

The Focused Analysis of COVID-19 RNA-based vaccines

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Abstract. The COVID-19 virus has rapidly spread over the whole world and has now been declared a pandemic since the first cases of SARS-CoV-2 infection, also known as COVID-19, were found in Wuhan, China. It is undeniable that the only effective method to reduce COVID-19-related morbidity and mortality is to prevent infection through widespread vaccination. Various COVID-19 vaccines have been developed to date, many of which have been approved for use in the prevention of COVID-19. The era of RNA vaccines has come since December 2020, when two RNA-based vaccines -- from German pharmaceutical company BioNTech and US pharmaceutical company Moderna respectively--received emergency use authorizations from regulators in several countries to fight COVID-19. RNA-based vaccines were the first to be created and approved for use in humans, and they continue to play a crucial role in the global effort to fight the illness. This essay will introduce the efficacy, adverse reactions, production, and encountered limitations of existing RNA-based COVID-19 vaccines, and also predict the possible development direction of RNA-based vaccines in the future.

Keywords: RNA-based Vaccines, mRNA Vaccines, COVID-19.

1. Introduction

The 2019 coronavirus disease (COVID-19) pandemic was triggered by infections with severe acute respiratory syndrome coronavirus 2. It has spread to 220 nations, territories, or places as of December 3rd, 2020. According to the World Health Organization (WHO), there have been more than 1.4 million fatalities and more than 63 million cases with laboratory confirmation, causing significant social and economic upheaval. To help bring an end to this pandemic, it is urgently necessary to produce a secure and efficient vaccination. Additionally, according to the World meters website, which compiles data on Covid-19, the total number of cases identified in Wuhan, Hubei Province, China, and worldwide exceeds 379 million [1].

As a member of the Beta coronavirus family, SARS-CoV-2 is a single-chain positive-sense RNA virus with two main parts: protein capsid and genomic RNA bundled into a nucleocapsid. A phospholipid bilayer comprises the hemagglutinin-esterase, and the spike glycoprotein trimmer (S) surrounds the nucleocapsid (HE) [2]. The RNA of SARS-CoV-2 is composed of four structural proteins-- sixteen non-structural proteins (nsp1–16), envelope (E), nucleocapsid (N), membrane (M), and spike (S) (Figure 1) [3].

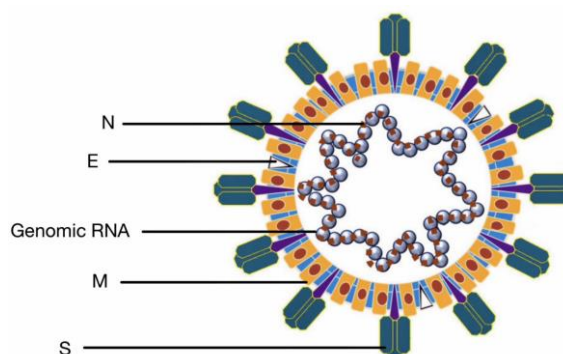


Figure 1. The structure of SARS-COV-2 virus [3].

Since the S glycoprotein is in charge of getting into host cells by interacting with the angiotensin-converting enzyme 2 (ACE2) cell receptor, it is a crucial target for therapeutics. The binding of the ACE2 and S glycoprotein on the cell surface allows the virus to enter human cells and be treated as a "friend" [4]. The virus's exterior membrane dissolves when it enters the human body, releasing its genetic material into the cytoplasm. Many different types of vaccines have been developed to combat this virus, among which the RNA-based vaccine has the best efficacy.

The development of RNA-based vaccines is the most promising and extensively researched method for creating new, safe, and effective vaccinations for prevention and treatment. It is a technique that enables the production of endogenous proteins by facilitating the transport of genetic material directly into the cell. When S glycoprotein is translated, messenger RNA, a tiny bit of nucleic acid that is separated from RNA in the nucleus, is produced. The mRNA of these vaccines is intended primarily to translate the new coronavirus's S glycoprotein [4].

