Mechanistic Exploration of Gold Nanoparticles as Adjuvants in Enhancing Vaccine Responses to Infectious Pathogens

Nida Wazir[1,*], Qamar un Nisa[2], Ayah Talal Zaidalkilani[3], Ammar AL-Farga[4] and Shahid Hussain Farooqi[5]

[1]Department of Pharmacology and Toxicology, University of Veterinary and Animal Sciences, Lahore, Pakistan
[2]Department of Pathology, University of Veterinary & Animal Sciences, Lahore, Pakistan
[3]Faculty of Pharmacy and Medical Sciences, Department of Nutrition, University of Petra, Amman, Jordan
[4]Department of Biological Sciences, College of Science, University of Jeddah, Jeddah, Saudi Arabia
[5]Department of Clinical Sciences, KBCMA College of Veterinary and Animal Sciences, Narowal, Sub-campus UVAS-Lahore, Pakistan
*Corresponding author: drnidawazir2023@gmail.com

Abstract

Improvement in the development and administration of vaccines has incorporated with the use of nanoparticles. In this regard, Gold Nanoparticles (GNPs) are considered for numerous good reasons. They are resistant to chemical reactions, can bind to small molecules and are relatively easy to produce hence being suitable for-vaccine engineering. Also, GNPs are useful in disease diagnosis and research because of their swift circulation in the body and their response to detection. Among their properties, the immunostimulant effects and increasing the efficiency of vaccines are most valuable. These GNPs can also incorporate antigens, which will guarantee the delivery of vaccines to a particular target tissue. When formulated with antibodies they can be targeted at delivering to a tissue or cell. These characteristics include size, structure, cost, and optical properties governance of their interaction with the immune system. Some findings indicate that GNPs can act to activate immune cells that include dendritic cells, T cells and B cells. This chapter's main goal is to demonstrate how GNPs are used to create nanovaccines that protect against a variety of infectious agents, including bacteria, viruses, and parasites. Even though GNPs have drawbacks and a low but noticeable amount of toxicity, their advantages make it worthwhile to devote more time and effort to creating innovative vaccine approaches.

Keywords: Nanoparticle immunology, Gold nanoparticle, GNP, Nanovaccine, Nanovaccine immunity

Cite this Article as: Wazir N, Nisa QU, Zaidalkilani AT, AL-Farga A and Farooqi SH, 2025. Mechanistic exploration of gold nanoparticles as adjuvants in enhancing vaccine responses to infectious pathogens. In: Nisa ZU, Altaf S, Zahra A and Saeed K (eds), Nanobiotech in Holistic Health: Innovations for Integrated Well-being. Unique Scientific Publishers, Faisalabad, Pakistan, pp: 162-169. https://doi.org/10.47278/book.HH/2025.370

A Publication of Unique Scientific Publishers

Chapter No: 25-024

Received: 09-Jan-2025
Revised: 01-Apr-2025
Accepted: 09-May-2025

Introduction

The vaccines have come a long way from the infectious diseases to cancer vaccines. This development is primarily attributed to the growth of nanotechnology. Nanoparticles (NPs) have been proven useful in vaccines production. Another crucial component of vaccine development is selecting the proper adjuvant — the substance used to jump start an immune reaction. Scientists have investigated various nanoparticles in order to evaluate their immunomodulatory potential in other disease models. These nanoparticles not only enhance the immunogenicity of the vaccine, but also have the function of delivery for the vaccine. Distribution technique, shape, size, and amalgamation of the antigen into the NP have an impact on the proficiency of it (Comber & Bamezai, 2015; Salazar-González et al., 2015). Much research has been conducted in the recent years to understand how nanoparticles modulate immune system and how they can enhance immunity, minimize adverse reactions and eliminate side effects (Frey et al., 2018; Kelly et al., 2019). Out of all the tremendous number of nanoparticles that have been involved in various vaccine endeavors, one can distinctively remember gold nanoparticles (GNPs). Because of their facility in their labeling with antigens & due to their ease in chemical point of view, GNPs are useful in preparation of vaccinations (Fai & Kumar, 2021). Some of the unique properties that are attributed to high surface area volume ratio of GNPs are peculiar plasmonic excitation, strong binding affinity and electronic structure (Personick et al., 2011). Additionally, it allows GNPs to engage with many types of ligands and functionality groups to improve the steadiness of the functionalized GNPs. Due to low toxicity of colloidal gold, magnetic and optical characteristics, it is applied in the treatment of multiple diseases (De Matteis et al., 2020). In some trials, multifunctional GNPs have been used in aggregation with FDA-endorsed antibiotics and antimicrobials (Fan et al., 2019). Also, GNPs conjugated with antigenic peptides have been able to effectively elicit high titer pathogen specific antibodies (Quach et al., 2018).

To achieve a specific and planned release at the intended target site, the researchers can employ GNP based systems to deliver drugs or antigens (Lee et al., 2018). These GNPs can be modified in a way that particular molecules or antibodies can attach themselves to the GNPs' surface in order to significantly increase the selectivity with which they bind target cells. Gold nanoparticles upon interaction with biological systems lead to some changes in that system as well as in themselves as indicated by figure 1. It increases the immune reaction at the area of

point injection while decreasing detrimental effects in other areas of the body (Kalishwaralal et al., 2020). Moreover, GNPs have additional immunoadjuvant effects that enhance the immune reaction. Researchers have the potential to authorize the immunogenicity of GNPs by altering shape, charge, size and surface disposition when explore a biological system (Dykman & Khlebtsov, 2019).

Key Characteristics and Roles of Gold Nanoparticles in Vaccine Development

As potent antigen trailers in immunization, gold nanoparticles (GNPs) are also extensively used. They could be considered perfect because they are easily formed, have unique chemical and physical properties and are reasonably non-toxic (Fan et al., 2020). Resultantly, these nanocarriers need to be directed straight at the definite immune cells or locations. The immunological response or vaccination requires the provocation of immune-related genes, antigen processing, antibody generation, cytokine creation and T cell activation, all of which are facilitated by this direct transport (Pati et al., 2018). GNPs fall under the class of tiny, highly surface-aread nanoparticles that can breach barriers and blood arteries. Their chances of attaining their destination are increased because they can be captivated by cells (W. Wang et al., 2020). Furthermore, GNPs are environmentally assisting because of their degradability in water and their chemical inertness. They may be modified with other peptides and compounds, thus they can be presented in various forms and to be slightly more sophisticated. Also, nanoparticles are stable, which enable broad areas of utilization as shown in figure 2 (Amina & Guo, 2020).

