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.

CHAPTER

25

Severe acute respiratory syndrome coronavirus-2: An era of struggle and discovery leading to the emergency use authorization of treatment and prevention measures based on computational analysis

Alisha Merchant, Vidal H. Tania, Mahaly Baptiste, Hashimul Ehsan and Gen Kaneko

Department of Biology, University of Houston-Victoria, Victoria, TX, United States

25.1 Introduction

The global pandemic of severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) originated in central China (Kaiser Family Foundation, 2021; World Health Organization, 2020) and has spread worldwide (Dey et al., 2021). On January 30, 2020, the World Health Organization (WHO) called it an international public health emergency (World Health Organization, 2020) and a pandemic was declared on March 11, 2020 (Dey et al., 2021). According to the Kaiser Family Foundation, there are 147,872,393 cases globally with 3,120,401 total deaths (Kaiser Family Foundation, 2021). On April 26, 2021, alone, there were 11,156 new deaths (Kaiser Family Foundation, 2021). Since January 2020, the United States was identified as the country with the highest number of cases (Kaiser Family Foundation, 2021). As of April 27, 2021, there have been 31,983,655 cases and 570,230 deaths in the United States (Centers for Disease Control and Prevention, 2021g). With the ongoing pandemic, there is a need for understanding the structure of SARS-CoV-2, which would serve as a basis for the development of

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treatment and preventative measures (Mishra et al., 2020). Computational approaches utilized for the analysis of biological data make this process efficient and time-saving (Mishra et al., 2020).

25.2 Severe acute respiratory syndrome-Coronavirus-2

25.2.1 Severe acute respiratory syndrome-Coronavirus-2 background

Seven types of Coronaviruses can infect humans and cause a variety of symptoms varying from mild symptoms to severe disease (Andersen et al., 2020; Centers for Disease Control and Prevention, 2020; Eastman et al., 2020). The Coronaviruses that cause mild symptoms are HKU1, NL63, HCoV OC43, and HCoV 229E (Centers for Disease Control and Prevention, 2020). Severe disease-causing Coronaviruses include the Middle East respiratory syndrome (MERS), Severe acute respiratory syndrome (SARS), and SARS-CoV-2 (Fig. 25.1) (Eastman et al., 2020). In the last two decades, the Coronaviruses have caused three different outbreaks: SARS-CoV in 2003, MERS-CoV in 2012, and SARS- CoV-2 in 2019 (Eastman et al., 2020).

The causative agent of COVID-19 is SARS-CoV-2 which belongs to a *Coronaviridae* family (Dey et al., 2021) of Coronaviruses that are known to infect humans and other animal species (Eastman et al., 2020). These are positive-sense, single-stranded, lipid-enveloped ribonucleic acid (RNA) betacoronaviruses (Eastman et al., 2020; Mishra et al., 2020) of roughly 30 kb in length (Mishra et al., 2020; Tavares et al., 2020). They contain several

