Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Biomedicine & Pharmacotherapy 137 (2021) 111254

Contents lists available at ScienceDirect

Biomedicine & Pharmacotherapy

journal homepage: www.elsevier.com/locate/biopha

Review

Targets and strategies for vaccine development against SARS-CoV-2

Jonaid Ahmad Malik [a,b], Almas Hanif Mulla [c], Tahmeena Farooqi [d], Faheem Hyder Pottoo [e,*], Sirajudheen Anwar [f], Kannan R.R. Rengasamy [g,h,i,*]

[a] Department of Pharmacology and Toxicology, National Institute of Pharmaceutical Education and Research, Guwahati, Assam, India
[b] Department of Biomedical Engineering, Indian Institute of Technology (IIT), Ropar, Punjab, India
[c] Department of Pharmacology, Goa College of Pharmacy, Goa, India
[d] Department of Pharmacology and Toxicology National Institute of Pharmaceutical Education and Research, Hyderabad Telangana, India
[e] Department of Pharmacology, College of Clinical Pharmacy, Imam Abdulrahman Bin Faisal University, P.O.BOX 1982, Dammam, 31441, Saudi Arabia
[f] Department of Pharmacology and Toxicology, College of Pharmacy, University of Hail, Hail, 81442, Saudi Arabia
[g] Institute of Research and Development, Duy Tan University, Da Nang, 550000, Viet Nam
[h] Faculty of Environment and Chemical Engineering, Duy Tan University, Da Nang, 550000, Viet Nam
[i] Indigenous Knowledge Systems Centre, Faculty of Natural and Agricultural Sciences, North-West University, Private Bag X2046, Mmabatho, 2745, North West Province, South Africa

ARTICLE INFO

Keywords:
Vaccine
COVID-19
Clinical trials
SARS-CoV-2 vaccine
Pandemic
Antigen targets

ABSTRACT

The SARS-CoV-2, previously called a novel coronavirus, that broke out in the Wuhan city of China caused a significant number of morbidity and mortality in the world. It is spreading at peak levels since the first case reported and the need for vaccines is in immense demand globally. Numerous treatment and vaccination strategies that were previously employed for other pathogens including coronaviruses are now being been adopted to guide the formulation of new SARS-CoV-2 vaccines. Several vaccine targets can be utilized for the development of the SARS-CoV-2 vaccine. In this review, we highlighted the potential of various antigenic targets and other modes for formulating an effective vaccine against SARS-CoV-2. There are a varying number of challenges encountered during developing the most effective vaccines, and measures for tackling such challenges will assist in fast pace development of vaccines. This review will give a concise overview of various aspects of the vaccine development process against SARS-CoV-2, including 1) potential antigen targets 2) different vaccination strategies from conventional to novel platforms, 3) ongoing clinical trials, 4) varying challenges encountered during developing the most effective vaccine and the futuristic approaches.

Abbreviations: Acad, academy; Ads, Adenoviruses; AdCs, chimpanzee adenoviruses; ADMP, adenovirus-derived multimeric protein; ADE, Antibody-dependent enhancement; ADDE, antibody-dependent disease enhancement; ARDS, acute respiratory distress syndrome; CD4+ (T cells), cluster of differentiation 4; CD8+ (T cells), cluster of differentiation 8; CTD, Clinical Trial Database; FP, membrane fusion peptide; E, Enveloped protein; FLSP, full length S protein; GP 96, glycoprotein-96; GNVAM, glycoprotein nanoparticle vaccine adjuvanted with Matrix M; HLA, Human leukocyte antigen; hACE 2, human angiotensin-converting enzyme 2; HCR, human cellular receptor; HIRs, host immune responses; Ins, Institute; LUNAR, Lipid-enabled and Unlocked Nucleomonomer Agent modified RNA; LVVV, Live viral vectored vaccine; LTAEs, longer-term adverse events; LNP, lipid nanoparticle; MMR, Measles Mumps Rubella; MVA, Modified vaccinia virus Ankara; MHC, Major histocompatibility complex; MIDRT, Moderna's infectious disease research team; MSA, Multiple Sequence alignment; MSC, mesenchymal stem cell; NIH, National Institute of health; NTD, terminal domain; NRVV, non-replicating viral vectors; ORF, open-reading frames; OPV, Oral polio vaccine; RBD, receptor binding protein; PEI, pulmonary eosinophilic infiltration; PLA, People's Liberation Army; RBM, Receptor-binding motif; RSV, respiratory syncytial virus; RSP, Recombinant spike protein; RDV, replication-deficient vectors; RDCA, replication-deficient chimpanzee adenovirus; SARS, COV-2 severe acute respiratory syndrome coronavirus 2; SARS, COV severe acute respiratory syndrome coronavirus; SPs, structural proteins; S Protein, spike glycoprotein; S2RBD, SARS-COV-2 receptor binding protein; SRBD, SARS-COV receptor binding protein; S2E, E protein of SARS-CoV-2; S2M, SARS-COV-2 Membrane protein; S2N, SARS-COV-2 nucleocapsid protein; STARR, Self-Transcribing and Replicating RNA; S2E, SARS-COV-2 envelope protein; SamRNA, self-amplifying mRNA; SSRV, Single-stranded RNA virus; SIN, severe interstitial pneumonia; SPs, Structural proteins; TCEBPI, T-cell epitope-based peptide immunization; TPA, tissue plasminogen activator; Uni, University; VLP, Virus-like particles; VSV, Vesicular stomatitis virus.

* Corresponding authors at: Institute of Research and Development, Duy Tan University, Da Nang 550000, Viet Nam.
E-mail addresses: fhpottoo@iau.edu.sa (F.H. Pottoo), rengasamyrrajakannan@duytan.edu.vn (K.R.R. Rengasamy).

https://doi.org/10.1016/j.biopha.2021.111254
Received 15 October 2020; Received in revised form 17 December 2020; Accepted 3 January 2021
Available online 28 January 2021

1. Introduction

The SARS-CoV-2, previously called as novel coronavirus, that broke out in the Wuhan city of China, is causing significant number of deaths and morbidity in the human population worldwide [1–4]. After China, countries that were affected disproportionately were South Korea, Iran, consequently the USA, India, Brazil, Spain, Italy, Germany, Turkey, Russia, United Kingdom and the France [5]. Thereafter 74,864,905 cases were reported in which 1,661,450 deaths took place worldwide as of December 17, 2020 [6]. The Beta-coronavirus is the genus and Coronavirdae is the family to which SARS-CoV-2 belongs. The SARS-CoV-2 is an SSRV having a 30 kb genome with fourteen ORF consisting of 4 major structural proteins that are nucleocapsid (N), Spike (S), membrane (M), and envelope (E) proteins [7–10]. *Rhinolophusaffinis* bat coronavirus RaTG13 has 93.1 % nucleotide sequence identical to that of Spike gene sequence of SARS-CoV-2 strains, however only less than 75 % nucleotide sequence similarity to that of SARS-CoV. The SARS-CoV-2 viral S sequences as compared to SARS-CoV have 3 extra short insertions in the N terminal domain & 4 out of 5 key residues changes in the RBM of S protein RBD [9,10]. The SARS-CoV-2 as well as SARS-CoV share similar HCR ACE-II [1,7,11]. For the development of vaccine against SARS-CoV2, the protein S that is a spikeprotein has become a major target, mainly based on the elicitation of viral neutralizing antibodies. The current scenario regarding the vaccine developmental process consists of, 1) 47 vaccine candidates in clinical trial 2) 155 vaccine candidates at pre-clinical stage [12]. The vaccines against MERS-CoV, SARS-CoV & other coronavirus vaccines have posed toxicity concerns based on previous clinical investigations with the application of Spike protein-based vaccines, including immunopathological and inflammatory effects, for example, PEI and ADDE following subsequent viral challenge of immunized animals [13–24] The macrophages expressing FcR receptor can uptake anti-spike antibodies for ADDE results in macrophage activation & the secretion of IL-6, IL-8 & MCP1 and loss of TGFβ [25]. For the inactivated vaccines of the RSV and MERS-CoV after viral challenge, it has been documented that the Th2 is associated with immunopathology [13,26–28]. It is therefore necessary that vaccines developed against SARS-CoV2 should be carefully assessed while conducting clinical trials [29]. The epitope-rich S1 or RBD part of S protein is considered as a target instead of a whole full-length Spike for the development of a vaccine against MERS-CoV.

2. Strategies for the SARS-CoV2 vaccine development

Owing to the dramatic rise in the number of cases of COVID-19 cases worldwide, numerous attempts have been stimulated to develop vaccines against this deadly SARS-CoV-2 virus. About 47 vaccines qualified for clinical-stage entry and more than 155 vaccines are under investigation in the pre-clinical stages [30]. None of the vaccines is having a license for any other coronavirus affecting human subjects like SARS or MERS. One of the significant reasons for the unavaliability of vaccines is due to financial constraints. However other reasons also exist, including the design of the vaccines, transient immune-responsiveness, and toxicity concerns in pre-clinical models [31]. Due to several challenges encountered in the development of the vaccine; discussed further in the section 5.0 of this paper, developing a vaccine against SARS-CoV2 is an onerous task to perform. Multiple methodologies are adopted for developing vaccines including both the next generation as well as conventional techniques. In the case of coronavirus, live attenuated vaccines have safety concerns however inactivated ones have been successful upto pre-clinical stages in the primate models [32]. In the case of SARS-CoV2, within seven days, the immunized macaques challenged with SARS-CoV-2 were shielded from extreme ailment & virus levels, while placebo-subjected macaques endured SIN.The human phase I and II clinical trials of the inactivated vaccine are ongoing in China [30].

