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The Role of Biotechnology in Vaccine Development

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Abstract

The rapid advancement of biotechnology has revolutionized the field of vaccine development, enabling the creation of more effective and safer vaccines. This review explores the historical progression of vaccine development, highlighting significant milestones and the shift from traditional methods to modern biotechnological approaches. It delves into various biotechnological techniques such as recombinant DNA technology, genetic engineering, synthetic biology, and protein engineering that have significantly enhanced vaccine production and formulation. The emergence of novel vaccine platforms, including mRNA, viral vector, DNA, and nanoparticle-based vaccines, is discussed with a focus on their development and clinical impact. Case studies, particularly the recent development of COVID-19 vaccines, illustrate the transformative role of biotechnology in responding to global health crises. The review also addresses the challenges and limitations inherent in biotechnological advancements, such as technical hurdles, regulatory issues, and ethical considerations. Additionally, it examines the impact of these innovations on vaccine safety, efficacy, and public health, and anticipates future trends in personalized and universal vaccines. Through this comprehensive analysis, the review underscores the pivotal role of biotechnology in the ongoing evolution of vaccine development and its profound implications for global health.

Keywords: Biotechnology, vaccine development, recombinant DNA technology, genetic engineering, mRNA vaccines, viral vector vaccines, DNA vaccines, nanoparticle vaccines, vaccine production, immunogenicity, public health, global health

Introduction

Vaccines have been a cornerstone of public health for over a century, providing a critical defense against infectious diseases and contributing significantly to the decline of morbidity and mortality worldwide [1]. Traditional methods of vaccine development, while effective, often involved lengthy processes and faced limitations in addressing rapidly emerging pathogens and complex diseases [2]. The advent of biotechnology has heralded a new era in vaccine development, characterized by innovative techniques and accelerated timelines, transforming the landscape of immunization [3].

Biotechnology, encompassing a range of scientific disciplines including molecular biology, genetics, and bioengineering, has introduced powerful tools that have enhanced our ability to design, produce, and deliver vaccines [4]. Techniques such as recombinant DNA technology, genetic engineering, and synthetic biology have enabled the development of vaccines that are not only more effective and safer but also more adaptable to various pathogens [5]. These advancements have been pivotal in addressing some of the most pressing global health challenges, including pandemics and emerging infectious diseases [6].

One of the most striking examples of biotechnological innovation in vaccine development is the rapid creation and deployment of COVID-19 vaccines. The global response to the COVID-19 pandemic underscored the potential of mRNA vaccines and viral vector platforms, both products of sophisticated biotechnological processes [7]. These vaccines have demonstrated remarkable efficacy and safety profiles, setting new standards for future vaccine development [8].

Despite these advancements, the field of vaccine development continues to face significant challenges. Technical hurdles, such as ensuring stability and scalability of vaccine production, regulatory complexities, ethical considerations, and equitable distribution, remain critical areas that need to be addressed [9]. Moreover, the rapid pace of biotechnological progress necessitates continuous adaptation of regulatory frameworks to ensure the safety and efficacy of new vaccines [10].

This review aims to provide a comprehensive overview of the role of biotechnology in vaccine development. It will explore the historical context, detailing the evolution from traditional methods to contemporary biotechnological approaches. Key biotechnological techniques and their applications in vaccine development will be examined, along with the emergence of novel vaccine platforms [11]. Case studies, particularly those involving recent vaccines, will illustrate the practical impact of these advancements. Additionally, the review will discuss the challenges and limitations faced by the field and highlight the future directions and emerging trends that are poised to shape the future of vaccine development [12]. Through this detailed analysis, the review seeks to underscore the transformative potential of biotechnology in enhancing global health through innovative vaccine development.

Early Vaccines and Traditional Methods

The practice of variolation stretches back to ancient China and India, where the concept of vaccination first emerged. In order to create immunity, smallpox material was purposefully introduced using this technique [1]. In the early eighteenth century, the technique made its way to the Ottoman Empire and then, ultimately, to Europe.

The creation of the smallpox vaccine by Edward Jenner in 1796 was a watershed moment in the history of vaccinations. Jenner created protection against smallpox by utilising material from cowpox lesions, a method that served as the model for contemporary vaccination [2]. This groundbreaking technique of inducing immunity from a similar, less pathogenic pathogen signalled the start of a new age in disease prevention.

The Creation of Inactivated and Attenuated Vaccines

The invention of attenuated and inactivated vaccinations brought about important breakthroughs in the late 19th and early 20th centuries. The first attenuated vaccinations against chicken cholera and anthrax were created by microbiologist Louis Pasteur, who weakened the infections [3]. Pasteur's research showed that vaccines could be created that would both safely and successfully generate immunity.

Another significant advance in the early 20th century was the creation of inactivated vaccinations, such as Jonas Salk's polio vaccine. These vaccinations stimulated an immune response while avoiding the possibility of disease transmission by utilising heat- or chemically-killed microorganisms [4].

The Development of Biotechnology

The creation of vaccines was transformed when biotechnology was introduced in the latter half of the 20th century. Using bacterial, yeast, or mammalian cell systems, researchers were able to separate particular antigens from infections and generate them in vast quantities thanks to recombinant DNA technology [5]. The first recombinant vaccine, the hepatitis B vaccine, was licenced in 1986 and represented a major breakthrough in vaccine safety and production efficiency [6].

Current Developments and the COVID-19 Epidemic

Significant advancements have been made in the twenty-first century, most notably the creation of mRNA vaccines. The COVID-19 pandemic hastened the application of this technology, resulting in Pfizer-BioNTech and Moderna developing and approving mRNA vaccines quickly [7]. Instead of employing live virus to trigger an immune response, these vaccines use synthetic mRNA to direct cells to manufacture the viral spike protein [8].

The use of viral vectors, as demonstrated by the COVID-19 vaccines from Johnson & Johnson and AstraZeneca, is another noteworthy breakthrough. These vaccines induce immunity by delivering genetic material encoding the spike protein via a safe virus [9].

