Concept of SARS-CoV-2 Vaccine Design to Fight COVID-19 Pandemic: A Review Insight

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Abstract

Cluster of pneumonia infection emerged in Wuhan, China due to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Additionally, more than 190 countries have confirmed 82 million cases of SARS-CoV-2 infection. Currently, there is a SARS-CoV-2 epidemic, and no effective prophylactic methods are available. A vaccine is considered as an effective method to restrict an epidemic. Several vaccine designing techniques have been established, which is enabling researchers from various institutes for developing vaccine towards SARS-CoV-2 infections. In this review, we condense the development of vaccine research against SARS-CoV-2.

Keywords: *COVID-19, SARS-CoV-2, Vaccine Design.*

Introduction

A cluster of pneumonia infections emerged in Wuhan Hospital, China which started from December 2019 up to now. The infection was reported to be caused due to a type of beta-coronavirus, which named by World Health Organization (WHO) as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2)[1,2]. As per reports till March 26th 2021, the pathogen had infected more than 120 million individuals and caused 2.7 million deaths worldwide[1].

SARS-CoV-2 had a material genetic of positive-stranded RNA and belongs to *Betacoronavirus* genera. Studies have revealed that SARS-CoV-2 genome consists of 29,700 nucleotides, and shares approximately 79.5% similarity to the genome of SARS-CoV[3,4]. Furthermore, SARS-CoV-2 is known to have an ORF1ab polyprotein at the 5′-end which had a role in coding for viral non-structural proteins. Genes at 3′-end had been known to encode the structural proteins such as spike (S), nucleocapsid (N), membrane (M), and envelope (E)[5,6]. Several studies have reported that SARS-CoV-2 binds to angiotensin converting enzyme 2 (ACE2) receptor on the host cell. The receptor played a key role in viral entry mechanism and related pathogenesis[7,8] (Figure 1).

A vaccine is considered as an effective method which had high economic significance to restrict an infectious disease outbreak[3,9]. The use of vaccines inducing neutralizing antibodies is the best option to increase the number of populations that are immune to SARS-CoV-2[10]. Thus, the development of SARS-CoV-2 vaccine became an essential need. As per few reports, about 40 pharmaceutical companies and academic institutions in the world have currently assigned the

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program for developing vaccines towards SARS-CoV-2. Therefore, we summarize the ongoing advance research in development of SARS-CoV-2 vaccine.