2.1. Spike protein or S protein

The S protein of SARS-CoV-2 is the subject of several efforts. The S protein is an essential molecule for viral entry into the host cells, being present on the outer layer of the virus. The antibodies targeting the S protein will prevent the virus from entering the host cell, thereby preventing the virus from replicating inside the host cell machinery [33]. On the 10th of January 2020, the full genetic sequence of S protein was released globally providing an idea for the development of a vaccine [4,10,33]. The SARS-CoV-2 vaccines in the UK are based on S protein. The researchers at the University of Oxford have modified a chimp adenovirus vector that carries genes encoding this Spike protein. Infecting human cells with this adenovirus will lead to the production of spike protein and became a valid target for the immune response. Approximately 1,000 subjects were recruited by the sponsor for phase 1 human clinical trial, initiated on 23 April 2020 [30]. A phase I/II preliminary study performed on the patient between 23 April and 21 May 2020 was published by the same sponsor in August. The vaccine has been reported to have a good safety profile, with both cellular and humoral responses produced [34]. As of 11th November 2020, the trial is under phase III, results of which are awaiting.The novel strategy for vaccine development is the use of mRNA; however, no licensed vaccines have previously used this methodology. The concept for mRNA is based on injecting mRNA encoding S protein and allowing host cell machinery to prepare the said protein. The merit of this methodology is allowing rapid scale production with a straight forward route. In the USA, mRNA vaccines against SARS-CoV2 have entered clinical trials and interim findings from phase III clinical trials have indicated 90 per cent effectiveness in participants with no previous exposure to SARS-CoV-2 [35].

2.2. Repurposing other vaccines

The vaccines which are already being developed for other ailments are repurposed for SARS-CoV2 infection as an alternative means of virus control. There are several vaccines for other ailments other than COVID-19 globally, which are being re-investigated for coronavirus control. The BCG vaccine which is already marketed against tuberculosis can boost the immune system offering some protection in many ailments, from influenza to bladder tumors [36,37]. Many investigations have suggested an epidemiological correlation between reduced national occurrence of COVID-19and community coverage of BCG [38,39]. There are about 5 clinical trials recruiting healthcare professionals to study whether BCG shows any protection against SARS-CoV-2 [30]. The other repurposed vaccines consist of OPV & MMR vaccine [40,41].

2.2.1. BCG

The scientists are now hypothesizing that BCG can offer protection against SARS-CoV2, and some publications support this claim [42]. The basic crux behind the hypothesis is that the US and Italy have suffered high mortality because of COVID-19 than BCG vaccinated countries like Japan, South Korea [43]. However,other multiple factors can give rise to SARS-CoV2 infection-related mortality in different places of the world. Many clinical investigations are ongoing to estimate BCG vaccination's effect on the possible responses from SARS-CoV-2 like in the Netherlands, US, and Australia in the high-risk subjects (health care workers and older), whether it might provide protection [44] from SARS-CoV2 [42]. A study in Germany are investigating VPM1000, which is an rDNA vaccine candidate derived from BCG, could prevent older or health care professionals from SARS-CoV-2 [42].

2.2.2. MMR

The relation of MMR vaccination and COVID-19 associated death rate has been suggested by the worldwide epidemiological data [44]. Several investigators did not agree to this statement for the live MMR vaccines other than BCG. Many countries like Latin American (Argentina, Ecuador, Chile) and Iran maintained more than ninety

percent vaccine coverage from 1985 till the high death rate from SARS-CoV2 [44]. The MMR is a live attenuated virus-based vaccine. MMR is currently in phase III clinical trial (NCT04357028) against SARS-CoV2. The hypothesis regarding the repurposing of the MMR vaccine is that antibodies can be generated against measles, which may cross-react with SARS-CoV2 [45].

2.3. Vaccination strategies

Much effort is being made to develop vaccine against SARS-CoV2 on accounts to tackle the current coronavirus pandemic. More than 150 firms or educational institutions are working on SARS-CoV2 vaccines with strategies that include DNA-based vaccines, RNA-based vaccines, non-replicating viral vector (NRVV), replicating viral vector (RVV), inactivated vaccines(IAcV), live-attenuated vaccines (LAVs), and protein subunits Fig. 1. Several groups are making use of computer-aided and machine learning technologies to study the interaction between the immune system and viral antigens to identify potential vaccine targets. These techniques had been already tried against numerous other pathogens, and are now being applied for the development of the vaccine against SARS-CoV-2. Tables 1 and 2 enlist the number of candidates, as of 11th November 2020, under different developmental stages and strategies respectively. As the SARS-CoV-2 shares similarities in the genetic makeup of two deadly coronaviruses, i.e. SARS and MERS, vaccine strategies used to combat SARS and MERS viruses are being adopted to guide the formulation and development of new SARS-CoV-2 vaccines [46].

2.4. Nucleic acid vaccines

Traditional vaccines (IAcV and LAVs) have a history of protecting from a massive number of infections such as poliovirus eradication from most of the countries. However, there are numerous disadvantages associated with these vaccines [47]. For example (a) booster shots are needed in case of inactivated vaccines to urge stronger immunity against diseases. (b) LAVs contain living organisms, there are high chances of increasing safety and stability concerns for instance i) Attenuated pathogens can revert to the original form and cause disease, ii) infection (BCG - local lymphadenitis), iii) Contamination of tissue culture [48]. It

Table 1
Number of vaccine candidates under different development stages.

Stage of development	Number of candidates	References
Pre-clinical	155	[12]
Phase I	22	
Phase I/II	14	
Phase II	2	
Phase I/II/III	0	
Phase III	10	

was, therefore a need to develop newer vaccine technologies to overcome the drawbacks of these conventional vaccines. Nucleic acid vaccines consist of only DNA or mRNA against specific structural viral protein and do not contain any disease-causing viral protein. DNA/RNA is taken up by cells and translated into viral antigen resulting in stimulation of an immune response against the viral antigen [49]. DNA and mRNA are gaining more attention than the conventional vaccine developmental approaches (inactivated, live-attenuated) owing to their safety, efficacy, and ease in mass manufacturing. This is evident from the number of researchers working on this technology. These vaccines can be administered by several methods such as commonly used injection needles into the skin, muscle tissues, various mucous membranes, and spleen. Delivery can also be done using the electroporation method [50]. There are some groups, on the other hand, working on a needle-free delivery system, for example, Immunomic Therapeutics, Inc./EpiVax, Inc./PharmaJet [51]. RNA-based vaccines are classified as conventional and self-amplifying RNAs. Traditionally mRNA-based vaccines encode the antigen of interest and contain untranslated regions from 5′ and 3′, whist self-amplifying RNAs (same RNAs) encode viral replication mechanism in addition to viral antigen permitting intracellular RNA amplification and profuse protein synthesis [52]. Not withstanding the numerous advantages of nucleotide vaccines, there are none approved mRNA/DNA vaccines so far available for the public use. If any vaccine against SARS-CoV2 happens to be from this category then it will be the first of its kind.

Two mRNA vaccine candidates, mRNA-1273(by Moderna/NIAID) and BNT162 a1, b1, b2, c2 (by BioNTech/FosunPharma/Pfizer) developed to protect from SARS-CoV2 entered phase III clinical trials after showing promising efficacy and safety in phase I and II [53,54]. The

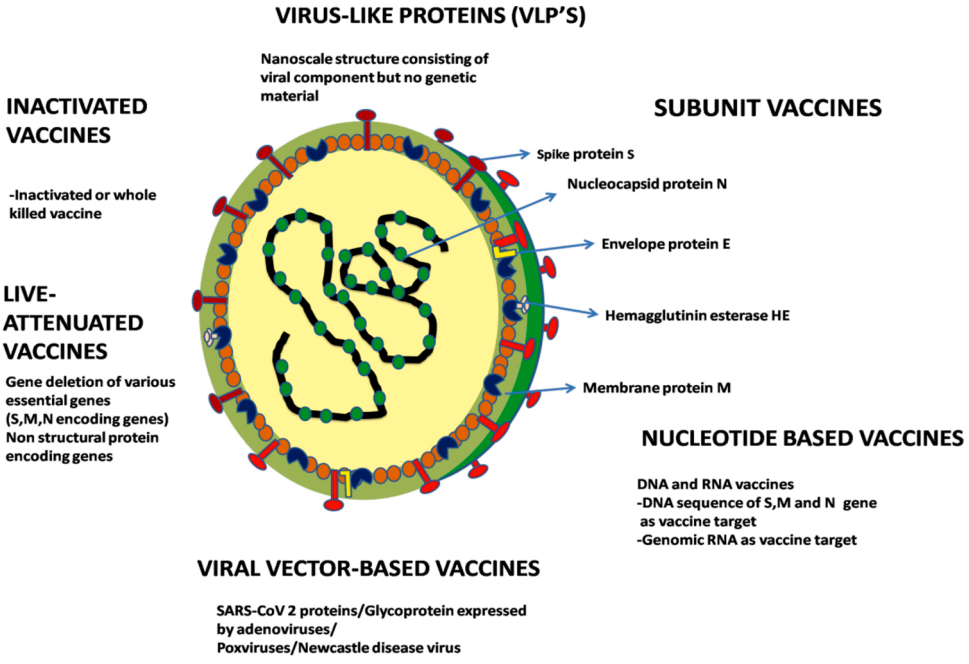

Fig. 1. Vaccination strategies for COVID 19.

Table 2
Numbers of vaccine candidates under different strategies.

Vaccine type	Pre-clinical	Phase 1	Phase I/II	Phase II	Phase I/II/III	Phase III	References
RNA	16	3	1	0	0	2 i-Moderna/NIAID ii-BioNTech/FosunPharma/Pfizer	
DNA	11	0	4	0	0	0	
Replicating viral vector	18	0	0	0	0	0	
Non-replicating viral vector	20	1	1	0	0	2 i-Uni of Oxford/AstraZeneca ii-CanSino Biological Inc./Beijing Ins of Biotechnology iii- Gamaleya Research Inst iv- Janssen Pharmaceutical Companies	[12]
Inactivated	9	0	2	0	0	3 i-Sinovac ii-Wuhan Ins of Biological Products/Sinopharm iii-Beijing Ins of Biological Products/Sinopharm	
Live attenuated	3	0	0	0	0	0	
Protein subunit	49	4	2	0	1	i- Novavax	
Virus-like particles	12	0	0	0	0	0	

mRNA-1273 is a new LNP-encapsulated mRNA-based vaccine that consists of prefusion-stabilized, FLSP of SARS-CoV-2. The NIH and MIDRT have confirmed the sequence for mRNA-1273 a few weeks after Chinese researchers shared the genetic sequence of SAR-CoV2 on a publicly available database. Moderna dispatched the first clinical batch of mRNA-1273 to the NIH for preceding the Phase 1 clinical trial on 24th February 2020. The process took just six weeks — the quickest turn around from the venture starts to immunization candidate in clinical history [55,56].