This essay will focus on analyzing the advantages and disadvantages of existing RNA-based vaccines, and predicting the future development direction of COVID-19 RNA-based vaccines.

2. Advantages of RNA-based vaccines

The mRNA comprises a 3' UTR, a 5' cap, a 5' untranslated region (UTR), and a poly(A) tail, which is also known as leader RNA and contains the stop signal coding sequence. With the assistance of ribosomes and transfer RNA, this molecule serves as a template in the cell's cytoplasm, allowing duplicates of the encoded protein to be synthesized from each mRNA template [4]. The RNA molecule does not need to cross an extra barrier into the nucleus of the nuclear membrane for transcription; rather, it is ready to translate the encoded protein immediately after injection into the cytoplasm. Each molecule gains a quantitative advantage over individual proteins as a result of this amplification, improving the stability of protein synthesis [5]. RNA molecules can directly proceed with their replication and translation within the host at the time of delivery. Therefore, more immunogens can be expressed.

RNA vaccines were first proposed in the 1990s. The French Scientists working for Sanofi Pasteur were the first to employ RNA to encode an influenza antigen in mice, resulting in an immunological response. However, the lipid delivery technique they employed was harmful to humans. Thanks to recent developments in RNA interference and lipid nanoparticle technology, researchers discovered a method to efficiently deliver nucleic acid vaccines and transcribe antigenic components in vivo [6]. In 2012, Geall's team at Novartis USA (Cambridge, MA) successfully inoculated rats with a respiratory virus vaccine by packaging RNA nucleotide fragments into tiny fat droplets called lipid nanoparticles (LNP). Vaccines "teach" the body to recognize pathogens and destroy them. Common vaccinations involve injecting weakened pathogens, surface proteins, or sugar fragments called antigens into the body to train the immune system to recognize the invader. mRNA vaccines, however, carry the instructions for making these invading proteins. The mRNA vaccines aim to enter human cells and induce the production of these antigens, effectively turning the body into a "vaccine factory."

The currently approved mRNA vaccines by FDA are Pfizer–BioNTech and Moderna vaccines (Table 1). The Pfizer-BioNTech and Moderna COVID-19 mRNA vaccines have 90 to 95% effectiveness rates. Trials of new COVID-19 mRNA vaccines demonstrated significant but brief reactogenic effects. Most people won't encounter serious side effects like fatigue and fever. The term "severe side effects" refers to those that limit daily activity.

Table 1. Current approved RNA-based vaccines by FDA

Name	Developers	Country	Type	Target	Efficacy	Safety	Reference
mRNA-1273	Moderna	USA	mRNA	S-2p	89.3–96.8%	Systemic adverse events were more common after the second vaccination.	[2]
Pfizer	BioNTech	USA	mRNA	S-2p	90.3–97.6%	The frequency of serious adverse events was low.	[7]

The vaccines that are effective now are plasmid DNA vaccines and RNA vaccines, but the plasmid DNA vaccines have some key disadvantages; Firstly, human immunogenicity is low, and several vaccine doses are required for optimal protection. Secondly, the risk of carcinogenesis due to the potential integration of cellular chromosomes [3].

Although mRNA vaccines are not the only COVID-19 vaccines on the market, they were the first to be created and approved for use in humans, and they continue to play a crucial role in the global effort to fight the illness. RNA vaccines have recently achieved significant success in the fight against COVID-19 with high protection rates above 90% for the Pfizer and Moderna vaccine candidates in the United States.

Creating mRNA vaccines is typically a simple process in general. Once a target antigen from an emerging disease has been discovered, the gene is sequenced, chemically synthesized with specified modifications, and targeted to specific cellular sites in preparation for cloning into plasmid DNA [8]. The production of RNA vaccines is very rapid. When the genetic sequence of a pathogen is known, researchers can quickly identify potential antigen-coding fragments from it. They insert the sequence into a DNA template to generate the appropriate RNA, package the vaccine, and vaccinate it in the human body. For example, Moderna, a famous biomedical company, did all these processes in just four days after getting the coronavirus genome sequence. In partnership with the National Institutes of Health, this company launched its first human trial in just two months.

