International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781

Overview of Recombinant Technology in the Development of Covid-19 Vaccine

Malathi Thiyagarajan*, Dr., DR. Surendra kumar
Affiliation : T.Malathi Thiyagarajan M.Pharm .,
Department of pharmacology, Senghundhar college of pharmacy.

Date of Submission: 20-08-2021 Date of Acceptance: 03-09-2021

ABSTRACT
The current review focuses on the recent advances in recombinant COVID-19 vaccine research and development and associated issues. Vaccines are the primary intervention strategy for control of corona virus transmission all over the world. Developing a new vaccine and its available for people it takes many years to compete all research procedure. Even though for the development of a vaccine against SARS-CoV-2, including the use of inactive or live-attenuated viruses, virus-like particles (VLPs),viral vectors and protein-based, DNA-based, and mRNA-based vaccines. However, the development of a vaccine typically spans 10–15 years. Therefore we need rapid identification and publication of the SARS-CoV-2 gene sequence, it was only a matter of months before the first vaccine candidate was ready for clinical testing. Based on advances in techniques for vaccine design, inactivated, live-vectored, nucleic acid, and recombinant COVID-19 vaccines are being developed and tested for their efficacy faster. Currently, more than 60 SARS-CoV-2 vaccines are developed at different clinical trial phases under following sections namely inactivated, live-attenuated, and recombinant vaccines.

KEYWORDS: Recombinant, Vaccine, SARS-CoV-2, SARS-CoV-1, COVID-19

I. INTRODUCTION:

COVID-19 is an unusual global health threat wherein the vaccine is needed immediately. Because of the high risk of collapsing healthcare systems in several countries, governments had to implement lockdown measures, including no international travel and other public containment measures in attempting to reduce the virus morbidity and mortality worldwide. Although effective, these actions led to vast economic devastation. [1, 3]

In this pandemic situation, it is crucial to ensure that rigorous and adequate clinical trials are performed to evaluate drugs with antiviral effects for avoiding the usage of ineffective and unsafe drugs. Current clinical trials of candidate vaccines are undergoing phase 1 or parallel 1/2 studies to initiate phase 3 at the earliest and decrease the time required for the development of the new vaccine.

Delta and Delta Plus variant of the SARS-CoV-2 virus have emerged as new threats to India's fight against the ongoing pandemic which witnessed several milestones of success with new infections and the number of vaccinations going up. Delta, first detected in India, is a global concern while Delta Plus — with 40 cases in India , Delta Plus is a variant of Delta.[1] While Delta was first reported in India, Delta plus first reported by Public Health England on its June 11 bulletin. It said that the new variant was present in six genomes from India as of June 7.

Delta has rapidly spread in the United Kingdom and reports said that Delta variant is responsible for 99 per cent of the UK's cases. In the UK itself, the number has surpassed 33,000 in a week. This variant is spreading to other countries as well. According to the Centre, as of June 18, 205 Delta Plus cases were detected worldwide, with the USA and the UK reporting more than half of the known cases. Delta variant of coronavirus has been held responsible for the second wave of the pandemic in India. Several other countries to consider , Delta as the factor behind a sudden surge of cases. [1, 2]

Delta Plus resists monoclonal antibody cocktail therapy in which artificial antibodies are produced in the body. But India's expert committee has confirmed that resistance to this does not mean that this variant is more virulent. mutations may enable the coronavirus to spread faster from person to person, and more infections can result in more people getting very sick or dying.

International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781

Vaccines are a very powerful tool in our fight against COVID-19.

Recombinant DNA technology:

Recombinant DNA technology is the joining together of DNA molecules from two different species. The recombined DNA molecule is inserted into a host organism to produce new genetic combinations that are of value to science, medicine, agriculture, and industry. Since the focus of all genetics is the gene, the fundamental goal of laboratory geneticists is to isolate, characterize, and manipulate genes. Recombinant DNA technology is based primarily on two other technologies, cloning and DNA sequencing. Cloning is undertaken in order to obtain the clone of one particular gene or DNA sequence of interest. The next step after cloning is to find and isolate that clone among other members of the library (a large collection of clones). Once a segment of DNA has been cloned, its nucleotide sequence can be determined. Knowledge of the sequence of a DNA segment has many uses.

Through recombinant DNA techniques, bacteria have been created that are capable of synthesizing human insulin, human growth hormone, alpha interferon, hepatitis B vaccine, and other medically useful substances.[4]

The **principle of recombinant DNA technology** involved four steps,

(1) Gene Cloning and Development of Recombinant DNA
(2) Transfer of Vector into the Host
(3) Selection of Transformed Cells and
(4) Transcription and Translation of Inserted Gene.

