encompassing both cellular and humoral immunity, and frequently confer prolonged immune protection following a single or a limited number of administrations.[20] The immunological principle of live vaccines is based on using attenuated or weakened pathogens. Despite their reduced virulence, attenuated vaccines retain live pathogens, thereby presenting a potential hazard of inducing illness in individuals with compromised immune systems.[21] Inactivated vaccines are devoid of live or infectious particles, thereby precluding the risk of inducing disease or reactivation. Despite their generally high safety profile, particularly among immunocompromised individuals, inactivated vaccines frequently exhibit diminished immunogenicity and shorter duration of protection compared to live vaccines, necessitating multiple administrations or adjuvants to bolster immunogenicity.[22]

The ongoing advancements in the field of vaccinology have led to the creation of more adaptable and effective methods for preventing and controlling diseases through the introduction of innovative vaccines. The introduction of subunit vaccines, peptides, DNA, and mRNA vaccines represents a significant milestone in contemporary vaccinology. By employing techniques from molecular biology and biotechnology, these vaccines are able to produce targeted pathogen components or direct cells to generate pertinent antigens, thereby stimulating the immune system to initiate protective responses. Subunit vaccines are based on a portion of the pathogen, such as proteins, sugars, or their combinations, to induce immune

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responses. Subunit vaccines, which contain only a small portion of the pathogen, are characterised by their high safety profile, making them particularly suitable for individuals with compromised immune systems.[23] However, in comparison to vaccines that utilize the entire pathogen, subunit vaccines may elicit weaker immune responses, necessitating the use of adjuvants to enhance immunogenicity. The development of subunit vaccines requires the utilisation of sophisticated technologies and methodologies for the identification, extraction, purification, and production of specific pathogen components, resulting in relatively higher development costs.[24] The hepatitis B virus (HBV) vaccine produced through DNA recombinant technology is an example of a subunit vaccine, but it has also imposed significant financial pressure on the health departments of developing countries (**Figure 2**). The development of peptide, DNA, and mRNA vaccines has made vaccine production more precise and efficient, simultaneously enhancing the ability to respond to different pathogens.[25,26] Peptide vaccines are based on protein fragments of the pathogen, selecting regions with strong antigenicity to induce immune responses.[27] DNA vaccines introduce DNA containing the target gene into host cells, prompting the production of pathogen-related antigens within the cells. mRNA vaccines deliver the pathogen's mRNA directly to host cells, allowing the cells to synthesise relevant proteins and thereby triggering immune responses.[28,29]

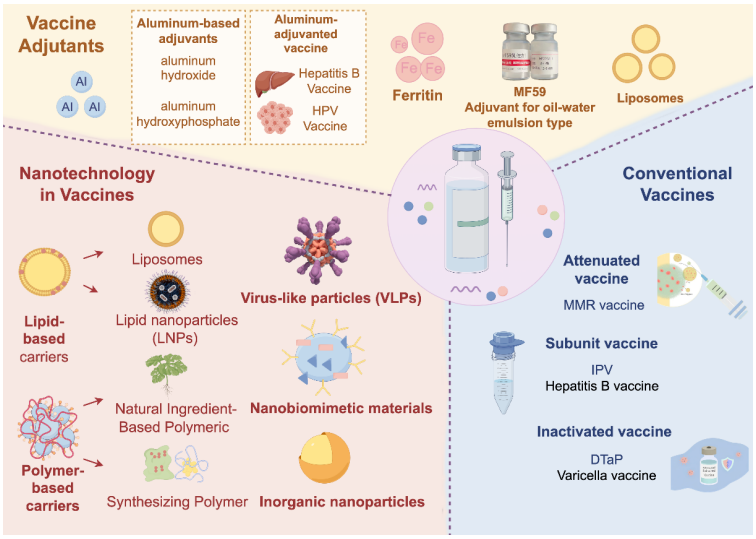

Figure 1. Innovative vaccine technologies: bridging traditional approaches and modern adjuvants with nanotechnology applications. It highlights the role of traditional vaccines and the essential function of modern adjuvants. It also explores the transformative applications of nanotechnology in vaccine development, including lipid carriers, polymer carriers, virus-like particles, and biomimetic materials. The review summarises advancements in vaccine technologies, with a particular focus on traditional vaccines and modern adjuvants, aimed at addressing future disease challenges. Created with Figdraw (https://www.figdraw.com). Al: aluminum; DTaP: diphtheria, tetanus, and acellular pertussis vaccine; HPV: human papillomavirus; IPV: inactivated poliovirus vaccine; LNPs: lipid nanoparticles; MMR Vaccine: measles, mumps, and rubella vaccine; VLPs: virus-like particles.

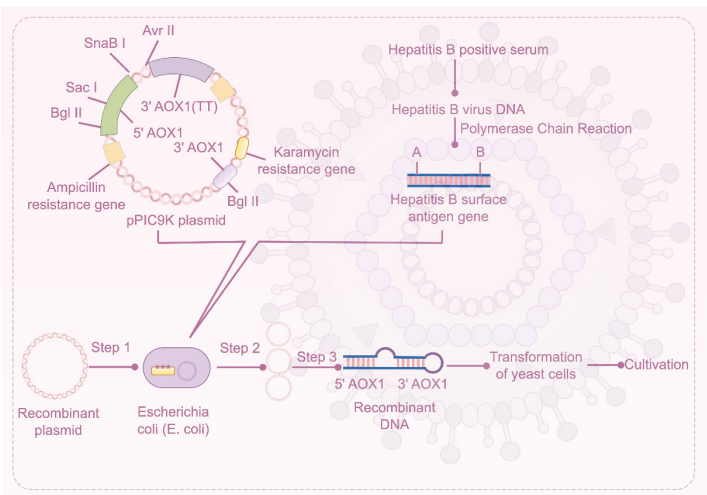

Figure 2. The preparation process of inactivated hepatitis B vaccine. Created with Figdraw (https://www.figdraw.com). 3′ AOX1: 3′ alcohol oxidase 1; 5′ AOX1: 5′ alcohol oxidase 1 promoter; Avr II: restriction enzyme; Bgl II: *Bacillus globigii* II; Sac I: *Streptomyces achromogenes* I; SnaB I: *Sphingomonas natatoria* B I.

The efficacy of innovative vaccines is dependent on the essential functions of adjuvants and delivery vectors.[30] Adjuvants play a critical role in augmenting immune responses, thereby enhancing the immunogenicity of vaccines.[31, 32] Delivery vectors, such as LNPs, facilitate the efficient transport of vaccine components into cells, thereby amplifying the efficacy of immune responses. The utilisation of these supplementary technologies allows vaccines to more effectively stimulate the immune system to produce antibodies and memory immune cells, resulting in heightened and enduring protection against pathogens.

