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Analyzing Toxicology Reports of the COVID-19 Vaccine

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ABSTRACT

In the development of vaccines for treatment against SARS-CoV-2, several categories have emerged and their benefits and concerns have become apparent. An ideal target, the Spike (S) protein, has emerged and become the focus of recombinant vaccines. The S protein is crucial for the infection process of SARS-CoV-2. Recombinant vaccines make use of this and focus on creating an immune response targeting the S protein. There are various subcategories of recombinant vaccines and all function in a fundamentally similar manner. Two other types of vaccines are live-attenuated and inactivated vaccines. They utilize weakened or inactivated coronavirus to create an immune response unlike recombinant vaccines. Nucleic acid-based coronavirus vaccines are a subcategory of recombinant vaccines and include DNA- and RNA-based vaccines. Recombinant vaccines make full use of the presented target and efficiently provide superior treatment against SARS-CoV-2.

Introduction

There have been over 25 million confirmed cases and close to one million deaths worldwide as a result of the COVID-19 pandemic. Vaccines have been in development, using several kinds of technology. Both mRNA and recombinant vaccines have been in the works, using research from SARS-CoV as a base. SARS-CoV-2 shares large amounts of genomic similarity with SARS-CoV, which first appeared in 2002–2003. For instance, the spike glycoprotein sequence has a similarity of 87.2%. “SARS-CoV-2 and SARS-CoV are both positive-sense single-stranded RNA viruses with a genome size of ~30 kilobases that encode several structural and non-structural proteins”(Yadav et al., 2021). A typical coronavirus has a viral envelope containing three proteins, the envelope protein (E), membrane protein (M), and the spike protein (S). These three structural proteins are accompanied by one other, nucleocapsid protein (N). The S protein is crucial for the infection of the virus and without it, viruses would not be capable of reacting to potential host cells. Due to the fact that the S protein is essential for the infection of coronaviruses, it has become the archetypal objective for vaccines. The S protein of SARS-CoV-2 also carries the large threat of being an exceptional neutralizer of the antibodies (NAbs) that are natural products of the body’s humoral immune system. A vaccine typically takes about ten to fifteen years to develop, however, due to the swift identification and publication of the SARS-CoV-2 gene sequence, the first vaccine candidate was fit for clinical testing in only a few months. Nonetheless, there are various methods of building immunity to SARS-CoV-2, and as such, research led to several types of vaccines to end this pandemic. While benefits and concerns emerge, some vaccines become more recommended due to their superiority. Recombinant vaccines were quickly developed and stand above the rest in terms of efficiency and safety. As of now, there are more than 60 different vaccines being developed at various clinical trial phases for treatment against SARS-CoV-2.