Recombinant Vaccines:

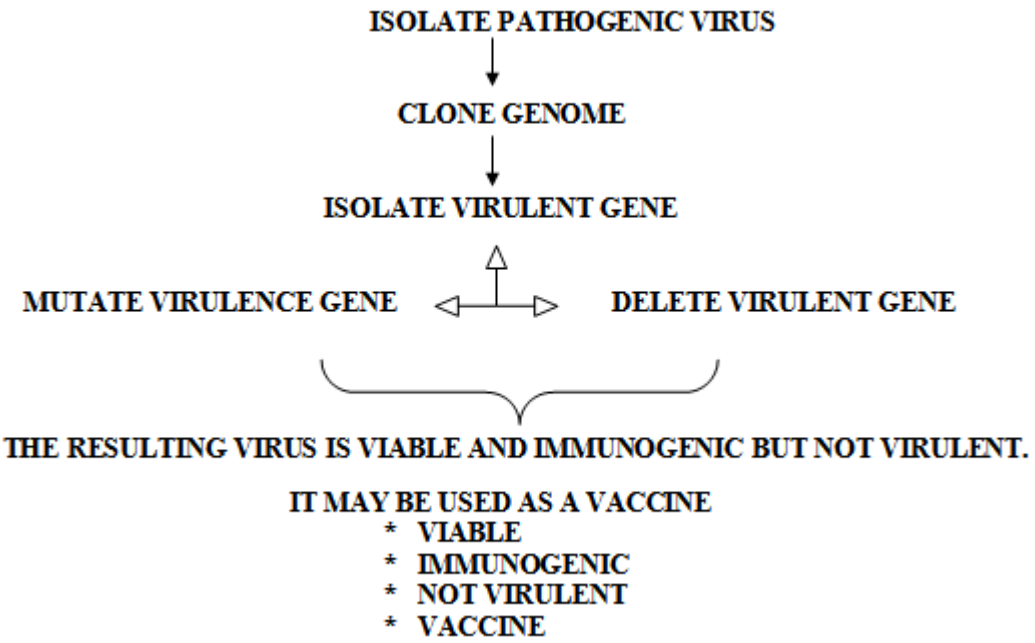
A recombinant vaccine is a vaccine produced through recombinant DNA technology. This involves inserting the DNA encoding an antigen (such as a bacterial surface protein) that stimulates an immune response into bacterial or mammalian cells, expressing the antigen in these cells and then purifying it from them.

The ChAdOx1-S/nCoV-19 [recombinant] vaccine is a replication-deficient adenoviral vector vaccine against coronavirus disease 2019 (COVID-19). The vaccine expresses the SARS-CoV-2 spike protein gene, which instructs the host cells to produce the protein of the S-antigen unique to SARS-CoV-2, allowing the body to generate an immune response and to retain that information in memory immune cells. Efficacy shown in clinical trials in participants who received the full series of vaccine (2 doses) irrespective of interval between the doses was 63.1%, based on a median follow-up of 80 days, but tended to be higher when this interval was longer. The data reviewed at this time support the conclusion that the known and potential benefits of ChAdOx1-S/nCoV-19 [recombinant] vaccine outweigh the known and potential risks. [3,4]

In addition to live vectored and inactivated viruses, novel recombinant technologies are being used in the development of COVID-19 vaccine. The advantage of recombinant vaccines is their greater response predictability and improved efficacy. The ChAdOx1-S/nCoV-19 [recombinant] vaccine is a replication-deficient adenoviral vector vaccine against corona virus disease 2019 (COVID-19).

SARS-CoV-2, the causative agent of COVID-19, has imposed a major public health threat, which needs effective therapeutics and vaccination strategies. Several potential candidate vaccines being rapidly developed are in clinical evaluation. Considering the crucial role of SARS-CoV-2 spike (S) glycoprotein in virus attachment, entry, and induction of neutralizing antibodies, S protein is being widely used as a target for vaccine development. Based on advances in techniques for vaccine design, inactivated, live-vectored, nucleic acid, and recombinant COVID-19 vaccines are being developed and tested for their efficacy. Phase3 clinical trials are underway or will soon begin for several of these vaccines. Assuming that clinical efficacy is shown for one or more vaccines, safety is a major aspect to be considered before deploying such vaccines to the public.