Vaccine Adjuvants

Adjuvants, essential components of vaccines, play a significant role in enhancing the vaccine's immunological response in terms of potency, speed, and duration.[15] The term "adjuvant" originates from the Latin word "adjuvare" meaning "to help".[33] The concept of adjuvants was introduced in 1925 by Gaston Ramon, who observed that sterile additives could boost antibody production in animals.[34] The following year, Alexander Glenny demonstrated the adjuvant effect of aluminum salts in immunotherapy by showing that aluminum salt-precipitated diphtheria toxoid induced a more robust immune response.[35-37] Later, in the 1940s, Freud and his colleagues[38, 39] developed an emulsion of water in oil, thereby creating the Freud adjuvant. However, due to its toxicity to the human body, Freud adjuvant was not approved for use in human vaccines. Similar to Freud adjuvant, the use of bacterial lipopolysaccharide adjuvants in human vaccines has also been restricted due to their local and systemic side effects.[40] In fact, from the 1920s to the 1990s, despite efforts to develop new adjuvants, only aluminum adjuvants were approved for use. It wasn't until 1997 that the oil-in-water emulsifier MF59 was approved in Europe as an adjuvant for influenza vaccines. Over the course of the following two decades, four additional adjuvants (AS04, AS03, AS01, and CpG ODN 1018) were authorised for use in vaccines, thereby diversifying the landscape of human vaccine adjuvants[41] (**Figure 3**). Additionally, during this period, many other compounds of various categories were evaluated as adjuvants, including mineral salts, microbial products, emulsions, saponins, synthetic small molecule stimulants, polymers, nanoparticles, and liposomes.[42] Adjuvants can be classified according to different criteria, with one classification based on their mechanism of action dividing them into delivery systems and immune enhancers. Simultaneously, they bind to antigens in the drug delivery system, acting as antigen carriers, and induce local inflammatory responses by activating the innate immune system, recruiting immune cells to the injection site.[43] Specifically, the antigen-adjuvant complex activates pattern-recognition receptor pathways by acting as pathogen-associated molecular patterns.[44] This causes the activation of innate immune cells with the production of cytokines and chemokines. The same pathway is directly activated by immune potentiators[45] (**Figure 4**).

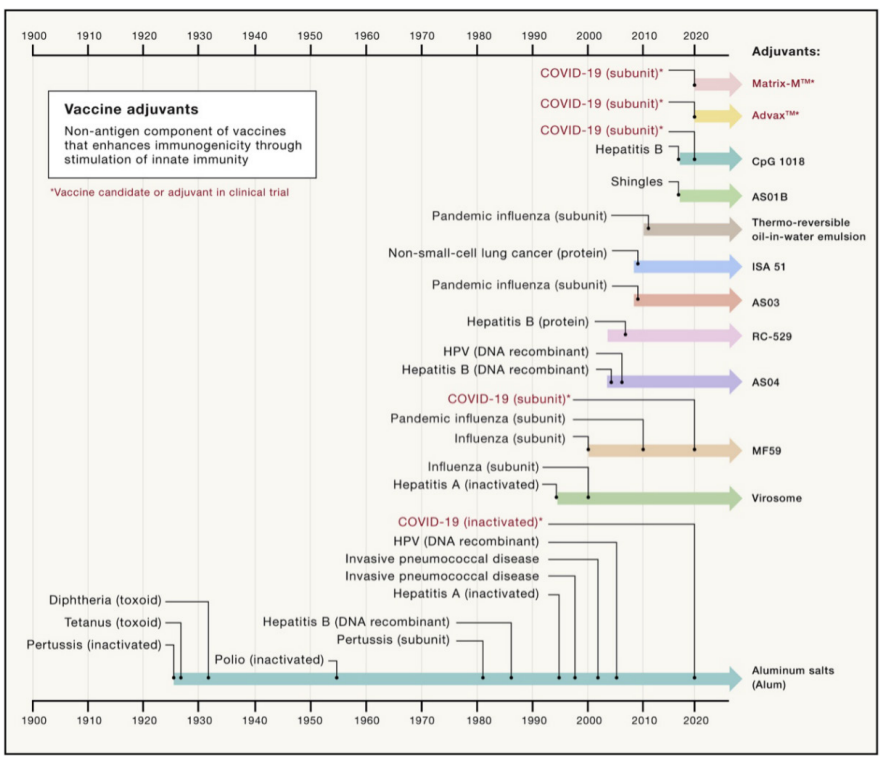

Figure 3. Timeline of adjuvant used in human vaccines. Adjuvants are non-antigen components of vaccines that stimulate the innate immune system. Adjuvants are indicated by bold arrows from the time of introduction. Vaccines that use the adjuvants are indicated as dots on the arrow at the earliest time of use. Reprinted from Iwasaki and Omer.[41] AS: adjuvant systems; AS01: liposome with MPL + QS-21; AS03: vitamin E/surfactant polysorbate 80/squalene; AS04: 3-deacyl-MPL; COVID-19: coronavirus disease 2019; CpG 1018: Toll-like receptor 9 agonist; HPV: human papillomavirus; MPL: monophosphoryl lipid A, a Toll-like receptor 4 agonist.

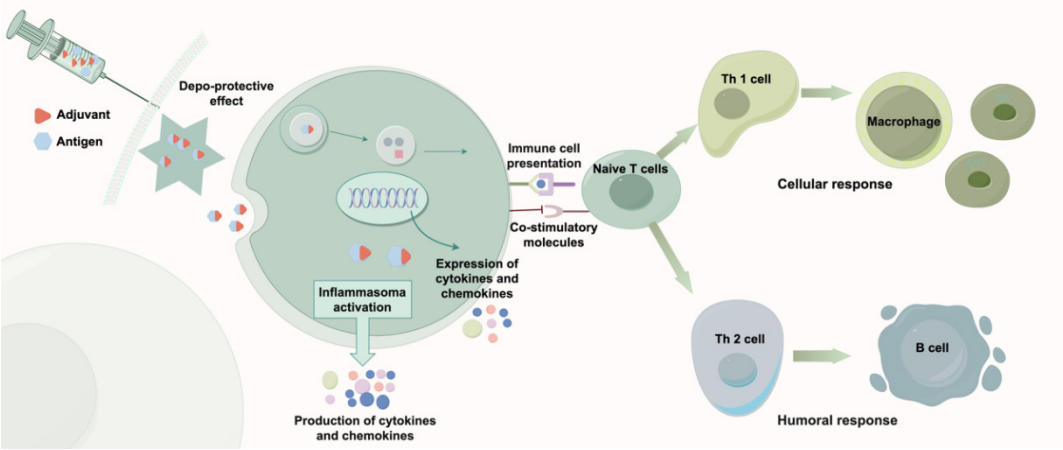

Figure 4. Mechanism of action of adjuvants. Reprinted from Facciolà et al.[44] Th 1 cell: type 1 T helper cell; Th 2 cell: type 2 T helper cell.

Aluminum-based adjuvants

Aluminum-based adjuvants, specifically aluminum hydroxide and aluminum phosphate, were the first adjuvants authorised for inclusion in human vaccines. These adjuvants have been shown to enhance the production of IgG1 and IgE antibodies by stimulating Th2 cell-mediated responses. However, the precise mechanisms by which aluminum adjuvants operate are complex, leading to ongoing scholarly debate. Presently, two distinct aspects of aluminum adjuvant function are acknowledged within the academic sphere.[46] One primary function of aluminum adjuvants is to act as a delivery mechanism that tightly binds to antigens, facilitating sustained release and thereby extending the bioavailability of antigens, ultimately enhancing antigen presentation. Aluminum-containing adjuvants are commonly used in the prevention and treatment of a range of diseases, including diphtheria, tetanus, meningitis, and HBV vaccines that have received approval from the U.S. Food and Drug Administration. Furthermore, aluminum is being considered as an adjuvant for vaccines currently undergoing clinical trials, such as the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) vaccine.[47, 48] Nevertheless, aluminum adjuvants exhibit certain drawbacks, such as challenges in stimulating a robust cellular immune response and the potential for adverse reactions, such as erythema and allergic responses. To enhance their efficacy, aluminum adjuvants can be optimised through improved formulation or by utilising nano-aluminum adjuvants.