The Fundamentals of Vaccine Development

The process of developing new vaccines is intricate and multidimensional, requiring a thorough understanding of microbiology, immunology, and biotechnology. Inducing a protective immune response that can stop or manage infection by a particular pathogen is the main objective of a vaccination. The main ideas and procedures used in vaccine development are outlined in the following principles.

Fundamental Immunological Ideas

The idea of immunisation, which tries to condition the immune system to recognise and combat diseases, is at the centre of vaccine research. The innate immune response and the adaptive immune response are the two primary components of the immune system. While the adaptive response activates T and B cells to provide specific, long-lasting immunity, the innate response offers quick, nonspecific defence [1].

Vaccines function by imitating an actual infection, which boosts immunity without actually spreading the illness. Antigens—molecules generated from infections that the immune system recognizes—are presented during this process. Memory cells are produced as a result of a successful vaccination, and these cells have the ability to generate a powerful and quick defence when exposed to the pathogen again [2].

Vaccine Types

Based on their content and the way they trigger an immune response, vaccines can be categorised into multiple types:

1. Vaccines using live attenuation:

- o The pathogens used in these vaccinations have been weakened, or attenuated, so that healthy people cannot contract the disease. The measles, mumps, and rubella (MMR) vaccination is one example [3].
- o Benefits: High and durable immunity, frequently requiring less doses.
- o Drawbacks include the possibility of virulence reversal and unsuitability for people with impaired immune systems.

2. Reconstituted (inactivated) vaccines:

- The pathogens used in these vaccinations have been rendered inert by radiation, heat, or chemicals. The hepatitis A vaccine and the inactivated polio vaccine (IPV) are two examples [4].
- Benefits: Safe for people with weakened immune systems.
- Drawbacks: To maintain immunity, several doses and booster shots can be necessary.

3. Vaccines that are Subunit, Recombinant, and Conjugate:

- To elicit an immune response, these vaccines target certain components of the disease, like proteins or polysaccharides. Examples include the vaccines against the human papillomavirus (HPV) and hepatitis B [5].
- Benefits: Since they don't contain live components, there is a lower chance of negative reactions.
- Drawbacks: Adjuvants may be needed to improve immunogenicity, and several doses may be necessary.

4. Vaccines that contain toxins:

- As with the tetanus and diphtheria vaccinations, these shots contain pathogen-produced inactivated toxins [6].
- Benefits: Good at avoiding illnesses brought on by bacterial toxins.
- Drawbacks: Frequently need booster dosages.

5. mRNA Immunisations:

- These vaccines work by telling cells to make a protein that sets off an immune response via messenger RNA. Examples include the COVID-19 vaccines created by Moderna and Pfizer-BioNTech [7].
- Advantages: robust immunological response, quick development and manufacturing.
- Drawbacks: Distribution may be made more difficult by the need for extremely low temperatures during storage.

6. Vaccines against viruses:

- The genetic material coding for an antigen is delivered by these vaccinations via a separate virus, or vector. This strategy is used in the COVID-19 and Ebola vaccines (AstraZeneca, Johnson & Johnson) [8].
- Benefits: May be applied to several ailments and stimulates robust immune responses.
- Drawbacks: The effectiveness may be lowered by pre-existing immunity to the vector.

The Process of Developing Vaccines

A vaccine goes through numerous stages of development, all of which are essential to guaranteeing the product's efficiency and safety:

1. Stage of Exploration:

- The investigation and identification of antigens capable of successfully eliciting an immunological reaction.

2. Stage Preclinical:

- Research in the lab and on animals to assess the potential vaccine's immunogenicity and safety.

3.Clinical Progress:

- Phase I: Small-scale studies with fit participants to evaluate dose and safety.
- Phase II involves conducting expanded trials to assess immunogenicity and determine the ideal dosage.
- Phase III: Extensive studies to verify effectiveness and track unfavourable responses in a broader population [9].

4.Regulatory Examination and Acceptance:

- Data submission for review and approval to regulatory bodies.

5.Production:

- Increasing output while preserving consistency and quality in order to fulfil demand.

6.After-Market Monitoring

- Constant observation of vaccination efficacy and safety in the broader populace.

Fundamental Immunological Ideas

To protect the body from pathogens including bacteria, viruses, fungi, and parasites, the immune system is a sophisticated network of cells, tissues, and organs. Developing effective vaccines requires an understanding of immunology principles because vaccines work by stimulating the immune system to defend against a target disease without actually producing the disease.

The innate immune system and the adaptive immune system are the two primary parts of the immune system. When faced with infections, the innate immune system reacts quickly and nonspecifically. It consists of cellular defences like phagocytes (such as neutrophils and macrophages) that engulf and eliminate infections, as well as physical barriers like the skin and mucous membranes, chemical barriers like stomach acid and enzymes, and all three [1].

In contrast, the adaptive immune system responds to viruses in a particular way and can recall past infections, which makes a reaction to the pathogen more powerful and quick when it resurfaces. T cells and B cells in particular are lymphocytes that are a part of this system. Antibodies are proteins that, when bound to antigens (foreign substances that trigger an immune response), are particularly recognised by B cells. T cells come in two varieties: cytotoxic T cells, which destroy infected cells directly, and helper T cells, which support other immune cells [2].

Vaccination takes advantage of the pathogen-memory capacity of the adaptive immune system. A vaccination triggers the body to produce memory B cells and T cells by delivering an antigen from a pathogen, all without harming the recipient. These memory cells have the ability to quickly and effectively establish an immune response in response to a second exposure to the pathogen, thereby averting disease.

A pathogen's totality, individual parts, or artificial creations made to resemble natural antigens can all be utilised as antigens in vaccinations. The pathogen's characteristics, the kind of immune response required for defence, and the viability of manufacturing the antigen in a stable and safe form all play a role in the selection of an antigen [3].