International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781



II. OBJECTIVE OF THE STUDY:

Coronavirus vaccines: development and achievement

Vaccines are the only one weapon to control of coronavirus transmission and infection. Several methods are available for the development of a vaccine against SARS-CoV-2, including the use of inactive or live-attenuated viruses, virus-like particles (VLPs), viral vectors, and protein-based, DNA-based, and mRNA-based vaccines. However, the development of a vaccine typically spans 10–15 years. However, owing to the rapid identification and publication of the SARS-CoV-2 gene sequence, it was only a matter of months before the first vaccine candidate was ready for clinical testing. [5]

Inactivated coronavirus vaccine

The development of inactivated vaccines requires a target virus to be initially inactivated, either chemically or by irradiation. This allows the nucleic acids of the virus to be destroyed, while keeping the viral antigens intact. The immunological characteristics and effectiveness of inactivated CoV vaccines were investigated in animal models during the emergence of the first SARS virus. An inactivated vaccine against SARS-CoV was first evaluated in rhesus monkeys, which was found to induce humoral and mucosal immunity, highlighting its potential for use in clinical trials. A double-inactivated, candidate whole-virus vaccine against SARS-CoV was also developed using sequential exposure to formaldehyde and ultraviolet radiation to ensure its safe use. The immunogenicity of this vaccine was verified using a mouse model, which showed high antibody titers against the CoV S protein and enhanced neutralizing antibodies, highlighting its potential for application as a platform for the development of a SARS-CoV-2 vaccine. [5,6]

Recently Gao et al (2020) developed PiCoVacc, a purified inactivated SARS-CoV-2 virus vaccine, that was found to incite SARS-CoV-2-specific neutralizing antibodies in mice, rats, and non-human primates. The generated antibodies were found to neutralize 10 representative strains of SARS-CoV-2, holding up its broad-ranged applicability against the virus. However, there is a potential public health risk associated with incomplete inactivation, which leads to undesired immune or inflammatory responses.

Live-attenuated coronavirus vaccine

Live-attenuated vaccines are being developed from live coronaviruses whose virulence has been reduced under laboratory conditions. This

International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781

technique allows for the virus to replicate in the host while producing only mild pathogenesis, if any. Live-attenuated vaccines are one of the basic technologies used for the development of licensed human vaccines. However, the spread of CoV via the feces of individuals who have received a live-attenuated vaccine and the risk of its recombination with wild-type CoV are among its major safety concerns. Another issue is its suitability for the older population, who are at a higher risk of severe disease.

The Serum Institute of India has allied with Codagenix Inc., a major US pharmaceutical company, to develop a live-attenuated vaccine against SARS-CoV-2, which is presently in the preclinical stage.However, owing to its safety concerns, in particular with regard to elderly individuals (at a higher risk of COVID 19), the use of live-attenuated virus vaccines is unlikely to represent the best approach.[8,9]

III. BACKGROUND OF THE STUDY:

Five Types of COVID-19 Vaccines

The vaccine roster includes traditional strategies like entire disabled viruses that fool the immune system and molecular approaches that instruct it. Proteins, DNA, or RNA can't cause infection and fit into platforms developed in recent years to treat other emerging infectious diseases. And DNA or RNA sequences can be tweaked – "optimized" – to elicit specific responses.[9, 10] .The candidate COVID vaccines can be grouped in various ways, lumping molecular techniques or defining them more granularly.

- **Inactivated virus**
- **Vectored viruses**
- **Protein**
- **DNA**
- **RNA**

Nucleic acid-based coronavirus vaccine :

The greatest advantage of DNA- and RNA-based vaccines is their potential for rapid development and reduced side effects. DNA vaccines have shown strong potential to trigger immune responses against CoVs in animal models. However, clinical data on the efficacy of DNA vaccines in humans remain limited. In a previous study on mice, a DNA vaccine encoding the S protein of SARS-CoV was found to induce T cells, a neutralizing antibody response, and protective immunity.A group of prototype DNA vaccines expressing various SARS-CoV-2 S proteins has been developed and tested in 35 rhesus macaques. The vaccinated macaques demonstrated specific humoral and cellular immune responses. Further upon being challenged with SARS-CoV-2, the animals showed a remarkable reduction of viral replication in the upper and lower respiratory tract. The data displayed the significant role of DNA vaccine against SARS-CoV-2 infection.