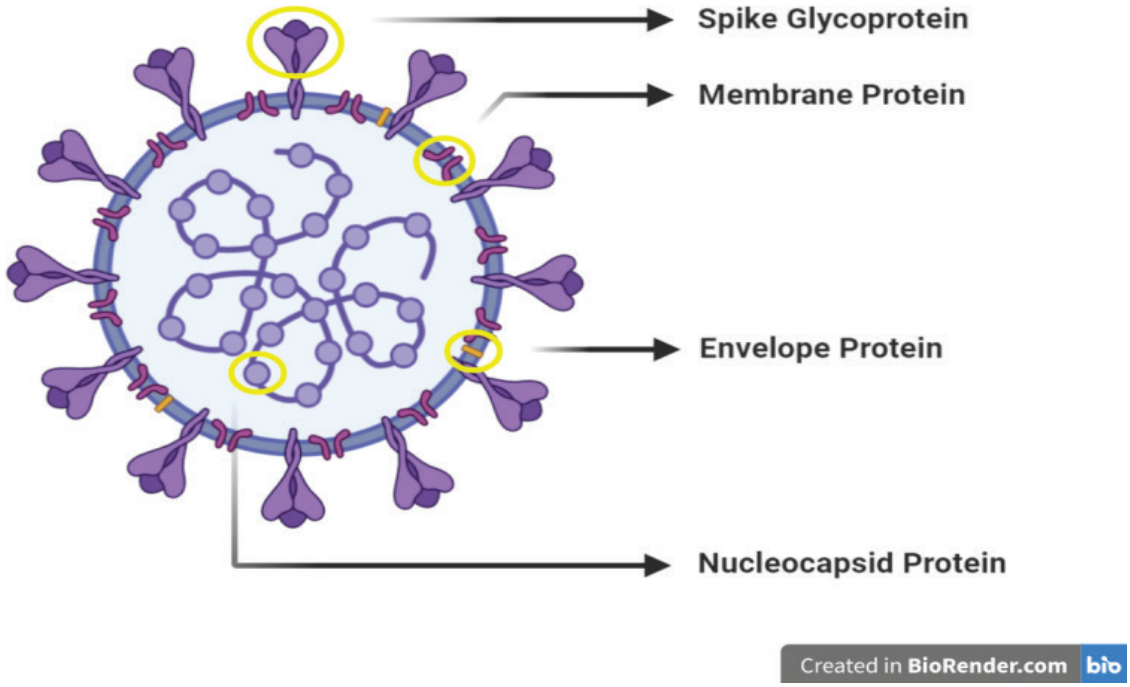

Figure 1. Structural part of SARS-CoV-2. The virion consists of spike glycoprotein (S), membrane protein (M), envelope protein (E), and nucleocapsid protein (N).

Antigenic Retrieval and Selection from SARS-CoV-2 Structural Protein

Various non-srtructural protein is common to be utilized for vaccine development. However, spike protein (S protein) is widely being investigated to be used for developing a vaccine against SARS-CoV-2. Studies have revealed that it is a viral surface protein, which is directly recognized by the host immune system[3,4]. Subsequently, it has been reported that S protein acts as a mediator for interaction between virus and host cell by binding to ACE2 receptor, and cause consequent pathogenicity[11]. The homologous structure of S protein is being used for vaccine development, which has proven to be effective in overcoming the previous outbreaks including SARS-CoV and Middle East respiratory syndrome coronavirus (MERS-CoV)[12].

The S protein monomer consists of 1273 amino acids in length and molecular weight about 140 kDa. Furthermore, it has ability to undergo the self-association mechanism and formed a homotrimer structure as a special feature of class I viral fusion protein. Structural studies showed that S protein is composed of two subunits, termed as S1 and S2 part. The S1 subunit can be organized into two domains, such as N-terminal domain (NTD) and C-terminal domain (CTD). In brief, the CTD is reported as receptor-binding domain (RBD). On the other hand, the S2 subunit had an essential part for membrane fusion which consist of internal membrane fusion peptide (FP), two heptad repeats (HR), membrane proximal external region (MPER), and transmembrane domain (TM)[13,14]. Recently, the S trimer structure of SARS-CoV-2 within the pre-fusion stage, and the RBD structure in convolution with ACE2 had been successfully determined[15].

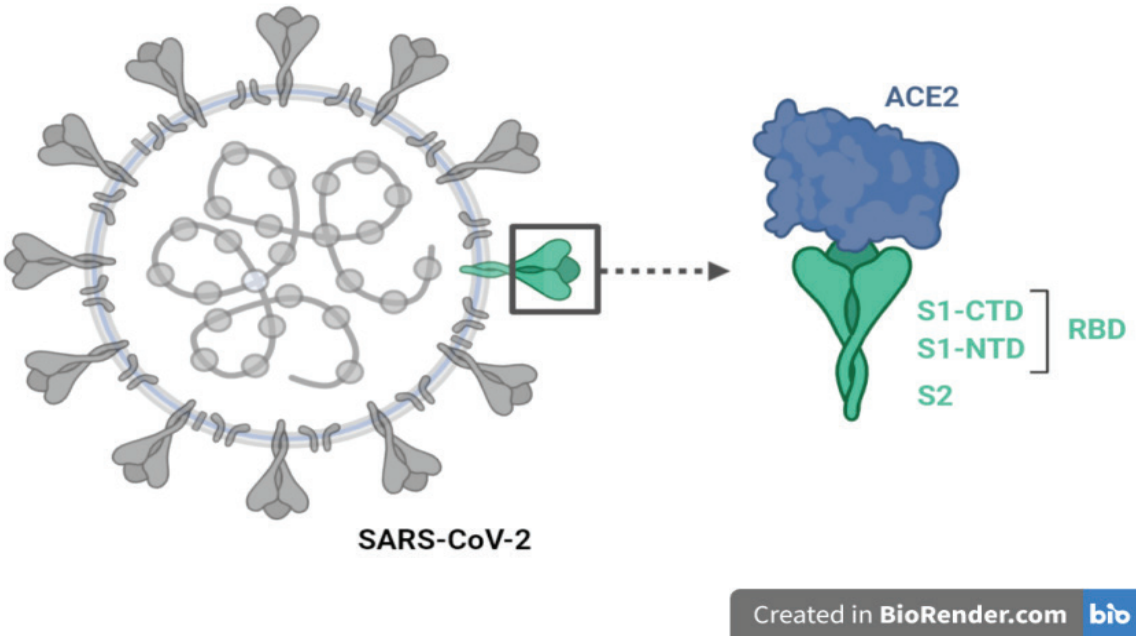

Figure 2. Brief structure of SARS-CoV-2 spike glycoprotein.