Ferritin

Ferritin is a 450 kDa self-assembling spherical protein commonly utilised as a platform for bioimaging and targeted drug delivery due to its biocompatibility and ability to encapsulate various payloads.[49-51] Ferritin vaccine adjuvant refers to a type of vaccine adjuvant typically used to enhance the immunogenicity and protective efficacy of vaccines. The design of this adjuvant is based on the structure and function of ferritin, aiming to enhance vaccine stability, immunogenicity, and antigen presentation, thereby boosting the immune response to vaccines in the human body. As a natural carrier protein, possesses excellent biocompatibility and controllability, thus being utilised in vaccine adjuvant research and development. It can bind with vaccine antigens to form stable complexes, enhancing antigen stability, solubility, and facilitating antigen presentation and uptake by the immune system. Additionally, ferritin itself may possess immunomodulatory properties, further enhancing vaccine immunogenicity. Ferritin vaccine adjuvants have demonstrated the ability to effectively enhance the immunoprotective effect of vaccines by improving vaccine immunogenicity, enhancing antigenic stability, promoting antigen presentation, and modulating the immune response. Recently, the Pfizer-BioNTech COVID-19 vaccine has utilised ferritin as an adjuvant.

Conventional adjuvants are typically compounds or molecules used to enhance the immunogenicity of vaccines and improve vaccine efficacy, while nanocarriers are a type of drug delivery system composed of nanoparticles. These nanoparticles have the ability to encapsulate, transport, and release drugs, enabling targeted delivery to specific cells and tissues through surface modifications. This targeted delivery enhances therapeutic effects and reduces side effects. Additionally, nanoparticles exhibit high drug loading capacity and controlled drug release capabilities.

Nanotechnology in Vaccines

With the increasing adoption of modern theoretical design patterns in vaccine development, the number of vaccines is on the rise.[52, 53] Most vaccines employ "minimalist" components, often exhibiting lower immunogenicity[54] (**Figure 5**[55]). Consequently, nanotechnology is playing an increasingly crucial role in vaccine development. As the trend in vaccine development is towards "minimal" formulations with lower immunogenicity, there is a growing need for formulations that enhance antigen potency. The utilisation of nanoparticles in vaccine formulations not only enhances the stability and immunogenicity of antigens but also facilitates targeted delivery and slow release.[56] Nanoparticle vaccines are vaccines characterised by their particulate morphology and size range from a few to several hundred nanometers[57] (**Figure 6**). Nanomaterials have demonstrated significant potential in vaccination because they can conveniently leverage parameters

such as size, shape, and surface modifications. This allows for customisation to resemble natural targets in the immune system,[58-60] or optimise biodistribution and interaction with immune cells.[61] Compared to monovalent vaccines, nanoparticle vaccines can induce stronger neutralising antibodies and cellular immune responses.[62] The synthesis origin, definition, and shape-shifting structure of nanomaterials, along with increasingly clear engineering design rules, offer a potential avenue for developing vaccines that must generate immune responses designed differently from natural infections. With the current global need for low-cost, easily storable vaccines, especially in remote tropical and developing regions, the synthetic and design characteristics of nanomaterials make them a promising tool to address challenges in global distribution. They contribute to improving vaccine stability, delivery efficiency, and biocompatibility.[63] Nanoparticles play a crucial role as carriers in stabilising and delivering vaccine components, particularly suitable for novel vaccine technologies such as nucleic acid vaccines. Their small size and unique surface properties enable nanoparticles to efficiently encapsulate and protect vaccine components, preventing degradation or decomposition in the body, thus extending their presence *in vivo*. Additionally, nanoparticles can serve as adjuvants by mimicking the morphology and structure of pathogens, activating the immune system, and promoting a stronger immune response. They can also regulate the delivery rate and distribution of vaccine components, enhancing vaccine delivery efficiency, and prolonging the immune system's exposure to vaccine components, further enhancing the persistence of vaccine immune effects. In summary, the versatile composition and architecture of nanoparticle systems provide new strategies and platforms for the development of rapid and efficient vaccines.[64, 65] **Table 1** lists some examples of clinical approved vaccines using nanotechnology for addressing cancer in China and data were collected from ClinicalTrials.gov.

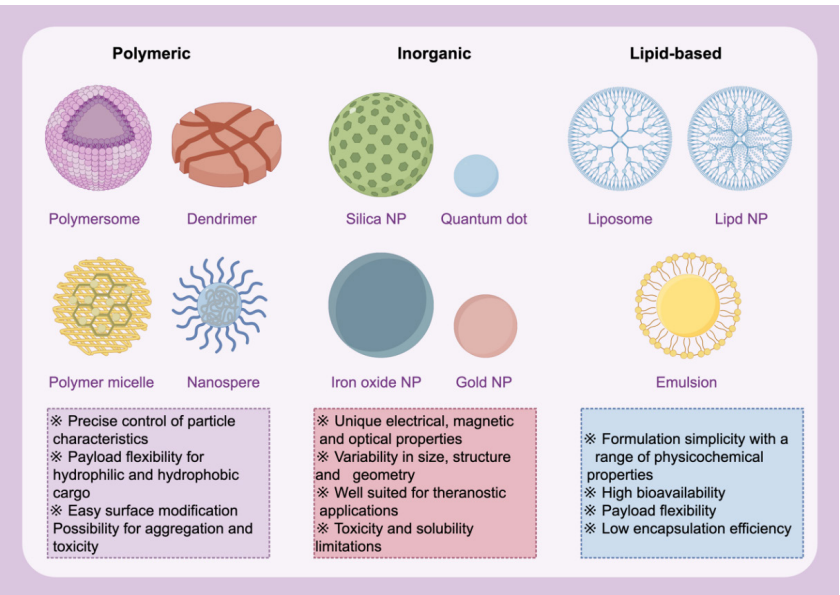

Figure 5. Classes of nanoparticles (NPs). Each class of NP features multiple subclasses, with some of the most common highlighted here. Each class has numerous broad advantages and disadvantages regarding cargo, delivery and patient response. Reprinted from Mitchell et al.[55]

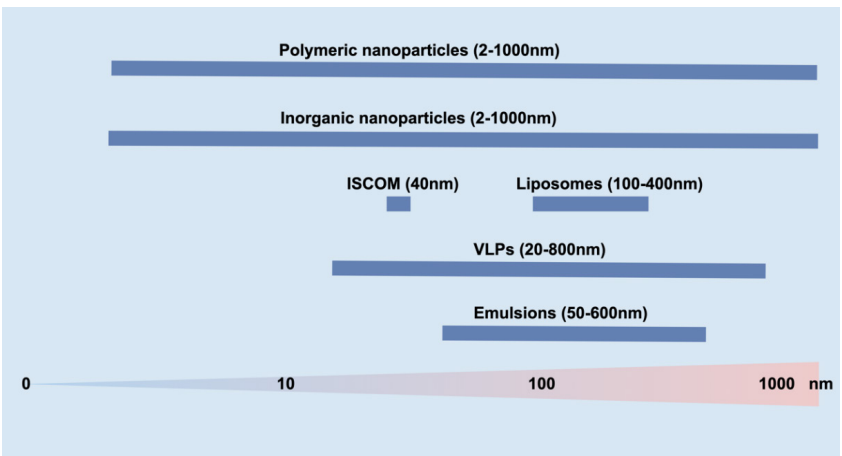

Figure 6. The size range of nanoparticles in nanovaccinology. Reprinted from Mitchell et al.[55] ISCOM: immunostimulating complex; VLPs: virus-like particles.