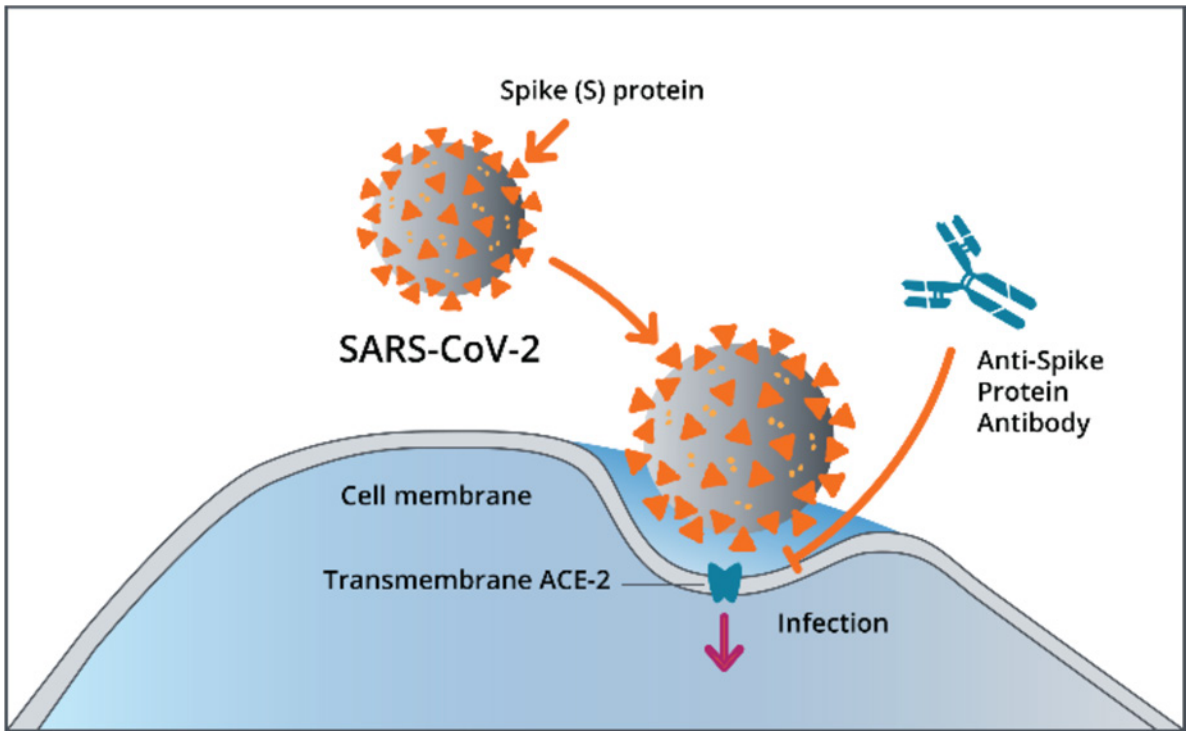

Figure 1. The interaction of the S protein for infection. (Berkeley Lights, 2020).

The Spike (S) protein

As aforementioned, the S protein is critical in the infection of coronaviruses, making it the ideal target for the treatment. The S protein can be divided into two major functional subunits, the head, labeled N-terminal S1 subunit, and the stalk, labeled C-terminal S2 subunit. The C-terminal S2 region is the section that is implanted directly into the viral envelope. Once a potential host cell is recognized, the S1 subunit will be responsible for binding to the host cell's receptors, while the S2 subunit completes the operation of fusing the virus's envelope to the membrane of the host cell. The S protein is also a note-worthy stimulator of neutralizing NAbs. NAbs commence their antiviral activity, by binding to a viral particle's surface epitope for the purpose of preventing their penetration of a host cell. 80R, CR3014, and CR3022 are all NAbs that have been classified to specifically attack the S1 domain of SARS-CoV. When further studies established the sensitivity of the S protein of SARS-CoV-2 to these particular NAbs, it piqued the interest of many researchers. The interest developed into a desire for standardized agents that have the ability to block the binding and attachment of the S protein of SARS-CoV-2 to host cells. However, conclusions regarding "the neutralization capability of CR3022 against the SARS-CoV-2 region binding domain (RBD) within the S1 subunit is uncertain"(Benedette Cuffari, 2021). Another previous study done on both Ebola and SARS viruses yielded the result of the information that a concoction of NAbs produces stronger neutralization capabilities than NAbs that are given alone. Research suggests that SARS-CoV-2 poses as great of a threat as it does due its angiotensin-converting enzyme 2 (ACE-2). Further studies reveal that the relation of SARS-CoV-2's S protein with ACE-2 is far more substantial than the S protein of its predecessor, SARS-CoV. This is what could have produced the result of the accelerated transmission and more infectious quality. The S1 subunit, located in the receptor-binding domain (RBD), makes first contact with the angiotensin-converting enzyme 2 receptor for connection, consequently penetrating the host cell by binding the membranes of the viral and host cells with the assistance of the S2 subunit. To such a degree, the S protein plays an indispensable role in, "the internalization of the virus, receptor binding, membrane fusion, tissue tropism, and host range and has thus emerged as an important target for vaccine development"(Yadav et al., 2021). Research

conducted for the development of a vaccine against SARS-CoV and the Middle East Respiratory Syndrome Coronavirus (MERS-CoV) further proves the magnitude of the S protein as the objective for potential treatments against SARS-CoV-2.