BioNTech/FosunPharma/Pfizer on the other hand are working on a unique combination of mRNA sequences as a vaccine candidate which contains two nucleoside modified mRNA candidates (BNT162a1 and BNT162b2). BNT162a1 is a uridine containing mRNA candidate while BNT162b2 is a samRNA. BNT162b1 consists of SARS-CoV-2 receptor-binding domain (S2RBD), while BNT162b2 contains the SARS-CoV-2 FLSP antigen. All of these mRNA constructs are packed within an LNP preparation. The spike protein is incorporated in two of the candidates and a smaller RBD fragment of the spike protein is incorporated in the other two candidates [57]. On 9th November 2020, Pfizer and BioNTech announced that their vaccine candidate demonstrated evidence of efficacy in 90 % of participants, who were not exposed to SARS-CoV-2 infection before injection,based on the first interim efficacy analysis from the Phase 3 clinical study [35].

2.5. Viral-vector based candidates

Vectored vaccines are constructed from a carrier such as adeno or pox virus which has been modified to contain a gene from the virus of interest [58]. The platform is broadly classified as replicating and NRVV.

Ads are extensively studied vectors for vaccine development attributable for its few potential benefits including their capacity to taint a wide range of hosts and to initiate elevated levels of transgene expression without the capability of the infectious viral gene being incorporated into the host genome; moreover, Ads can be fabricated safely and cheaply. Adenoviral vectors can be made non-replicating by removal of the fundamental gene for replication (E1 genes). Replication-enabled adenoviral vectors can be rendered with the deletion of E3 genes.As the E1 gene stays intact, as an outcome, these vectors have restricted ability for foreign gene insertion in comparison to RDV [59].

Extensively studied Ads are human Ads serotype 5(AdHu5) but because of the high frequency of AdHu5 neutralizing antibodies (NAs) found across the human populace, the efficiency of gene transfer by the vector is affected leading to diminishing potency of the vaccine. To beat this issue, Ads procured from numerous different species are tried to fill in as potential immunization vectors. Ads derived from chimpanzees were found to be advantageous as AdCs can be easily produced in a human cell line. Moreover, in comparison to human serotypes of Ad (AdHus), their seroprevalence in the human population is low, because they rarely circulated in humans [60].

AdCs are currently being utilized for the development of the SARS-CoV2 vaccine, for instance, a single dose of a vaccine candidate, ChAdOx1 nCoV-19 (Oxford University/ AstraZeneca) has demonstrated to protect six rhesus macaques from the incidence of pneumonia invoked by the virus. The vaccine manifested safety profile within an acceptable range and subsequent booster doses increased antibody immune responses in phase I/II single-blind, randomized control trials [34]. There are many more vectors being investigated for their efficiency as vaccine candidates (Table 3).

2.6. Subunits vaccines

Subunit vaccines incorporate just the parts, or antigens, that best invigorate the immune system framework. Since there is no live fragment involve, there is no danger of prompting a disease [61]. Subunit antibodies can be additionally classified into protein-based subunit vaccines, polysaccharide vaccines, and conjugate subunit vaccines [48]. Protein subunit vaccines are more steady and safer than live attenuated and inactivated/killed vaccines. They can be manufactured in a more cost-efficient manner as compared to other types of vaccines [47]. A shortcoming of this strategy is that if isolated proteins get denatured, may bind to different antibodies than the targeted protein of the

Table 3
Replicating and non-replicating viral vectors investigated as vaccine candidates against SARS-CoV2.

Replicating	Non- replicating	References
YF17D Vector Measles Vector Horsepox vector LVVV based on attenuated influenza virus backbone Influenza vector Replication-competent VSV chimeric virus technology Newcastle disease virus vector Avian paramyxovirus vector	Sendai virus vector Adenovirus-based MVA encoded VLP Replication defective Simian Adenovirus adenovirus-based NasoVAX adenovirus-based + HLA-matched peptides Inactivated Flu-based SARS-CoV2 vaccine + Adjuvant Influenza A H1N1 vector parainfluenza virus 5 -based vaccine Recombinant deactivated rabies virus Dendritic cell-based vaccine	[12]

pathogen [48].

SPs comprising spike (S), envelope (E), membrane (M), and nucleocapsid (N) that are expressed by SARS-CoV2 are viral antigens actuate neutralizing antibodies and generate a protective immune response [49]. Most exploited structural proteins are S and N proteins of coronaviruses. Companies like Intravacc.in collaboration with EpiVax are working on the outer membrane vesicle (OMV) delivery platform with synthetically produced SARS-CoV2 epitopes. It is one of the diverse platforms that are being investigated to produce a subunit vaccine.The candidate is currently under pre-clinical evaluation. Other diverse novel platforms that are being investigated under this strategy include GP-96 backbone and li-key peptide [51].

2.7. Virus-like-particles

VLP's are the newest vaccine development platform. The technology was previously utilized against several infections including HIV, influenza, Norwalk virus, enterovirus 71, rotavirus, etc, and is now being tried by several groups as SARS-CoV2 vaccine strategy [51]. VLPs are nanoscale structures comprising of viral proteins components assembled in a way that they mimic the original virus but do not contain the viral genetic material [62]. Therefore, cannot cause an infection like an actual virus. They are not contagious and non-replicative yet hold the ability to find entry into cytomembranes, enabling it pertinent for target delivery of antigens to antigen-presenting cells. Besides, the external surfaces and internal shells of VLPs give fantastic alteration locales to comply with the prerequisites for drug delivery [63]. Moreover, VLP's have an essential ability to instigate humoral and cell-mediated responses. They are further classified as enveloped and non-enveloped VLP's (eVLP's) based on their structure. Enveloped VLP's consist of the cell membrane of the host cell called an envelope and this envelope contains integrated target antigens displayed on the surface [64].

VBI vaccines Inc. are evaluating its multivalent eVLPvaccine candidate that encode antigenic protein components of SARS-CoV-2, SARS-CoV, and MERS-CoV proteins together on the same particle. As the construct consists of 3 different protein components, it is said to be trivalent (Fig. 2). It is beneficial as it allows for the production of antibodies possessing broad reactivity, offering protection from strains of SARS-CoV2 that may get mutated over time [65]. Another collaborates; Imophoron Ltd and Bristol University's Max Planck Centre are testing their unique ADDomerTM multiepitope display vaccine tech in animals. ADDomer is an ADMP-based self-assembling nanoparticle scaffold constructed to promote the plug-and-play display of numerous immunogenic epitopes from pathogens. What is unique about ADDomer-based vaccines is that it is thermostable thus making it free from cold storage requirements [66]. However, safety and efficacy in preventing SARS-CoV2 infection are not proved yet.

2.7.1. Clinical trials of BCG vaccine

Double blind, Phase III, randomized placebo-controlled trial is done to compare the severity of COVID-19 with efficacy of BCG vaccination to that of placebo. BCG (Bacille Calmette – Guerin) protects from various respiratory tract infections including those of the viral origin. Various clinical trials are designed to determine the impact of BCG vaccine on other clinically relevant respiratory tract infections or COVID-19, and also the immune system enhancement in those respiratory illness. COVID-19 related medical illness and the sponsors of BCG vaccine to compare its effectiveness has been listed in the table below:

Sponsor	Medical condition	References
LUMC, Department Infection Disease	BCG vaccination/immune response	[67]
Assistance publique Hopitaux de Paris	COVID-19 Health workers	
National Koranyi Institute of Pulmonology	Healthy volunteers working with SARS-Cov-2 infection	
University of Southern Denmark	Immune system activation for COVID-19 patients	
University of Rzeszow	Placebo - controlled Phase III, randomized double blind	
Hellenic Institute for the study of sepsis	Activate II trial- randomized trial to prevent infections by COVID-19	
Radboudumc	SARS-CoV-2 infection	
University Medical Center	SARS-CoV-2 infection	

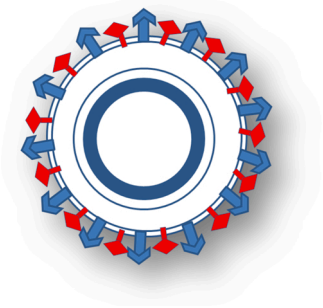

Fig. 2. Trivalent eVLP containing three different structural protein components.

2.8. Passive immunization

The passive immunization is an old procedure, however regrading SARS-CoV2 it is gaining scientific importance. The non availability of therapies against COVID-19 has triggered a pandemic, and now every scientific way is is being utilized to tackle this crisis. The available reports of passive immunization against SARS-CoV2 was initiated by Chinese and South Korean researchers [68]. A lot of publications have reported that passive immunization could be a great alternative unless and until some effective treatments are available like vaccines [69]. It was reported by Shen et al. that the application of convalescent plasma could be an available approach against COVID-19 subjects suffering from respiratory failure and with this approach they reported success in JAMA. In about five patients the passive immunization was effective with a decrease in viral load within 12 days after the transfusion [69].

2.9. Immune cell therapy

It has been reported that MSC therapy could act as a potential candidate forCOVID-19 by treating ARDS and the hypercytokinemia [70]. There are about thirty-one clinical trials registered on the NIH CTD which are going on, that are selectively for COVID-19 despite the absence of any pre-clinical evidences in covid models [70]. Due to COVID-19 the death rate is rising, the MSC are being investigated in the clinical settings to find out that whether it can show some proming outcome in the corona positive subjects. The subjects who received the MSC demonstrated lower TNF- α and high IL-10 levels as compared to placebo, but only three subjects were included in the study, so MSC can have a great potential in treating COVID-19 [70].

3. Potential targets for SARS-CoV2 vaccines

Structural proteins that are covering the SARS-CoV-2 surface serve as potential vaccination targets. These structures include the protein S, the protein E, protein M and the protein N [46].