Protein-based coronavirus vaccine

The S protein is a good candidate target for vaccine development. Tazehkand and Hajipour (2020) fused an envelope and nucleocapsid protein with multi epitopes (B and MHC I epitopes) derived from the S protein and RNA-dependent RNA polymerase to construct a fusion vaccine. Although the vaccine was verified for its structural stability as well as physicochemical and immunological properties during a preliminary screening, the authors anticipated the need for further experiments with laboratory animals. [11]

Vectored vaccines against coronavirus

Viral vectors represent one of the prospective strategies for the CoV vaccine platform. Their utility depends on their ability to infect cells. The main advantage of this platform is its efficient and gene-specific delivery as well as its initiation of healthy immune responses. A recombinant adenovirus type-5 (Ad5) vectored COVID-19 vaccine expressing S protein of SARS-CoV-2 was assessed for phase 1 trial at Wuhan, China. The increase in specific neutralizing antibodies and T cell response were observed on day 14 after vaccination. The results remained promising and expect further evaluation. Several groups have reported the results of preclinical trials of SARS-CoV-2 vaccines using other viruses as vectors, including measles replicating viral vector (Zydus Cadila, Institut Pasteur/Themis/University of Pittsburgh Center for Vaccine Research), influenza vector expressing RBD (University of Hongkong), and non-replicating viral vector adenovirus-based NasoVAX expressing the SARS-CoV-2 spike protein (Altimmune).[12]

Artificially synthesized protein-microarray

A recent study reported the development of a microneedle array-based recombinant SARS-CoV-2 S1 subunit vaccine. This vaccine was tested for its immunogenicity in vivo and was found to be able to induce an effective antigen-specific antibody response within two weeks post-immunization.

International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781

Virus-like particle-based vaccine

Virus-like particles (VLPs) are multi-protein supra-molecular preparations with features equivalent to those of viruses. They represent a resourceful platform for vaccine development owing to their flexible immunological features, including suitable size, repetitive surface geometry, and stimulation of innate and adaptive immune responses. VLP-based vaccines target B lymphocytes and induce potent antibody responses, resulting in T helper cell activation and their presentation on MHC class II molecules via antigen-presenting cells (APCs). Medicago Inc., a leading US-based biopharmaceutical company, recently developed a VLP-based vaccine against SARS-CoV-2. This vaccine is currently undergoing preclinical studies to determine its safety and efficacy. Currently, the existing expression method for foreign proteins in plants represents a potential platform for the generation of suitable vaccine candidates against SARS-CoV-2 with reservations.[12, 13]]

IV. CURRENT STATUS AND FUTURE ASPECTS:

The current COVID-19 pandemic has substantially accelerated the demands for efficient vaccines. A wide spectrum of approaches includes live attenuated and inactivated viruses, protein subunits and peptides, viral vector-based delivery, DNA plasmids, and synthetic mRNA. Preclinical studies have demonstrated robust immune responses, reduced viral loads and protection against challenges with SARS-CoV-2 in rodents and primates. Vaccine candidates based on all delivery systems mentioned above have been subjected to clinical trials in healthy volunteers. Phase I clinical trials have demonstrated in preliminary findings good safety and tolerability. Evaluation of immune responses in a small number of individuals has demonstrated similar or superior levels of neutralizing antibodies in comparison to immunogenicity detected in COVID-19 patients. Both adenovirus- and mRNA-based vaccines have entered phase II and study protocols for phase III trials with 30,000 participants have been finalized. [13, 14, 15]

Although advances in genetic sequencing and other technological developments have speed up the establishment of various vaccine platforms, several uncertainties still remain. Questions have arisen regarding the mutation rates of SARS-CoV-2, which could lead to immune evasion. Understanding mutations in the coding and non-coding regions, genetic diversity, pathogenicity, and host-pathogen interactions is essential. Mutations in the S protein seem to induce conformational changes, which may alter antigenicity and, thus, may affect the vaccine design. Moreover, previous studies suggested that various mutations in the target proteins of the coronaviruses can be associated with drug resistance and changes in the protein structures of target proteins that may lead to vaccine inefficacy.

It is crucial to highlight that the uncertainty over long-lasting protection against COVID-19 still remains. Patients with reinfection have been reported, and it is necessary to determine how long a protective immune response can be maintained in an individual. Considering the immunogenicity of the

NAME OF THE VACCINE	COVAXIN	COVISHIELD	MODERNA	SPUTNIK
MANUFACTURER / DEVELOPER:	Bharat Biotech	AstraZeneca, Serum Institute of India	Moderna, NIAID	Gamaleya Research Institute
RESEARCH NAME	BBV152	AZD1222 (ChAdOx1)	mRNA-1273	Gam-COVID-Vac
VACCINE TYPE	Inactivated	Non-Replicating Viral Vector	RNA	Non-Replicating Viral Vector
ADMINISTRATION METHOD	Intramuscular injection	Intramuscular injection	Intramuscular injection	Intramuscular injection

Side effects of the coronavirus vaccines

It's quite normal to experience side effects after the administration of vaccine. Vaccine get activate our body's immune system to defend itself from the pathogen. Therefore some of the people get side effects.