The S trimer protein structure had been reported to act as an epitope and had ability to induce a high immunogenicity. Coleman *et al*. showed that the S trimer protein could be produced within baculovirus insect cells, and it could be transformed into nanoparticles[16]. Furthermore, it was formulated with an adjuvant (alum) and injected into the mice. Moreover, the immunized mice were observed to obtain the data about titers level of specific antibodies after the nanoparticle administration[16,17]. Recently, the Clover Biopharmaceutical has announced that they have designed SARS-CoV-2 S-trimer based-vaccine which processed into the preclinical testing stage[9].

RBD has been known to interact directly with ACE2 receptors of the host cells. Thus, many studies have suggested RBD for immunization to investigate the antibody formation. It estimated for having ability to inhibit the viral recognition of ACE2 receptor, and prevent the subsequent pathogenesis[18,19]. Accordingly, many RBD-antigens based SARS-CoV-2 subunit vaccines are currently being developed. A study showed that the recombinant RBD, which was isolated from different types of virus serotypes, could trigger the multi-conformational by neutralizing antibodies towards SARS-CoV-2. This might be due to the presence of multiple epitopes that are recognized by specific immune systems[20].

As previously reported, S1 subunit has RBD as well as NTD, which suggests its role in binding S protein to the host receptor[21]. Subsequently, these domains can be widely utilized for vaccine development. As comparison, previous studies showed that MERS-SoV S1 subunit with MF59 adjuvant could protect the human dipeptidyl peptidase 4 transgenic (hDPP4-Tg) mice against infections[22]. Furthermore, FP in S2 subunit has been reported to be involved in membrane fusion which is essential step for viral pathogenicity[23,24]. At present, Tianjin University is constructing RDB-FB-based vaccines and producing high titers of neutralizing antibodies in mice[9].

In other hand, N protein is abundantly expressed by the coronavirus and it has molecular weight about 50 kDa. It has been reported for promoting several functions such as nucleocapsid formation, initiation of viral budding transduction signal, RNA replication, and mRNA transcription[25,26]. Furthermore, N protein

has been suggested to have high antigenicity as about 80% of patients with COVID-19 have been observed to produce antibodies specific to SARS-CoV N protein[27].

Moreover, they have been observed to significantly reduce the viral load of vaccinia virus[28]. In addition, previous studies reported that bronchitis virus-related N protein could induce the activation of cytotoxic T lymphocytes (CTLs) by attenuating infection and thereby, protecting lungs from viral pathogenicity[29,30].

Moreover, M protein is a glycoprotein located in the transmembrane of virus and it has molecular weight about 25 kDa. In brief, it has a crucial role for viral assembly. Furthermore, previous report exhibited that it abundantly expressed on the viral surface[31,32]. Full length M protein has been previously evaluated as a vaccine, which is reported could be neutralizing antibodies in patients with SARS. M proteins also consisted T cell epitope clusters and it was evaluated to be immunogenic[33,34]. In addition, it also identified for having high conserved domain[32,35].

E protein has been reported to have about 76-109 amino acids from studies in different types of coronaviruses. Moreover, it has been demonstrated to have limited immunogenicity compared to S, N, and M proteins. Thus, E protein is not predicted as an immunogen. However, E protein knockout models have been observed to significantly reduce secretion of inflammatory cytokine such as interleukin 1β (IL-1β), tumor necrosis factor (TNF), and IL-6[36].

Recent Developments in SARS-CoV-2 Vaccine Technology

Studies have shown that whole-cell killed or live-attenuated vaccine exhibit all antigenic components to the host and it could potentially trigger different immunological responses to fight the pathogen[37,38]. Traditional vaccines could be developed using advanced technology to design the foremost SARS-CoV-2 vaccine that may have prompt applications in healthcare. Wuhan Institute of Virology, Chinese Academy of Sciences, Zhejiang University, and other companies have successfully isolated SARS-CoV-2 and they have started developing vaccines. Recently, Codagenix Inc. collaborated with Serum Institute of India Ltd. developed live-attenuated SARS-CoV-2 vaccine[9].