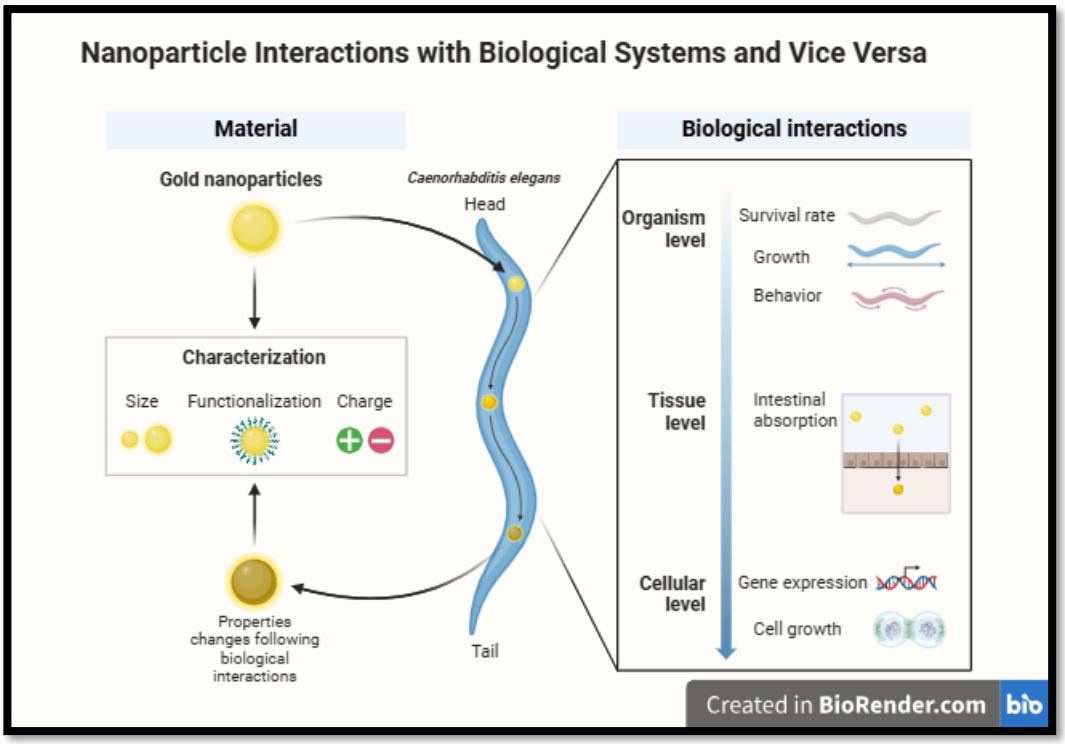

Fig. 1: Change in properties of Gold Nanoparticles upon interation with Biological Systems and vice versa

Viral genes can be utilised in vaccine production to incorporate GNPs into viral structures called virus like particles (VLPs) (Wang et al., 2016). It can also be adsorbed to proteins and polysaccharides before use as an antigen. According to past research, the gold sugar nanoparticles have immunomodulatory effects. Furthermore, GNPs also boost the immune system which makes them preferable in preparing vaccines. Due to these characteristics, GNPs are presented as the media technology desired to be applicable in medication delivery, injection, and disease diagnosis (Dykman, 2020).

Impact of Gold Nanoparticle Morphology on Immune Responses

The size and form of the GNPs have great influence on the manner in which they behave in a host's immune system. Most of these characteristics determine the manner of functioning of these proteins as the enhancer and in modulation of the immune system for various illnesses (Kohout et al., 2018). For example, in bone marrow-derived dendritic cells (BMDCs), the rod-shaped GNPs increase interleukins, particularly IL-1b and IL-18. However, when normal cells, granulocyte macrophage colony-stimulating factor (GM-CSF), IL-6, and IL-17 are present, the spherical GNPs are active in activating cytokines (Cai et al., 2022). (Niikura et al., 2013) studied on the impact of shape of GNPs; spherical, rod and cube on the immune system. Comparing their study's findings, the scientists noted that these nanoparticles' capacity to generate antibodies or TNFα depends critically on their surface area to size ratio. When it came to cytokines like IL-6 and IL-12, the largest GNPs-about 40 nm in size-were more effective than smaller or other nanoparticles. The antibodies that GNPs form, are similarly prejudiced by their chemical makeup. It is conjectured that changes to the particle surface, such as roughness, antigen coating or hydrophobicity influence the type and degree of immunity (Farfán-Castro et al., 2021). Besides, the location of GNPs within cells is also inclined by their size. For instance, the larger nanoparticles, with a size of between 5.5 and 8.2 nm, aggregate in the cytoplasm; and 2.4 nm nanoparticles are internalized through endocytosis. However, GNPs with size >18-20 nm do not enter the cells at all (Mulens-Arias et al., 2020).

Effect of GNPs on Dendritic Cells, Macrophages, and Natural Killer Cells

Dendritic Cells

Dendritic cells (DCs) are critically important for initiating immune response and GNPs exert a robust immunomodulatory effect on DCs. Researchers can attach several chemicals to the GNPs surface in order to thus support the immune response system and make it even more clumsy to the target DCs. (Cunha-Matos et al., 2016). In one study, animals containing *Listeria monocytogenes* antigens received DCs which have been loaded with GNP. This strategy generated an effective Th1 immunity which was attracting CD8+T cells and killer cells and the vaccine was much more effective than conventional approaches, proving the GNP system's capability of strengthening the immune system (Rodriguez-Del Rio et al., 2015). An endotoxin- like effect is observed in BMDCs that release proinflammatory cytokines such as IL-6, TNF-α, and IFN-γ when they take up GNPs. These cytokines participate in initiating as well as maintaining immunological reactions (El-Sayed et al., 2021). In addition, GNPs can stimulate neutrophil to release extracellular decoy that causes immune reaction through DNA receptors such as Toll-like receptor 9 (TLR9) (Staroverov et al., 2019). It has become evident that GNP coatings interfere with how well they interact with dendritic cells. Namely, it points at GNPs that may interact with monocyte-derived dendritic cells (MDDCs), if the latter are coated with polyethylene glycol (PEG) or polyvinyl alcohol (PVA) or both. Although the PEG coating improved the aspect of cells creating TNF-α, it reduced the quantity of GNPs these cells could uptake. On the other hand, GNPs conjugated with PVA or PEG-PVA blend caused higher levels of IL-1b release which was in equal proportion to the amount produced. However, neither of these coatings seemed to have a direct effect on the MDCs themselves; namely, their phenotype or functionality (Fytianos et al., 2015)