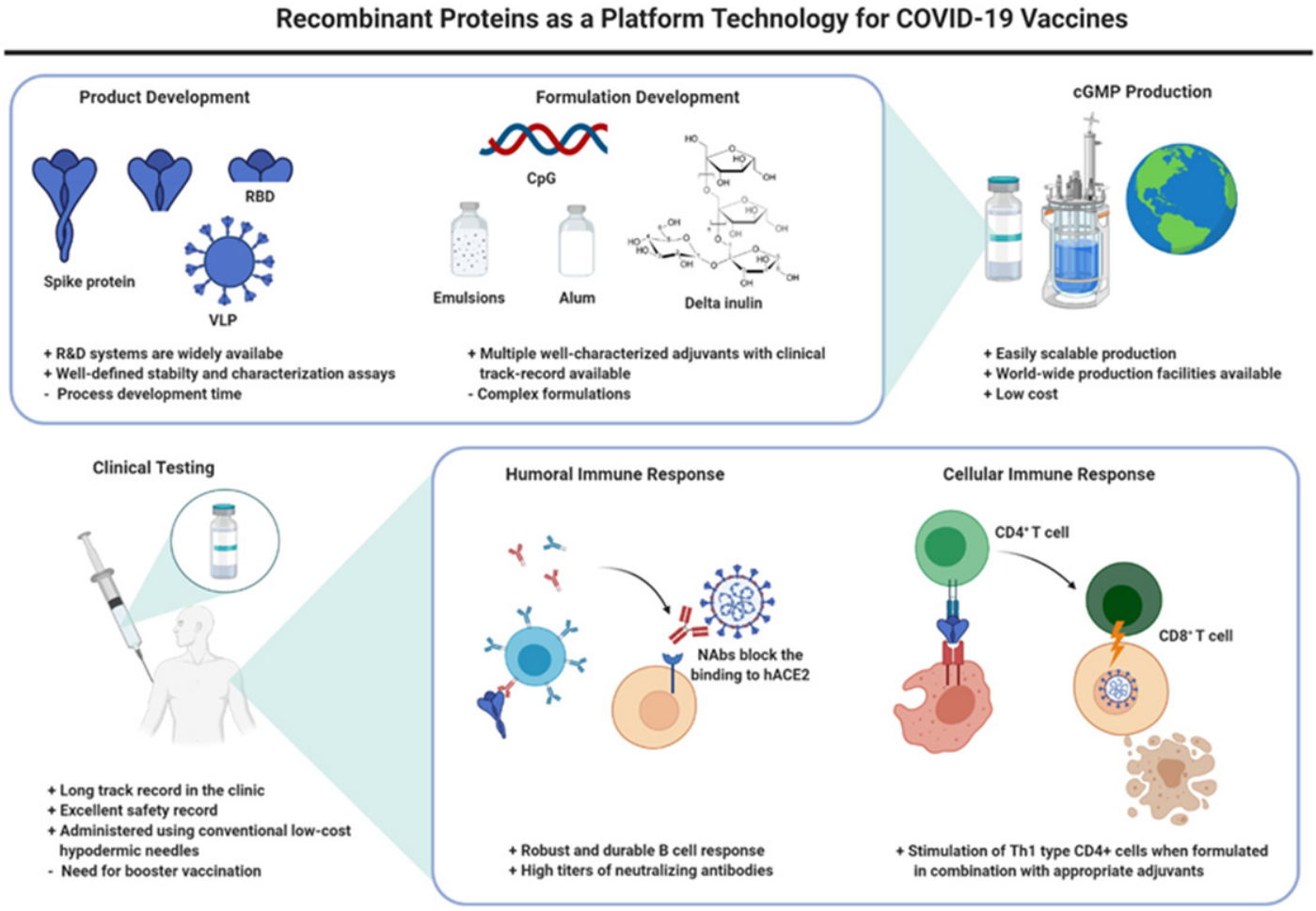

Figure 2. Recombinant proteins are a platform for the development of COVID-19 vaccine technology. (Pollet, J., Chen, W., & Strych, U., 2020).

Recombinant SARS-CoV-2 vaccines

Recombinant vaccines fall under one category of the COVID-19 vaccine. Recombinant vaccines only use individual parts of the germ, such as its protein, sugar, or capsid. Because these vaccines use only specific pieces of the germ, they give a very strong immune response that's targeted to key parts of the germ"("Vaccine Types", 2021). The vaccine functions by instructing host cells to assemble the protein of the S-antigen distinctive to SARS-CoV-2. By creating the protein, memory immune cells retain the necessary information and create an immune response to the virus. As aforementioned, the S protein is critical in the infection of coronaviruses. In the proposed recombinant vaccine, the antibodies produced to oppose the S protein are predicted to inhibit its fusion with angiotensin-converting enzyme 2 and neutralize the virus. The types of recombinant vaccines are seemingly endless, the most prominent of them being protein-based coronavirus vaccines, vectored coronavirus vaccines, artificially synthesized protein-microarray coronavirus vaccines, virus-like particle-based coronavirus vaccines, and nucleic acid-based coronavirus vaccines.