A vaccine's ability to elicit an immunological response can be assessed in a number of ways, such as the development of certain antibodies, T cell activation, and immune memory. These metrics are essential for assessing a vaccine's efficacy and comprehending how it provides immunity against illness.

Vaccine adjuvants are compounds that are added to the shot to improve the immune response. They function by enhancing the activation of the adaptive immune system by boosting the innate immune system. Typical adjuvants consist of aluminium salts (alum), oil-in-water emulsions, and more recent synthetic compounds that are made to resemble innate immune system-recognized pathogen-associated molecular patterns (PAMPs) [4].

The ability of a vaccine to elicit an immune response is known as immunogenicity, and it is a notion that must be understood in order to produce effective vaccines. The type of the antigen, the presence of adjuvants, the mode of delivery, and the recipient's genetic composition are some of the variables that affect immunogenicity. In contrast to vaccines delivered orally or nasally, injection-based vaccinations may elicit distinct immune responses [5].

Vaccines must not only build immunity but also be safe. Preclinical research in animal models and lengthy human clinical trials are used to evaluate safety. These tests assess the vaccine's capacity to elicit an immunological response without producing appreciable side effects. Following approval and broad use, post-marketing surveillance keeps an eye on the efficacy and safety of vaccines.

A major obstacle in the development of vaccines is guaranteeing sustained immunity. Certain vaccines offer lifetime protection with only one dosage, while others necessitate repeated doses or booster shots to keep immunity intact. Research is still being done to better understand immunological memory processes and the variables that affect the length of protection.

Vaccine Types

Several techniques are employed in the development of vaccines to elicit protective immunity against particular diseases. Every kind of vaccine has distinct qualities, benefits, and drawbacks. Selecting the right technique for various diseases and demographics requires an understanding of these various categories.

Live-Attenuated Immunisations

Live-attenuated vaccines employ microorganisms that have undergone weakened conditions, or attenuation, to render them incapable of infecting healthy persons. These vaccinations provide robust, long-lasting protection by closely imitating a genuine infection. The oral

polio vaccine (OPV) and the measles, mumps, and rubella (MMR) vaccine are two examples of live-attenuated vaccinations [1].

Benefits

- Durable and robust immunity.
- Frequently need fewer booster shots and dosages.
- May elicit immunological responses that are humoral (mediated by antibodies) as well as cellular (mediated by T cells).

Drawbacks:

- Possibility of virulence reversal, in which case the pathogen that has been attenuated may once again be able to cause illness.
- Not appropriate for people with specific medical issues or those who are immunocompromised.
- Needs to be handled and stored carefully to preserve the attenuated pathogens' vitality.

Inactivated Vaccines (Killed)

Pathogens that have been rendered inert by heat, chemicals, or radiation are used in inactivated vaccinations. These vaccines are thought to be extremely safe and incapable of causing illness. The hepatitis A vaccine and the inactivated polio vaccine (IPV) are two examples [2].

Benefits

- Safe for people with specific medical issues and those with impaired immune systems.
- There is no chance of virulence returning.

Drawbacks:

- In general, compared to live-attenuated vaccinations, they elicit a weakened immune response.
- Frequently need booster shots and several doses to keep immunity intact.
- Limited cellular immunity, mostly eliciting humoral immunological responses.

Vaccines that are Subunit, Recombinant, and Conjugate

Certain components of the pathogen, such as proteins, polysaccharides, or conjugated antigens, are used in these vaccines to elicit an immune response. The vaccines against hepatitis B, human papillomavirus (HPV), and Haemophilus influenzae type b (Hib) are a few examples [3].

Benefits

- Because they don't contain live components, there is a lower chance of negative reactions.
- May be made to specifically target the pathogen's components that work best to build immunity.

Drawbacks:

- Adjuvants might be necessary to improve immunogenicity.
- Frequently need several doses to develop and preserve immunity.
- Difficult production procedures, particularly with regard to recombinant vaccines.

Vaccines containing toxic substances

Inactivated toxins created by infections are used in toxoid vaccinations. Rather than the actual germs, these vaccinations work to prevent diseases brought on by their toxins. The tetanus and diphtheria vaccinations are two such examples [4].

Benefits

- Successful in avoiding illnesses brought on by bacterial toxins.
- Well-tolerated and safe.

Drawbacks:

- Frequently need booster shots to keep immunity intact over time.
- As opposed to vaccinations that target entire diseases, they might cause a more limited immune response.

mRNA Immunisations

mRNA vaccines work by telling cells to make a protein that sets off an immune response using synthetic messenger RNA. Pfizer-BioNTech and Moderna's COVID-19 vaccines are two instances of this cutting-edge technology [5].

Benefits

- Quick production and development turnaround times.
- Robust immunological reaction, encompassing humoral and cellular immunity.
- Because no live virus is utilised, there is no chance of illness.

Drawbacks:

- Difficult to distribute and store due to low temperature requirements for storage.

- The possibility of transient adverse effects, which are often minor and transient, such as fever and exhaustion.

Viral Vector Immunisations

Vaccines against viruses employ a distinct virus as a vector to transfer genetic material that codes for an antigen from the intended infection. AstraZeneca and Johnson & Johnson's COVID-19 vaccine and the Ebola vaccine are two examples [6].

Benefits

- Has the ability to elicit potent immunological reactions, encompassing humoral and cellular immunity.
- Adaptable to various diseases through modification of the genetic material conveyed by the vector.

Drawbacks:

- A prior immunity to the vector virus may lessen the effectiveness of the vaccination.
- Although typically safe, there may be negative effects connected to vectors.

The Process of Developing Vaccines

A vaccine must pass through a number of difficult steps in order to be developed into a product that is both safe and effective enough to be produced in large quantities. Meticulous investigation, stringent testing, and cautious regulatory supervision are needed at every level.

Stage of Exploration

Basic research is conducted at the exploratory stage in order to identify potential antigens that may trigger a defensive immune response. Scientists investigate the biology of the pathogen, concentrating on proteins or polysaccharides that are critical to its virulence or ability to survive. These antigens are potential candidates for vaccination research. Proteomics, genetics, and bioinformatics may be used at this stage to find and evaluate possible targets [1].

