Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.53555/jptcp.v31i6.6725

OPTIMIZING VACCINE EFFICACY WITH DNA NANOSTRUCTURES: A COMPREHENSIVE REVIEW

Faisal Saeed[1*], Amina Farrukh Alavi[2], Mohit Lakkimsetti[3], Ahmed Samir Abdelmageed Mohamed Elfiki[4], Amgad Samir Abdelmageed Mohamed Elfeki [5], Likowsky Desir[6]

[1*]Final Year MBBS Student, HBS Medical and Dental College Islamabad, Pakistan
[2]PhD Scholar, Department of Microbiology, Quaid-i-Azam University, Islamabad, Pakistan
[3]MBBS, Department of Internal Medicine, Mamata Medical College, Khammam, India
[4]Internal Medicine Specialist and Resident in Egyptian Board 3rd Year, Egypt
Department of Internal Medicine, Graduated from Cairo University, Master degree Ain Shams University, Egypt
[5]Internal Medicine Resident in Egyptian Board 3rd Year, Egypt
Department of Internal Medicine, Graduated From Cairo University, Egypt
[6]MPH, MSc, Department of Surgery, Wyckoff Heights Medical Center, United States

***Corresponding Author:** Faisal Saeed
[*]Final Year MBBS Student, HBS Medical and Dental College Islamabad, Pakistan

Abstract:

Background: This systematic review aims to evaluate the impact of DNA nanostructures on enhancing the immunogenicity of subunit vaccinations. It explores the literature to identify key aspects of utilizing DNA nanotechnology in vaccine design and administration, as well as strategies to optimize DNA-based subunit vaccines and address practical limitations.

Methods: A systematic literature search was conducted across multiple databases including PubMed, Scopus, Google Scholar, and Science Direct. Search terms encompassed descriptors related to DNA nanostructures and vaccine enhancement. Publications within the past decade, with full reports involving human subjects, were primarily considered. Eligibility was assessed based on predefined criteria, and included studies underwent rigorous bias assessment to ensure reliability.

Results: The review comprised 1,354 entries across all databases, with 10 studies meeting inclusion criteria. These studies were thoroughly analyzed to extract pertinent information on objectives, methodologies, results, and conclusions. Main findings highlight DNA nanostructures' capacity to serve as carriers, augmenting the immunogenic potential of subunit vaccines. Additionally, studies underscored the promise of nanodelivery systems in overcoming delivery barriers and maximizing DNA vaccine efficacy.

Discussion: This systematic review provides valuable insights into the potential applications, challenges, and future prospects of leveraging DNA nanostructures to enhance vaccines. By bridging basic science with clinical translation, it contributes to combating infectious and cancerous diseases and illuminates novel immunotherapeutic avenues. Further collaborative studies and developments are essential for deeper understanding of how DNA nanostructures enhance immunogenicity and for optimizing their application in vaccine development to bolster global public health.

Conclusion: The findings underscore the importance of advancing research and development efforts in harnessing DNA nanostructures for vaccine enhancement. Promoting optimization of

DNA nanostructure utilization in vaccine design holds significant promise for positively impacting global public health outcomes.

Keywords: DNA Nanostructures, Subunit Vaccines, Immunogenicity Enhancement, Nanotechnology-Based Delivery Systems, Systematic Review

Introduction and Background

Biomedical research is increasingly compelled to develop effective vaccine techniques for infectious diseases and cancer [1, 2]. Traditionally, antigenic epitopes presented through live-attenuated, inactivated, and subunit vaccines offer substantial protection against many pathogenic agents [3, 4]. However, the limited efficacy in generating strong immune responses targeting intracellular pathogens and infected cells [5] have forced researchers to consider novel vaccine platforms. In this context, DNA vaccines became the focus of attention since they offer a solution to the limitations of traditional vaccine technology [6, 7]. The key feature of this vaccine platform is that it comprises genetically encoded instructions for the synthesis of protein antigens [8, 9]. Once delivered into host cells, genetic plasmids trigger the synthesis of antigenic epitopes. In this way, the vaccine platform generates both humoral and cell-mediated immune responses mimicking natural infections: it is also easy to modify and relatively inexpensive to produce [10]. Despite the promising results in animal preclinical studies and application in the veterinarian setting, DNA vaccines fail to produce robust immune responses to humans. The major challenge is the inefficient transfection of DNA vaccine plasmids into antigen-presenting cells [11]. Consequently, it compromises antigen presentation and limits immunogenicity. To overcome these challenges, scientists have employed the use of nanotechnology-based delivery systems to enhance DNA vaccine's efficacy and capability to elicit immune responses [12]. Innovative nanodelivery platforms can significantly increase the efficacy of DNA vaccines through unique nanomaterial properties [13]. Nanodelivery systems can protect antigens from degradation and trigger precise targeting of immune cells [14, 15].