3.1. Spike glycoprotein

Spike protein is the principal antigenic component out of all viral proteins that are responsible for instigating HIRs and the generation of neutralizing antibodies which induce preventive immunity against viral infections. Spike protein has therefore been selected as an essential target for immunization against SARS-CoV2 [46,71] majority of the

ongoing pre-clinical and clinical vaccine development researches are therefore based on spike protein. Out of 47 vaccine candidates currently under clinical trials, around 35 vaccine candidates are spike protein-based utilizing varying technology platforms (Table 4). The potential components of S protein for use as antigens include the FLSP, the RBD domain, the S1 subunit, the S2 subunit, the N-terminal domain (NTD), and membrane fusion peptide (FP) [72].

The S protein of coronaviruses is essential in promoting viral entry into target cells. Several researchers have discovered S2RBD in S protein and found that the RBD protein firmly binds to hACE2 receptors. The investigation further affirmed SARS-CoVRBD(S-RBD) generated antibodies that cross-react with S2-RBD protein, and S-RBD-actuated antisera cross-kill SARS-CoV-2, recommending the possibility to create SARS-CoV RBD-based immunizations for counteraction of SARS-CoV2 infection [73].

Most advanced spike protein-based candidate, *ChAdOx 1* vaccine developed by the University of Oxford Jenner Institute. The vaccine contains the optimized full-length surface S glycoprotein sequence of SARS-CoV-2, with a TPA asa leader sequence. It uses a RDCA to deliver a SARS-CoV-2 S protein to induce a protective immune response [34]. The evidence for the effectiveness of the *ChAdOx 1* was increased after researchers confirmed that the single-dose protected 6 rhesus macaques from the incidence of pneumonia invoked by the SAR-COV-2 [74] (Table 5).

3.2. The nucleocapsid protein

No vaccine that is explicitly focusing on nucleocapsid protein has entered clinical trials, however, ImmunityBio, Inc. &NantKwest Inc. vaccine candidate that is a Human Adenovirus Type 5 Vector (hAd5) encoding Spike (S) + Nucleocapsid (N) have entered phase I of a human clinical trial [12]. With the utilization of machine learning and reverse vaccinology (Vaxign reverse vaccinology (RV) and Vaxign-ML machine learning), analysts have found that the SARS-CoV-2 N protein (S2N protein) is more conserved with SAS-CoV and MERS-CoV than the other four human coronaviruses. N protein is found within the viral envelope, in the course of virion assembly, it wraps the RNA to shape the helical nucleocapsid. This protein is more conserved than the S protein and was accounted to initiate a humoral and cellular immune response against SARS-CoV-2 [78]. There are likewise new investigations showing the crystalline structure closeness of S2N protein with other revealed coronaviruses with distinct surface electrostatic features [79]. Institutes like National Research Centre in Egypt have proposed a candidate targeting multiple epitopes including S, N, M, and S1 protein. The candidate is still under a pre-clinical trial.

3.3. Membrane protein (M Protein)

The M protein of coronavirus has a vital role in virus assemblage [80]. M is positioned amidst S proteins in the virus envelope along side modest quantities of E and is the prime component in initiating the virus budding process. In the course of viral assembly, M interacts with several other SPs including N protein, with E protein, S protein, and also with itself [81]. Significant CD4+ and CD8+ T cells mediated immune responses were identified against S2M protein during the recent studies conducted in the virus-infected and recovered patients. S2M was recognized significantly and notable reactivity was observed [82]. Past examinations have shown that synthetic peptides obtained from immunodominant epitopes instigated strong antibody-induced immune reactions in the immunized rabbits, featuring the immunogenicity of SARS-CoV M protein [83]. Among the developers of the SARS-CoV2 vaccine, company OncoGen has proposed a synthetic long peptide vaccine candidate targeting S and M proteins of SARS-CoV-2, results of the pre-clinical studies are not yet revealed.

Table 4
Number of confirmed COVID-19 vaccine targets.

Target	Strategy	References
Spike glycoprotein S1,S2,RBD	DNA, RNA, replicating/non replicating viral vectors, subunit, VLP's, inactivated, live attenuated. Gp 96 backbone, OMV technology	[12]
Nucleocapsid protein	Subunit vaccine	
Membrane protein	Subunit protein	

4. Potential targets that are not under clinical/pre-clinical phases but require attention

4.1. Envelope protein

S2E can exist in monomeric and homo-pentameric form. It is a 75 amino acid protein. Past mutagenesis-based investigations have shown its significant function in viral pathogenesis [84]. A group recently designed the TCEBPI for SARS-CoV2 using the envelope protein as a target by utilizing an immunoinformatics approach. The designed vaccine however requires thorough testing to guarantee its safety and immunogenic profile [85]. S2E protein is 100 % homologous to bat coronavirus and the pangolin coronavirus. Also, less than 95 % of sequence similarity was noted between the S2E protein and the E protein sequence of SARS-CoV [84]. It is therefore suggested to carry out further investigation to determine and confirm the immunogenicity and efficacy of E protein as a potential vaccine target.

4.2. Nsp3

Nsp3 contains several functional domains that are important in assisting viral pathogenesis. During the phylogenetic and MSA study of nsp3 protein, It was reported that this protein in SARS-CoV-2 was more analogous to the human coronaviruses (SARS and MERS), and bat coronaviruses (HKU3, HKU4, and HKU9). Nps3 can serve as a potential vaccine target, further, an investigation is therefore suggested to validate its potential [78].

5. Challenges in vaccine development

5.1. An ideal vaccine

i) Should be safe, even in immunocompromised people. ii) Should be highly effective and optimally induce 'sterilizing' immunity [86]. iii) Should retail immunogenicity despite adverse storage. iv) Inexpensive v) Free from toxicity and adverse effects vi) Should give long term protection vii) Should have high thermal stability. Vaccine development is a lengthy, expensive process and many challenges arise during the development, manufacturing, and mass distribution.

5.2. Time

The major challenge in developing SARS-CoV2 vaccine is the fast-tracking of every step in the discovery, development, and evaluation process. This was not the case before the pandemic, as for any new vaccine to enter a commercial market and be widely available in the public domain, follows extensive safety and efficacy evaluation which usually requires a minimum duration of 5 years. However, due to an expedited increase in the number of COVID-19 cases worldwide, vaccine regulatory authorities both at international and national levels are forced to fast track every process of development to meet the world's immediate vaccine requirement. The scientific communities are using multiple approaches to shorten development phase including overlapping clinical phase and using advanced computer-aided and biotechnological tools.

Table 5
List of S protein-based vaccine candidates under clinical trial.

Vaccine	Status	Developers	Technology	References
ChAdOx 1	Phase III	Oxford uni/ AstraZeneca	Non-replicating viral vector	[12]
mRNA-1273	Phase III	Moderna/NIAID	LNP-encapsulated mRNA	
BNT162 a1, b1, b2, c2	Phase III	BioNTech/FosunPharma/Pfizer	mRNA candidates (BNT162a1 and BNT162b2); uRNA candidate; and a SAMRNA candidate. BNT162b1 encodes an optimized S2RBD antigen, while BNT162b2 encodes an optimized SARS-CoV-2 full-length S protein.	[57]
Ad5-nCoV	Phase III	CanSino biological	Adenovirus type 5 vector that encodes S protein	
Unnamed	Phase III	Anhui ZhifeiLongcom Biopharmaceutical/ Institute of Microbiology, Chinese Acad of Sciences	Protein Subunit Adjuvanted recombinant protein (RBD-Dimer	
Ad26COVS1	Phase III	Janssen Pharmaceutical Companies	Non-replicating viral vector	[12]
INO-4800	Phase I/II	Inovio Pharmaceuticals	Electroporation of DNA INO-4800 encoding S protein	
AG0301-COVID19	Phase I/ II	Osaka Uni/ AnGes/ Takara Bio	DNA + Plasmid against spike protein	
ZyCoV-19	Phase I/II	ZydusCadila	DNA plasmid vaccine	
GX-19	Phase I/II	Genexine Consortium	DNA vaccine encoding S-protein antigen	[75]
NVX-CoV2373	Phase III	Novavax	Full-length SARS CoV-2 GNVAM	[12]
KBP-COVID-19	Phase I/II	Kentucky Bioprocessing, Inc	Protein subunit RBD based	
LUNAR COV19	Phase I/II	Arcturus/Duke-NUS	STARR™, a combination of self-replicating RNA that encodes for the prefusion spike protein with LUNAR®	[76]
Gam-COVID-Vac Lyo	Phase I	Gamaleya Research Ins	Non-Replicating Viral Vector. Adeno type 26	
COVAX-19	Phase I	Vaxine Pty Ltd/Medytox	RSPwith Advax™ adjuvant	[12]
SCB-2019	Phase I	Clover Biopharmaceuticals Inc./GSK/Dynavax	Trimeric subunit Spike Protein vaccine	
NVX-CoV2373	Phase I	Uni of Queensland (Brisbane, Australia)	Protein subunit- Molecular clamp stabilized S protein with MF59 adjuvant	
LNP-nCoVsaRNA	Phase I	Imperial College of London	samRNA encoding S protein within anLNP	[77]
CVnCoV	Phase I	CureVac	mRNA	
ARCoV	Phase I	PLA Acad of Military Sciences/Walvax Biotech.	mRNA	
Unnamed	Phase I	Ludwig-Maximilians - University of Munich	MVA-SARS-2-S	
FINLAY- FR-2 anti SARS-CoV-2	Phase I	Instituto Finlay de Vacunas, Cuba	rRBD produced in CHO-cell conjugated chemically to tetanus toxoid	
MVC-COV1901	Phase I	Medigen Vaccine Biologics Corporation/ NIAID/Dynavax	Subunit protein - stabilized spike protein	
FINLAY- FR-1 anti SARS – CoV – 2	Phase I	Instituto Finlay de Vacunas, Cuba	Subunit protein – RBD + Adjuvant	[12]
Sf9	Phase I	West China Hospital, Sichuan University	Subunit protein - RBD	
UB-612	Phase I	COVAXX / United Biomedical Inc. Asia	Subunit protein- Multitope peptide-based S1-RBD-protein vaccine	
V590	Phase I	Merck Sharp & Dohme/IAVI	Replicating viral vector- VSV delivering the SARS-CoV-2 Spike	
COVID-19-101	Phase I	Institute Pasteur/Themis/Univ. of Pittsburg CVR/Merck Sharp & Dohme	RVV	
DelNS1-2019-nCoV-RBD-OPT1	Phase I	Beijing Wantai Biological Pharmacy/ Xiamen University	Intranasal flu-based-RBD	
Unnamed	Phase I	Sanofi Pasteur/GSK	Protein subunit	
Unnamed	Phase I	ImmunityBio, Inc. &NantKwest Inc.	Human Adenovirus Type 5 Vector (hAd5) Spike (S) + Nucleocapsid (N)	
BacTRL-Spike	Phase I	Symvivo	DNA	
GRAd-COV2	Phase I	ReiThera/LEUKOCARE/Univercells	Non-Replicating Viral Vector	
Unnamed	Phase I	SpyBiotech/Serum Institute of India	VLP	