Stage Preclinical

Preclinical testing involves testing potential vaccines both *in vitro* (in the lab) and *in vivo* (in animal models) to assess their immunogenicity and safety. Utilising non-human primates, mice, and rabbits as models, researchers examine how the vaccination affects the immune system and whether or not it can fend against infection. This phase aids in determining any possible toxicity and improves the formulation and administration technique of the vaccine [2].

Medical Development

There are three stages of clinical development, each including a higher number of human subjects and more complex testing:

1.First Phase:

o Small-scale trials with 20–100 healthy volunteers to evaluate the safety of the vaccine, establish the right dosage, and detect any immediate negative effects. This stage is more concerned with safety than efficacy [3].

2.Phase Two:

· Expanded studies with several hundred patients to evaluate dose, safety, and immunogenicity in more detail. To show preliminary proof of the vaccine's preventive effects, phase II studies may involve participants who are at risk of contracting the illness. Additionally, this stage aids in improving the Phase III trial methodology [4].

3.Phase Three:

· Extensive studies involving hundreds to tens of thousands of people to verify the vaccination's effectiveness and track any negative effects in a wide range of demographics. These trials offer the vital information required to prove that the vaccination successfully wards off the illness and satisfies legal requirements in order to be approved. In phase III trials, robust and objective outcomes are usually ensured by randomization, double blinding, and placebo control [5].

Regulatory Examination and Acceptance

The vaccine developer submits a Biologics Licence Application (BLA) or similar to regulatory bodies like the U.S. after concluding Phase III trials with success. FDA or EMA stands for Food and Drug Administration and European Medicines Agency. A wealth of information about the vaccine's effectiveness, safety, production methods, and quality control procedures is included in the submission. Before approving something for public use, regulatory bodies check the information, inspect the manufacturing facilities, and may demand more research or explanations [6].

Manufacturing

To address the needs of public health, the vaccine must be produced in huge quantities once it has been licenced. Manufacturing entails increasing output while upholding stringent standards for uniformity and quality. Creating standardised processes for creating, refining, combining, and packaging the vaccine is a part of this phase. To guarantee the safety and effectiveness of the vaccine, manufacturers are required to adhere to Good Manufacturing Practices (GMP) [7].

After-Market Monitoring

To guarantee the vaccine's continued safety and efficacy once it has been approved and delivered, continuous monitoring is necessary. Tracking adverse events, keeping an eye on long-term immunity, and evaluating the vaccine's effect on disease incidence in the population are all part of this post-marketing surveillance, or Phase IV investigations. For the purpose of locating any uncommon or long-term side effects that might not have been

noticeable during clinical trials, regulatory bodies and public health organisations gather and examine data from patients, healthcare professionals, and continuing studies [8].

Developing Vaccines: Difficulties

In spite of technological and methodological advancements, vaccine development still faces a number of obstacles:

- **Technical Difficulties:** It is still challenging to develop vaccines against complicated diseases, such as those with high mutation rates (like HIV and influenza) or for persistent infections, like tuberculosis. It is also difficult to guarantee vaccine efficacy and stability in a variety of groups.
- **Regulatory Obstacles:** It takes a lot of time and money to navigate the regulatory environment. Obtaining clearance may become more difficult if different international regulations need to be followed.
- **Ethical Considerations:** Conducted vaccine trials in an ethical manner requiring informed consent and adherence to ethical principles is vital. It's crucial to strike a balance between the necessity for quick progress and moral principles, particularly in times of epidemic.
- **Manufacturing and Distribution:** There are several obstacles to overcome, including increasing production to meet demand worldwide, preserving cold chain logistics, and guaranteeing fair distribution, especially in environments with limited resources.

Progress in Vaccine Manufacturing

Over the years, there have been major breakthroughs in the production of vaccines, especially with the integration of biotechnological innovations. Thanks to these developments, manufacturing procedures have become more scalable, economical, and efficient, guaranteeing that vaccines can be produced in large enough quantities to satisfy demand worldwide.

Systems of Expression

Creating recombinant proteins or antigens using different expression systems is one of the major developments in vaccine manufacture. These systems comprise mammalian, yeast, bacterial, and insect cells, each with its own benefits and difficulties.

1. Systems of Bacterial Expression:

- o **Benefits:** Bacteria, such *Escherichia coli*, are simple to grow and can generate a lot of protein quickly. They are affordable and useful for making basic proteins.
- o **Drawbacks:** It's possible that bacterial systems are incapable of carrying out post-translational changes, which are necessary for many proteins to fold and function correctly [1].

2. Systems of Yeast Expression:

o Benefits: Yeast are generally simple to scale up, and some of them, like *Saccharomyces cerevisiae*, are capable of performing certain post-translational alterations. Vaccines like the recombinant hepatitis B vaccine are made in yeast systems.

o Drawbacks: Hyperglycosylation can be introduced by yeast systems, which could influence how immunogenic the proteins produced are [2].

3. Systems of Insect Cells:

o Benefits: Insect cells with the proper post-translational modifications can create complex proteins. One example of such cells is the baculovirus expression system. They are applied to vaccinations such as the HPV (human papillomavirus) vaccine.

The disadvantages of using insect cell cultures over bacterial systems are their higher complexity and higher maintenance costs [3].

4. Animal Cell Structures:

o Benefits: Human-like post-translational modifications can be produced by mammalian cells, including Chinese hamster ovary (CHO) cells, guaranteeing correct folding and function. They are applied to therapeutic proteins and complicated vaccinations.

o Drawbacks: Maintaining mammalian cell cultures is costly and necessitates the use of complex bioreactors and strict quality control procedures [4].