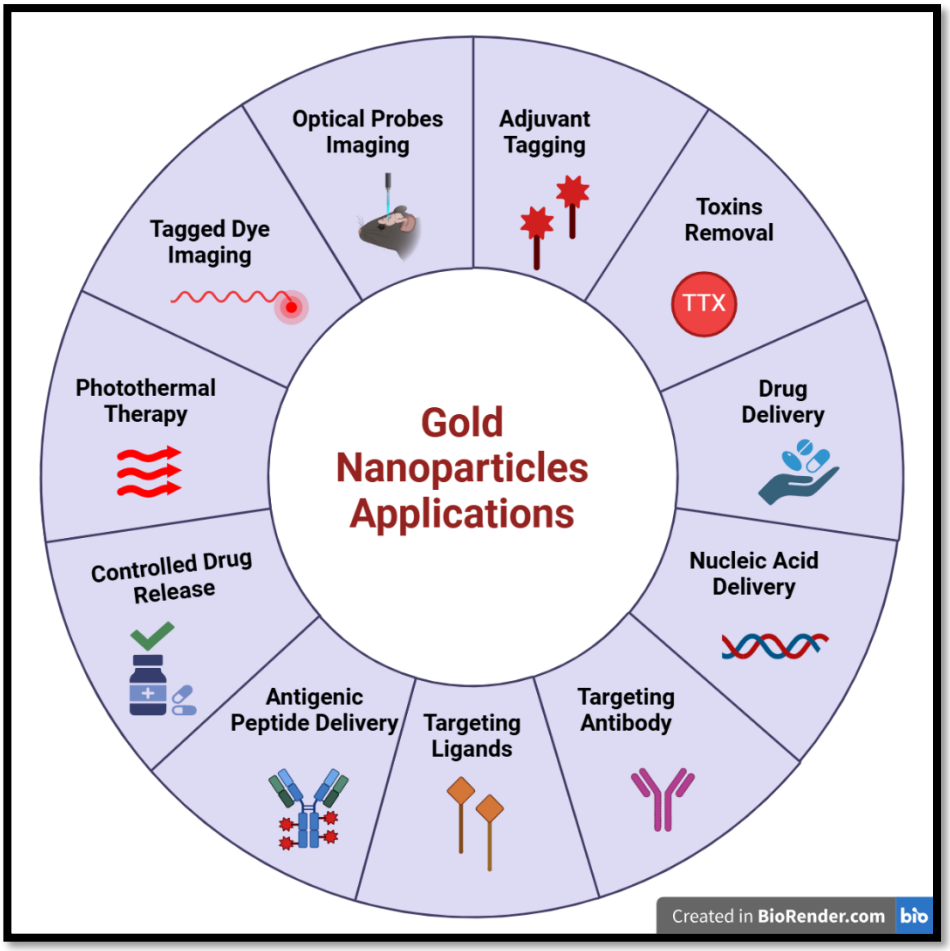

Fig. 2: Applications of Gold Nanoparticles

Macrophages

Macrophages have a versatile role in distinguishing and creating the immune system, as well as preventing diseases from arising. (L. Wang et al., 2020). It has also been discovered that the internalization of GNPs enhances the macrophage ability to communicate with other cells, a key characteristic of tissue repair and regeneration (Luan et al., 2020). Notably, GNPs inhibit the ability of macrophages to store proinflammatory cytokines. GNPs were reported to have similar impact on the spleen cells that are sensitized with LPS, which attacked the immune system, or show that they can modulate excess inflammation (Kingston et al., 2016). The capture of free radicals from these cells is again an important attribute of macrophages of GNPs. For instance, GNPs have been found to reduce the levels of Reactive Oxygen Species (ROS) in LPS stimulated mouse macrophages in a concentration dependent-manner. This decline is related to the decrease of the synthesis of other cytokines, such as TNF-α and IL-17 involved in inflammation. They describe how GNPs may function as guardians of inflammation and present a new treatment approach (Jiao et al., 2016).

Natural Killer Cells

In the circulation, natural killer cells (NK) originating from the lymphoid progenitor lineage are strictly required for immunological shadowing. To induce infected cell death, they release perforins and granzymes. To guide the targeting of GNPs to NK cells, the mechanism of NK cell mediated antibody-dependent cellular cytotoxicity (ADCC) has been planned out. This is the manner through which antibody-tagged GNPs utilize as well as interact with NK cell receptors. This enhances the activation of NK cells to deliver performances (Jiao et al., 2016). A new approach of immunotherapy is a technique for employing PEGylated polyamidoamine dendrimers confined in GNPs to direct human ferritin heavy string into NK cells (Qu et al., 2019). Such an agent, when used to treat NK cells in vitro, gives GNPs a new shape that behaves anti-inflammatory, excitedly reducing IFN-γ emission (Elbagory et al., 2019).

Use of GNP in Antiviral Immunization

The researchers specifically prefer to use errand gold nanoparticles because of their multidisciplinary and viral vaccine characteristics. Their capacity to deliver and address particular cells makes them a unique method for vaccine manufacturing. In addition to enabling the transport of antigens, these nanoparticles provide the attached or core formulations optimal antiviral protection and enhance immune response. Besides their immunomodulatory properties, GNPs support the release and delivery of antigens for continuous effective immunization. Due to their qualities, they are an indispensable component of today's vaccines and a dynamic factor in the virus disease control system.