Vaccine subunits that comprise of more than one antigen with high immunogenicity could trigger the host immune response. Good quality of vaccines should be safe and easy to produce; however, it commonly requires adjuvants to exhibit the strong immune response[39]. Several companies have established programs to develop SARS-CoV-2 subunit-based vaccine, and most of them have been reported utilizing protein S as an antigen. The utilization of "molecular clamp" technology by University of Queensland and "Trimer-Tag" technology by Clover Biopharmaceuticals Inc. are several examples of subunit vaccines. Recently, Novavx Inc. announced nanotechnology-based vaccine candidates that have been evaluated in animal models while further testing in humans is needed. Furthermore, Pasteur Institute has collaborated with Chongqing Zhifei Biology Co. Ltd. and started developing SARS-CoV-2 subunit-based vaccine[9,40].

Advanced technology in the synthesis, modification, and delivery of mRNA vaccines has been developed for past two decades. The mRNA vaccine is a notable alternative for conventional vaccine since it known for remarkable efficiency, fast production, low cost, and safe consumption[41,42]. In general, the mRNA vaccine development consists of several steps, including antigen selection, sequence optimization, nucleotide modification, delivery system, evaluation of immune response, and evaluating vaccine safety[43]. Recently, the mRNA vaccine against SARS-CoV-2 had been developed by Moderna and it is qualified for further analysis. Similarly, the mRNA vaccine based on protein S and RBD is being assessed into in vivo study under the collaboration of Fudan University, Shanghai Jiaotong University, and Bluebird Biopharmaceutical[9].

Heterologous antigen expression is the principal action for live vector vaccines. It commonly produces stronger immunogenicity and trigger higher cellular

immunity[44]. Houston-based Greffex Inc. has been successful in developing live vector-based SARS-CoV-2 vaccine using adenovirus vectors, and it is evaluated in animals. Moreover, Tonix Pharmaceuticals has recently announced the development of SARS-CoV-2 vaccine using Horsepox Virus (TNX-1800)[9].

Another kind of vaccine, such as synthetic peptide-based vaccines are also developed. It generated from small fragments or peptides which synthesized through chemically process. However, it has been reported that it exhibited a low immunogenicity effect due to the low molecular weight and complex structure. As a result, synthetic peptide vaccines require a few modifications, such as structural modification, presence of a delivery system, and adjuvants formulation[45]. Currently, researchers from Hong Kong University of Science and Technology are developing a peptide vaccine based on B and T cell epitopes from S and N proteins of SARS-CoV[46].

Conclusion

We hope that several vaccines that have been developed could be used as an effective treatment for COVID-19 outbreak. Different methodology of vaccine development had both advantages and disadvantages. Furthermore, current advancements in technology may assist researchers to contribute in development of SARS-CoV-2 vaccine.

Conflict of Interest: The author declare that they have no conflict of interest.