Table 1. Examples of clinical approved vaccines using nanotechnology for addressing cancer in China in the last 3 years

Application	Type	Trial ID	Phase	Start date
Solid tumour	mRNA vaccine (AFN18)	ChiCTR2400090447	Early Phase 1	September 2024
Advanced solid tumour	mRNA tumour vaccine InnoPCV	ChiCTR2400088149	N/A	August 2024
Advanced HPV-positive oral squamous cell carcinoma	Recombinant adenovirus vaccine	ChiCTR2400084773	Phase 1	May 2024
Solid tumour	Neoantigen mRNA vaccine	NCT06195384	Phase 1	May 2024
Recurrent pancreatic cancer	XP-004 personalised mRNA tumour vaccine combined with PD-1 inhibitor	NCT06496373	Phase 1	April 2024
Pancreatic cancer	Neoantigen mRNA vaccines	NCT06326736	Early Phase 1	April 2024
Digestive system neoplasms	mRNA vaccine: iNeo-Vac-R01	NCT06019702 NCT06026800 NCT06026800	Phase 1	September 2023
Advanced pancreatic cancer	mRNA vaccine	ChiCTR2300077339	Phase 1	August 2023
Advanced hepatocellular carcinoma	mRNA vaccine	ChiCTR2300073495	Early Phase 1	July 2023
Advanced solid tumour	mRNA-0217/S001 vaccine	NCT05916248	Phase 1	May 2023
Advanced pancreatic cancer	mRNA-0217/S001 vaccine	NCT05916261	Early Phase 1	April 2023
Hepatocellular carcinoma	HBV mRNA vaccine	NCT05738447	Phase 1	February 2023
Malignant tumours	EBV mRNA vaccine	NCT05714748	Phase 1	November 2022
Recurrent or metastatic bladder cancer	Chimeric exosomal tumour vaccines	NCT05559177	Early Phase 1	September 2022

Note: EBV: Epstein-Barr virus; HPV: human papillomavirus; N/A: not applicable; PD-1: programmed death-1.

Virus-like particles

Researchers have used computational design methods to create a self-assembling protein nanoparticle, termed as self-assembling nanoparticles.[66] These nanoparticles can display a pre-fusion state trimer of the respiratory syncytial virus F protein, known as DS-Cav1, which is a major neutralising antibody target. Through this design, the nanoparticles can repetitively present antigens on their surface, thereby enhancing the immune response. Virus-like particles (VLPs) can be considered a specific type of self-assembling nanoparticles[67] (**Figure 7**[68]). VLPs are nanoscale protein particles composed of viral structural proteins (such as coat or envelope) but lack the viral genome, making them unable to replicate in cells or host organisms, thus not causing infection.[69-71] These non-infectious particles can naturally form during the infection process or be produced on a large scale in the laboratory through genetic engineering techniques. VLPs have been directly used for the delivery of tumour-associated antigens, and vaccination with VLPs can be combined with radiation therapy, chemotherapy, or immunotherapy.[3, 72]

Vipin Kumar Deo and colleagues demonstrated the successful induction of immune responses in mice without the use of any adjuvants using VLPs-NcSRS2, which is formed by the dimerisation of the respiratory syncytial virus-gag protein consisting of 701 amino acids, forming VLPs of 100-200 nm on the cell membrane.[73] Vaccines such as Engerix (HBV) and Cervarix (human papillomavirus, HPV) from GlaxoSmithKline, as well as Recombivax HB (HBV) and Gardasil (HPV) from Merck, have demonstrated the role of VLPs as drug delivery systems.[74] VLPs have a wide range of applications and provide a unique approach to gene therapy. Using recombinant VLPs targeting the human immunodeficiency virus (HIV) CD4-specific receptor and delivering thymidine kinase, it has been shown that VLPs can selectively kill HIV-infected cells under ganciclovir treatment. Furthermore, using HIV VLPs, it is possible to target resting CD4 T cells and manipulate genes through gene silencing techniques, demonstrating the modifiability of VLPs[75] (**Figure 8**[68]).

VLPs vaccines have shown significant progress and potential in clinical development. Since the 1980s, VLP-based vaccines have been used for the prevention of various diseases, including HBV, malaria, HPV, and influenza. These vaccines mimic the external structure of viruses without containing the viral genetic material, thus eliciting protective immune responses against specific viruses.[76-79] The third generation HBV VLP vaccine, Sci-B-Victim, developed from the initial vaccine by Blumberg in the early 1980s, has been approved for use in Israel and 14 other countries in East Asia. Compared to the previous two generations, the third-generation vaccine shows better immune responses in elderly individuals, obese patients, immunocompromised patients, and those with kidney failure, undergoing dialysis, or transplantation. It includes three HBV surface antigens (S, Pre-S1, and Pre-S2) and is expressed in CHO cells, demonstrating the ability to induce high-titer anti-HBsAg antibodies with low doses.[80-83] VLPs, as vaccine candidates, exhibit significant advantages in terms of safety, immunogenicity, long-lasting immune protection, cross-protective ability, multifunctionality, no need for frozen storage, rapid processing, low effective dose, reduced risk of genetic recombination, and distinguishing between vaccine administration and virus infection. They have become a powerful tool in developing new vaccines, especially in the face of global public health emergencies.[84-86] Furthermore, VLPs serve as an innovative vaccine adjuvant, demonstrating unique advantages in various vaccine developments. According to a study by Park et al.[87] published in *Immunity &*

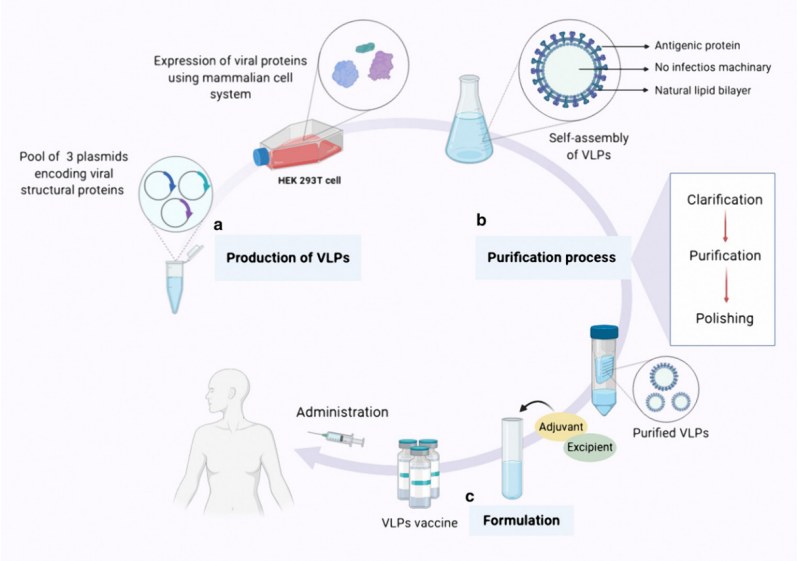

Figure 7. Overview of VLP-based vaccine expression, purification and formulation. In general, the process of manufacturing VLP-based vaccine consists of three stages. (a) Production stage: Includes cloning of the viral structural genes of interest and expression of viral proteins with self-assembling ability in a suitable expression platform (HEK293T cell line, a mammalian expression system). At the end of this stage, the VLPs are collected in the form of particles that do not have infectious properties. (b) Purification stage: Briefly consists of downstream processing such as clarification, purification and polishing to finally obtain purified intact VLPs without residual host debris. (c) Formulation stage: Where adjuvant and additional ingredients are added to the vaccine formulation to finally achieve a safe, efficient and effective product for vaccination. Reprinted from Nooraei et al.[68] VLPs: virus-like particles.