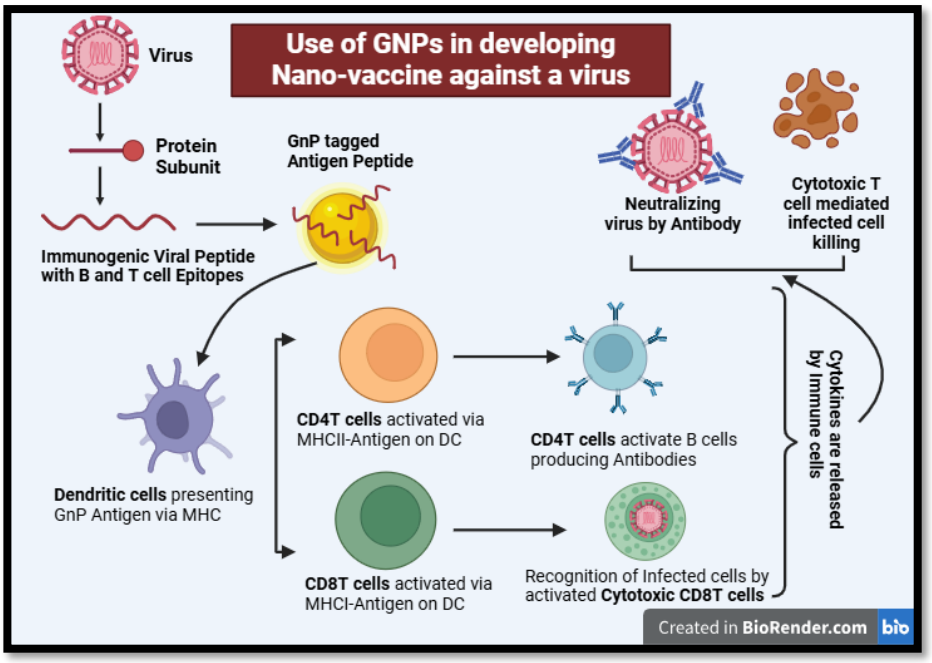

Fig. 3: A schematic representation of the use of GNPs in developing nanovaccines against a virus

Figure 3 explains how any conserved, immunogenic peptide that is separated from a virus and attached to GNPs to form a nanovaccine is studied on mice. Even when the vaccine is given, dendritic cells transport the peptide to CD4 helper T cells and CD8 cytotoxic T cells, resulting in immunological initiation. This causes B cells to become activated and helper T cells to thrive clonally, which produces certain antibodies. While cytokines produced during this immune response help in forming either Th1 or Th2 responses depending on the degree of inflammation, cytotoxic T cells trace and eradicate infected cells. Long-lasting immunity is recognized by the development of memory B and T cells and the efficient neutralization of the virus by the formed antibodies. This ensures a rapid immune response to future infections by the same virus, possibly averting severe illness (Carabineiro, 2017).

1. HIV

The envelope glycoprotein of the human immunodeficiency virus (HIV) known as gp120, has a significant cluster of mannose-rich glycans, which is identified by 2G12-like antibodies. Gold nanoparticles that could link with 2G12 were formed using a monolayer covering of self-assembled oligomannosides, which looked like gp120 (Abia et al., 2013). Furthermore, GNPs coupled to thiol-terminated oligosaccharides have been employed in the formation of HIV vaccines (Chiodo et al., 2015). When coated with a synthetic fractional structure of several mannosidases, GNPs with a size of 2 nm show exceptional binding to anti-HIV antibody-like 2G12. Alpha helices or beta strands can be formed by conjugating the third variable region (V3 peptide) of HIV1's gp120 to GNP. In addition to manufacturing a large quantity of particular neutralizing antibodies in rabbits, this increases their steadiness against peptidase dilapidation in any form (Xu et al., 2012).

2. Hepatitis B

Gold nanoparticles (GNPs) have been publicized to be an auspicious platform for diagnosis and cure of hepatitis B. One such ground-breaking tactic involves extraction of hepatitis B virus (HBsAg) DNA from epidermal cells with a GNP coated gene gun that has been shown to be a calming strategy (Negahdari et al., 2019). The second important application involves the use of GNPs as an adjuvant with plasmid DNA

encoding HBsAg, injected into mice. Incorporation of GNPs not only increases the immune response but also expedites antibody production such that animals reach peak antibody levels sooner than with standard methods. Additional in vitro studies have also shown further advantages. When RAW 264.7 macrophages were treated with HBsAg conjugated gold nanocages, antigen uptake and processing capacity were increased. Along with this secretion of interleukin-4 (IL-4) further implies an increased immune response. In addition, new progress shows that GNPs are particularly useful for the detection and identification of HBsAg, which represents a new and powerful method for monitoring hepatitis B infection (Kim et al., 2018).

3. Hepatitis C

Using conductive plasmonic gold nanoparticles (GNPs), a research team has unveiled a new approach to a highly effective hepatitis C DNA vaccine. The formation of pores in the cell membrane allows this unique opening, which greatly increases absorption of the DNA vaccine. The findings were surprising: The mice in the group that got GNP had the gene at 100 times the level of the control group that didn't get GNP. Mice treated with GNPs also have enhanced immune responses, with markedly increased humoral (antibody-mediated) and cell mediated immunity (Draz et al., 2017).

4. Dengue

Domain III envelope glycoprotein derived from serotype 2 of the dengue virus to bind gold nanoparticles (GNPs) of different sizes (20, 40 and 80 nm) was used. Subcutaneous administration to BALB/C mice 3 times was done. The results were encouraging: Mice developed serotype specific neutralizing antibodies and were highly immunized. The researchers then judiciously adjusted the height and size of the GNP, and they found this transformed the vaccination level in the animals. Moreover, the treatment results in profound changes in the immune system: spleen cell proliferation and activation as well as helper T cell and cytotoxic T cell expansion. In mice, synthesis of important cytokines such as IL-4 and IFN-γ was also increased (Quach et al., 2018). In another experiment, GNPs were joined with small interfering RNA (siRNA) that targeted dengue virus. This approach increases the anti-inflammatory effect by improving the stability and delivery of siRNA (Paul et al., 2014).