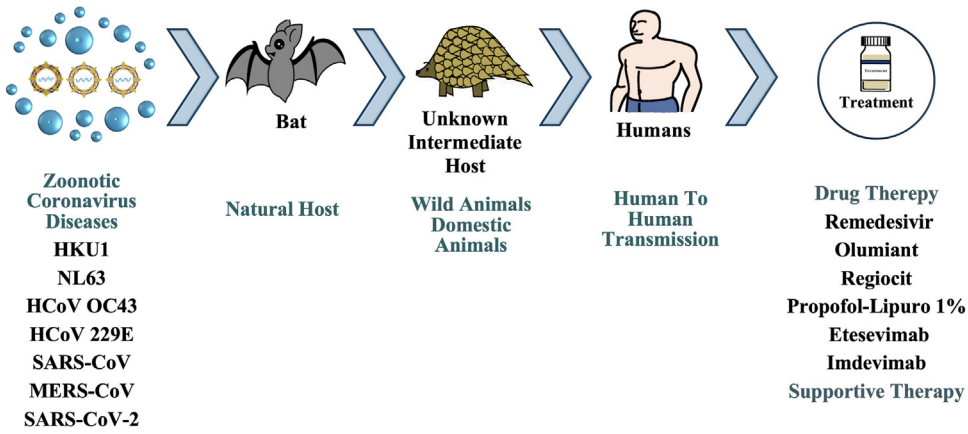

FIGURE 25.1 Overview of disease transmission and treatment. An overview of disease transmission and treatment of Coronavirus disesase-2019 demonstrates seven coronaviruses. They include HKU1, NL63, HCoV OC43, HCoV 229E, severe acute respiratory syndrome-Coronavirus, Middle East respiratory syndrome-Coronavirus, and severe acute respiratory syndrome-Coronavirus-2. The transmission of the virus to humans is through bats and an unknown intermediate host. There are several suspected hosts: one of which is Pangolin. Infected individuals may be asymptomatic or symptomatic. Depending on the severity of the symptoms, Veklury—remdesivir (only Food and Drug Administration-approved drug) may be prescribed. Other potential treatments include Olumiant, Convalescent Plasma, Regiocit, Propofol-Lipuro 1%, Etesevimab, Imdevimab, and other supportive therapy.

different spike proteins [ectodomain (receptor-binding subunits S1 and S2), transmembrane domain, and intracellular tail]. These viruses have been given the name Coronavirus because of their crown-like surface appearance (Eastman et al., 2020). The transmission of the virus to humans is known to be through bats and an unknown intermediate host. There are several suspected hosts: one of which is pangolin (Zhao et al., 2020). Depending on the severity of the symptoms different treatments may be prescribed (Fig. 25.1) (The United States Food and Drug Administration, 2021a).

25.2.2 Computational analysis of severe acute respiratory syndrome-Coronavirus-2

Computational approaches such as in silico analysis can be used to characterize viral genomes. The antigen analysis, for example, helps to determine immunological epitopes that can trigger an immune response without causing the reversal of viral pathogenesis (Dhama et al., 2020; Mishra et al., 2020) In silico proteome analysis of the SARS-CoV-2 genome (Mishra et al., 2020; Wu et al., 2020) identified 29 encoded proteins (Baruah et al., 2020). There are 14 open reading frames (ORFs): ORF1a, ORF1b, ORF 14, 9b, 8b, 7b, 7a, p6, 3a (papain-like protease), and 3b (Baruah et al., 2020; Sanami et al., 2021). ORF1a and ORF1b encode 16 different nonstructural proteins (NSPs) (Baruah et al., 2020; Tan et al., 2005). Other proteins that are encoded by the virus genome are the envelope protein (E), the membrane protein (M), spike glycoprotein S, and the nucleocapsid protein (N) (Baruah et al., 2020; Sanami et al., 2021; Tan et al., 2005).

ORF1ab, which consists of 7096 amino acids, is the largest multifunctional polyprotein rich in leucine and valine (Baruah et al., 2020). This protein is involved in the replication and translation of SARS-CoV-2 viral RNAs (Baruah et al., 2020). It includes 15 proteins including NSPs 1, NSP10, EndoRNAse, NSP6, helicase, NSP8, NSP4, RNA-dependent RNA polymerase (RdRp), NSP3, NSP2, 3C-like proteinase, 3′-to-5′ exonuclease, NSP7, NSP9, and 2′-O-ribose methyltransferase (Baruah et al., 2020). RdRp was also discovered to be involved in the replication and transcription of SARS-CoV-2 viral RNAs (Baruah et al., 2020).

Four proteins are critical for replication and transcription processes in SARS-CoV-2 (Baruah et al., 2020). These proteins include S, E, M, and N proteins. Spike glycoprotein S consists of 1273 amino acids (Dey et al., 2021) and are rich in leucine and serine (Baruah et al., 2020). They play a vital role in receptor-mediated SARS-CoV-2 attachment and fusion (Baruah et al., 2020). E proteins are rich in leucine and valine (Baruah et al., 2020). These proteins are crucial for virus assembly, pathogenesis, and morphogenesis (Baruah et al., 2020). Furthermore, M proteins were identified to be the most abundant SARS-CoV-2 structural protein, which played a crucial role in protein−protein interactions, viral fusion, assembly, and budding (Baruah et al., 2020). Likewise, N proteins are rich in glycine, alanine, and serine (Baruah et al., 2020). These proteins have three conserved domains: the N-terminal domain, C-terminal domain, and N-3 region (Baruah et al., 2020).