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References

1. Zhu N, Zhang D, Wang W, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med. 2020;382(8):727-733.
2. Hui DS, I Azhar E, Madani TA, et al. The continuing 2019-nCoV epidemic threat of novel coronaviruses to global health — The latest 2019 novel coronavirus outbreak in Wuhan, China. Int J Infect Dis. 2020;91:264-266.
3. Kharisma VD, Ansori ANM. Construction of Epitope-Based Peptide Vaccine Against SARS-CoV-2: Immunoinformatics Study. J Pure Appl Microbiol. 2020;14(suppl 1):999-1005.
4. Ansori ANM, Kharisma VD, Muttaqin SS, Antonius Y, Parikesit AA. Genetic Variant of SARS-CoV-2 Isolates in Indonesia: Spike Glycoprotein Gene. J Pure Appl Microbiol. 2020;14(suppl 1):971-978.
5. Khailany RA, Safdar M, Ozaslan M. Genomic characterization of a novel SARS-CoV-2. Gene Rep. 2020;19:100682.
6. Phan T. Novel coronavirus: From discovery to clinical diagnostics. Infect Genet Evol. 2020;79:104211.
7. Zhang H, Penninger JM, Li Y, Zhong N, Slutsky AS. Angiotensin-converting enzyme 2 (ACE2) as a SARS-CoV-2 receptor: molecular mechanisms and potential therapeutic target. Intensive Care Med. 2020;46(4):586-590.
8. Wina Nurtias LY, Turista DDR, Puspitasari E. Human immune response to SARS-CoV-2 infection. J Teknol Lab. 2020;9(1):29-40.
9. Zhang J, Zeng H, Gu J, Li H, Zheng L, Zou Q. Progress and Prospects on Vaccine Development against SARS-CoV-2. Vaccines. 2020;8(2):153.
10. Speiser DE, Bachmann MF. COVID-19: Mechanisms of Vaccination and Immunity. Vaccines. 2020;8(3):404.
11. Padron-Regalado E. Vaccines for SARS-CoV-2: Lessons from Other Coronavirus Strains. Infect Dis Ther. 2020;9(2):255-274.
12. Du L, He Y, Zhou Y, Liu S, Zheng B-J, Jiang S. The spike protein of SARS-CoV — a target for vaccine and therapeutic development. Nat Rev Microbiol. 2009;7(3):226-236.
13. Wang Q, Zhang Y, Wu L, et al. Structural and Functional Basis of SARS-CoV-2 Entry by Using Human ACE2. Cell. 2020;181(4):894-904.e9.
14. Li F. Structure, Function, and Evolution of Coronavirus Spike Proteins. Annu Rev Virol. 2016;3(1):237-261.

15. Wrapp D, Wang N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science. 2020;367(6483):1260-1263.
16. Coleman CM, Liu YV, Mu H, et al. Purified coronavirus spike protein nanoparticles induce coronavirus neutralizing antibodies in mice. Vaccine. 2014;32(26):3169-3174.
17. Muthumani K, Falzarano D, Reuschel EL, et al. A synthetic consensus anti–spike protein DNA vaccine induces protective immunity against Middle East respiratory syndrome coronavirus in nonhuman primates. Sci Transl Med. 2015;7(301):301ra132-301ra132.
18. Zhu X, Liu Q, Du L, Lu L, Jiang S. Receptor-binding domain as a target for developing SARS vaccines. J Thorac Dis. 2013;5:7.
19. Chen W-H, Strych U, Hotez PJ, Bottazzi ME. The SARS-CoV-2 Vaccine Pipeline: an Overview. Curr Trop Med Rep. 2020;7(2):61-64.
20. Lan J, Yao Y, Deng Y, et al. Recombinant Receptor Binding Domain Protein Induces Partial Protective Immunity in Rhesus Macaques Against Middle East Respiratory Syndrome Coronavirus Challenge. EBioMedicine. 2015;2(10):1438-1446.
21. Turista DDR, Islamy A, Kharisma VD, Ansori ANM. Distribution of COVID-19 and Phylogenetic Tree Construction of SARS-CoV-2 in Indonesia. J Pure Appl Microbiol. 2020;14(suppl 1):1035-1042.
22. Wang Y, Tai W, Yang J, et al. Receptor-binding domain of MERS-CoV with optimal immunogen dosage and immunization interval protects human transgenic mice from MERS-CoV infection. *Hum* Vaccines Immunother. 2017;13(7):1615-1624.
23. Tang T, Bidon M, Jaimes JA, Whittaker GR, Daniel S. Coronavirus membrane fusion mechanism offers a potential target for antiviral development. Antiviral Res. 2020;178:104792.
24. Lai AL, Millet JK, Daniel S, Freed JH, Whittaker GR. The SARS-CoV Fusion Peptide Forms an Extended Bipartite Fusion Platform that Perturbs Membrane Order in a Calcium-Dependent Manner. J Mol Biol. 2017;429(24):3875-3892.
25. Malik YA. Properties of Coronavirus and SARS-CoV-2. Malays J Pathol. 2020;42(1):3-11.
26. Li S, Lin L, Wang H, et al. The Epitope Study on the SARS-CoV Nucleocapsid Protein. Genomics Proteomics Bioinformatics. 2003;1(3):198-206.
27. Leung DTM, Tam FCH, Ma CH, et al. Antibody Response of Patients with Severe Acute Respiratory Syndrome (SARS) Targets the Viral Nucleocapsid. J Infect Dis. 2004;190(2):379-386.
28. Kim TW, Lee JH, Hung C-F, et al. Generation and Characterization of DNA Vaccines Targeting the Nucleocapsid Protein of Severe Acute Respiratory Syndrome Coronavirus. J Virol. 2004;78(9):4638-4645.
29. Collisson E. Cytotoxic T lymphocytes are critical in the control of infectious bronchitis virus in poultry. Dev Comp Immunol. 2000;24(2-3):187-200.
30. Seo SH, Pei J, Briles WE, Dzielawa J, Collisson EW. Adoptive Transfer of Infectious Bronchitis Virus Primed αβ T Cells Bearing CD8 Antigen Protects Chicks from Acute Infection. Virology. 2000;269(1):183-189.
31. Astuti I, Ysrafil. Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2): An overview of viral structure and host response. Diabetes Metab Syndr Clin Res Rev. 2020;14(4):407-412.
32. Neuman BW, Kiss G, Kunding AH, et al. A structural analysis of M protein in coronavirus assembly and morphology. J Struct Biol. 2011;174(1):11-22.
33. Liu J, Sun Y, Qi J, et al. The Membrane Protein of Severe Acute Respiratory Syndrome Coronavirus Acts as a Dominant Immunogen Revealed by a Clustering Region of Novel Functionally and Structurally Defined Cytotoxic T□Lymphocyte Epitopes. J Infect Dis. 2010;202(8):1171-1180.
34. Pang H, Liu Y, Han X, et al. Protective humoral responses to severe acute respiratory syndrome-associated coronavirus: implications for the design of an effective protein-based vaccine. J Gen Virol. 2004;85(10):3109-3113.
35. Arndt AL, Larson BJ, Hogue BG. A Conserved Domain in the Coronavirus Membrane Protein Tail Is Important for Virus Assembly. J Virol. 2010;84(21):11418-11428.
36. Nieto-Torres JL, DeDiego ML, Verdiá-Báguena C, et al. Severe Acute Respiratory Syndrome Coronavirus Envelope Protein Ion Channel Activity Promotes Virus Fitness and Pathogenesis. Denison MR, ed. PLoS Pathog. 2014;10(5):e1004077.
37. Sharma A, Krause A, Worgall S. Recent developments for Pseudomonas vaccines. Hum Vaccin. 2011;7(10):999-1011.
38. Ansori ANM, Kharisma VD, Antonius Y, Tacharina