5. Influenza

Gold nanoparticles (GNPs) have been subjected to significant research in developing anti-influenza drugs. The conserved N-terminal extracellular domain of the influenza virus matrix protein 2 (M2e) peptide was used and linked to 12 nm GNPs (Tao et al., 2017). BALB/C mice were twice vaccinated with GNP conjugates. The results were encouraging: Mice that produced more IgG1 and IgG2 antibodies had better protection when challenged with the deadly PR8-H1N1 virus (Tao et al., 2014).

Many other vaccination processes have been developed to combat viral pathogens. A few of these are highlighted in Table 1.

Table 1: Use of GNP-based Nanovaccines against Viral Pathogens

No.	Antigen Type	AuNP Formulation	Immune Reaction Observed
1	Spike protein from Avian Coronavirus	Virus-like particles synthesized with 100 nm AuNPs	Improved antigen targeting to lymphoid tissues, enhanced spleen T-cell activation, higher antibody titers, and mitigated infection symptoms (Chen et al., 2016).
2	Gastroenteritis virus proteins	Conjugated with 15 nm AuNPs	Elevated levels of IL-6, IFN-γ, and IL-1β, enhanced macrophage and lymphocyte activity, and stronger humoral immunity through spleen follicle increase (Staroverov et al., 2011).
3	Glycoprotein from respiratory syncytial virus	Nanorods	Stimulated immune cell expansion, including activation of primary T-cells by dendritic cells (Stone et al., 2013).
4	Rabies virus glycoprotein (Moscow strain 3253)	Conjugated with 15 nm AuNPs	Elicited production of virus-neutralizing antibodies with high specificity (Bawage et al., 2016).
5	West Nile virus	Various shapes: 20–40 nm nanospheres, nanorods, and nanocubes	40 nm nanospheres produced the highest antibody titers; nanorods showed greater cellular uptake by dendritic cells and macrophages (Niikura et al., 2013).

Use of GNP in Antibacterial Immunization

Gold nanoparticles (GNPs) have been developed as an important vaccine and antigen delivery tool against a variety of bacterial diseases. A few of them are highlighted in Table 2. In some cases, they act as boosters to boost immune responses. For example, antibody against the N-terminal domain of the flagellin domain of *Pseudomonas aeruginosa* in combination with Freund's adjuvant and GNPs significantly augmented the IgG response. Similarly, combining 15 nm GNPs with two antigens from *Francisella tularensis* induced anti-tularemia sera by eliciting antibodies. GNPs were also used for the first time as an adjuvant to generate antibodies against surface antigens of *Yersinia pseudotuberculosis* (Staroverov et al., 2003). In addition, F1 antigens were coated with 15 nm GNPs and studies were performed on *Yersinia pestis*. This process led to high levels of IgG2a, interferon gamma and Th1 cell infiltration and hence improved immune responses (Gregory et al., 2012). Surface antigens of *Salmonella typhimurium* were coated with GNPs and were shown to have improved immunogenic properties after viral clearance. Additionally, GNPs from the capsular polysaccharide of *Neisseria meningitidis* were treated with non-immunogenic sugars to increase T cell activity, MHCII expression and overall immune responses (Fallarini et al., 2013).

Use of GNP in Anti-Parasitic Immunization

Stimulating the host's immune response, GNPs have become an important area of research on parasitic diseases. Many other vaccination

processes have been developed to combat parasitic pathogens. A few of them are explained in Table 3. In one study it was found that GNPs are capable of activating MHC I and II which are responsible for antigens presentation to the immune system. This stimulation activates a response from the leading actors of the immune system, CD4+ and CD8+ T cells. It was touched that the GNPs could enhance the host's immunity against parasitic infections and synthesize high titer of species specific antibody (Parween et al., 2011; Kumar et al., 2015).

Table 2: Use of GNP based Nanovaccine against Bacterial Pathogens

No.	Antigen Type	AuNP Formulation	Immune Reaction Observed
1	Listeriolysin O peptide (LLO91-99) from *L. monocytogenes*	Conjugated with AuNP	Increased CD4+ and CD8+ T-cell populations, NK cells, and dendritic cells; stimulated cytokine production (IL-12, TNF-α, IFN-γ, MCP-1) (Calderón-Gonzalez et al., 2016).
2	Outer membrane vesicles from *E. coli*	Conjugated with 30 nm AuNPs	Prompted rapid maturation of dendritic cells, higher-quality antibodies, and strong Th1/Th17 immune responses via IFN-γ and IL-17 (Gao et al., 2015).
3	Tetanus toxoid (*Clostridium tetani*)	25 nm AuNPs combined with saponin-based adjuvants	Oral delivery significantly improved mucosal immune responses in the presence of plant-derived adjuvants (Liu et al., 2018).
4	Tuberculin (antigens from various mycobacteria)	Conjugated with 15 nm AuNPs	Promoted robust antibody responses against diverse mycobacterial strains (Staroverov & Dykman, 2013).

Table 3: Use of GNP based Nanovaccine against Parasitic Pathogens

No.	Antigen Type	AuNP Formulation	Immune Reaction Observed
1	Recombinant protein from *Schistosoma mansoni*	Gold nanorods conjugated	Activated Th1 response; increased IFN-γ production by CD4+ and CD8+ T-cells; up-regulated MHCI/MHCII expression and IL-1β synthesis (Assis et al., 2018).
2	Surface protein Pfs25 from *P. falciparum*	Gold nanospheres, nanostars, nanocages, and nanoprisms	Produced high-affinity antibodies using nanospheres and nanostars with the highest reactivity; This demonstrated efficient suppression of parasite migration to mosquitoes (Kumar et al., 2015).
3	Merozoite surface protein fragment (19 kDa) from *P. falciparum*	17 nm AuNP combined with Alhydrogel®	Stimulated immune reactions against low immunogenicity peptides; preventing the invasion of parasites in-vitro (Parween et al., 2011).