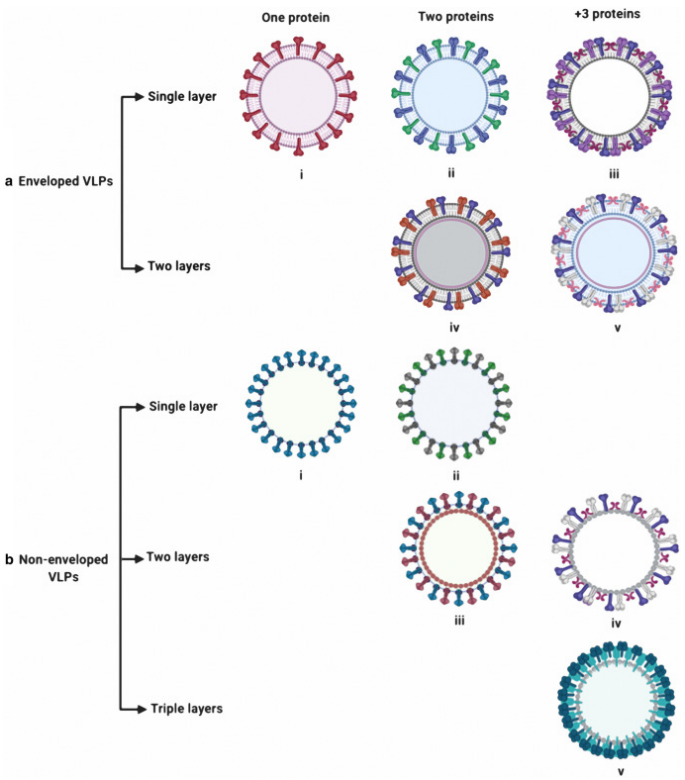

Figure 8. Classification of various VLP structures. (a) For enveloped VLPs, expression of one (i) or two (ii) glycoproteins will form a single layer, as demonstrated by the expression of influenza virus haemagglutinin alone or co-expression of both haemagglutinin and neuraminidase, respectively. Expression of more than three glycoproteins (iii) will also form a single layer VLP. Double layered enveloped VLPs can be formed by the multiple glycoproteins on their surface that can have two (iv) or more than three glycoproteins (v). (b) For non-enveloped VLPs, single layered non-enveloped VLPs can be assembled from a single protein (i) or two proteins (ii). Double layered non-enveloped VLPs can be assembled from two proteins (iii) or more than three proteins (iv). Triple layered VLPs have been assembled from more than three proteins (v). Reprinted from Nooraei et al.[68] VLPs: virus-like particles.

Ageing in 2023, VLPs play a crucial role in enhancing immune protection against influenza viruses in the elderly. In this study, researchers combined the haemagglutinin VLPs of influenza virus with glycosylphosphatidylinositol-anchored cytokines to prepare a novel influenza vaccine, termed as cytokine-conjugated VLP vaccine. The results revealed that this vaccine significantly enhanced protection against both homologous and heterologous influenza viruses in elderly mouse models. Park et al.'s study[87] underscores the potential of VLPs as adjuvants in enhancing the effectiveness of influenza vaccines in the elderly, particularly in boosting T-cell responses and providing cross-protection.

Lipid-based carriers

Lipid-based nano-delivery systems are commonly utilised as non-viral carriers for nucleic acids due to their stable nanostructures in physiological environments and their ability to fuse with negatively charged endosomal membranes, facilitating the effective delivery of nucleic acids. Lipids are characterised by their amphiphilic nature, consisting of a polar head group, a hydrophobic tail region, and linkers connecting the two domains. These lipid-based nanomedicine systems often incorporate additional lipid components, such as phospholipids, cholesterol, or polyethylene glycol components. The primary distinctions among these nanoparticles are contingent upon their lipid composition, synthesis parameters, and techniques employed for nucleic acid encapsulation.

LNPs and nanostructured lipid carriers have emerged as promising adjuvants in vaccine development and drug delivery. These carriers can protect encapsulated therapeutic agents from degradation, improve their stability, and facilitate controlled release at the target site, thereby enhancing their immunogenicity and bioavailability. It has been demonstrated that using lipid carriers as adjuvants can enhance the efficacy of mRNA vaccines, such as the successful SARS-CoV-2 mRNA vaccine, achieved by lipidising ionisable lipids with mRNA adjuvants, thereby enhancing the overall immunogenicity of the vaccine.[88] Additionally, nanostructured lipid carriers , as a binary lipid-based nanocarrier, comprising a mixture of solid and liquid lipids, have been shown to improve the delivery of lipophilic active ingredients, thereby enhancing their functionality and bioavailability.[89] The preparation process of mRNA vaccines typically involves combining the mRNA encoding the target protein with suitable lipid carriers to form LNPs. Subsequently, purification and processing are conducted through appropriate methods to obtain formulations for vaccine administration. These advancements underscore the potential of lipid carriers not only as drug delivery systems but also as versatile platforms for adjuvants and vaccine formulations.

Liposomes

Liposomes consist of phospholipids featuring polar head groups and non-polar tails, along with stabilising agents like cholesterol, enabling them to undergo spontaneous self-assembly into vesicles owing to their amphiphilic properties. Cationic lipids and zwitterions commonly exploit electrostatic interactions with negatively charged nucleic acids to create cationic liposomes, enhancing encapsulation efficiency. Smaller liposomes (≤ 100 nm) are more prone to evading phagocyte uptake. Nevertheless, the positive charge on nanoparticle surfaces may result in non-specific serum protein binding and immunostimulation, potentially causing toxicity. In response, pegylated cationic liposomes have been devised as alternative formulations. Three common techniques for liposome preparation include membrane hydration, solvent injection, and reversed-phase evaporation. These methods have been shown to result in efficient drug encapsulation, uniform particle size distribution, and prolonged stability. However, despite the benefits of liposomes as carriers for nucleic acids, their production necessitates intricate processes and the utilisation of organic solvents, potentially hindering their scalability for mass production.[13, 90-92]