5.3. Toxicity and adverse effects

The fast-tracking of vaccine development processes highten the risk of increased side effects. There is an immense possibility that some essential data might go missing or unnoticed at this accelerated speed of development. Researchers are concerned about the risk to public health if any important evaluation goes under notice. The use of animal models as a requirement of preclinical evaluation is usually needed to assess the safety and access risk associated with the new vaccine candidates or combination vaccines before starting first-in-human trials. Thorough pre-clinical and laboratory testing have to go in compliance with good laboratory practice guidelines and with national guidelines on animal experimentations. These studies are also necessary to establish characteristics (physical, chemical, and biological) of the vaccine candidates. Some research groups have omitted these essential animal testing studies in the face of global health crisis while others are running parallel pre-clinical and first-in-human trials. This break from the usual protocol is worrisome and a challenge that requires prime attention. Note that the translation of animal studies to humans may not 100 % with regards to toxicity studies. However, some animal models such as the mice, ferrets, syrian hamster, and rhesus macaques are demonstrating symptoms similar to humans upon exposure to SARS-CoV-2 [38].

Safety assessment of very recent technologies like DNA, RNA, VLP's is to be given more importance as even if there is evidence of safety and efficacy, there are very few/no vaccine so far licensed and used in a large

population. Serious adverse events and allergies were previously reported due to protein, non-protein impurities found in vaccines therefore thorough quality check during the manufacturing process is mandatory before distribution. A lamentable occurrence had occurred on account of polio immunization in 1955 because the cycle of inactivating the live virus was flawed. There were reports of paralysis and within a month the mass immunization program against polio was deserted. It was later uncovered that the vaccine had resulted in causing 40,000 instances of polio, leaving 200 youngsters with differing degrees of paralysis and several deaths [87]. Therefore manufacturing methodology has to be extremely audited and validated; manufacturers should be extremely meticulous in producing large quantities of vaccine doses. This will be challenging during the present race to license the first SARS-CoV2 vaccine and mass-produce a large number of doses.

5.4. Long term protection

Ideally, vaccination should provide long term protection. However, immunization induced resistance blurs after some time and the loss of protection varies with every disease [88]. Two doses of inactivated polio antibody (IPV) are 90 % effective or more against polio and three doses are 99 %–100 % effective and the duration of protection lasts for several years to decades [89]. Most promising SARS-CoV2 vaccine candidates in clinical trials require booster doses. It is too early to say any of it provides long term protection.

Reinfection is another major aspect affecting the protection period. A very recent study has confirmed reinfection with genomic evidence. It was concluded that SARS-CoV-2 might flow among the human populace regardless of crowd insusceptibility on account of general infection or immunization. Additional monitoring of patients with reinfection will help optimized vaccine design against SARS-CoV2 [90].

5.5. Mutations

In the early pandemic situation, there was concern among the scientific community over mutations arising in SARS-CoV-2. However recent studies have indicated no cause for concern.The outcomes of phylogenetic examination of various SARS-CoV-2 strains procured from various nations showed that all the glycoproteins of various strains of SARS-CoV-2, obtained from various nations were strongly related to each other; hence antibody structured against one strain would be successful against the various strains of SARS-CoV-2 from various nations. Nevertheless, it is essential to continuously monitor genomic sequence given the knowledge of previous experience on virus mutation rate [91].

5.6. ADE

ADE virus infection is a condition in which antibodies targeting the virus enhance the entry of virus or viral replication into the host cell (monocytes/granulocytes), by interacting with Fc receptors [92]. ADE of ailment is an overall worry for the development of immunizations and treatments since it possibly intensifies the infection or triggers dangerous immunopathology [93]. A serious ADE was seen in the case of the dengue vaccine. The rate of hospitalization was increased in vaccinated children than in non-vaccinated children. It was discovered that the antibody imitated the primary infection and that a reduction in the immunity presented a few youngsters to the danger of ADE in case of a subsequent secondary infection. ADE is believed to be liable for causing COVID-19. So far there are none in-vitro, in-vivo, or clinical proof of ADE happening in COVID-19 patients nonetheless, ADE may represent some serious results during the regular course of the illness [94]. ADE in SARS and MERS has been demonstrated in animal models. Therefore careful monitoring of ADE over several years post-vaccination is necessary especially during the mass vaccination program as ADE is evident when enough people have been vaccinated.

5.7. Cost

Even if a single vaccine is proven safe and efficacious, large scale manufacturing and distribution will be challenging especially if vaccine candidate involves novel technologies as very few manufacturing plant have previous experiences in mass production. The establishment will have to comply with the GLP guidelines for the particular vaccine candidate. Setting up new premises and infrastructures for vaccine production which is meeting complete quality guidelines will have cost involving. Also in the current global rush to develop a vaccine, there is a possibility that this very crucial compliance step might miss adequate attention; posing a potential danger. The challenge is to vaccinate the entire world population. Experts worry that this might be physically difficult to achieve owing to resource scarcity. Also, there should be production balanced against the need for other vaccines. The kind of infrastructure needed for production will depend on the type of vaccine. The world's governments and companies need to invest enough money so that vaccines can be made quickly available. Financial losses may also occur if the pandemic is ended before development phases are completed as experienced from previous epidemics like SARS.

6. Prospects

So far researchers have assured vaccines for SARS-CoV2 will be ready within a few months. If the first licensed vaccine happens to be from a novel technology platform, it will lead to a revolution in the vaccine development landscape. Since these platforms utilize machine learning, immunoinformatics, biotechnological tools, genome sequencing, and other in-silico approaches to arrive at the most promising hit molecule, aiding in the reduction of the duration of vaccine development significantly. It will change how we will respond to future emerging virus infection. Some of these candidates have loosened animal toxicity studies and started directly dosing in humans. If such vaccines are successful in justifying such a break from the usual protocol, usage of animal models will also change. International and national vaccine regulatory authorities might be required come up with newer guidelines in conducting pre-clinical and clinical evaluation studies with the condition that novel vaccine candidate is successful in providing immunity against the virus for a more extended period.

Post-licensure vaccine safety assessment is a long term assessment of adverse event occurring post-vaccination. It is necessary for detecting vaccine-related rare and LTAEs and keeping up people's certitude in vaccines and following immunization schedules [95]. Such surveillance has to be conducted meticulously using either active or passive methods to determine AEFI/ADE or if there is a decrease of immunity.

Keeping entire hope on a vaccine to end the pandemic should not be encouraged. Because of the wide challenges highlighted above, it will not be a quick process to reach the vaccine shots to each individual. According to WHO, world leaders and the public must follow and come up with a novel social measure to reduce viral concentrates within the human populace.

7. Conclusion

More studies are required urgently to reach the most successful vaccine candidate in order to minimize the growing number of COVID-19 cases (74,864,905 worldwide to date 17th, December 2020). The need for thorough review of testing methodologies is just as critical as its urgent obligation to ensure that vaccines produced are free from long-term and short-term toxicity and adverse effects. Since the vaccine alone cannot combat the pandemic, modern preventive social strategies are needed so that the world can battle the present pandemic and be able to face another pandemic if it occurs in the future.

Declaration of Competing Interest

None.

Acknowledgments

The authors; Jonaid Ahmad Malik, Almas Mulla & Tehmeena Farooqi aknowledge NIPER-Guwahati ((Ministry of Chemicals and Fertilizers, Govt. of India), AICTE (All India Council for Technical Education), and NIPER-Hyderabad (Ministry of Chemicals and Fertilizers, Govt. of India) resp. for them providing fellowships.

References

[1] J.F.W. Chan, S. Yuan, K.H. Kok, K.K.W. To, H. Chu, J. Yang, F. Xing, J. Liu, C.C. Y. Yip, R.W.S. Poon, H.W. Tsoi, S.K.F. Lo, K.H. Chan, V.K.M. Poon, W.M. Chan, J. D. Ip, J.P. Cai, V.C.C. Cheng, H. Chen, C.K.M. Hui, K.Y. Yuen, A familial cluster of pneumonia associated with the 2019 novel coronavirus indicating person-to-person transmission: a study of a family cluster, Lancet (2020), https://doi.org/10.1016/S0140-6736(20)30154-9.

[2] C. Huang, Y. Wang, X. Li, L. Ren, J. Zhao, Y. Hu, L. Zhang, G. Fan, J. Xu, X. Gu, Z. Cheng, T. Yu, J. Xia, Y. Wei, W. Wu, X. Xie, W. Yin, H. Li, M. Liu, Y. Xiao, H. Gao, L. Guo, J. Xie, G. Wang, R. Jiang, Z. Gao, Q. Jin, J. Wang, B. Cao, Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China, Lancet (2020), https://doi.org/10.1016/S0140-6736(20)30183-5.

[3] S.C. Wu, Progress and concept for COVID-19 vaccine development, Biotechnol. J. (2020), https://doi.org/10.1002/biot.202000147.