Discussion and Future Perspectives

This chapter contends that gold nanoparticles hold a great promise in preventing and managing many diseases. GNPs have been on the rise in the synthesis and use in nano vaccine research and are considered as a possible solution to address some of the world's most deadly diseases. For this reason, GNPs are considerably safe for use by humans and this makes them a common ingredient in vaccines against many diseases ranging from infectious to cancerous diseases. GNPs are exceptional, and their affluence of application and manipulability are tremendously attractive to researchers. For example, GNPs are conjugated to isotopes, fluorescent tags or optical probes, and high-resolution optical imaging techniques are used to protect specific cells with antibodies or ligands. GNPs have also been shown as effective for use in biosensors and microscopy for detecting bacterial infections. GNP conjugated vaccines have been shown to give 100% survival rates against lethal viruses in animal studies. Yet many obstacles remain for the successful application of GNPs in vaccine development and clinical practice. First, we need to have a uniform, large scale production system for GNPs. It has been previously mentioned that charge, size and shape of GNPs can impact cell interaction, and thus these variables need to be considered carefully when manufacturing. GNPs from 15 to 50 nm have been most studied and nano shells have been used in many of these studies, but other shapes such as nanocages, nanorods and nano cubes have not been explored. This leaves a missing piece of our understanding of how different GNPs of different sizes and types impact the system.

One area that needs further research is the way that GNPs affect immune cells. Many exciting questions will be answered by detailed studies of how functional and nonfunctional GNPs interact with immune cells. GNPs biodistribution needs to be further investigated, particularly on non-targeted organs and cells. Studies tend to focus on organs, cells or infected pathogens, but overlook the possibility that GNP might accumulate in other organs. This brings us to the next important consideration: nanotoxicology. As GNPs are not irreversible, methods to reduce their toxicity or improve their elimination from the body need to be developed.

Conclusion

The peculiarities of GNPs such as increased number of accessible surface for a given volume, chemical inertness, low biotoxicity, and ability to modulate immune reaction make them a breakthrough in immunotherapy and vaccinology. Due to their diverse roles, these molecules may serve as immune enhancing agents and antigen carriers that enhance the efficacy of vaccines and decrease toxicity. Moreover, varying the size, morphology, and surface properties of GNPs, it is possible to stimulate powerful immune reactions with different types of immune cells such as natural killer cells, dendritic cells or macrophages. GNPs are incorporated into antiviral vaccines such as the flu vaccine, Hepatitis, Dengue and HIV, via enhancing antigen dispersion, promoting immune stimulation and increasing the duration of protection. Furthermore, because of their flexibility, they can couple with parasitic and bacterial antigens and encapsulate into virus like particles, which affords them potential for future enhancement in therapy as well as diagnosis. However, there are some challenges that have to be faced, for instance, one needs to produce GNP with high uniformity and scaled up production, toxicity and biodistribution, avoiding biocorona formation, which may compromise the efficacy of the nanoparticles. Further, for its broader applications, further research is required to understand more about the effects of GNP with immune cells and efficiency trials are needed to determinate the right dose and protocol to follow in order to avoid risks.

References

Abia, I., Peng, T., Mains, S., & Pohl, N. (2013). Design and synthesis of thiol-terminated oligosaccharides for attachment on gold nanoparticles: Toward the development of an HIV vaccine. Abstracts of Papers of the American Chemical Society, Amer Chemical SOC 1155 16th ST, NW, Washington, DC, 20036 USA.

Amina, S. J., & Guo, B. (2020). A review on the synthesis and functionalization of gold nanoparticles as a drug delivery vehicle. *International Journal of Nanomedicine*, *15*, 9823-9857.

Assis, N. R., Caires, A. J., Figueiredo, B. C., Morais, S. B., Mambelli, F. S., Marinho, F. V.,Oliveira, S. C. (2018). The use of gold nanorods as a new vaccine platform against schistosomiasis. *Journal of Controlled Release*, *275*, 40-52.

Bawage, S. S., Tiwari, P. M., Singh, A., Dixit, S., Pillai, S. R., Dennis, V. A., & Singh, S. R. (2016). Gold nanorods inhibit respiratory syncytial virus by stimulating the innate immune response. *Nanomedicine: Nanotechnology, Biology and Medicine*, *12*(8), 2299-2310.

Cai, F., Li, S., Huang, H., Iqbal, J., Wang, C., & Jiang, X. (2022). Green synthesis of gold nanoparticles for immune response regulation: Mechanisms, applications, and perspectives. *Journal of Biomedical Materials Research Part A*, *110*(2), 424-442.

Calderón-Gonzalez, R., Terán-Navarro, H., Frande-Cabanes, E., Ferrández-Fernández, E., Freire, J., Penadés, S., & Yañez-Díaz, S. (2016). Pregnancy vaccination with gold glyco-nanoparticles carrying Listeria monocytogenes peptides protects against listeriosis and brain-and cutaneous-associated morbidities. *Nanomaterials*, *6*(8), 151.

Carabineiro, S. A. C. (2017). Applications of gold nanoparticles in nanomedicine: recent advances in vaccines. *Molecules*, *22*(5), 857.

Chen, H.-W., Huang, C.-Y., Lin, S.-Y., Fang, Z.-S., Hsu, C.-H., Lin, J.-C.,Hu, C.-M. J. (2016). Synthetic virus-like particles prepared via protein corona formation enable effective vaccination in an avian model of coronavirus infection. *Biomaterials*, *106*, 111-118.

Chiodo, F., Enríquez-Navas, P. M., Angulo, J., Marradi, M., & Penadés, S. (2015). Assembling different antennas of the gp120 high mannose-type glycans on gold nanoparticles provides superior binding to the anti-HIV antibody 2G12 than the individual antennas. *Carbohydrate Research*, *405*, 102-109.

Comber, J., & Bamezai, A. (2015). Gold nanoparticles (AuNPs): A new frontier in vaccine delivery. *Journal of Nanomedicine and Biotherapeutic Discovery*, *5*(4), e139.

Cunha-Matos, C. A., Millington, O. R., Wark, A. W., & Zagnoni, M. (2016). Real-time assessment of nanoparticle-mediated antigen delivery and cell response. *Lab on a Chip*, *16*(17), 3374-3381.

De Matteis, V., Rizzello, L., Cascione, M., Liatsi-Douvitsa, E., Apriceno, A., & Rinaldi, R. (2020). Green plasmonic nanoparticles and bio-inspired stimuli-responsive vesicles in cancer therapy application. *Nanomaterials*, *10*(6), 1083.

Draz, M. S., Wang, Y. J., Chen, F. F., Xu, Y., & Shafiee, H. (2017). Electrically oscillating plasmonic nanoparticles for enhanced DNA vaccination against hepatitis C virus. *Advanced Functional Materials*, *27*(5), 1604139.

