Cost-Effective SARS-CoV-2 Vaccine targeting Novel Variants

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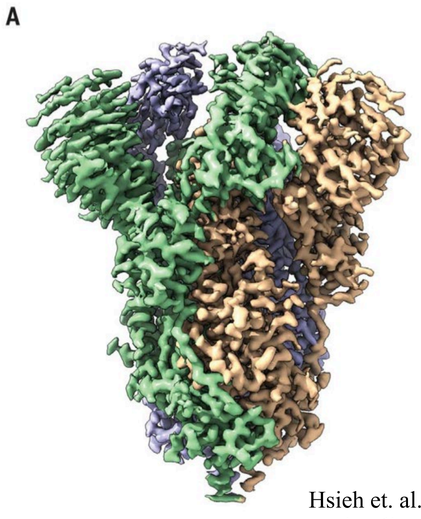
Abstract

Ever since they were first discovered, vaccines have allowed for the advancement of humanity through its use as a preventative measure against various infectious diseases. Vaccine development has recently advanced to unprecedented levels with the advent of SARS-COV-2 mRNA vaccines. Current SARS-COV-2 mRNA vaccines house genetically engineered mRNA sequences, coding for the viral fusion spike protein (S), inside of nano lipid particles to train the immune system to recognize and react to S protein during actual infection. However, the issue arises when these vaccines aren't available to developing countries that can't afford the current successful vaccines, leading to the persistence of the pandemic and evolution of the S protein affecting the transmissibility of SARS-COV-2. As places with populations nearing herd immunity become reinfected with an evolved variant of SARS-COV-2, such as the population of Manaus, Brazil, secondary outbreaks can be as disastrous as the initial outbreak of COVID-19. Virus Like Particle (VLP) vaccines are a possible remedy to this issue and work by using a viral vector with a target viral antigen that is associated with membrane binding on a host cell. This project proposes a theoretical cost-effective VLP vaccine housing a genetically engineered form of S protein, HexaPro, with the hopes that it induces protection against novel COVID-19 variants arising in developing nations.

Introduction

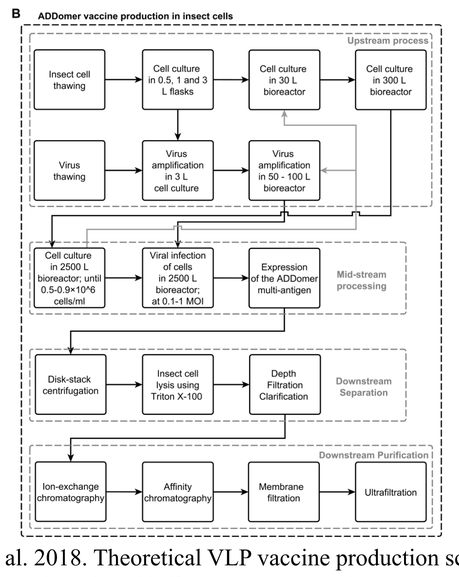
Vaccines have been able to prevent many life-threatening diseases. By preparing the body for an infection that normally would have led to disease, vaccines allow for immunity to develop in people who are at risk for serious complications when achieving natural immunity. Natural immunity is the process by which a person is infected by SARS-CoV-2, develops symptoms, and recovers after the adaptive immune system creates T-cells specific to SARS-CoV-2 and B-cells that produce anti-SARS-CoV-2 antibodies. Immunity can be achieved in individuals by training B-cells to create antibodies against the invading pathogen that either make SARS-CoV-2 easier to be engulfed by phagocytic cells that destroy the virus or neutralize the virus. This is done by antibodies binding to the S protein on the virus which blocks the virus' ability to bind to angiotensin-converting enzyme 2 (ACE2) receptors on human cells and initiate virus entry. Before SARS-CoV-2 enters a cell, it needs to overcome the free energy requirement that is necessary for membrane fusion. This is accomplished by the S protein binding to ACE2 which triggers a conformational change from its metastable pre-fusion state to a stable post-fusion state, leading to viral and cell membrane fusion and viral entry. This crucial characteristic of the S protein makes it a prime target for vaccines. Currently, mRNA vaccines are groundbreaking forms of vaccines that have been rapidly developed with a high success rate percentage in clinic trials. With success rates of around 95% from Phase 3 Clinical Trials for vaccines developed by the pharmaceutical companies Moderna and Pfizer, these vaccines have been distributed to much of the developed world to work towards herd immunity, or the point when around 67% of the population is immune to SARS-CoV-2. However, due to the current vaccine cost per dose created from storage requirements and production, the Moderna and Pfizer vaccines are estimated to cost around 34-35 dollars for a two dose-regimen. These costs make distribution to less developed countries difficult as these countries can't afford to buy enough doses or store them under the cold temperature requirements of Moderna's -20 ° C or Pfizer's -80 ° C that are required for the vaccines to be most stable until use. If not enough vaccines are distributed to countries who can't afford enough doses to achieve herd immunity, the population will have to go through natural immunity to be protected against re-infection by the variant that caused the initial outbreak. However, this approach to herd immunity is very unreliable and deadly, which is exemplified in Manaus, Brazil. 76% of the population of this city was estimated to have been infected with SARS-CoV-2 by October 2020, which is above the theoretical threshold for herd immunity to occur, but a second wave of infection as deadly as the first was still observed. The new variant discovered in the region, P1, was thus a result of the continued circulation of the virus in the population due to some lineages of the original variant having pre-existing immune evasion as well as mutations in the S protein that have been suggested to increase transmissibility. P1 thus not only became more infectious, but it was able to reinfect the already recovered population due in part to waning SARS-CoV-2 neutralizing antibodies in the population as well as the mutation of the S protein that the previous antisera of survivors was not able to neutralize. Doses are being donated to developing countries through organizations such as the World Health Organization (WHO), through the COVID-19 Vaccines Global Access initiative (COVAX), but to acquire herd immunity in these countries, more action is needed. With a Virus Like Particle (VLP) based vaccine, doses can become more cost-effective due to the abundant and robust nature of VLP production. VLP platforms are particles that mimic a virus by displaying target viral antigens on a protein capsid that is devoid of viral genetic material and can be made abundantly with insect cells using the Insect cell – Baculovirus expression system (IC-BES). Thus, with a theoretical vaccine using a VLP-based platform, an effective and room temperature stable option may be available for countries in need.