[4] N. Zhu, D. Zhang, W. Wang, X. Li, B. Yang, J. Song, X. Zhao, B. Huang, W. Shi, R. Lu, P. Niu, F. Zhan, X. Ma, D. Wang, W. Xu, G. Wu, G.F. Gao, W. Tan, A novel coronavirus from patients with pneumonia in China, 2019, N. Engl. J. Med. (2020), https://doi.org/10.1056/NEJMoa2001017.

[5] J.A. Malik, M. Maqbool, COVID-19: An overview of cuurent scenario, CellMed Orthocellular Med. Pharm. Assoc. 10 (2020), https://doi.org/10.5667/CellMed.2020.0021.

[6] Worldometer, Coronavirus Update (Live): 74,864,905 Cases and 1,661, 450 Deaths From COVID-19 Virus Pandemic - Worldometer, Worldometer, 2020.

[7] N. Chen, M. Zhou, X. Dong, J. Qu, F. Gong, Y. Han, Y. Qiu, J. Wang, Y. Liu, Y. Wei, J. Xia, T. Yu, X. Zhang, L. Zhang, Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study, Lancet (2020), https://doi.org/10.1016/S0140-6736(20)30211-7.

[8] R. Lu, X. Zhao, J. Li, P. Niu, B. Yang, H. Wu, W. Wang, H. Song, B. Huang, N. Zhu, Y. Bi, X. Ma, F. Zhan, L. Wang, T. Hu, H. Zhou, Z. Hu, W. Zhou, L. Zhao, J. Chen, Y. Meng, J. Wang, Y. Lin, J. Yuan, Z. Xie, J. Ma, W.J. Liu, D. Wang, W. Xu, E. C. Holmes, G.F. Gao, G. Wu, W. Chen, W. Shi, W. Tan, Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding, Lancet (2020), https://doi.org/10.1016/S0140-6736(20)30251-8.

[9] Y. Wan, J. Shang, R. Graham, R.S. Baric, F. Li, Receptor recognition by the novel coronavirus from Wuhan: an analysis based on decade-long structural studies of SARS coronavirus, J. Virol. (2020), https://doi.org/10.1128/jvi.00127-20.

[10] P. Zhou, X. Lou Yang, X.G. Wang, B. Hu, L. Zhang, W. Zhang, H.R. Si, Y. Zhu, B. Li, C.L. Huang, H.D. Chen, J. Chen, Y. Luo, H. Guo, R. Di Jiang, M.Q. Liu, Y. Chen, X. R. Shen, X. Wang, X.S. Zheng, K. Zhao, Q.J. Chen, F. Deng, L.L. Liu, B. Yan, F. X. Zhan, Y.Y. Wang, G.F. Xiao, Z.L. Shi, A pneumonia outbreak associated with a new coronavirus of probable bat origin, Nature (2020), https://doi.org/10.1038/s41586-020-2012-7.

[11] Q. Li, X. Guan, P. Wu, X. Wang, L. Zhou, Y. Tong, R. Ren, K.S.M. Leung, E.H.Y. Lau, J.Y. Wong, X. Xing, N. Xiang, Y. Wu, C. Li, Q. Chen, D. Li, T. Liu, J. Zhao, M. Liu, W. Tu, C. Chen, L. Jin, R. Yang, Q. Wang, S. Zhou, R. Wang, H. Liu, Y. Luo, Y. Liu, G. Shao, H. Li, Z. Tao, Y. Yang, Z. Deng, B. Liu, Z. Ma, Y. Zhang, G. Shi, T.T.Y. Lam, J.T. Wu, G.F. Gao, B.J. Cowling, B. Yang, G.M. Leung, Z. Feng, Early transmission dynamics in Wuhan, China, of novel coronavirus-infected pneumonia, N. Engl. J. Med. (2020), https://doi.org/10.1056/NEJMoa2001316.

[12] WHO, Draft Landscape of COVID-19 Candidate Vaccines- November 3, World Heal. Organ., 2020 (n.d.).

[13] A.S. Agrawal, X. Tao, A. Algaissi, T. Garron, K. Narayanan, B.H. Peng, R.B. Couch, C.T.K. Tseng, Immunization with inactivated Middle East Respiratory Syndrome coronavirus vaccine leads to lung immunopathology on challenge with live virus, Hum. Vaccines Immunother. (2016), https://doi.org/10.1080/21645515.2016.1177688.

[14] B.M.Y. Cheung, L.S. Chan, I.J. Lauder, C.R. Kumana, Detection of body temperature with infrared thermography: accuracy in detection of fever, Hong Kong Med. J. (2012).

[15] H. Weingartl, M. Czub, S. Czub, J. Neufeld, P. Marszal, J. Gren, G. Smith, S. Jones, R. Proulx, Y. Deschambault, E. Grudeski, A. Andonov, R. He, Y. Li, J. Copps, A. Grolla, D. Dick, J. Berry, S. Ganske, L. Manning, J. Cao, Immunization with modified vaccinia virus ankara-based recombinant vaccine against severe acute respiratory syndrome is associated with enhanced hepatitis in ferrets, J. Virol. (2004), https://doi.org/10.1128/jvi.78.22.12672-12676.2004.

[16] Z.Y. Yang, H.C. Werner, W.P. Kong, K. Leung, E. Traggiai, A. Lanzavecchia, G. J. Nabel, Evasion of antibody neutralization in emerging severe acute respiratory syndrome coronaviruses, Proc. Natl. Acad. Sci. U. S. A. (2005), https://doi.org/10.1073/pnas.0409065102.

[17] M. Czub, H. Weingartl, S. Czub, R. He, J. Cao, Evaluation of modified vaccinia virus Ankara based recombinant SARS vaccine in ferrets, Vaccine (2005), https://doi.org/10.1016/j.vaccine.2005.01.033.

[18] D. Deming, T. Sheahan, M. Heise, B. Yount, N. Davis, A. Sims, M. Suthar, J. Harkema, A. Whitmore, R. Pickles, A. West, E. Donaldson, K. Curtis, R. Johnston, R. Baric, Vaccine efficacy in senescent mice challenged with recombinant SARS-CoV bearing epidemic and zoonotic spike variants, PLoS Med. (2006), https://doi.org/10.1371/journal.pmed.0030525.

[19] Y. He, Y. Zhou, P. Siddiqui, J. Niu, S. Jiang, Identification of immunodominant epitopes on the membrane protein of the severe acute respiratory syndrome-associated coronavirus, J. Clin. Microbiol. (2005), https://doi.org/10.1128/JCM.43.8.3718-3726.2005.

[20] Y.W. Kam, F. Kien, A. Roberts, Y.C. Cheung, E.W. Lamirande, L. Vogel, S.L. Chu, J. Tse, J. Guarner, S.R. Zaki, K. Subbarao, M. Peiris, B. Nal, R. Altmeyer, Antibodies against trimeric S glycoprotein protect hamsters against SARS-CoV challenge despite their capacity to mediate FcγRII-dependent entry into B cells in vitro, Vaccine (2007), https://doi.org/10.1016/j.vaccine.2006.08.011.

[21] C.W. Olsen, W.V. Corapi, C.K. Ngichabe, J.D. Baines, F.W. Scott, Monoclonal antibodies to the spike protein of feline infectious peritonitis virus mediate antibody-dependent enhancement of infection of feline macrophages, J. Virol. (1992), https://doi.org/10.1128/jvi.66.2.956-965.1992.

[22] T. Takano, S. Yamada, T. Doki, T. Hohdatsu, T. Vopat, Pathogenesis of oral type I feline infectious peritonitis virus (FIPV) infection: antibodydependent enhancement infection of cats with type I FIPV via the oral route, J. Vet. Med. Sci. (2019), https://doi.org/10.1292/jvms.18-0702.

[23] C. Te Tseng, E. Sbrana, N. Iwata-Yoshikawa, P.C. Newman, T. Garron, R.L. Atmar, C.J. Peters, R.B. Couch, Immunization with SARS coronavirus vaccines leads to pulmonary immunopathology on challenge with the SARS virus, PLoS One (2012), https://doi.org/10.1371/journal.pone.0035421.

[24] R.C. Weiss, F.W. Scott, Antibody-mediated enhancement of disease in feline infectious peritonitis: comparisons with dengue hemorrhagic fever, Comp. Immunol. Microbiol. Infect. Dis. (1981), https://doi.org/10.1016/0147-9571(81)90003-5.

[25] L. Liu, Q. Wei, Q. Lin, J. Fang, H. Wang, H. Kwok, H. Tang, K. Nishiura, J. Peng, Z. Tan, T. Wu, K.W. Cheung, K.H. Chan, X. Alvarez, C. Qin, A. Lackner, S. Perlman, K.Y. Yuen, Z. Chen, Anti-spike IgG causes severe acute lung injury by skewing macrophage responses during acute SARS-CoV infection, JCI Insight (2019), https://doi.org/10.1172/jci.insight.123158.

[26] T.R. Johnson, R.A. Parker, J.E. Johnson, B.S. Graham, IL-13 Is Sufficient for Respiratory Syncytial Virus G Glycoprotein-Induced Eosinophilia After Respiratory Syncytial Virus Challenge, J. Immunol. (2003), https://doi.org/10.4049/jimmunol.170.4.2037.

[27] A.Z. Kapikian, R.H. Mitchell, R.M. Chanock, R.A. Shvedoff, C.E. Stewart, An epidemiologic study of altered clinical reactivity to Respiratory Syncytial (RS) virus infection in children previously vaccinated with an inactivated RS virus vaccine, Am. J. Epidemiol. (1969), https://doi.org/10.1093/oxfordjournals.aje.a120954.

[28] H.W. Kim, J.G. Canchola, C.D. Brandt, G. Pyles, R.M. Chanock, K. Jensen, R. H. Parrott, Respiratory syncytial virus disease in infants despite prior administration of antigenic inactivated vaccine, Am. J. Epidemiol. (1969), https://doi.org/10.1093/oxfordjournals.aje.a120955.

[29] S. Jiang, Don't rush to deploy COVID-19 vaccines and drugs without sufficient safety guarantees, Nature. (2020), https://doi.org/10.1038/d41586-020-00751-9.

[30] S. Caddy, Developing a vaccine for covid-19, BMJ (2020), https://doi.org/10.1136/bmj.m1790.