MR, Rantam FA. Immunobioinformatics analysis and phylogenetic tree construction of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in Indonesia: spike glycoprotein gene. J Teknol Lab. 2020;9(1):13-20.

39. Takashima Y, Osaki M, Ishimaru Y, Yamaguchi H, Harada A. Artificial Molecular Clamp: A Novel Device for Synthetic Polymerases. Angew Chem Int Ed. 2011;50(33):7524-7528.
40. Liu H, Su D, Zhang J, et al. Improvement of Pharmacokinetic Profile of TRAIL via Trimer-Tag Enhances its Antitumor Activity in vivo. Sci Rep. 2017;7(1):8953.
41. Pardi N, Hogan MJ, Porter FW, Weissman D. mRNA vaccines — a new era in vaccinology. Nat Rev Drug Discov. 2018;17(4):261-279.
42. Zhang C, Maruggi G, Shan H, Li J. Advances in mRNA Vaccines for Infectious Diseases. Front Immunol. 2019;10:594.
43. Jahanafrooz Z, Baradaran B, Mosafer J, et al. Comparison of DNA and mRNA vaccines against cancer. Drug Discov Today. 2020;25(3):552-560.
44. Yurina V. Live Bacterial Vectors—A Promising DNA Vaccine Delivery System. Med Sci. 2018;6(2):27.
45. Azmi F, Ahmad Fuaad AAH, Skwarczynski M, Toth I. Recent progress in adjuvant discovery for peptide-based subunit vaccines. Hum Vaccines Immunother. 2014;10(3):778-796.
46. Ahmed SF, Quadeer AA, McKay MR. Preliminary Identification of Potential Vaccine Targets for the COVID-19 Coronavirus (SARS-CoV-2) Based on SARS-CoV Immunological Studies. Viruses. 2020;12(3):254.