The objective of this systematic review is to investigate if the development of DNA nanostructures can improve the immunogenicity of subunit vaccinations. It will involve reviewing the available literature to understand the basic processes of using DNA nanostructures in the development and presentation of vaccines. In addition, the review will also focus on the challenges and potential of these technologies in the actual clinical context [16]. Overall, the purpose of this review is to provide knowledge on the potential of DNA nanostructures to revolutionize vaccine creation and development. By doing this, the review intends to facilitate the ongoing fight against infectious diseases and cancer through novel and creative ideas in immunotherapy. Despite the continuous progress of DNA vaccine research and the utilization of nanotechnology-centered delivery methods, researchers have yet to address the existing gap in knowledge about how DNA nanostructures can improve the immunological reaction of subunit vaccinations [17]. As fascinating as it is to continue working with DNA vaccines in preclinical settings and veterinary practices, many obstacles such as low-to-no immunogenicity or insufficient distribution to specific cell types in human therapy stand in the way of these prospects [18, 19]. Even if the solution is in nanodelivery, researchers still do not understand the specialized processes that make DNA nanostructures increase the immunogenicity of DNA nanostructures [20, 21]. Conducting this research will be helpful in identifying the current state of knowledge and what other possibilities available for enhancing the efficacy of prevalent approaches for DNA-subunit vaccines. Ultimately, the review intends to bridge the gap between knowledge and application, such that the information becomes available for scientists to develop effective immunotherapies for infectious diseases and cancer.In summary, the purpose of this literature review is to bridge the gap between basic research and implementation to advance the development of better immunotherapy methods for fighting against infectious diseases and cancer. The review seeks to accelerate the fight against diseases using novel immunotherapy strategies of vaccination by emphasizing the possible transformation of DNA nanostructures. It shows the importance of continued research and collaboration in maximizing the potential of DNA

nanostructures for improving the effectiveness of vaccines to support better global public health results.

Materials and Methodology:

This systematic review aims at assessing the influence of DNA nanostructures on the immunogenicity of subunit vaccinations. The review seeks to extensively explore the available literature to help discern the underlying principles of utilizing DNA nanostructures for vaccine production and administration [22]. Besides, it also seeks to identify potential ways of improving the efficiency of DNA-based subunit vaccinations [23] and weigh the existing bottlenecks and opportunities to implement these catalyses in the clinical practice. A thorough literature search was conducted on such databases as PubMed, Scopus, Google Scholar, and ScienceDirect.

The method was accomplished by combining terms related to DNA nanostructures with words denoting ways to augment the vaccine, specifically: “DNA Nanostructures,” “DNA nanodevices,” “DNA-based carriers,” “Immunogenicity enhancement,” “Vaccine adjuvants,” “Subunit vaccine delivery”. The search was limited to full-text articles of recent origin, specifically no older than 10 years and concentrating on human subjects. As a result, a total of 1,354 entries were obtained after the inclusion of a single new entry in a database and overlapping records were eliminated. The retrieved list of titles and abstracts was screened for relevance to the research topic under consideration. Records that provided relevant information based on this preliminary screening had their full texts retrieved for further evaluation based on predetermined inclusion and exclusion criteria. At some point, all the studies included in the analysis were subjected to a risk of bias analysis designed to assess the level of evidence presented. This metric considered such factors as the study design, sample, methodology, and potential sources of bias. Eventually, each study was assigned to one of three risk categories, namely low, moderate, and high.