Hsieh et. al., 2020. Trimeric HexaPro visualized as cryo-EM structure.

Methodology

The VLP platform that would be used for this vaccine would be produced from a genetically engineered insect virus, baculovirus, whose DNA will be recombined using the MultiBac system. This system involves the use of a progenitor baculovirus genome that contains the genes encoding for ADDomer particles expressing HexaPro, which are placed into an artificial bacterial chromosome (BAC) that is then taken up by E. coli in culture. Benefits of this system include the flexibility of the encoded antigen that is eventually expressed on ADDomer VLPs, which results from baculovirus genome modification creating antigens that may induce immune responses against future mutations in the S protein. E. coli production can be scaled up to produce numerous BACs that are then transfected into insect cells to create baculovirions. These live baculovirions can then be cultured in insect cells on a mass scale to produce the target ADDomer particles. ADDomer protein scaffolds originate from adenovirus serotype 3 and are VLPs that self-assemble from protomers into pentameric protein complexes, or pentons. 12 pentons create highly stable ADDomer VLPs and express up to 360 antigens, proteins, and/or protein scaffolds, up to 200 amino acids in length, which are genetically encoded from the recombined baculovirus. ADDomer VLPs can also be stabilized with disulfide bonds leading to protein stability at temperatures between 37 ° C and 45 ° C for several months, an advantage for countries where cold storage is not easily achieved. These VLPs also have similar sizes to viruses which adds to the immunogenicity of the vaccine. HexaPro is a better antigen than the S-2P used in prevalent mRNA vaccines due to its greater stabilization and expression of S protein in the prefusion state. The stabilization of this prefusion state increases the amount of viral fusion glycoproteins, which are superior immunogens to wild type counterparts, and so, a stabilized prefusion state is a desirable form for the S protein in vaccines. S-2P is named as such due to the substitution of 2 prolines in the S2 subunit of the S protein, and with these two proline substitutions, the prefusion state of S protein was expressed and stabilized at an adequate level for effective vaccine production. HexaPro differs from S-2P by having four additional proline substitutions that lead to the greater stabilization and thus overall greater expression of S protein in the prefusion state. The stability allows for the prefusion state to stay in this crucial form throughout the process of protein purification in the mass production process. When more of this effective antigen are expressed on the ADDomer VLP, vaccine production increases as a result of doses requiring less protein to induce protection against SARS-CoV-2. HexaPro also has a similar ACE2 binding affinity and antigenicity to S-2P, demonstrating that similar protection from current SARS-CoV-2 vaccines using S-2P may be achieved in this theoretical vaccine. Intramuscular injection would be the primary delivery method of the vaccine. The shortcoming of this vaccine is that the facilities in developing countries may not accommodate for mass insect cell production and there may still be a need to rely on distribution from countries that harbor the necessary biotechnological infrastructure. Purifying the multiple components of the vaccine production and keeping them free from contamination also complicates mass production, which may increase the production cost. However, with the production of vaccines made in a similar fashion to this proposed vaccine, such as the FluBlok Influenza vaccine, doses are estimated to be produced in the amount of 1 billion doses in a 6-9 month period. These are based on assumptions made from FluBlok manufacture in 3 large bio-facilities, with 15 micrograms of vaccine per dose. This is in comparison to Pfizer and Moderna having the ability to produce 500 million and 400 million doses in 2021, respectively, but as two doses are required these numbers are halved to 250 million and 200 million full immunizations, respectively. It is important to note that these companies are ramping up production that may see production near 1 billion doses, or 500 million full immunizations, for 2021.



Kis et. al. 2018. Theoretical VLP vaccine production scheme based on FluBlok production scheme.

Conclusions

Vaccines are helpful tools for preventing infectious diseases, and as the COVID-19 pandemic continues, they have been indispensable factors in preventing further loss of life. However, this life saving technology's effectiveness is limited by the cost of current mRNA-based vaccine doses. In order to assist less developed countries with variants that are evolving to reinfect previously infected individuals, an ADDomer VLP based vaccine vector expressing HexaPro may be an option. This would remedy the high production costs seen from prevalent mRNA vaccines and allow for room temperature stable vaccines to be mass produced at scales necessary to achieve herd immunity in populations where more transmissible SARS-CoV-2 variants arise. The ease of antigen adaptability in the production of the VLPs created for this vaccine will keep up with the pace of which new variant lineages are evolved to surpass previously acquired immunity.

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