International Journal of Pharmaceutical Research and Applications
Volume 6, Issue 5 Sep-Oct 2021, pp: 17-23 www.ijprajournal.com ISSN: 2249-7781

Most of these are mild and short term. They may include:

- having a painful, heavy feeling and tenderness in the arm where you had your injection
- headache or muscle ache
- joint pain
- chills
- nausea or vomiting
- feeling tired
- fever (temperature above 37.8°C).

Many of them have flu-like symptoms with episodes of shivering and shaking for a day or two. These common side effects are much less serious than developing coronavirus or complications associated with coronavirus and they usually ride of within a few days. Millions of people worldwide are hoping to receive a vaccination against the coronavirus in the near future. Vaccines have been found safe and approved by health authorities in many countries. India has administered 17 crore total doses of anti-covid across the country reported from Ministry of Health And Family Welfare. At the same time, many people are ambivalent because, while they want to protect themselves against infection, they also fear possible side effects from vaccination. They have doubts as to whether the vaccines are actually safe, given the rapid pace of development, and whether possible side effects have been adequately studied.

V. FUTURE PERSPECTIVES:

The great advantage of the current advancements in nucleic acid-based technologies for vaccine development is the short time required from the design to clinical trials. Therefore, it may be soon possible to test together, in the same vaccine, different variants of antigens that cover circulating mutations. This would represent a major step forward in vaccine development against rapidly emerging threats such as the current SARS-CoV-2 pandemic. Therefore, developing vaccines against COVID-19 require further studies on gene mutations and how to avoid vaccine failure because of them. [16]

With huge information available on vaccine development for COVID-19, there are still some aspects to be focused on while designing a potential recombinant vaccine. The efficiency and safety are the two major characteristics of a vaccine that demand a number of preclinical and clinical trials. Therefore, we need to investigate more animal models and volunteers with varied health conditions and age. However, the period of trials can be deliberately reduced with the help of modern biotechnology platforms that may result in fast and effective vaccine development in this type of utmost emergency. [15, 16]

VI. CONCLUSION:

There is crisis need for a safe and effective COVID-19 vaccine. Vaccines innovation takes year to discover it come across experimental trials in animal models, followed by human clinical trials in subjects of various age groups before they can be approved for mass production then it reach to the public. The recurrence of coronaviruses in the last decades suggests possible future outbreaks and potential to become pandemic and therefore the process of vaccine development should focus on wide host-range against several of the circulating CoVs to get flexible products within a short period of time. This necessitates the alliance of recombinant platforms to bring more accuracy and predictable efficacy to the current vaccine technology. The ultimate goal of current research should be the easy availability with minimal side effects and access of vaccines to the lower section of society.

REFERENCE:

[1]. https://www.hindustantimes.com/india-news/what-is-the-difference-between-covid-s-delta-variant-and-delta-plus-variant-101624435169704.html
[2]. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7711739/#idm140346187948208title
[3]. https://clinicaltrials.gov/ct2/show/study/NCT04368728?term=vaccine&cond=covid-19&draw=3
[4]. http://www.isrctn.com/ISRCTN89951424
[5]. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7749647/
[6]. https://www.frontiersin.org/articles/10.3389/fgeed.2020.579297/full
[7]. https://dnascience.plos.org/2020/09/10/how-the-various-covid-vaccines-work/
[8]. WHO-World-health-organization coronavirus disease (COVID-19)
[9]. https://www.who.int/emergencies/diseases/novel-coronavirus-2019.
[10]. https://www.pharmaceutical-business-review.com/news/sinovac-biotech-covid-19-vaccine-trial/
[11]. https://www.who.int/publications/m/item/draft-landscape-of-covid-19-candidate-vaccines

[12]. https://ir.oncosec.com/press-releases/detail/2042/oncosec-collaborates-with-providence-cancer-institute-to
[13]. https://www.nih.gov/news-events/news-releases/nih-clinical-trial-investigational-vaccine-covid-19-begins
[14]. https://investors.modernatx.com/news-releases/news-release-details/moderna-advances-late-stage-development-its-vaccine-mrna-1273
[15]. http://www.ncbi.nlm.nih.gov/pubmed/20504275
[16]. https://www.nature.com/subjects/recombinant-vaccine