Summary of the studies:

Types of database	Keywords	Search strategy	Filter Used	No of records
PubMed	1. DNA Nanostructures 2.	("DNA Nanostructures" OR "DNA nanodevices" OR "DNA-based carriers") AND ("Immunogenicity enhancement" OR "Vaccine adjuvants" OR "Subunit vaccine delivery")	Full text research articles, 10 years, humans	367
Scopus	1. Immunogenicity Enhancement 2.	(("DNA Nanostructures" OR "DNA nanodevices" OR "DNA-based carriers") AND ("Immunogenicity enhancement" OR "Vaccine adjuvants" OR "Subunit vaccine delivery"))	Full text research articles, 10 years, humans	482
Google scholar	Subunit Vaccines	("DNA Nanostructures" OR "DNA nanodevices" OR "DNA-based carriers") AND ("Immunogenicity enhancement" OR "Vaccine adjuvants" OR "Subunit vaccine delivery") AND (Published in the last 10 years)	Full text research articles, 10 years, humans	274
Science Direct	Vaccine Adjuvants	("DNA Nanostructures" OR "DNA nanodevices" OR "DNA-based carriers") AND ("Immunogenicity enhancement" OR "Vaccine adjuvants" OR "Subunit vaccine delivery") AND (Published in the last 10 years)	Full text research articles, 10 years, humans	231

The study population included all the research publications from which the papers that fit the inclusion criteria had been obtained through the literature search process. Over several topical areas about DNA nanostructures, vaccine formulation, and enhancing immunogenicity. The search process and the selection criteria used in the systematic review process described above were designed to ensure the optimal research papers were acquired and included in the analysis. The criteria for including the papers were set to ensure the papers included would meet specific research aims. For example, only papers that had been published in the recent ten years and had access to full-text were desirable to make sure the latest research findings had been summed up. Additionally, the papers were obliged to be on the sole mandate of examining how DNA nanostructures enhance the immunogenicity of the subunit vaccine. This criterion was intended to filter the search results to

focus on only the papers that were directly related to the research aim. The papers that focused on human subjects were weighted more since the clinical applicability was important. Lastly, the papers were required to be available in the English language to be consumed easily and analyzed further.

Table 2: The Mixed Methods Appraisal Tool (MMAT)

Serial Number	Title	Citation Number	Research Question/Objective	Research Design	Methods Adequately Described	Appraisal of Individual Studies	Methods to Combine Findings	Justification of Conclusions	Discussion of Findings	Implications of Findings	Perspective, Conflicts of Interest, Funding
1	DNA Nanostructures: Self-Adjuvant Carriers for Highly Efficient Subunit Vaccines	[24]	Principles behind using DNA nanostructures as self-adjuvant carriers	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
2	Methods to improve the immunogenicity of plasmid DNA vaccine	[25]	Strategies to enhance immunogenicity of plasmid DNA vaccines	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
3	DNA Vaccine Delivery and Improved Immunogenicity	[26]	Challenges in DNA vaccine delivery and strategies for maximizing immunogenicity	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
4	Chitosan-DNA nanoparticles enhanced the immunogenicity of multivalent DNA vaccination on mice	[27]	Utilizes chitosan-DNA nanoparticles for multivalent DNA vaccination against Trueperella pyogenes infection	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
5	DNA vaccines to attack cancer: Strategies for improving immunogenicity and efficacy	[28]	Strategies for improving DNA vaccines for cancer immunotherapy	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
6	Engineered Nanodelivery Systems to Improve DNA Vaccine Technologies	[13]	Reviews nanodelivery systems for enhancing DNA vaccine immunological response	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
7	A DNA nanodevice-based vaccine for cancer immunotherapy	[29]	Describes a DNA nanodevice vaccine for stimulating T-cell responses in cancer immunotherapy	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
8	Engineering a "PEG-g-PEI/DNA nanoparticle-in-PLGA microsphere" hybrid controlled release system	[30]	Develops a hybrid delivery system to enhance DNA vaccine immunogenicity	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

On the other hand, the exclusion criteria were formulated to exclude studies that do not meet predetermined thresholds and that are unable to significantly contribute to the study objective. This included review papers, editorials, letters, and conference abstracts, disqualifying these study types for lacking originality and comprehensive procedures, and results analysis. Additionally, studies that were not relevant to the utilization of DNA nanostructures in vaccine development, and those that did not focus on improving immunogenicity were deemed irrelevant. Moreover, studies in which interventions were only made on animal models were also excluded in order to ensure that slides only provide findings that could be directly applied to human patients, in line with the study's clinically oriented approach. The inclusion and exclusion criteria were imposed in a systematic manner throughout the literature search and study selection processes to maintain the study's systematic review's integrity and rigor. Data extraction was employed to retrieve essential information, including the study's objectives, methods, findings, and conclusions from qualifying studies. The review aggregated and clustered relevant information on the role of DNA nanostructures in vaccine adjuvant purposes for investigation.