Like most recombinant vaccines, protein-based vaccines utilize the convenience of the S protein as an ideal target. One such protein-based vaccine is the result of a joint effort by Tazehkand and Hajipour to create a fusion vaccine. It involves the fusion of multi epitopes (B and MHC I epitopes) copied from the S protein and an RNA-dependent RNA polymerase with a nucleocapsid protein and an envelope. The vaccine's physicochemical and immunological aspects and structural stability were tested and verified during an initial screening, however, the authors expect that additional experiments with laboratory animals will be necessary. The effectiveness of the vectored vaccine strategy relies on the viral vector's ability to infect cells. The vectored vaccine's primary advantage is the efficiency and gene-specific delivery, in addition to 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"(Yadav et al., 2021). The results of the vaccine showed great potential and encouraged further evaluation. In regards to an artificially synthesized protein-microarray vaccine, recent research reports that a microneedle array-based recombinant SARS-CoV-2 S1 subunit vaccine was created. This vaccine brought the results of being capable of inducing a potent antigen-specific antibody response in the span of two weeks after the vaccination after being examined for its immunogenicity *in vivo*. Another recombinant vaccine type are virus-like particle-based coronavirus vaccines. "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)"(Yadav et al., 2021). The cooperative work conducted for the reason of acquiring the foundational knowledge of SARS-CoV-2 has led to an accelerated pace of technological advances that will prove to be useful in developing an effective vaccine in defense of the coronavirus.

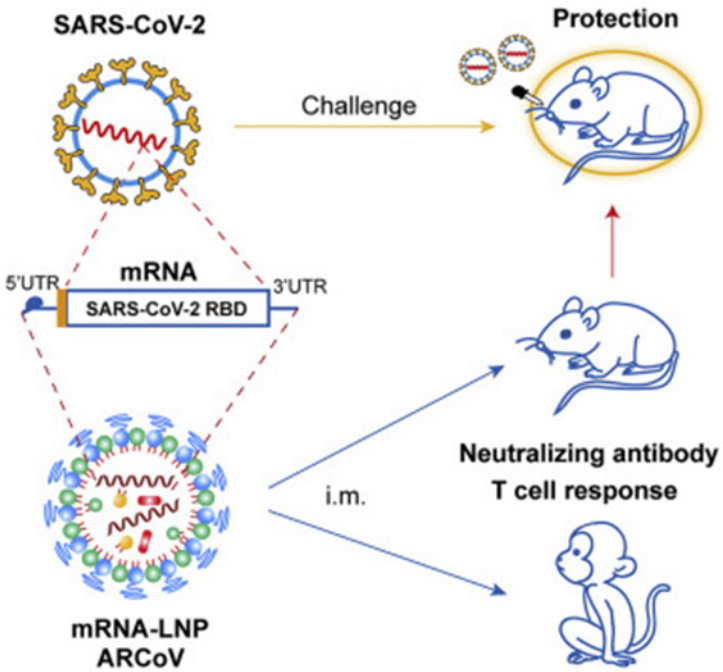

Figure 3. The animal models used for mRNA vaccines. (Zhang, 2020).