Contrarily, conventional vaccine production techniques demand expensive and time-consuming stages designed for each vaccine candidate. In the process of developing RNA vaccines, vaccine manufacturers can identify the effective antigen more quickly. RNA vaccines are helpful for basic research because of their fast-manufacturing nature.

Overall, RNA-based vaccines have high expression stability, and the manufacturing process is straightforward. However, there are still some disadvantages to these vaccines.

3. Limitation and improvement of mRNA vaccines

Despite the benefits of RNA-based vaccines, the development and preparation phase of identifying SARS-CoV-2 and vaccines presents obstacles to research and policy development. Reports of few incidences of mild to severe responses to prospective mRNA vaccines have aroused safety concerns. Therefore, it is essential to comprehend the potential dangers associated with RNA vaccine platforms, such as systemic and local inflammatory reactions, the durability of induced antigen expression, the need for auto-reactivity to enable antibody formation, and the toxicity of delivery components. Even if the S protein is a reasonable immunogenic antigen, the optimal antigen needs to be constructed in order to generate the desired immune system response. Glycosylation is a factor that could diminish the vaccine's efficacy [4]. However, SARS-CoV-2 has an excessive amount of glycosylation sites, making the task of developing a vaccine extremely challenging. Notably, the RNA-based technology targets only the S protein, as opposed to the whole viral particle, and could create antibodies specific to the S protein. Therefore, more research is required to identify the physiological, immunological, and structural characteristics of the structural proteins employed in the production of mRNA vaccines. Due to the fact that conventional methods of vaccine development incorporating novel viral targets,

unique vaccine platforms, and innovative development models are possible to be laborious and dangerous, increasing the pace while retaining safety remains a crucial challenge in the pandemic age.

Notably, more than 70% of active vaccine candidates are actually produced by the commercial sector, with the rest developed by research institutions, the public sector, and other nonprofit organizations. While a handful of famous multinational developers, such as GlaxoSmithKline, Janssen, Pfizer, Janssen, and Sanofi, have commissioned COVID-19 vaccines. Most of these companies are producing on a small scale, which means they lack experience in large-scale production. To meet the global demand, there must be effective international coordination between production and supply, organization and distribution, and between citizens and governments [9].

A further difficulty related to RNA-based vaccinations is that licensed mRNA vaccines may need to be stored at a low temperature throughout the manufacturing, transportation, storing, and distribution processes. For example, the distribution of BNT612b2 and mRNA-1273 within authorized ranges necessitates shallow temperatures ($-80 \sim -20$ °C), which restricts their availability to countries and regions with super-duper equipment not commonly accessible. The availability of steady energy and the high cost of refrigeration equipment is uncomfortable and impracticable in many developing nations. In addition to economic and logistical difficulties, these peculiar storage conditions may also have a substantially detrimental effect on the environment. Therefore, it is essential to develop thermostable RNA-based vaccinations that do not require costly refrigeration equipment, severe storage conditions, or a delivery network. All nations afflicted by COVID-19 must employ strategic planning to resist the virus.

Some technical advances, such as thermostable lipid nanoparticles in the RNA system and freeze-drying procedure may be able to resolve this issue [10]. ARCT-021 is a lyophilized vaccine that retains therapeutic efficacy without requiring an ultra-cold (less than -70°C) supply chain. This vaccine was developed in conjunction with Duke–NUS Medical School. Lyophilization may potentially add to the mRNA vaccines' long-term storage requirements. To maintain potency, sucrose or trehalose must be added to the LNP in this instance [6].

Short supply and uneven market distribution are other significant challenges for governments in recommending RNA-based vaccines. Current RNA-based vaccines for COVID-19 typically require two doses to be effective, and the current manufacturing is insufficient, especially given the necessity for booster doses [11].

Furthermore, because most RNA-based vaccine production facilities are located in high-income nations, middle- and low-income countries must import these vaccines and make adjustments outside of the cold chain to distribute them internally.

To provide the market demand quickly and fairly, new production techniques are needed.