Lipid nanoparticles

LNPs commonly consist of ionisable and cationic lipids, cholesterol, phospholipids, and polyethylene glycols, with ionisable lipids playing a crucial role in shielding nucleic acids from enzymatic degradation. Additionally, supplementary lipids such as phospholipids and cholesterol contribute to the stability of the formulation and facilitate membrane fusion, necessitating approximately 30–40 mol% parts of accessory lipids for efficient embedding of small interfering RNAs in LNPs. Polyethylene glycol lipids are created by conjugating hydrophilic polyethylene glycol polymers with hydrophobic lipid anchors to enhance circulation half-life and stability, as well as to prevent LNP clearance. Polyethylene glycol lipids are synthesised through the conjugation of hydrophilic polyethylene glycol polymers with hydrophobic lipid anchors, with the aim of enhancing the half-life and stability of LNPs and inhibiting their clearance. Low-molecular-weight polyethylene glycol lipids have been shown to decrease the binding of non-specific proteins. Furthermore, the proportion of polyethylene glycol lipids in the formulation influences the size of the particles.[93, 94]

In contrast to liposomes, LNPs have a micelle structure within the core of the particle. In addition, LNPs exhibit better kinetic stability and harder morphology compared to liposomes. Large-scale commercial preparation methods can yield more homogeneous LNPs. Ionisable LNPs have a near-neutral charge at physiological pH, but the amine groups on ionisable lipids become protonated and positively charged at low pH, allowing assembly with negatively charged phosphate groups on nucleic acids. After complexation, the pH can be adjusted to neutral or physiological pH for administration. Following *in vivo* injection, ionisable LNPs have the ability to exit the bloodstream and target specific tissues. These LNPs can subsequently bind to the surface of cells and be internalised through endocytosis. The presence of positively charged ionisable lipids aids in the escape from endosomes and interacts with the negatively charged lipid membrane of the endosome, leading to destabilisation and facilitating the release of nucleic acids.

Following extensive research, LNPs have been utilised in the administration of diverse vaccines, demonstrating a significant enhancement in both humoral and cellular immune responses. LNPs have been widely investigated as a potential mRNA

vaccine delivery platform in various clinical trials, including those for COVID-19, tumour, and influenza vaccines. In the year 2020, Pfizer's BNT162b2 vaccine, also referred to as Comirnaty, and Moderna's mRNA1273 vaccine, known as Spikevax, were granted emergency use authorisation for marketing.[95, 96] These mRNA vaccines, utilising LNP delivery of mRNA antigens, have played a pivotal role in combating the COVID-19 pandemic. Furthermore, the advancement of mRNA vaccine technology was recognised with the Nobel Prize in 2023. While LNPs have been extensively utilised in vaccine delivery, there remains a need for additional enhancements in addressing the toxicity, accumulation, and instability of liposomes in dynamic environments. These areas are currently the focal points of ongoing research efforts.[97, 98]

Inorganic nanoparticles

Inorganic nanomaterials function as innovative platforms that facilitate the connection between drugs and therapeutic targets. Inorganic nanoparticles are usually stable and non-degradable, and some of them are already in preclinical studies for use as vaccine delivery systems. Additionally, the loading and release of material molecules from inorganic nanoparticles can be controlled by internal or external factors such as temperature, pH, light, and magnetic fields. Gold, aluminum nanoparticles, and mesoporous silica can be used in vaccine delivery systems. Furthermore, inorganic nanomaterials can provide scaffolds for other biomaterials, such as polymers and lipids, offering unique structural and kinetic properties to build more robust and effective delivery carriers.[99] In practical biomedical applications, certain inorganic nanomaterials exhibit challenges in biodegradability within the body, leading to prolonged residence times and subsequent adverse effects. Consequently, research and development efforts focused on biodegradable inorganic nanomaterials are crucial for ensuring their safe utilisation in the biomedical field.

Gold nanoparticles are frequently employed as inorganic nanomaterial delivery systems.[100-102] Gold nanoparticles are one of the commonly used inorganic nanomaterials delivery systems.[103, 104] Gold nanoparticles have garnered attention as a potential antigen delivery system due to their ability to be precisely manipulated in terms of physicochemical properties through material synthesis and surface chemical modification. This allows for the shaping of specific immune responses. Gold nanoparticles facilitate antigen presentation and enhance adaptive immune responses by safeguarding antigens from degradation and forming nanovaccines with optimal particle size for efficient transport of antigens to lymph nodes.[105] Furthermore, it has been demonstrated that gold nanoparticles have the capability to augment antigen-specific antibody production through the activation of NLRP3 inflammasome and the stimulation of Th2-type cytokine production.[106] When employed as an adjuvant in the SARS-CoV-2 protein vaccine, gold nanoparticles were observed to enhance the production of antigen-specific IgG antibodies, yet were unable to elicit a cellular immune response adequate for combating viral infection.[107] Consequently, in order to broaden the application of gold nanoparticles, they are frequently utilised in conjunction with supplementary immunostimulants or subjected to surface modifications to bolster T-cell immune responses. In the realm of clinical research, the efficacy of utilising gold nanoparticles as adjuvants in dengue and SARS-CoV-2 vaccines is currently under investigation (NCT04935801, and NCT05113862).

However, there remain certain concerns regarding the use of gold nanoparticles. The cell toxicity of these nanoparticles has been frequently observed, with the level of toxicity being dependent on factors such as size, zeta potential, and surface functionalisation. It has been found that nanoparticles with diameters less than 12 nm can penetrate the blood-brain barrier, while those less than 30 nm can be taken up by cells through endocytosis. In particular, gold nanoparticles with diameters of 4, 12, and 18 nm have been shown to be non-toxic in human leukaemia cell lines. Conversely, nanoparticles with diameters as small as 1.4 nm have been found to induce oxidative stress, mitochondrial damage, and necrosis in cells.[108-111]

Mesoporous silica nanoparticles (MSNs), known for their high loading capacity and facile surface modification, have also garnered attention as potential vaccine delivery systems.[112] Through the integration of iron oxide nanoparticles, Lee et al.[113] have successfully developed hollow MSNs with enlarged mesopores for enhanced vaccine delivery. Poly(ethyleneimine) was used to modify the surface of MSNs, enabling efficient loading of proteins and enhancing antigen presentation. This system increased production of antigen-specific cytotoxic T lymphocytes, inhibited tumour growth, and improved survival in mice when used as a cancer vaccine delivery system. MSN-based materials have been investigated in research as potential delivery systems for various antigens. It has been observed that MSNs are gradually being explored in clinical trials as carriers for different pharmaceutical agents.[114, 115] Given the demonstrated efficacy of MSNs as a vaccine delivery platform in preclinical investigations, it is anticipated that vaccines utilising MSNs as a delivery system will soon undergo approval for clinical trials.