Recombinant nucleic acid-based SARS-CoV-2 vaccines

Under the nucleic acid-based coronavirus vaccines category, fall DNA- and RNA-based vaccines. DNA- and RNA-based vaccines have many advantages, the greatest being their reduced side effects and their potential for rapid development. DNA vaccines have shown great potential efficacy in animals, however, clinical data regarding humans is minimal. In a study using mice, it was discovered that T cells, a neutralizing antibody response, and protective immunity were achieved by a DNA-based vaccine that encoded the S protein of SARS-CoV. In another study, this one using 35 rhesus macaques, a group of prototype DNA vaccines expressing numerous S proteins of SARS-CoV-2 were developed and tested, resulting in the vaccinated macaques demonstrating distinct humoral and cellular immune responses. When tested with SARS-CoV-2, they showed an exceptional abatement of viral replication in both the upper and lower respiratory tract. The clinical data presented validates the remarkable role of DNA vaccines against SARS-CoV-2. mRNA vaccines utilize newer technology and are considered to be even more advantageous than DNA-based vaccines. This is due to the fact that mRNA vaccines do not necessitate access into the nucleus of the host cell nucleus in order to be transcribed. mRNA vaccines differ from the majority of other vaccines in their contents. Most vaccines use an inactive or weak germ to trigger an immune response, however, mRNA vaccines don't contain these germs. mRNA vaccines instruct cells to make a protein which then goes on to trigger an immune response. The immune response is what makes the antibodies, as opposed to most recombinant vaccines which directly teach host cells to produce antigens. As entry into the nucleus of the host cell is unnecessary for mRNA vaccines, "a lower dose can be used, without the need for any special delivery mechanisms. Moreover, mRNA-based vaccines avoid the risk of integration with the host cell genome and are able to produce pure viral protein"(Yadav et al., 2021). "Moderna Inc., a biotechnology firm, recently declared that mRNA-1273, a COVID-19 mRNA vaccine, has entered in phase 3 clinical trials with ~30,000 subjects. This vaccine program was funded by the Coalition for Epidemic Preparedness Innovations (CEPI) in association with the NIAID. mRNA-1273 encodes for a stable form of the SARS-CoV-2 S protein. One of the RNA-based vaccines developed by Pfizer/Biotech BNT162b1 and BNT162b2 has entered in large phase 3 clinical trial with 29481 participants. BNT162b1 is nucleoside-modified mRNA encoding trimerized SARS-CoV-2 spike glycoprotein receptor-binding domain (RBD) has been prepared as a lipid nanoparticle-formulated vaccine"(Yadav et al., 2021). RNA- and DNA-based are classified as nucleic acid-based coronavirus vaccines and, as such, are types of recombinant vaccines.

Live-attenuated and Inactivated SARS-CoV-2 vaccines

Recombinant vaccines are not alone in the development for treatment against SARS-CoV-2, live-attenuated coronavirus vaccines and inactivated coronavirus vaccines are also present. The live-attenuated vaccines require the use of live coronaviruses, albeit less infectious. Laboratory conditions see to the decreasing of the virulence of the live viruses. The vaccine functions through the replication of the live coronavirus within the host, however, due to the reduced infectiousness, the only reaction that can be induced is minimal pathogenesis. Live-attenuated coronavirus vaccines carry the risks of CoV spreading through the bodily waste of the individuals vaccinated with the live-attenuated vaccine and the advisability for the geriatric population, who have a reduced immune defense against severe disease. Inactivated coronavirus vaccines are developed through the use of an inactivated virus. The target virus can be inactivated by either irradiation or chemically with the result of the virus's nucleic acids being eradicated but leaving the antigens unimpaired. Inactivated CoV vaccines were tested during the advancement of SARS-CoV using animal models. The first evaluation used rhesus monkeys and yielded favorable results. "An inactivated vaccine against SARS-CoV… was found to induce humoral and mucosal immunity, highlighting its potential for use in clinical trials"(Yadav et al., 2021). However, there are safety concerns about incomplete inactivation, which can result in displeasing immune or inflammatory responses.

Conclusion

Worldwide cooperation has pushed along the development of COVID-19 vaccines at a rapid pace. Different types of vaccines have been created and improved, each focusing on various approaches for treatment against SARS-CoV-2. While most recombinant vaccines focus on the S protein as a target, the multiple techniques employed create a number of categories, such as, protein-based coronavirus vaccines, vectored coronavirus vaccines, artificially synthesized protein-microarray coronavirus vaccines, virus-like particle-based coronavirus vaccines, nucleic acid-based coronavirus vaccines, as well as RNA- and DNA-based vaccines. The S protein is an ideal target due to the fact that it plays the major role in infection via its two subunits. However, live-attenuated vaccines and inactivated vaccines function by different means. They employ the use of live viruses and inactivated viruses, respectively. However, further research and studies must be done in order to better discover the full scope of the safety risks of all vaccines. Even so, the efficacy and development of the vast subcategories of recombinant vaccines is superior to that of live-attenuated and inactivated vaccines. All vaccines deal with opportunities and obstacles, yet recombinant vaccines utilize an appropriate target and provide the most effective treatment. To combat the rising levels of SARS-CoV-2 infections, even more effort must be made to solve safety concerns, increase suitability for the population, and reinforce efficacy.

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