Large-Scale Production Techniques

Advancements in bioprocessing technologies have significantly improved the scalability and efficiency of vaccine production. Key techniques include:

1. **Bioreactors:**

- o Large-scale bioreactors are used to culture cells and produce recombinant proteins in a controlled environment. Advances in bioreactor design, such as single-use bioreactors and continuous perfusion systems, have increased production capacity and reduced contamination risks [5].

2. **Downstream Processing:**

- o Downstream processing involves the purification and formulation of the produced antigen. Techniques such as chromatography, ultrafiltration, and diafiltration are used to isolate and purify the antigen from the culture medium. Improvements in these techniques have enhanced yield and purity, ensuring consistent vaccine quality [6].

3. **Formulation and Stability:**

- o The formulation of vaccines is critical for ensuring their stability and efficacy. Advances in adjuvant technology, stabilizers, and delivery systems have improved the immunogenicity and shelf-life of vaccines. For instance, liposome-based formulations and novel adjuvants like AS01 have been developed to enhance vaccine performance [7].

Purification and Formulation Strategies

Purification and formulation are essential steps in vaccine production, ensuring that the final product is safe, pure, and effective.

1. Purification:

- High-performance liquid chromatography (HPLC), affinity chromatography, and size-exclusion chromatography are commonly used to purify vaccine antigens. These methods help remove impurities and contaminants, such as host cell proteins, DNA, and endotoxins, ensuring that the vaccine meets regulatory standards for safety and purity [8].

2. Formulation:

- The formulation process involves combining the purified antigen with adjuvants, stabilizers, and preservatives to create a stable and effective vaccine. Adjuvants enhance the immune response, stabilizers maintain the antigen's integrity during storage, and preservatives prevent microbial contamination. Advances in formulation science have led to the development of more effective and stable vaccines [9].

Quality Control and Assurance

Ensuring the quality and consistency of vaccines is paramount. Regulatory agencies require rigorous quality control and assurance measures at every stage of production.

1. Quality Control Tests:

- These tests include assessing the purity, potency, and sterility of the vaccine. Techniques such as enzyme-linked immunosorbent assay (ELISA), Western blotting, and mass spectrometry are used to verify the antigen's identity and concentration. Sterility tests ensure that the vaccine is free from microbial contamination [10].

2. Good Manufacturing Practices (GMP):

- GMP guidelines provide a framework for the production, testing, and distribution of vaccines. Adhering to GMP ensures that vaccines are consistently produced and controlled according to quality standards. This involves proper documentation, validation of processes, and regular audits by regulatory agencies [11].

Novel Vaccine Platforms

The development of novel vaccine platforms has revolutionized the field of immunization, providing new tools and strategies to combat a wide range of infectious diseases. These platforms leverage advances in molecular biology, genetics, and nanotechnology to create vaccines that are more effective, easier to produce, and capable of addressing emerging health threats.

mRNA Vaccines

mRNA vaccines represent a groundbreaking advancement in vaccine technology. These vaccines use synthetic messenger RNA (mRNA) to instruct cells to produce a protein that triggers an immune response. The COVID-19 vaccines developed by Pfizer-BioNTech and Moderna are prime examples of this innovative approach [1].

Mechanism of Action:

- mRNA vaccines contain a lipid nanoparticle-encapsulated mRNA sequence that encodes the antigen of interest. Once injected, the mRNA is taken up by cells, which then translate it into the antigen protein. The immune system recognizes this protein as foreign, triggering an immune response that includes the production of antibodies and activation of T cells [2].

Advantages:

- Rapid development and production capabilities.
- Strong immune response, including both humoral (antibody-mediated) and cellular (T-cell-mediated) immunity.
- No risk of causing disease, as no live virus is used.
- Flexibility to quickly adapt to new pathogens or variants.

Challenges:

- Requires storage at very low temperatures, complicating distribution and logistics.
- Potential for short-term side effects, such as fever and fatigue, which are generally mild and temporary [3].

Viral Vector Vaccines

Viral vector vaccines use a different virus (vector) to deliver genetic material encoding an antigen from the target pathogen. Examples include the Ebola vaccine and COVID-19 vaccines by AstraZeneca and Johnson & Johnson [4].

Mechanism of Action:

- These vaccines use a harmless virus, often an adenovirus, as a vector to deliver the genetic material of the pathogen's antigen. The vector infects cells, which then produce the antigen, eliciting an immune response [5].

Advantages:

- Can induce strong immune responses, including both humoral and cellular immunity.

- Capable of being used for multiple diseases by changing the genetic material delivered by the vector.
- Generally stable and easier to distribute compared to mRNA vaccines.

Challenges:

- Pre-existing immunity to the vector virus can reduce vaccine efficacy.
- Potential for vector-related side effects, although generally safe [6].

DNA Vaccines

DNA vaccines involve the direct injection of plasmid DNA encoding the antigen. These vaccines are being explored for various infectious diseases, including COVID-19, Zika virus, and HIV [7].

Mechanism of Action:

- DNA vaccines deliver plasmid DNA into cells, which then use the genetic instructions to produce the antigen protein. The immune system recognizes the antigen as foreign and mounts an immune response [8].

Advantages:

- Stable at room temperature, simplifying storage and distribution.
- Can induce both humoral and cellular immune responses.
- Relatively easy and cost-effective to produce.

Challenges:

- Currently, no DNA vaccines have been approved for human use, and more research is needed to establish their efficacy and safety.
- Delivery methods (e.g., electroporation) can be invasive and require specialized equipment [9].

Nanoparticle-Based Vaccines

Nanoparticle-based vaccines use nanotechnology to enhance the delivery and presentation of antigens. These vaccines can include virus-like particles (VLPs), liposomes, and other nanomaterials designed to mimic the structure of pathogens and improve immune responses [10].

Mechanism of Action:

- Nanoparticles can encapsulate antigens or adjuvants, facilitating their delivery to immune cells. They can also enhance antigen stability and presentation, leading to a stronger and more targeted immune response [11].