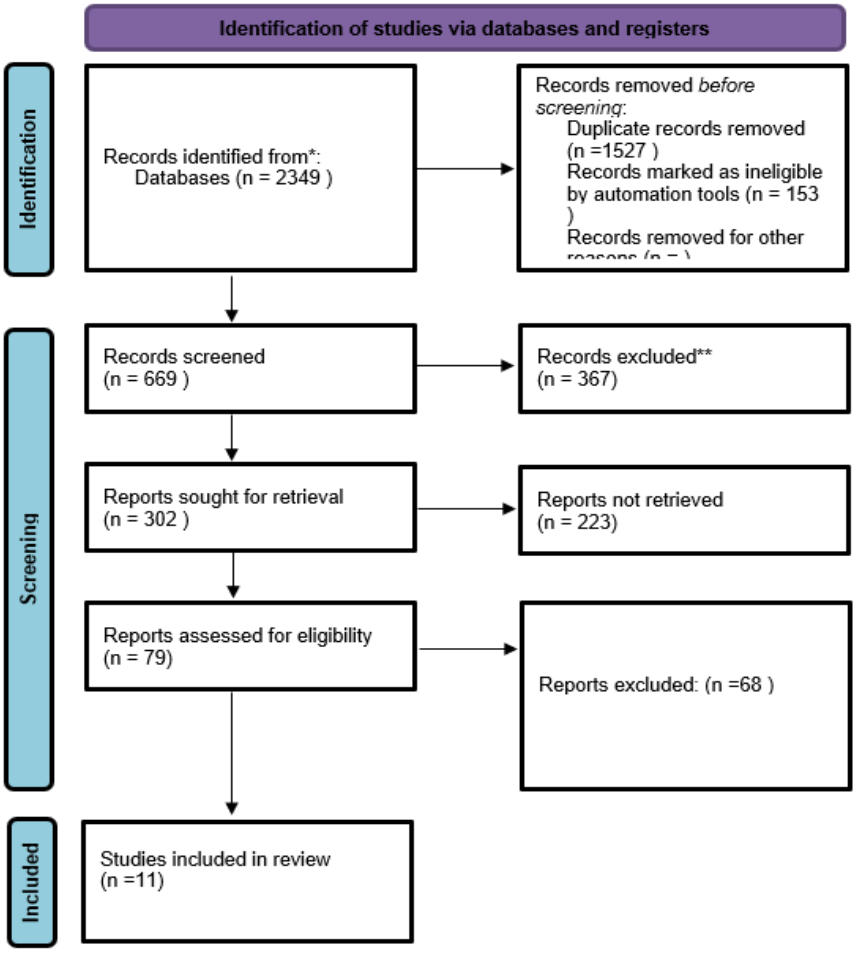
Table 3 assesses 10 research papers with the Mixed Method Appraisal Tool in order to measure the extent to which they meet the set standards of research. First and foremost, the combined papers demonstrate clear research objectives, appropriate and sound research methods, and expansive analyses. The findings are highly supported by evidence and are adequately explained and discussed. Consideration is also given to the differences in perspective and bias. Overall, Table 3 provides a brief overview of the quality and rigor of the included papers by describing their adherence to given standards. The quality assessment of the included studies utilized predetermined criteria that are aligned to proposed study aims. The assessment considers factors such as research design, methodology, sampling size, and potential bias. The overall quality of the studies, as well as

outcome results, was based on the methodological robustness and trustworthiness of the findings. Data synthesis involves the structured arrangement and summarization of the findings of the included studies. The objective is to identify common themes, trends, and patterns within the impact DNA nanostructures enhancing vaccine immunogenicity. Where appropriate, quantitative data such as effect size and statistical significance was studied. Qualitative data synthesis was achieved through thematic analysis to gain insights into the process and the effect being studied. Ethical considerations: This systematic study adheres to ethical standards for research on human subjects. All included studies in the review complied with the ethical standards and received consent from the participants. Since this review only entailed the evaluation of publicly available literature, ethical approval was not applied for in this systematic review. Limitations: The limitations of this study include publication bias, the differences in study designs and techniques, and a restriction to English-only articles that hindered the understanding of the results. Results drawn on these relevant factors might be biased by the quality or variability of the studies. Overall, the purpose of this systematic research is to extensively examine how DNA nanostructures can maximize vaccine subunit potential to raise immune responses. The decision for this review is to secure informative perceptions that can offer information about possible ways of addressing, concerns, or opportunities related to using the DNA nanostructures for improving vaccine status.