[31] F. Amanat, F. Krammer, SARS-CoV-2 vaccines: status report, Immunity (2020), https://doi.org/10.1016/j.immuni.2020.03.007.

[32] Q. Gao, L. Bao, H. Mao, L. Wang, K. Xu, M. Yang, Y. Li, L. Zhu, N. Wang, Z. Lv, H. Gao, X. Ge, B. Kan, Y. Hu, J. Liu, F. Cai, D. Jiang, Y. Yin, C. Qin, J. Li, X. Gong, X. Lou, W. Shi, D. Wu, H. Zhang, L. Zhu, W. Deng, Y. Li, J. Lu, C. Li, X. Wang, W. Yin, Y. Zhang, C. Qin, Rapid development of an inactivated vaccine for SARS-CoV-2, BioRxiv (2020), https://doi.org/10.1101/2020.04.17.046375, 2020.04.17.046375.

[33] A.C. Walls, Y.J. Park, M.A. Tortorici, A. Wall, A.T. McGuire, D. Veesler, Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein, Cell. (2020), https://doi.org/10.1016/j.cell.2020.02.058.

[34] P.M. Folegatti, K.J. Ewer, P.K. Aley, B. Angus, S. Becker, S. Belij-Rammerstorfer, et al., Safety and immunogenicity of the ChAdOx1 nCoV-19 vaccine against SARS-CoV-2: a preliminary report of a phase 1/2, single-blind, randomised controlled trial, Lancet (2020), https://doi.org/10.1016/S0140-6736(20)31604-4.

[35] Pfizer, BioNTech, Pfizer and BioNTech Announce Vaccine Candidate against COVID-19 Achieved Success in First Interim Analysis from Phase 3 Study, Pfizer, 2020.

[36] S. Guallar-Garrido, E. Julián, Bacillus Calmette-Guérin (BCG) therapy for bladder cancer: an update, Immunotargets Ther. (2020), https://doi.org/10.2147/itt.s202006.

[37] S.J.C.F.M. Moorlag, R.J.W. Arts, R. van Crevel, M.G. Netea, Non-specific effects of BCG vaccine on viral infections, Clin. Microbiol. Infect. (2019), https://doi.org/10.1016/j.cmi.2019.04.020.

[38] B. Deb, H. Shah, S. Goel, Current global vaccine and drug efforts against COVID-19: Pros and cons of bypassing animal trials, J. Biosci. (2020), https://doi.org/10.1007/s12038-020-00053-2.

[39] A. Miller, M.J. Reandelar, K. Fasciglione, V. Roumenova, Y. Li, G.H. Otazu, Correlation between universal BCG vaccination policy and reduced mortality for