Dykman, L., & Khlebtsov, N. (2019). Gold nanoparticles in chemo-, immuno-, and combined therapy: review. *Biomedical Optics Express*, *10*(7), 3152-3182.

Dykman, L. A. (2020). Gold nanoparticles for preparation of antibodies and vaccines against infectious diseases. *Expert Review of Vaccines*, *19*(5), 465-477.

El-Sayed, N., Korotchenko, E., Scheiblhofer, S., Weiss, R., & Schneider, M. (2021). Functionalized multifunctional nanovaccine for targeting dendritic cells and modulation of immune response. *International Journal of Pharmaceutics*, *593*, 120123.

Elbagory, A. M., Hussein, A. A., & Meyer, M. (2019). The in vitro immunomodulatory effects of gold nanoparticles synthesized from hypoxis hemerocallidea aqueous extract and hypoxoside on macrophage and natural killer cells. *International Journal of Nanomedicine*, *14*, 9007-9018.

Fai, T. K., & Kumar, P. V. (2021). Revolution in the synthesis, physio-chemical and biological characterization of gold nanoplatform. *Current Pharmaceutical Design*, *27*(21), 2482-2504.

Fallarini, S., Paoletti, T., Battaglini, C. O., Ronchi, P., Lay, L., Bonomi, R.,Lombardi, G. (2013). Factors affecting T cell responses induced by fully synthetic glyco-gold-nanoparticles. *Nanoscale*, *5*(1), 390-400.

Fan, J., Cheng, Y., & Sun, M. (2020). Functionalized gold nanoparticles: synthesis, properties and biomedical applications. *The Chemical Record*, *20*(12), 1474-1504.

Fan, Y., Pauer, A. C., Gonzales, A. A., & Fenniri, H. (2019). Enhanced antibiotic activity of ampicillin conjugated to gold nanoparticles on PEGylated rosette nanotubes. *International Journal of Nanomedicine*, *14*, 7281-7289.

Farfán-Castro, S., García-Soto, M. J., Comas-García, M., Arévalo-Villalobos, J. I., Palestino, G., González-Ortega, O., & Rosales-Mendoza, S. (2021). Synthesis and immunogenicity assessment of a gold nanoparticle conjugate for the delivery of a peptide from SARS-CoV-2. *Nanomedicine: Nanotechnology, Biology and Medicine*, *34*, 102372.

Frey, M., Bobbala, S., Karabin, N., & Scott, E. (2018). Influences of nanocarrier morphology on therapeutic immunomodulation. *Nanomedicine*, *13*(14), 1795-1811.

Fytianos, K., Rodriguez-Lorenzo, L., Clift, M. J., Blank, F., Vanhecke, D., von Garnier, C.,Rothen-Rutishauser, B. (2015). Uptake efficiency of surface modified gold nanoparticles does not correlate with functional changes and cytokine secretion in human dendritic cells in vitro. *Nanomedicine: Nanotechnology, Biology and Medicine*, *11*(3), 633-644.

Gao, W., Fang, R. H., Thamphiwatana, S., Luk, B. T., Li, J., Angsantikul, P.,Zhang, L. (2015). Modulating antibacterial immunity via bacterial membrane-coated nanoparticles. *Nano Letters*, *15*(2), 1403-1409.

Gregory, A., Williamson, E. D., Prior, J., Butcher, W., Thompson, I., Shaw, A., & Titball, R. W. (2012). Conjugation of Y. pestis F1-antigen to gold nanoparticles improves immunogenicity. *Vaccine*, *30*(48), 6777-6782.

Jiao, P., Otto, M., Geng, Q., Li, C., Li, F., Butch, E. R.,Yan, B. (2016). Enhancing both CT imaging and natural killer cell-mediated cancer cell killing by a GD2-targeting nanoconstruct. *Journal of Materials Chemistry B*, *4*(3), 513-520.

Kalishwaralal, K., Luboshits, G., & Firer, M. (2020). Synthesis of gold nanoparticle: peptide–drug conjugates for targeted drug delivery. *Drug Delivery Systems*, *2059*, 145-154.

Kelly, H. G., Kent, S. J., & Wheatley, A. K. (2019). Immunological basis for enhanced immunity of nanoparticle vaccines. *Expert Review of Vaccines*, *18*(3), 269-280.

Kim, J., Oh, S. Y., Shukla, S., Hong, S. B., Heo, N. S., Bajpai, V. K., & Huh, Y. S. (2018). Heteroassembled gold nanoparticles with sandwich-immunoassay LSPR chip format for rapid and sensitive detection of hepatitis B virus surface antigen (HBsAg). *Biosensors and Bioelectronics*, *107*, 118-122.

Kingston, M., Pfau, J. C., Gilmer, J., & Brey, R. (2016). Selective inhibitory effects of 50-nm gold nanoparticles on mouse macrophage and spleen cells. *Journal of Immunotoxicology*, *13*(2), 198-208.

Kohout, C., Santi, C., & Polito, L. (2018). Anisotropic gold nanoparticles in biomedical applications. *International Journal of Molecular Sciences*, *19*(11), 3385.

Kumar, R., Ray, P. C., Datta, D., Bansal, G. P., Angov, E., & Kumar, N. (2015). Nanovaccines for malaria using Plasmodium falciparum antigen Pfs25 attached gold nanoparticles. *Vaccine*, *33*(39), 5064-5071.

Lee, J.-H., Cho, H.-Y., Choi, H. K., Lee, J.-Y., & Choi, J.-W. (2018). Application of gold nanoparticle to plasmonic biosensors. *International Journal of Molecular Sciences*, *19*(7), 2021.

Liu, J., Wang, J., Li, Z., Meng, H., Zhang, L., Wang, H., & Qu, L. (2018). A lateral flow assay for the determination of human tetanus antibody in whole blood by using gold nanoparticle labeled tetanus antigen. *Microchimica Acta*, *185*, 1-7.

Luan, Y., van der Mei, H. C., Dijk, M., Geertsema-Doornbusch, G. I., Atema-Smit, J., Ren, Y., Busscher, H. J. (2020). Polarization of macrophages, cellular adhesion, and spreading on bacterially contaminated gold nanoparticle-coatings in vitro. *ACS Biomaterials Science Engineering*, *6*(2), 933-945.