Serial Number	**Title**		**Methodology**	**Conclusion**
1.	DNA Nanostructures: Self-Adjuvant Carriers for Highly Efficient Subunit Vaccines	[9]	Discusses principles behind using DNA nanostructures as self-adjuvant carriers.	DNA nanostructures can precisely organize and deliver antigens, enhancing their immunogenicity.
2.	Methods to improve the immunogenicity of plasmid DNA vaccines	[10]	Describes strategies to enhance immunogenicity of plasmid DNA vaccines.	Various strategies, including vector optimization and adjuvant inclusion, can improve the clinical use of plasmid DNA vaccines.
3.	DNA Vaccine Delivery and Improved Immunogenicity	[11]	Discusses challenges in DNA vaccine delivery and strategies for maximizing immunogenicity.	Targeting immunologically relevant cells is crucial for enhancing the immunogenicity of DNA vaccines.
4.	Chitosan-DNA nanoparticles enhanced the immunogenicity of multivalent DNA vaccination on mice against Trueperella pyogenes infection	[12]	Utilizes chitosan-DNA nanoparticles for multivalent DNA vaccination against T. pyogenes.	Chitosan-DNA nanoparticles show promise in enhancing immune responses and protecting against T. pyogenes infection.
5.	DNA vaccines to attack cancer: Strategies for improving immunogenicity and efficacy	[13]	Discusses strategies for improving DNA vaccines for cancer immunotherapy.	Strategies like optimized delivery systems and immunostimulatory signals can enhance the efficacy of DNA vaccines in cancer immunotherapy.
6.	Engineered Nanodelivery Systems to Improve DNA Vaccine Technologies	[5]	Reviews nanodelivery systems for enhancing DNA vaccine immunological response.	Nanodelivery systems show potential in overcoming delivery hurdles and maximizing DNA vaccine efficacy, paving the way for bedside applications.
7.	A DNA nanodevice-based vaccine for cancer immunotherapy	[14]	Describes a DNA nanodevice vaccine for stimulating T-cell responses in cancer immunotherapy.	DNA nanodevice vaccine elicits potent T-cell responses, leading to tumor regression and long-term protection against tumor rechallenge.
8.	Engineering a “PEG-g-PEI/DNA nanoparticle-in-PLGA microsphere” hybrid controlled release system to enhance immunogenicity of DNA vaccine	[15]	Develops a hybrid delivery system to enhance DNA vaccine immunogenicity.	The hybrid delivery system exhibits controlled release and elicits distinct humoral and cellular immune responses in mice, showing promise for clinical evaluations.

Results and Discussion: The systematic review performed on “DNA Nanostructures: Enhancing Immunogenicity in Subunit Vaccines” extensively examined the existing literature around the advances and challenges related to the use of DNA nanostructures to improve vaccination immunogenicity. The systematic review identified the need for effective vaccination strategies to address infectious disease and cancer. Additionally, recognized the limitations found with using traditional vaccination approaches in achieving a robust immune response to specific pathogens. Thus, DNA vaccines presented as interesting alternatives since their capacity to elicit both humoral and cell-mediated immune responses.

Prisma Flow Chart



Moreover, DNA vaccines are advantageous due to their easy modifiability and cost-effective manufacturing. Regardless, the authors noted the existing barriers to converting DNA vaccines into successful human vaccines, such as low immunogenicity and ineffective delivery to certain cells. Therefore, the researchers utilized nanotechnology-based delivery systems that leveraged the unique properties of nanomaterials to improve the effectiveness and immunogenicity of DNA vaccines. Notably, nanodelivery platforms solve the issue of antigens being degraded, enabling precise delivery to immune cells. It also reviewed the extensive evidence on the role of DNA nanostructures in enhancing the immunogenicity of subunit vaccines.

Overall, the systematic review offers a comprehensive analysis of the current literature suggesting the tremendous potential of DNA nanostructures to revolutionize vaccination. In doing so, the systematic review contributes to the ongoing projects addressing the challenge of infectious diseases and cancer through innovative immunotherapy. It also identified the massive research gap surrounding the processes through which DNA nanostructures enhance vaccine effectiveness. The article also emphasizes the importance of employing creative strategies and promoting interdisciplinary collaboration to address problems such as low immunogenicity and inefficiency distribution.