Polymer-based carriers

Polymer nanoparticles represent a colloidal system with sizes typically ranging from 5 to 1000 nm, with a more common size range of 100–500 mn[116, 117] (**Figure 6**). This term is a collective noun used to describe any type of nanosized polymer particles, particularly polymer nanospheres and nanocapsules. Polymer nanospheres are matrix particles, meaning the entire particle mass is solid, and they can serve as carriers for other bioactive molecules that can adsorb to the particle surface or be encapsulated within the particle.[118] These bioactive materials include drugs, genes, nucleic acids, fluorescence, and other functional materials. In contrast, nanocapsules are vesicle systems where the bioactive agent is confined to a water core and surrounded by a polymer shell.[119, 120] The advantages of polymer nanoparticles as active substance delivery systems include high drug encapsulation efficiency, higher cellular uptake compared to other particle delivery systems, enhanced stability of encapsulated active substances, and excellent biocompatibility with tissues and cells when prepared using biocompatible or biodegradable polymer materials.[55] Importantly, polymer nanoparticles can be designed to effectively deliver the drug to a target site and thus increase therapeutic results, minimising

the side effects.[117] Disadvantages of polymer nanoparticles include possible nonbiodegrad-ability, fragileness, higher manufacturing costs, toxic solvent residuals among others.[121] Polymer nanoparticles can be divided into two types, one is natural polymer nanoparticles, such as proteins and polysaccharides, which possess excellent biocompatibility and biodegradability, and are commonly used as vaccine carriers or packaging materials.[122, 123] The other category is synthetic polymer nanoparticles, which are typically composed of synthetic polymers and can achieve specific drug release kinetics and immunomodulatory effects through precise engineering design.[55] These two types of nanoparticles play crucial roles in vaccine development, providing important technological support for the development of safer and more effective vaccines. Generally, most of the natural polymers are biodegradable while some of the synthesis polymers are not.[117] Polymer carriers are highly regarded as adjuvants because they can activate multiple pattern recognition receptors. It has been demonstrated that when these carriers are used as vaccine adjuvants, they can synergistically induce both specific humoral and cellular immune responses. A notable example is a polymer that can simultaneously activate the Toll-like receptor pathway and the cyclic guanosine monophosphate–adenosine monophosphate synthase-stimulator of interferon genes pathway.[124] By conjugating this polymer with protein antigens, researchers have created an antigen delivery system for subunit vaccines that can elicit robust antigen-specific humoral and cellular immune responses without the need for additional adjuvants.[125] This innovative approach not only enhances vaccine efficacy but also simplifies the design and manufacturing process of vaccines, providing a promising strategy for inducing antigen-specific immunity to prevent infectious diseases.

Natural ingredient-based polymeric nanoparticles

Natural polymers are derived from various sources such as animals, plants, bacteria, and fungi, and are typically categorised into polysaccharides and protein polymers. Extensive research has been conducted on these polymers for their potential applications in drug delivery, as they are capable of forming scaffolds and serving as essential components of the extracellular matrix in living organisms.[124] Common examples of natural polymer nanomaterials include chitosan, dextran, cellulose derivatives, and protein nanoparticles. These natural polymers are known for their biocompatibility, which can help reduce immune system reactions to vaccines, and their ability to be degraded in the body to minimise toxin accumulation. However, functionalisation and chemical modification of natural polymers may be limited compared to synthetic polymers, affecting their performance in specific applications.[126-128]

Synthesising polymer nanomaterials

Synthetic polymer nanomaterials have demonstrated tremendous potential. These materials can serve as carriers for vaccines, facilitating precise delivery and sustained release, thereby enhancing vaccine immunogenicity and reducing side effects. Recent research indicates that synthetic polymer nanomaterials can modulate their immunogenicity through surface modification and functionalisation, better adapting to the host immune system's requirements.[129] Additionally, they can act as carriers to accurately deliver vaccine components to target cells or tissues, improving vaccine targeting and efficacy.[63, 99, 130] Compared to natural polymers, synthetic polymers offer higher controllability and stability. They can precisely control the vaccine release process by adjusting their structure and chemical composition. The relatively simple manufacturing process and lower production costs make scaled-up vaccine production feasible. Through synthetic polymer nanomaterials, safer and more effective vaccines can be developed, customisable for different diseases and immunological needs, offering broad application prospects.

Poly(lactic-co-glycolic acid) (PLGA) is a commonly used synthetic polymer material known for its good biocompatibility and biodegradability.[131] It finds wide application in the vaccine field, where it serves as a carrier to enhance vaccine stability and immunogenicity. Additionally, PLGA nanoparticles can be utilised in drug delivery systems to achieve sustained drug release, thereby enhancing therapeutic efficacy. PLGA nanoparticles can effectively target dendritic cells (DCs) with antigens and immune stimulatory molecules (such as adjuvants), owing to the crucial role of DCs in initiating anti-tumour immunity. By targeting DCs, PLGA nanoparticles can promote robust, specific, and durable anti-tumour T-cell responses. Its physicochemical properties, such as molecular weight, hydrophilic/hydrophobic balance, and crystallinity, can be modulated by altering its composition and preparation process.[132] This tunability enables PLGA nanoparticles to customarily control the release rate of antigens and adjuvants, thereby optimising immune responses. The mechanisms by which PLGA nanoparticles are taken up by DCs include uptake through phagocytosis into the cells and subsequent release of encapsulated antigens within acidic endosomes and lysosomes. PLGA holds significant potential, particularly in enhancing vaccine efficacy, reducing the number of injections, and lowering costs. Examples such as HPV vaccines, influenza vaccines,[133] COVID-19 vaccines,[134] and hepatitis vaccines demonstrate the use of synthetic polymer nanomaterials as carriers or auxiliary materials, enhancing vaccine stability, immunogenicity, and targeting. These instances underscore the widespread application of synthetic polymer nanomaterials in the vaccine domain, showcasing their potential in improving vaccine efficacy, enhancing stability, and achieving targeted delivery.

Nonbiomimetic materials

The strategic integration of nanomaterials and biomimetic approaches has the potential to alter the immunogenicity of various nanomaterials by transitioning them from a distinct state to a concealed state, thereby evading detection by the host's immune system. Recent studies have shown that this concealment can be achieved through the attachment of specific recognition molecules, such as aptamers, peptides, and antibodies, onto nanoparticle surfaces. However, the intricate nature of biomolecules may present obstacles to accurately replicating their functions in nanomedicine, prompting the

exploration of membrane biomimetic technologies. Biofilm serves as a nanodrug carrier, preserving the intrinsic physical and chemical characteristics of nanomaterials while imparting distinctive biological properties. Recent studies on membrane-derived research suggest that the laborious process of surface modifications and engineering can be streamlined through the use of membrane coatings. Furthermore, the selection of membrane type as a biological interface allows for the customisation of properties in nanomedicines. Furthermore, the manipulation of membrane types serves as a valuable biological interface, imparting various properties to nanomedicines. Specifically, red blood cell membrane-coated nanoparticles exhibit prolonged *in vivo* circulation, platelet membranes provide targeting capabilities, leukocyte membrane-shielded nanoparticles demonstrate endothelial delivery properties, and cancer cell membrane coatings enable homologous tumour targeting in nanomedicine. This targeting strategy holds significant importance in the field. Moreover, in addition to membrane biomimetic technology, various alternative strategies such as extracellular vesicle biomimetic methods,[135, 136] biomacromolecule-mediated biomimetic strategies,[137, 138] viral biomimetic vectors,[139] fungus-based systems,[140] and three-dimensional printing strategy[141] have been employed to achieve precise and effective utilisation of nanomaterials.