Mulens-Arias, V., Balfourier, A., Nicolás-Boluda, A., Carn, F., & Gazeau, F. (2020). Endocytosis-driven gold nanoparticle fractal rearrangement in cells and its influence on photothermal conversion. *Nanoscale*, *12*(42), 21832-21849.

Negahdari, B., Darvishi, M., & Saeedi, A. A. (2019). Gold nanoparticles and hepatitis B virus. *Artificial cells, Nanomedicine and Biotechnology*, *47*(1), 455-461.

Niikura, K., Matsunaga, T., Suzuki, T., Kobayashi, S., Yamaguchi, H., Orba, Y.,Ninomiya, T. (2013). Gold nanoparticles as a vaccine platform: influence of size and shape on immunological responses in vitro and in vivo. *American Chemical Society Nano*, *7*(5), 3926-3938.

Parween, S., Gupta, P. K., & Chauhan, V. S. (2011). Induction of humoral immune response against PfMSP-119 and PvMSP-119 using gold nanoparticles along with alum. *Vaccine*, *29*(13), 2451-2460.

Pati, R., Shevtsov, M., & Sonawane, A. (2018). Nanoparticle vaccines against infectious diseases. *Frontiers in Immunology*, *9*, 2224.

Paul, A. M., Shi, Y., Acharya, D., Douglas, J. R., Cooley, A., Anderson, J. F., & Bai, F. (2014). Delivery of antiviral small interfering RNA with gold nanoparticles inhibits dengue virus infection in vitro. *Journal of General Virology*, *95*(8), 1712-1722.

Personick, M. L., Langille, M. R., Zhang, J., & Mirkin, C. A. (2011). Shape control of gold nanoparticles by silver underpotential deposition. *Nano Letters*, *11*(8), 3394-3398.

Qu, Y., Li, Y., Liao, S., Sun, J., Li, M., Wang, D., & Luo, K. (2019). Linear and core-crosslinked glycopolymer-gadolinium conjugates: Preparation and their behaviors as nanoscale magnetic resonance imaging contrast agents. *Journal of Biomedical Nanotechnology*, *15*(8), 1637-1653.

Quach, Q. H., Ang, S. K., Chu, J.-H. J., & Kah, J. C. Y. (2018). Size-dependent neutralizing activity of gold nanoparticle-based subunit vaccine against dengue virus. *Acta Biomaterialia*, *78*, 224-235.

Rodriguez-Del Rio, E., Marradi, M., Calderon-Gonzalez, R., Frande-Cabanes, E., Penadés, S., Petrovsky, N., & Alvarez-Dominguez, C. (2015). A gold glyco-nanoparticle carrying a listeriolysin O peptide and formulated with Advax™ delta inulin adjuvant induces robust T-cell protection against listeria infection. *Vaccine*, *33*(12), 1465-1473.

Salazar-González, J. A., González-Ortega, O., & Rosales-Mendoza, S. (2015). Gold nanoparticles and vaccine development. *Expert Review of Vaccines*, *14*(9), 1197-1211.

Staroverov, S., & Dykman, L. (2013). Use of gold nanoparticles for the preparation of antibodies to tuberculin, the immunoassay of mycobacteria, and animal vaccination. *Nanotechnologies in Russia*, *8*(11), 816-822.

Staroverov, S., Ermilov, D., Shcherbakov, A., Semenov, S., SIu, S., & Dykman, L. (2003). Generation of antibodies to Yersinia pseudotuberculosis antigens using the colloid gold particles as an adjuvant. *Zhurnal Mikrobiologii, Epidemiologii i Immunobiologii*, *1*(3), 54-57.

Staroverov, S., Vidyasheva, I., Gabalov, K., Vasilenko, O., Laskavyi, V., & Dykman, L. (2011). Immunostimulatory effect of gold nanoparticles conjugated with transmissible gastroenteritis virus. *Bulletin of Experimental Biology and Medicine*, *151*(4), 436.

Staroverov, S. A., Volkov, A. A., Mezhenny, P. V., Domnitsky, I. Y., Fomin, A. S., Kozlov, S. V.,Guliy, O. I. (2019). Prospects for the use of spherical gold nanoparticles in immunization. *Applied Microbiology and Biotechnology*, *103*, 437-447.

Stone, J. W., Thornburg, N. J., Blum, D. L., Kuhn, S. J., Wright, D. W., & Crowe Jr, J. E. (2013). Gold nanorod vaccine for respiratory syncytial virus. *Nanotechnology*, *24*(29), 295102.

Tao, W., Hurst, B. L., Shakya, A. K., Uddin, M. J., Ingrole, R. S., Hernandez-Sanabria, M.,Tarbet, E. B. (2017). Consensus M2e peptide conjugated to gold nanoparticles confers protection against H1N1, H3N2 and H5N1 influenza A viruses. *Antiviral Research*, *141*, 62-72.

Tao, W., Ziemer, K. S., & Gill, H. S. (2014). Gold nanoparticle–M2e conjugate coformulated with CpG induces protective immunity against influenza A virus. *Nanomedicine*, *9*(2), 237-251.

Wang, L., Zhang, H., Sun, L., Gao, W., Xiong, Y., Ma, A.,Yang, H. (2020). Manipulation of macrophage polarization by peptide-coated gold nanoparticles and its protective effects on acute lung injury. *Journal of Nanobiotechnology*, *18*, 1-16.

Wang, W., Wang, J., & Ding, Y. (2020). Gold nanoparticle-conjugated nanomedicine: design, construction, and structure–efficacy relationship studies. *Journal of Materials Chemistry B*, *8*(22), 4813-4830.

Wang, Y., Wang, Y., Kang, N., Liu, Y., Shan, W., Bi, S.,Zhuang, G. (2016). Construction and immunological evaluation of CpG-Au@ HBc virus-like nanoparticles as a potential vaccine. *Nanoscale Research Letters*, *11*, 1-9.

Xu, L., Liu, Y., Chen, Z., Li, W., Liu, Y., Wang, L., & Zhao, Y. (2012). Surface-engineered gold nanorods: promising DNA vaccine adjuvant for HIV-1 treatment. *Nano Letters*, *12*(4), 2003-2012