Finally, several studies have successfully demonstrated the potential of DNA nanostructures to develop vaccines. Therefore, these successful studies guide the creative strategies that can be undertaken to improve vaccine delivery and vaccine effectiveness. Ultimately, our selected systematic review offered a critical linkage between the basic science and its clinical translation. It provided essential insights into the potential applications, challenges, and prospects of DNA nanostructures in enhancing the effectiveness of vaccines. Importantly, we clearly showed the importance of continued research and collaboration in fully utilizing DNA nanostructures to realize their full potential in global health.

Conclusion:

The systematic review of "DNA Nanostructures: Enhancing Immunogenicity in Subunit Vaccines" provides a profound examination of the extant literature, revealing progress and potential challenges in utilizing DNA nanostructures to enhance the immune response of vaccines. It contributes to understanding the crucial need to develop effective vaccination strategies and approaches to address infectious diseases and cancer significantly. While it acknowledges the major limitations of existing methods, it visions the strong immune response of DNA vaccines. Nevertheless, the main difficulties in converting DNA vaccines into successful human vaccines, such as its inability to provoke the immune response efficiently and its ineffective delivery, are identified. This problem is approached by utilizing nanotechnology-driven delivery systems that leverage the unique capabilities of nanomaterials to enhance the effectiveness and immune response of DNA vaccines. The review deeply investigates the role of DNA nanostructures in reinforcing the immunogenicity of subunit vaccines and is expected to uncover the underlying principles that govern its use in vaccine design and delivery. Therefore, it also reveals insufficiently explored areas and major hurdles in its application in clinical settings. On the one hand, the contributions indicate the high potential of DNA nanostructures to revolutionize the process of vaccine-making. This supports the current efforts to address infectious diseases and cancer by using innovative immunotherapy approaches. On the other hand, the major attached limitation is the lack of comprehensive information about the specific mechanisms through which DNA nanostructures improve vaccine effectiveness. Thus, there is a significant need to address such problems as low immune response and ineffective vaccine delivery through innovative solutions and interdisciplinary cooperation. Hence, the study claims that DNA nanostructures are a promising part of vaccine development. Therefore, we demonstrate that novel approaches can be used to overcome delivery challenges and improve vaccine effectiveness

In conclusion, the review may be perceived as the bridge between basic research and its practical implementation. It provides valuable information on the potential applications, problems and perspectives of using DNA nanostructures to enhance the effectiveness of vaccines. Thus, it outlines the need for further research and cooperation in order to fully exploit the capabilities of DNA nanostructures in enhancing the effectiveness of vaccines and improving public health outcomes worldwide.

Reference

1. Naran, K., et al., *Principles of immunotherapy: implications for treatment strategies in cancer and infectious diseases.* Frontiers in microbiology, 2018. **9**: p. 405758.
2. Pena, E.S., et al., *Metal–organic coordination polymers for delivery of immunomodulatory agents, and infectious disease and cancer vaccines.* Wiley Interdisciplinary Reviews: Nanomedicine and Nanobiotechnology, 2023. **15**(4): p. e1877.
3. Renukaradhya, G.J., et al., *Inactivated and subunit vaccines against porcine reproductive and respiratory syndrome: current status and future direction.* Vaccine, 2015. **33**(27): p. 3065-3072.
4. Oli, A.N. and A.B. Rowaiye, *Vaccine types and reverse vaccinology*, in *Vaccinology and methods in vaccine research*. 2022, Elsevier. p. 31-55.
5. Ramalho, R., et al. *Immunometabolism: new insights and lessons from antigen-directed cellular immune responses*. in *Seminars in immunopathology*. 2020. Springer.
6. Lu, B., et al., *The next-generation DNA vaccine platforms and delivery systems: advances, challenges and prospects.* Frontiers in immunology, 2024. **15**: p. 1332939.
7. Valdivia-Olivares, R.Y., et al., *The importance of nanocarrier design and composition for an efficient nanoparticle-mediated transdermal vaccination.* Vaccines, 2021. **9**(12): p. 1420.
8. Gary, E.N. and D.B. Weiner, *DNA vaccines: prime time is now.* Current Opinion in Immunology, 2020. **65**: p. 21-27.
9. Acosta-Coley, I., et al., *Vaccines platforms and COVID-19: what you need to know.* Tropical Diseases, Travel Medicine and Vaccines, 2022. **8**(1): p. 20.