COVID-19, MedRxiv. (2020), https://doi.org/10.1101/2020.03.24.20042937, 2020.03.24.20042937.
[40] R. Franklin, A. Young, B. Neumann, R. Fernandez, A. Joannides, A. Reyahi, Y. Modis, Homologous protein domains in SARS-CoV-2 and measles, mumps and rubella viruses: preliminary evidence that MMR vaccine might provide protection against COVID-19, MedRxiv (2020), https://doi.org/10.1101/2020.04.10.20053207, 2020.04.10.20053207.
[41] S.E. Wenzel, J.V. Fahy, C. Irvin, S.P. Peters, S. Spector, S.J. Szefler, T.B. Casale, M. M. Cloutier, J.A. Elias, M.C. Liu, V. Taggert, Proceedings of the ATS workshop on refractory asthma: current understanding, recommendations, and unanswered questions, Am. J. Respir. Crit. Care Med. (2000), https://doi.org/10.1164/ajrccm.162.6.ats9-00.
[42] G. Redelman-Sidi, Could BCG be used to protect against COVID-19? Nat. Rev. Urol. 17 (2020) 316–317, https://doi.org/10.1038/s41585-020-0325-9.
[43] Miller: Correlation between universal BCG vaccination… - Google Scholar, (n.d.). https://scholar.google.com/scholar_lookup?journal=medRxiv&title=Correlation+between+universal+BCG+vaccination+policy+and+reduced+morbidity+and+mortality+for+COVID-19:+an+epidemiological+study&author=A+Miller&publication_year=2020&doi=10.1101/2020.03.24.20042937&#d=gs_cit&u=%2Fscholar%3Fq%3Dinfo%3AwQWjrPo0y3cJ%3Ascholar.google.com%2F%26output%3Dcite%26scirp%3D0%26hl%3Den (accessed December 5, 2020).
[44] Ö. Özdemir, Measles-mumps-rubella vaccine and COVID-19 relationship, MBio 11 (2020) 1–2, https://doi.org/10.1128/mBio.01832-20.
[45] R. Mirzaei, R. Mohammadzadeh, F. Mahdavi, F. Badrzadeh, S. Kazemi, M. Ebrahimi, F. Soltani, S. Kazemi, A.S. Jeda, M. Darvishmotevalli, R. Yousefimashouf, H. Keyvani, S. Karampoor, Overview of the current promising approaches for the development of an effective severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) vaccine, Int. Immunopharmacol. 88 (2020) 106928, https://doi.org/10.1016/j.intimp.2020.106928.
[46] G. Salvatori, L. Luberto, M. Maffei, L. Aurisicchio, L. Aurisicchio, G. Roscilli, G. Roscilli, F. Palombo, E. Marra, E. Marra, SARS-CoV-2 spike protein: an optimal immunological target for vaccines, J. Transl. Med. (2020), https://doi.org/10.1186/s12967-020-02392-y.
[47] M. Wang, S. Jiang, Y. Wang, Recent advances in the production of recombinant subunit vaccines in Pichia pastoris, Bioengineered (2016), https://doi.org/10.1080/21655979.2016.1191707.
[48] World Health Organization, MODULE 2- Types of Vaccine and Adverse Reactions, WHO, 2018, pp. 41–42.
[49] S.C. Pandey, V. Pande, D. Sati, S. Upreti, M. Samant, Vaccination strategies to combat novel corona virus SARS-CoV-2, Life Sci. (2020), https://doi.org/10.1016/j.lfs.2020.117956.
[50] W.W. Leitner, H. Ying, N.P. Restifo, DNA and RNA-based vaccines: principles, progress and prospects, Vaccine (1999), https://doi.org/10.1016/S0264-410X(99)00271-6.
[51] World Health Organization, Draft Landscape of COVID-19 Candidate Vaccines - 28th July, Who, 2020.
[52] N. Pardi, M.J. Hogan, F.W. Porter, D. Weissman, mRNA vaccines-a new era in vaccinology, Nat. Rev. Drug Discov. (2018), https://doi.org/10.1038/nrd.2017.243.
[53] ClinicalTrials.gov[Internet].ModernaTX, a Study to Evaluate Efficacy, Safety, and Immunogenicity of mRNA-1273 Vaccine in Adults Aged 18 Years and Older to Prevent COVID-19 - Full Text View - ClinicalTrials.gOv, Clinicaltrials.Gov., 2020.
[54] L.A. Jackson, E.J. Anderson, N.G. Rouphael, P.C. Roberts, M. Makhene, R.N. Coler, M.P. McCullough, J.D. Chappell, M.R. Denison, L.J. Stevens, A.J. Pruijssers, A. McDermott, B. Flach, N.A. Doria-Rose, K.S. Corbett, K.M. Morabito, S. O'Dell, S. D. Schmidt, P.A. Swanson, M. Padilla, J.R. Mascola, K.M. Neuzil, H. Bennett, W. Sun, E. Peters, M. Makowski, J. Albert, K. Cross, W. Buchanan, R. Pikaart-Tautges, J.E. Ledgerwood, B.S. Graham, J.H. Beigel, An mRNA vaccine against SARS-CoV-2 — preliminary report, N. Engl. J. Med. (2020), https://doi.org/10.1056/nejmoa2022483.
[55] A. Dance, Coronavirus vaccines get a biotech boost, Nature (2020), https://doi.org/10.1038/d41586-020-02154-2.
[56] Moderna, Moderna's Work on a COVID-19 Vaccine Candidate, Moderna, Inc., Moderna, 2020.
[57] Genetic Engineering and Biotechnology News (GEN), BioNTech, Pfizer, and Fosun Pharma - BNT162, Mary Ann Liebert, Inc., 2020.
[58] P.D. Minor, Live attenuated vaccines: historical successes and current challenges, Virology (2015), https://doi.org/10.1016/j.virol.2015.03.032.
[59] Y. Choi, J. Chang, Viral vectors for vaccine applications, Clin. Exp. Vaccine Res. (2013), https://doi.org/10.7774/cevr.2013.2.2.97.
[60] J. Guo, M. Mondal, D. Zhou, Development of novel vaccine vectors: chimpanzee adenoviral vectors, Hum. Vaccines Immunother. (2018), https://doi.org/10.1080/21645515.2017.1419108.
[61] NIH: NIAID, vaccine types | NIH: national institute of allergy and infectious diseases, NIH natl, Inst. Allergy Infect. Dis. (2019), https://doi.org/10.1016/0021-8707(56)90057-0.
[62] B.D. Hill, A. Zak, E. Khera, F. Wen, Engineering virus-like particles for antigen and drug delivery, Curr. Protein Pept. Sci. (2017), https://doi.org/10.2174/1389203718666161122113041.
[63] C. Yang, K. Huang, Clinical applications of virus-like particles: opportunities and challenges, Curr. Protein Pept. Sci. 20 (2019) 488–489, https://doi.org/10.2174/1389203720051903271207 52.
[64] B. Sarkar, S.S. Islam, U.S. Zohora, M.A. Ullah, Virus like particles - a recent advancement in vaccine development, Korean J. Microbiol. (2019), https://doi.org/10.7845/kjm.2019.9089.
[65] VBI VACCINES, VBI-2901: Pan-Coronavirus Vaccine Candidate Overview, VBI VACCINES, 2020, pp. 6–9.
[66] C. Vragniau, J.C. Bufton, F. Garzoni, E. Stermann, F. Rabi, C. Terrat, M. Guidetti, V. Josserand, M. Williams, C.J. Woods, G. Viedma, P. Bates, B. Verrier, L. Chaperot, C. Schaffitzel, I. Berger, P. Fender, Synthetic self-assembling ADDomer platform for highly efficient vaccination by genetically encoded multiepitope display, Sci. Adv. (2019), https://doi.org/10.1126/sciadv.aaw2853.
[67] Home - ClinicalTrials.gov, (n.d.). https://clinicaltrials.gov/ (accessed December 17, 2020).
[68] E.M. Bloch, S. Shoham, A. Casadevall, B.S. Sachais, B. Shaz, J.L. Winters, C. Van Buskirk, B.J. Grossman, M. Joyner, J.P. Henderson, A. Pekosz, B. Lau, A. Wesolowski, L. Katz, H. Shan, P.G. Auwaerter, D. Thomas, D.J. Sullivan, N. Paneth, E. Gehrie, S. Spitalnik, E.A. Hod, L. Pollack, W.T. Nicholson, L. A. Pirofski, J.A. Bailey, A.A.R. Tobian, Deployment of convalescent plasma for the prevention and treatment of COVID-19, J. Clin. Invest. 130 (2020) 2757–2765, https://doi.org/10.1172/JCI138745.
[69] J.C. Fischer, K. Zänker, M. Van Griensven, M. Schneider, D. Kindgen-Milles, W. T. Knoefel, A. Lichtenberg, B. Tamaskovics, F.J. Djiepmo-Njanang, W. Budach, S. Corradini, U. Ganswindt, D. Häussinger, T. Feldt, H. Schelzig, H. Bojar, M. Peiper, E. Bölke, J. Haussmann, C. Matuschek, The role of passive immunization in the age of SARS-CoV-2: an update, Eur. J. Med. Res. 25 (2020), https://doi.org/10.1186/s40001-020-00414-5.
[70] B.L. Yen, M.L. Yen, L.T. Wang, K.J. Liu, H.K. Sytwu, Current status of mesenchymal stem cell therapy for immune/inflammatory lung disorders: gleaning insights for possible use in COVID-19, Stem Cells Transl. Med. 9 (2020) 1163–1173, https://doi.org/10.1002/sctm.20-0186.
[71] Sino biologicals, Spike Protein / S protein, Sino Biol. (2020).
[72] J. Zhang, H. Zeng, J. Gu, H. Li, L. Zheng, Q. Zou, Progress and prospects on vaccine development against sars-cov-2, Vaccines (2020), https://doi.org/10.3390/vaccines8020153.
[73] W. Tai, L. He, X. Zhang, J. Pu, D. Voronin, S. Jiang, Y. Zhou, L. Du, Characterization of the receptor-binding domain (RBD) of 2019 novel coronavirus: implication for development of RBD protein as a viral attachment inhibitor and vaccine, Cell. Mol. Immunol. (2020), https://doi.org/10.1038/s41423-020-0400-4.
[74] N. van Doremalen, T. Lambe, A. Spencer, S. Belij-Rammerstorfer, J. N. Purushotham, J.R. Port, V.A. Avanzato, T. Bushmaker, A. Flaxman, M. Ulaszewska, F. Feldmann, E.R. Allen, H. Sharpe, J. Schulz, M. Holbrook, A. Okumura, K. Meade-White, L. Pérez-Pérez, N.J. Edwards, D. Wright, C. Bissett, C. Gilbride, B.N. Williamson, R. Rosenke, D. Long, A. Ishwarbhai, R. Kailath, L. Rose, S. Morris, C. Powers, J. Lovaglio, P.W. Hanley, D. Scott, G. Saturday, E. de Wit, S.C. Gilbert, V.J. Munster, ChAdOx1 nCoV-19 vaccine prevents SARS-CoV-2 pneumonia in rhesus macaques, Nature (2020), https://doi.org/10.1038/s41586-020-2608-y.
[75] C. Gov. Genexine, Safety and Immunogenicity Study of GX-19, a COVID-19 Preventive DNA Vaccine in Healthy Adults - Full Text View - ClinicalTrials.gOv, ClinicalTrials.Gov., 2020.
[76] Genetic Engineering and Biotechnology News (GEN), Arcturus therapeutics and Duke-NUS - LUNAR-COV19, Mary Ann Liebert, Inc., 2020.
[77] Imperial College of London, How the Vaccine Works | Faculty of Medicine | Imperial College London, Imp. Coll., London, 2020.
[78] E. Ong, H. Wang, M.U. Wong, M. Seetharaman, N. Valdez, Y. He, M.L. Vaxign, Supervised machine learning reverse vaccinology model for improved prediction of bacterial protective antigens, Bioinformatics (2020), https://doi.org/10.1093/bioinformatics/btaa119.
[79] S. Kang, M. Yang, Z. Hong, L. Zhang, Z. Huang, X. Chen, S. He, Z. Zhou, Z. Zhou, Q. Chen, Y. Yan, C. Zhang, H. Shan, S. Chen, Crystal structure of SARS-CoV-2 nucleocapsid protein RNA binding domain reveals potential unique drug targeting sites, Acta Pharm. Sin. B (2020), https://doi.org/10.1016/j.apsb.2020.04.009.
[80] B.W. Neuman, G. Kiss, A.H. Kunding, D. Bhella, M.F. Baksh, S. Connelly, B. Droese, J.P. Klaus, S. Makino, S.G. Sawicki, S.G. Siddell, D.G. Stamou, I.A. Wilson, P. Kuhn, M.J. Buchmeier, A structural analysis of M protein in coronavirus assembly and morphology, J. Struct. Biol. (2011), https://doi.org/10.1016/j.jsb.2010.11.021.
[81] E.A.J. Alsaadi, I.M. Jones, Membrane binding proteins of coronaviruses, Future Virol. (2019), https://doi.org/10.2217/fvl-2018-0144.
[82] A. Grifoni, D. Weiskopf, S.I. Ramirez, J. Mateus, J.M. Dan, C.R. Moderbacher, S. A. Rawlings, A. Sutherland, L. Premkumar, R.S. Jadi, D. Marrama, A.M. de Silva, A. Frazier, A.F. Carlin, J.A. Greenbaum, B. Peters, F. Krammer, D.M. Smith, S. Crotty, A. Sette, Targets of T cell responses to SARS-CoV-2 coronavirus in humans with COVID-19 disease and unexposed individuals, Cell (2020), https://doi.org/10.1016/j.cell.2020.05.015.
[83] Y. He, Y. Zhou, H. Wu, B. Luo, J. Chen, W. Li, S. Jiang, Identification of immunodominant sites on the spike protein of severe acute respiratory syndrome (SARS) coronavirus: implication for developing SARS diagnostics and vaccines, J. Immunol. (2004), https://doi.org/10.4049/jimmunol.173.6.4050.
[84] B. Tilocca, A. Soggiu, M. Sanguinetti, G. Babini, F. De Maio, D. Britti, A. Zecconi, L. Bonizzi, A. Urbani, P. Roncada, Immunoinformatic analysis of the SARS-CoV-2 envelope protein as a strategy to assess cross-protection against COVID-19, Microbes Infect. (2020), https://doi.org/10.1016/j.micinf.2020.05.013.
[85] M.I. Abdelmageed, A.H. Abdelmoneim, M.I. Mustafa, N.M. Elfadol, N.S. Murshed, S.W. Shantier, A.M. Makhawi, Design of a multiepitope-based peptide vaccine against the e protein of human COVID-19: an immunoinformatics approach, Biomed Res. Int. (2020), https://doi.org/10.1155/2020/2683286.
[86] G.L. Ada, The ideal vaccine, World J. Microbiol. Biotechnol. (1991), https://doi.org/10.1007/BF00328978.

[87] M. Fitzpatrick, The cutter incident: how America's first polio vaccine led to a growing vaccine CrisisOffitPaul a pages 240pp price $27.50 ISBN 0-300-10864-10868New Haven/London: yale university press, J. R. Soc. Med. (2006), https://doi.org/10.1177/014107680609900320.
[88] J. Cohen, How long do vaccines last? The surprising answers may help protect people longer, Science 80 (2019), https://doi.org/10.1126/science.aax7364.
[89] CDC, Polio Vaccine Effectiveness and Duration of Protection, Centers Dis. Control Prev., 2018.
[90] K.K.-W. To, I.F.-N. Hung, J.D. Ip, A.W.-H. Chu, W.-M. Chan, A.R. Tam, C.H.-Y. Fong, S. Yuan, H.-W. Tsoi, A.C.-K. Ng, L.L.-Y. Lee, P. Wan, E. Tso, W.-K. To, D. Tsang, K.-H. Chan, J.-D. Huang, K.-H. Kok, V.C.-C. Cheng, K.-Y. Yuen, COVID-19 re-infection by a phylogenetically distinct SARS-coronavirus-2 strain confirmed by whole genome sequencing, Clin. Infect. Dis. (2020), https://doi.org/10.1093/cid/ciaa1275.
[91] S. Duffy, Why are RNA virus mutation rates so damn high? PLoS Biol. (2018) https://doi.org/10.1371/journal.pbio.3000003.
[92] S.M.C. Tirado, K.-J. Yoon, Antibody-dependent enhancement of virus infection and disease, Viral Immunol. 16 (2003) 69–86, https://doi.org/10.1089/088282403763635465.
[93] A.M. Arvin, K. Fink, M.A. Schmid, A. Cathcart, R. Spreafico, C. Havenar-Daughton, A. Lanzavecchia, D. Corti, H.W. Virgin, A perspective on potential antibody-dependent enhancement of SARS-CoV-2, Nature (2020), https://doi.org/10.1038/s41586-020-2538-8.
[94] F. Negro, Is Antibody-dependent Enhancement Playing a Role in COVID-19 Pathogenesis? Swiss Med. Wkly., 2020 https://doi.org/10.4414/smw.2020.20249.
[95] P.L. Moro, P. Haber, M.M. McNeil, Challenges in evaluating post-licensure vaccine safety: observations from the Centers for Disease Control and Prevention, Expert Rev. Vaccines (2019), https://doi.org/10.1080/14760584.2019.1676154.