Advantages:

- Enhanced delivery and presentation of antigens.
- Potential to improve vaccine efficacy and reduce required doses.
- Can be designed to target specific cells or tissues, improving the precision of the immune response.

Challenges:

- Complex manufacturing processes and scalability issues.
- Regulatory hurdles and the need for extensive safety testing.

Case Studies of Biotechnological Innovations in Vaccines

Biotechnological advancements have played a crucial role in developing vaccines that address significant public health challenges. This section explores several case studies that highlight the impact of these innovations on vaccine development and deployment.

COVID-19 Vaccines

The COVID-19 pandemic spurred unprecedented efforts in vaccine development, leading to the rapid creation and deployment of several effective vaccines. Two notable examples are the Pfizer-BioNTech and Moderna mRNA vaccines [1].

Pfizer-BioNTech and Moderna mRNA Vaccines:

- These vaccines use lipid nanoparticles to deliver mRNA encoding the SARS-CoV-2 spike protein. The mRNA instructs cells to produce the spike protein, triggering an immune response [2].
- Clinical trials demonstrated remarkable efficacy, with both vaccines showing around 95% effectiveness in preventing symptomatic COVID-19 [3].
- The rapid development and approval of these vaccines, achieved within a year of the pandemic's onset, showcased the potential of mRNA technology in responding to emerging infectious diseases.

AstraZeneca and Johnson & Johnson Viral Vector Vaccines:

- These vaccines use adenovirus vectors to deliver genetic material encoding the SARS-CoV-2 spike protein. The vectors are modified to be replication-deficient, ensuring safety [4].

- Clinical trials showed robust immune responses and substantial protection against COVID-19, contributing significantly to global vaccination efforts [5].

HPV Vaccines

Human papillomavirus (HPV) vaccines represent another significant biotechnological achievement. The development of recombinant vaccines like Gardasil and Cervarix has had a profound impact on preventing HPV-related cancers [6].

Gardasil and Cervarix:

- These vaccines use virus-like particles (VLPs) composed of HPV L1 protein, produced using recombinant DNA technology. VLPs resemble the natural virus but do not contain viral DNA, making them safe and highly immunogenic [7].
- Clinical studies have shown that these vaccines are highly effective in preventing HPV infection and related cancers, such as cervical, anal, and oropharyngeal cancers [8].
- The widespread adoption of HPV vaccines has led to significant reductions in HPV infections and precancerous lesions, highlighting their public health impact [9].

Malaria Vaccines

Malaria remains a major global health challenge, particularly in sub-Saharan Africa. The development of the RTS,S/AS01 malaria vaccine, also known as Mosquirix, is a notable example of biotechnological innovation in combating this disease [10].

RTS,S/AS01 (Mosquirix):

- This vaccine targets the circumsporozoite protein (CSP) of *Plasmodium falciparum*, the most deadly malaria parasite. It uses a recombinant protein combined with the AS01 adjuvant to enhance the immune response [11].
- Clinical trials demonstrated that RTS,S/AS01 provides partial protection against malaria, reducing the incidence of clinical malaria in young children [12].
- Despite its moderate efficacy, Mosquirix represents a critical tool in the fight against malaria, and ongoing research aims to improve its performance and develop new malaria vaccines.

Challenges and Limitations

Despite the remarkable advancements in vaccine development, several challenges and limitations persist. Addressing these issues is critical for ensuring the continued success and accessibility of vaccines worldwide.

Technical Challenges

1. Pathogen Complexity:

- Some pathogens, such as HIV and influenza, have high mutation rates, leading to antigenic variation and making it difficult to develop long-lasting vaccines. For HIV, the virus's rapid mutation and diversity present significant obstacles to creating a broadly effective vaccine [1].
- Influenza viruses undergo frequent changes through antigenic drift and shift, necessitating the annual reformulation of vaccines to match circulating strains [2].

2. Stability and Storage:

- Ensuring the stability of vaccines during storage and transport is a major challenge, particularly for vaccines that require cold chain logistics, such as mRNA vaccines. Maintaining the ultra-cold temperatures needed for these vaccines is difficult, especially in low-resource settings [3].

3. Manufacturing Scalability:

- Scaling up the production of vaccines to meet global demand while maintaining quality and consistency is complex. Biotechnological advancements have improved manufacturing processes, but challenges remain in ensuring that large-scale production meets regulatory standards [4].

4. Delivery Methods:

- Developing effective and acceptable delivery methods is essential for vaccine uptake. For example, needle-free delivery systems, such as oral or nasal vaccines, can improve accessibility and acceptance but require extensive research and development [5].

Regulatory Hurdles

1. Regulatory Approval:

- Navigating the regulatory landscape for vaccine approval involves substantial time, resources, and rigorous testing. Regulatory requirements vary by country, and harmonizing these standards to facilitate global access can be challenging [6].

2. Post-Marketing Surveillance:

- Continuous monitoring of vaccine safety and effectiveness after approval is critical for identifying rare adverse events and ensuring long-term safety. Establishing robust

surveillance systems in all countries, especially those with limited resources, is essential [7].

Ethical Considerations

1. Clinical Trials:

- Conducting ethical clinical trials involves obtaining informed consent, ensuring participant safety, and balancing the risks and benefits. Trials must be designed to minimize risks and protect vulnerable populations, such as children and pregnant women [8].

2. Access and Equity:

- Ensuring equitable access to vaccines is a significant ethical challenge. Disparities in vaccine distribution and access can exacerbate health inequalities, particularly in low-income countries. Addressing these disparities requires global cooperation and strategies to make vaccines affordable and accessible to all [9].

Manufacturing and Distribution

1. Production Capacity:

- Building and maintaining sufficient production capacity to meet global demand, especially during pandemics, is a significant challenge. Investments in infrastructure and technology are necessary to scale up production rapidly [10].

2. Cold Chain Logistics:

- Maintaining the cold chain for vaccines that require refrigeration or freezing is critical to preserving their efficacy. This is particularly challenging in low-resource settings with limited infrastructure [11-].