4. Conclusion

RNA has now become a very effective way to make vaccines and is a promising way to advance and diversify. During the COVID-19 outbreak, many biotechnology companies have begun to work on transforming RNA molecules. Because of the rapid development of RNA technology, there are unlimited combinations of high safety, high efficacy, and rapid production processes for RNA vaccines to make better progress. However, considering the traditional RNA vaccine production strategies, improving the stability and efficiency of its protein production and related factors is still a significant research and development topic. It is widely established that the distribution of cytoplasmic mRNA is essential for evoking a powerful and long-lasting immune response to viral infections. To address the current need for vaccination, RNA vaccines are a good choice because they provide increased and prolonged in vivo antigen synthesis and possess a substantial innate immune activation effect. However, there is still room for improvement, and innovations focused on formulation technology, and next-generation delivery and preservation systems for vaccines could help improve their efficacy. A new RNA vaccine against COVID-19 would protect the human body and prevent further transmission if it has a significant effect. Alternatively, if a pandemic suddenly

disappears before a more effective vaccine has been produced, scientists and vaccine makers should continue to put effort into finding the most effective RNA vaccine and plan for continued research and effective usage in preparation for future outbreaks. If a pandemic suddenly disappears before a more effective vaccine has been developed, developers and scientists should continue to put efforts on finding the most effective RNA vaccine and plan for continued research and emergency usage in preparation for future outbreaks. So global efforts to produce COVID-19 vaccines should be led with improved manufacturing speed, scale deployment, distribution equity, and global supply equity. Effectively improve RNA vaccine to help people get through this difficult time.

References

- [1] Dai L, Gao G F. Viral targets for vaccines against COVID-19 [J]. *Nature Reviews Immunology*, 2020, 21: 73–82.
- [2] Islam N, Sheils N E, Jarvis M S, et al. Comparative effectiveness over time of the mRNA-1273 (Moderna) vaccine and the BNT162b2 (Pfizer-BioNTech) vaccine [J]. *Nature Communications*, 2022, 13(1): 2377.
- [3] Machado B A S, Hodel K V S, Fonseca L M, et al. The Importance of RNA-Based Vaccines in the Fight against COVID-19: An Overview [J]. *Vaccines*, 2021, 9(11): 1345.
- [4] Borah P, Deb P K, Al-Shar'i N A, et al. Perspectives on RNA Vaccine Candidates for COVID-19 [J]. *Frontiers in Molecular Biosciences*, 2021, 8: 63524.
- [5] Jain S, Venkataraman A, Wechsler M E, et al. Messenger RNA-based Vaccines: Past, Present, and Future Directions in the Context of the COVID-19 Pandemic [J]. *Advanced Drug Delivery Reviews*, 2021, 179: 114000.
- [6] Ball R, Bajaj P, Whitehead K. Achieving long-term stability of lipid nanoparticles: examining the effect of pH, temperature, and lyophilization [J]. *International Journal of Nanomedicine*, 2016, 12: 305–315.
- [7] Polack F P, Thomas S J, Kitchin N, et al. Safety and Efficacy of the BNT162b2 mRNA Covid-19 Vaccine [J]. *New England Journal of Medicine*, 2020, 383(27): 2603–2615.
- [8] Liu M A. A Comparison of Plasmid DNA and mRNA as Vaccine Technologies [J]. *Vaccines*, 2019, 7(2): 37.
- [9] Pepini T, Pulichino A-M, Carsillo T, et al. Induction of an IFN-Mediated Antiviral Response by a Self-Amplifying RNA Vaccine: Implications for Vaccine Design [J]. *Journal of Immunology*, 2017, 198(10): 4012–4024.
- [10] Fraiman J, Erviti J, Jones M, et al. Serious adverse events of special interest following mRNA COVID-19 vaccination in randomized trials in adults [J]. *Vaccine*, 2022, 40: 5798-5805.
- [11] Lundstrom K. Latest development on RNA-based drugs and vaccines [J]. *Future Science OA*, 2018, 4(5): FSO300.