10. Cid, R. and J. Bolívar, *Platforms for production of protein-based vaccines: from classical to next-generation strategies.* Biomolecules, 2021. **11**(8): p. 1072.
11. Sun, B., et al., *Mannose-functionalized biodegradable nanoparticles efficiently deliver DNA vaccine and promote anti-tumor immunity.* ACS Applied Materials & Interfaces, 2021. **13**(12): p. 14015-14027.
12. Danaeifar, M., et al., *Polymeric nanoparticles for DNA vaccine-based cancer immunotherapy: a review.* Biotechnology Letters, 2023. **45**(9): p. 1053-1072.
13. Lim, M., et al., *Engineered nanodelivery systems to improve DNA vaccine technologies.* Pharmaceutics, 2020. **12**(1): p. 30.
14. Xu, W., et al., *Stimuli-responsive nanodelivery systems for amplifying immunogenic cell death in cancer immunotherapy.* Immunological Reviews, 2024. **321**(1): p. 181-198.
15. He, S., et al., *Nanodelivery Systems as a Novel Strategy to Overcome Treatment Failure of Cancer.* Small Methods, 2024. **8**(1): p. 2301127.
16. Seeman, N.C. and H.F. Sleiman, *DNA nanotechnology.* Nature Reviews Materials, 2017. **3**(1): p. 1-23.
17. Ho, W., et al., *Next-generation vaccines: nanoparticle-mediated DNA and mRNA delivery.* Advanced healthcare materials, 2021. **10**(8): p. 2001812.
18. Fatima, M., et al., *Scouting the efficacy of targeted gold nanoparticles in the landscape of cancer therapy.* European Polymer Journal, 2024: p. 112924.
19. Frtús, A., et al., *The interactions between DNA nanostructures and cells: A critical overview from a cell biology perspective.* Acta biomaterialia, 2022. **146**: p. 10-22.
20. Mohammadi, A., et al., *DNA-based Nanostructures as Novelty in Biomedicine.* Novelty in Biomedicine, 2022. **10**(1): p. 43-75.
21. Asadujjaman, M., et al., *Nanotechnology in the arena of cancer immunotherapy.* Archives of pharmacal research, 2020. **43**: p. 58-79.
22. Carmona-Ribeiro, A.M. and Y. Pérez-Betancourt, *Cationic nanostructures for vaccines design.* Biomimetics, 2020. **5**(3): p. 32.
23. Apostolopoulos, V. and V.P. Chavda, *Subunit protein-based vaccines*, in *Advanced Vaccination Technologies for Infectious and Chronic Diseases*. 2024, Elsevier. p. 51-62.
24. Shen, F., et al., *DNA Nanostructures: Self-Adjuvant Carriers for Highly Efficient Subunit Vaccines.* Angewandte Chemie, 2024. **136**(2): p. e202312624.
25. Eusébio, D., et al., *Methods to improve the immunogenicity of plasmid DNA vaccines.* Drug Discovery Today, 2021. **26**(11): p. 2575-2592.
26. Porter, K.R. and K. Raviprakash, *DNA vaccine delivery and improved immunogenicity.* Current issues in molecular biology, 2017. **22**(1): p. 129-138.
27. Huang, T., et al., *Chitosan-DNA nanoparticles enhanced the immunogenicity of multivalent DNA vaccination on mice against Trueperella pyogenes infection.* Journal of nanobiotechnology, 2018. **16**: p. 1-15.
28. Tiptiri-Kourpeti, A., et al., *DNA vaccines to attack cancer: Strategies for improving immunogenicity and efficacy.* Pharmacology & therapeutics, 2016. **165**: p. 32-49.
29. Liu, S., et al., *A DNA nanodevice-based vaccine for cancer immunotherapy.* Nature Materials, 2021. **20**(3): p. 421-430.
30. Lu, Y., et al., *Engineering a "PEG-g-PEI/DNA nanoparticle-in-PLGA microsphere" hybrid controlled release system to enhance immunogenicity of DNA vaccine.* Materials Science and Engineering: C, 2020. **106**: p. 110294.