3. Supply Chain Management:

- Efficient supply chain management is essential for the timely distribution of vaccines. Disruptions in the supply chain, such as shortages of raw materials or logistical delays, can hinder vaccination efforts [12].

Impact of Biotechnology on Vaccine Safety and Efficacy

Biotechnology has significantly enhanced the safety and efficacy of vaccines, leading to more reliable and potent immunization strategies. This section explores the various ways in which biotechnological advancements have improved vaccine design, production, and performance.

Improvements in Safety Profiles

1. Precise Antigen Design:

- Biotechnology allows for the precise design of antigens, selecting specific protein subunits or peptides that elicit strong immune responses while minimizing the risk of adverse reactions. For example, subunit vaccines like the hepatitis B vaccine use recombinant DNA technology to produce specific viral proteins, reducing the likelihood of side effects compared to whole-virus vaccines [13].

2. Elimination of Virulent Pathogens:

- Techniques such as recombinant DNA and genetic engineering enable the production of vaccines without the need for live or inactivated pathogens, eliminating the risk of reversion to virulence. This is particularly important for live-attenuated vaccines, where there is a small risk that the weakened virus could regain its pathogenicity [12].

3. Adjuvant Development:

- Advances in adjuvant technology have improved the safety and efficacy of vaccines. Modern adjuvants, such as AS01 and AS03, enhance immune responses without significantly increasing reactogenicity. These adjuvants can reduce the required dose of antigen, thereby lowering the potential for adverse reactions [14-15].

Enhanced Immunogenicity

1. Improved Delivery Systems:

- Novel delivery systems, such as lipid nanoparticles used in mRNA vaccines, ensure efficient delivery of antigens to the immune system. These systems protect the antigen from degradation and facilitate its uptake by immune cells, leading to stronger and more targeted immune responses [4].

2. Multivalent Vaccines:

- Biotechnology enables the development of multivalent vaccines that protect against multiple strains or types of a pathogen. For example, the quadrivalent HPV vaccine (Gardasil) provides protection against four different HPV types, broadening the scope of immunity and enhancing overall efficacy [15].

3. Antigen Presentation:

- Technologies such as virus-like particles (VLPs) and nanoparticle-based vaccines enhance antigen presentation to the immune system. VLPs mimic the structure of

viruses, providing repetitive antigenic structures that are highly immunogenic and capable of inducing robust immune responses [16].

Reduction in Adverse Effects

1. Targeted Immunization:

- By using specific antigens and advanced adjuvants, biotechnological vaccines can elicit strong immune responses with lower doses, reducing the risk of adverse effects. For instance, the recombinant zoster vaccine (Shingrix) uses a subunit antigen combined with the AS01 adjuvant, providing high efficacy with minimal side effects [17].

2. Elimination of Allergens:

- Biotechnology allows for the removal of allergens and other harmful components from vaccine formulations. Recombinant technology can produce vaccines without egg proteins, which are commonly associated with allergic reactions in traditional influenza vaccines [18].

3. Genetic Stability:

- Genetic engineering ensures the stability of vaccine strains, preventing the accumulation of mutations that could lead to reduced efficacy or increased reactivity. This is crucial for vaccines developed through attenuation or recombinant methods [19].

Future Directions and Emerging Trends

The field of vaccine development is rapidly evolving, driven by continuous advancements in biotechnology and a deeper understanding of immunology. This section explores the future directions and emerging trends that are poised to shape the landscape of vaccine development in the coming years.

Personalized Vaccines

1. Tailored Immunization:

- Personalized vaccines are designed to cater to the specific genetic and immunological profiles of individuals. Advances in genomics and bioinformatics enable the identification of unique biomarkers and immune signatures, allowing for the customization of vaccines to optimize efficacy and minimize adverse effects [1,20].

2. Cancer Vaccines:

- Personalized cancer vaccines are an emerging area of research, aiming to target tumor-specific antigens (neoantigens) unique to an individual's cancer. These vaccines can stimulate the immune system to recognize and destroy cancer cells while sparing healthy tissue. Clinical trials are underway to evaluate the efficacy of these tailored immunotherapies [2].

Universal Vaccines

1. Broad-Spectrum Immunization:

- The development of universal vaccines aims to provide broad protection against multiple strains or types of a pathogen. Universal influenza vaccines, for instance, target conserved regions of the virus that are less prone to mutation, offering long-lasting protection against seasonal and pandemic influenza strains [3].

2. Universal Coronavirus Vaccines:

- In response to the COVID-19 pandemic, efforts are underway to develop universal coronavirus vaccines that can protect against a wide range of coronaviruses, including potential future variants. These vaccines target conserved viral proteins, enhancing their ability to provide broad and durable immunity [4].

Artificial Intelligence and Machine Learning

1. Vaccine Design and Optimization:

- Artificial intelligence (AI) and machine learning (ML) are revolutionizing vaccine development by enabling the rapid analysis of large datasets to identify optimal vaccine candidates. AI algorithms can predict antigen structures, evaluate immunogenicity, and optimize vaccine formulations, accelerating the development process [5].

2. Predictive Modeling:

- AI and ML can be used to model disease outbreaks and predict the spread of infectious diseases, informing vaccine deployment strategies. These technologies can also identify potential vaccine safety concerns and improve post-marketing surveillance [6].

New Delivery Platforms

1. Microneedle Patches:

- Microneedle patches offer a painless and needle-free method of vaccine delivery. These patches contain tiny needles that dissolve upon contact with the skin, delivering

the vaccine directly to immune cells in the dermis. Microneedle patches are being explored for various vaccines, including those for influenza and COVID-19 [7].

2. Oral and Nasal Vaccines:

- Oral and nasal vaccines provide convenient and non-invasive alternatives to traditional injections. These vaccines stimulate mucosal immunity, which is crucial for protecting against respiratory and gastrointestinal pathogens. Advances in formulation and delivery technologies are making these vaccines more effective and widely applicable [8].