HBV is a primary aetiological factor for the development of cirrhosis and liver cancer. Vaccination with the HBV vaccine has been shown to be an effective measure in preventing the transmission of the virus and reducing the incidence of liver cancer. Wang et al.[142] have innovatively enhanced the traditional HBV vaccine by incorporating a bionic nano-platform to design the NP-pre S1 vaccine using bionic nano-ferritin material. This novel vaccine formulation targets different SIGNR1$^+$ antigen presenting cells to enhance the immunogenicity of the pre S1 domain of the HBV surface protein, leading to increased antibody production. In chronic HBV mouse models, this vaccine demonstrates promising preventive and therapeutic effects. Wang et al.[143] employed polydopamine to functionalise biomimetic nanoparticles with viral antigens and Toll-like receptor agonists on the surface of red blood cells, resulting in a notable augmentation of the antiviral immune response within the organism. Given the ability of red blood cells to migrate to the spleen and engage with antigen-presenting cells, the attachment of biomimetic viral nanoparticles to red blood cells facilitates the effective delivery of viral antigens to DCs. In comparison to the control group, the utilisation of biomimetic nanoparticle-modified red blood cells effectively induced maturation and activation of DCs, production of S1-specific IgG antibodies, and elicited T cell immune responses in mice. Due to its ease of manipulation and versatility in accommodating diverse molecular structures, polydopamine can be utilised as a versatile platform for the development of vaccines targeting a wide range of diseases. Nonbiomimetic nano-vaccines are anticipated to emerge as the predominant form of nanovaccines in the next generation. However, their intricate preparation procedures, challenges in large-scale production, complex composition, and elevated production expenses currently impede their widespread clinical utilisation.[144]

Other applications of nanotechnology in vaccines

In the field of vaccine development, the application of technologies beyond nanoparticles is also crucial. These technologies can significantly enhance the immunogenicity, stability, and targeting of vaccines. For instance, the combination of nanoemulsions and nanocarriers can effectively improve the release efficiency of antigens, while nanofibres and nanogels show great potential in drug delivery and tissue engineering. Nanogels are nanometer-scale hydrogel structures made from polymer materials, characterised by their high water-holding capacity. Typically ranging in size from 1 to 1000 nm, nanogels have high water content and flexible structures, making them suitable for delivering vaccine components such as proteins, peptide antigens, and nucleic acids. Chitosan nanogels and polyethylene glycol-modified nanogels can effectively enhance vaccine stability and immunogenicity while achieving targeted delivery through surface modifications. The unique advantages of nanogels, including sustained and controlled release and targeting capabilities, make them an ideal choice for vaccine development. In 2023, Ji et al.[145] proposed using nano-crosslinking technology to construct modular hydrogel vaccines, which effectively inhibited tumour growth and metastasis, showing particular promise in preventing cancer recurrence and metastasis after surgery. By incorporating pH-sensitive groups, chitosan nanogels can facilitate the slow or controlled release of vaccine components, thereby enhancing immune responses. For example, phosphorylated chitosan, a water-soluble pH-sensitive variant, has been used to deliver ovalbumin antigens, with experimental results indicating its ability to form a gel network containing ovalbumin, promoting sustained antigen release and long-term immune stimulation.[146] Moreover, nanofibres are recognised for their high specific surface area and excellent mechanical properties. Typically made from polymers, inorganic materials, or biomaterials, they offer a wide range of applications. Professor Joel H. Collier and his team at Duke University designed a self-assembling peptide nanofibre material, Q11, and their research found that combining the HIV envelope protein gp120 with the self-assembling peptide nanofibre Q11 could induce the production of antibodies with greater breadth and functionality.[147] These innovative nanomaterials provide new avenues for vaccine development, advancing the pursuit of safer and more effective vaccine strategies.

In summary, VLPs can elicit a robust immune response, are non-replicating and non-infectious, and offer a foundation for large-scale manufacturing (**Table 2**). Nevertheless, viral entities have the potential to induce significant inflammation, resulting in tissue necrosis and other detrimental consequences. Lipid-based nanovaccines exhibit favorable biosafety profiles, well-established industrialisation technologies, and efficient mass production capabilities. Nevertheless, challenges persist in the clinical implementation of liposome nanovaccines, including suboptimal immune carrier adjuvant functionality, limitations in loading large molecule drugs, and inefficient cellular uptake. Current research efforts are concentrated on regulating the lipid composition of liposomes and modifying their surface properties to address these issues. Polymer nanoparticles exhibit biodegradability, favorable biocompatibility, safety, and controlled release properties. Additionally, certain polymers can

serve as adjuvants to augment the immune activation efficacy of vaccines. Biomimetic nanomaterials exhibit versatility in effectively transporting antigens to specific locations, offering the benefits of superior biocompatibility and prolonged *in vivo* circulation. Nonetheless, the intricate and costly nature of their preparation process often hinders mass production.

This review is subject to certain limitations. Firstly, while there exists a wide array of nanoparticles, this paper focuses solely on the predominant types, omitting some lesser-known variants. Secondly, the discussion on the applications of nanotechnology in vaccines is limited to nanoparticles, with other potential applications receiving minimal attention.

Table 2. Advantages and limitations of various nanotechnologies

Type	Advantage	Limitation
Virus-like particles	High immunogenicity; Self-assembly; Relatively safe (no viral DNA)	Higher production costs; Complex manufacturing
Lipid carriers		
Liposomes	Good biocompatibility; Strong drug encapsulation; Immunoadjuvant	Stability issues; Complex production; Low uptake
Lipid nanoparticles	Protect nucleic acids; Enhanced stability	Insufficient adjuvant function; Complex manufacturing
Inorganic nanoparticles	Enhanced stability; Controllable release	Biocompatibility issues; Long-term toxicity unclear
Polymer-based carriers		
Natural polymer nanoparticles	High loading; Biodegradable	Limited functionality; Production costs
Synthesised polymer nanomaterials	Targeted delivery; Low costs	Potential toxicity; Non-biodegradable
Nonbiomimetic materials	Modifies immunogenicity; Unique biological properties	Complexity in biomolecule replication; Surface modification challenges
Nanogels	High water content; Sustained release; Enhanced stability	Complex preparation; Biocompatibility issues
Nanofibres	High surface area; Multifunctional	Complex manufacturing; Stability issues

Conclusion

Conventional vaccines exhibit limitations such as inadequate stability, low immunogenicity, challenges in targeted delivery, and the potential for adverse reactions due to the necessity of large doses of adjuvants, thereby hindering their ability to fully address societal demands. The emergence of nano-based vaccines, facilitated by advancements in materials science, has enabled the rapid development of vaccines capable of eliciting durable and potent immune responses through strategies such as carrier design and surface ligand modification. This innovative approach holds promising clinical utility. Researchers continue to encounter several challenges that must be addressed. Primarily, optimising the biological characteristics of nanomaterials and mitigating potential risks are essential hurdles to overcome. Additionally, the mechanisms through which various nanomaterials elicit immune responses in organisms remain poorly understood. Establishing correlations between the physicochemical properties of nanomaterials and their immunological effects is crucial for advancing research and applications in this field. Moreover, there is currently a deficiency in precise and efficient methods of observation and research systems for investigating the biological impacts of nanomaterials. It is imperative to advance the development of pertinent in situ characterisation techniques, advocate for the systemic immunology concept, conduct comprehensive analyses of immune responses to vaccination utilising high-throughput techniques, systematically elucidate the mechanisms by which existing nanomaterials affect immunity, and establish a dependable theoretical foundation for the exploration and creation of novel nanomaterials with immunological properties. As nanotechnology advances and production processes are optimised, the potential for a wider range of vaccines to be developed increases. The ongoing clinical transformation of nanovaccines offers hope for addressing currently challenging clinical diseases.

Author contributions

MC, YC, CX and GR conceived the review; LG, JH and XZ supervised the review; and MC, YC, and GR wrote the manuscript; YC prepared the figures. All authors read and approved the final version of the manuscript.

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Conflicts of interest statement

The authors declare that they have no conflicts of interest.

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