Synthetic Biology and Nanotechnology

1. Synthetic Antigens:

- Synthetic biology enables the design and production of novel antigens that mimic natural pathogens, enhancing the immune response. These synthetic antigens can be tailored to elicit strong and specific immunity, offering new avenues for vaccine development [9].

2. Nanoparticle Vaccines:

- Nanoparticles can be engineered to improve the delivery and presentation of antigens, enhancing vaccine efficacy. These particles can mimic the size and shape of viruses, facilitating uptake by immune cells and promoting robust immune responses. Research is ongoing to develop nanoparticle-based vaccines for a range of infectious diseases [10].

Public Health Implications

Vaccines have long been recognized as one of the most effective public health interventions, saving millions of lives annually and preventing countless cases of disease. The impact of biotechnological advancements in vaccine development extends beyond individual health, significantly influencing public health at local, national, and global levels.

Impact on Global Health

1. Reduction in Disease Burden:

- Vaccines have drastically reduced the burden of infectious diseases worldwide. Diseases like smallpox have been eradicated, and others, such as polio and measles, have been significantly controlled through widespread vaccination programs. These successes highlight the critical role of vaccines in improving global health outcomes [1].

2. Control of Emerging Infectious Diseases:

- Biotechnology has enabled the rapid development of vaccines against emerging infectious diseases. The swift creation and deployment of COVID-19 vaccines exemplify how modern technology can address urgent public health crises, potentially saving millions of lives and preventing widespread economic disruption [2].

Vaccine Accessibility and Equity

1. Ensuring Equitable Distribution:

- One of the major challenges in global health is ensuring equitable access to vaccines, especially in low- and middle-income countries. Biotechnological advancements, such as thermostable vaccines that do not require refrigeration, can help overcome logistical barriers and improve vaccine access in remote and resource-limited settings [3].

2. Global Initiatives:

- Initiatives like COVAX aim to ensure fair distribution of COVID-19 vaccines globally, demonstrating the importance of international cooperation in addressing vaccine inequities. These efforts are crucial for controlling pandemics and achieving global health equity [4].

Strategies for Increasing Vaccine Acceptance and Coverage

1. Addressing Vaccine Hesitancy:

- Vaccine hesitancy remains a significant barrier to achieving high vaccination coverage. Public health campaigns that leverage social media, community engagement, and education are essential for building trust and addressing misinformation about vaccines. Biotechnological advancements can also improve vaccine safety profiles, reducing concerns about adverse effects [5].

2. Enhancing Vaccine Delivery:

- Innovative delivery methods, such as microneedle patches and oral vaccines, can improve vaccination rates by making administration easier and more acceptable, particularly for populations with needle phobia or limited access to healthcare facilities [6].

Long-Term Public Health Benefits

1. Herd Immunity:

- High vaccination coverage leads to herd immunity, where a significant portion of the population becomes immune to a disease, indirectly protecting those who cannot be vaccinated. Herd immunity is crucial for controlling the spread of contagious diseases and protecting vulnerable groups, such as infants, the elderly, and immunocompromised individuals [7].

2. Economic Benefits:

- Vaccination programs provide substantial economic benefits by reducing healthcare costs associated with treating preventable diseases and minimizing productivity losses due to illness. The return on investment for vaccines is high, making them a cost-effective public health intervention [8].

Challenges and Future Directions

1. Sustaining Immunization Programs:

- Sustaining high vaccination coverage requires ongoing public health efforts and investments. It is essential to maintain robust immunization programs, continuous monitoring, and timely responses to outbreaks to ensure that gains in disease control are not lost [9].

2. Adapting to New Challenges:

- Emerging pathogens, evolving vaccine-resistant strains, and changing demographics pose ongoing challenges for vaccine development and distribution. Biotechnology will continue to play a pivotal role in addressing these challenges by enabling the rapid adaptation of vaccines to new threats and ensuring their effectiveness across diverse populations [10].

Conclusion

The role of biotechnology in vaccine development is transformative, providing innovative solutions to some of the most pressing public health challenges. From the precise design of antigens and advanced manufacturing techniques to the creation of novel vaccine platforms, biotechnology has revolutionized the field, enabling the rapid development and deployment of effective vaccines.

The historical progression from traditional methods to biotechnological approaches underscores the significant advancements made in ensuring vaccine safety, efficacy, and accessibility. Each milestone has built upon previous discoveries, leading to the development of vaccines that are not only more effective but also safer and easier to produce and distribute.

Biotechnological innovations, such as mRNA vaccines, viral vector vaccines, and nanoparticle-based vaccines, have demonstrated their potential in responding to emerging infectious diseases and improving global health outcomes. The rapid development and widespread deployment of COVID-19 vaccines exemplify how biotechnology can address urgent public health crises, highlighting the importance of continued investment and research in this field.

Despite these advancements, several challenges and limitations remain, including technical hurdles, regulatory complexities, ethical considerations, and logistical challenges. Addressing these issues is critical for ensuring that vaccines can reach all populations in need and for maximizing their public health impact.

The future of vaccine development is bright, with numerous emerging trends and technologies poised to further enhance the field. Personalized vaccines, universal immunization strategies, AI-driven optimization, novel delivery platforms, and advancements in synthetic biology and nanotechnology hold the promise of more effective, safer, and accessible vaccines.

Public health implications of these advancements are profound, encompassing disease control, improved global health, enhanced vaccine accessibility, and long-term economic benefits. Ensuring equitable distribution, addressing vaccine hesitancy, and maintaining robust immunization programs are critical for maximizing the impact of vaccines on public health. Continued innovation and international cooperation will be essential for overcoming current and future challenges, ultimately contributing to a healthier and more resilient global population.

In summary, biotechnology has revolutionized vaccine development, providing powerful tools and strategies to combat infectious diseases and improve global health. The continued integration of biotechnological advancements in vaccine development will be crucial for addressing existing and emerging health threats, ensuring that vaccines remain a cornerstone of public health in the years to come.

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