25.2.3 Mechanism of action

The replication of SARS-COV-2 is a multistep process. Once the SARS-CoV-2 virus enters the host body, it binds via the spike glycoprotein S to the host cell receptor at the

angiotensin-converting enzyme 2 (ACE2) (Boopathi et al., 2020). S2 fuses with the transmembrane protease serine 2 (TMPRSS-2) on the host and viral membranes forming the viral replication complex (Fig. 25.2A) (Eastman et al., 2020; Yesudhas et al., 2021). This begins the replication process, and the binding allows the virus to attach itself and release its RNA into the host cell by the process of receptor-mediated endocytosis (Li, Li et al., 2021; Yesudhas et al., 2021). RNA is released into the cytoplasm through membrane fusion forming a vesicle (Fig. 25.2B). Consequently, the host ribosome binds to the viral RNA (Yesudhas et al., 2021).

Viral replicase polyproteins such as pp1a and pp1ab are produced by translation. The proteolysis of these polyproteins forms NSPs (Li, Li et al., 2021). The positive viral RNA makes the enzyme RdRp or RNA replicase and is replicated to a negative RNA template strand (Li, Li et al., 2021). These are utilized during transcription to produce several

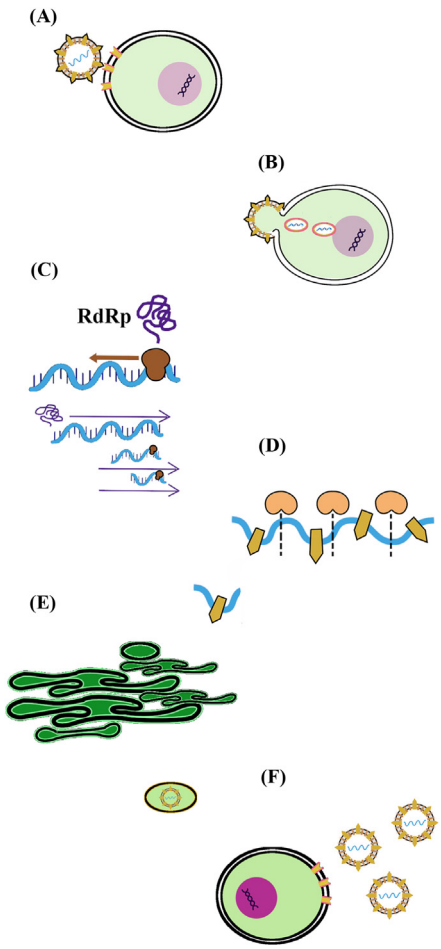

FIGURE 25.2 Severe acute respiratory syndrome-Coronavirus-2 mechanism of action. (A) Host is infected with SARS-CoV-2 virus. (B) The virus binds via spike glycoprotein S to the angiotensin-converting enzyme 2 receptors of the cell. (C) RNA translation produces RdRp. RdRp produces subgenomic RNAs, which are translated into proteins. (D) Proteases cut the proteins into individual structural proteins. (E) Replicated viral RNA and proteins are sent to the Golgi apparatus. (F) New virus is released from the cell and ready to infect another cell.

subgenomic mRNAs of different strand sizes (Fig. 25.2C) and are translated by the host ribosomes into structural proteins S, E, N, and M proteins (Fig. 25.2D) (Eastman et al., 2020). At the Golgi apparatus and endoplasmic reticulum, these proteins are assembled into virions, packaged by the endoplasmic reticulum forming the Golgi intermediate compartment (Fig. 25.2E) (Li, Li et al., 2021). This complex is then released from the cell via exocytosis where they will eventually infect other cells by repeating the entire process all over again (Fig. 25.2F) (Boopathi et al., 2020; Eastman et al., 2020; Li, Li et al., 2021; Yesudhas et al., 2021). There will be an eventual buildup of viral particles in the host which will lead to an immune response.

25.2.4 Clinical outcomes

COVID-19 is a contagious disease (Pollock & Lancaster, 2020) that primarily infects the respiratory system and intestinal tracts (Eastman et al., 2020; Zhang et al., 2020). The cells affected by the disease include epithelial cells of the nose, pneumocytes, alveolar macrophages, and enterocytes (Eastman et al., 2020). Other organ systems that can be potentially infected by the virus are the urogenital system, circulatory system, and central nervous system (Zhang et al., 2020). The clinical presentation of SARS-CoV-2 is diverse, varying from asymptomatic cases to death. Common symptomology experienced by patients includes hypoxia, cough, fever (temperature $\geq$ 100.4°F), feeling feverish, dyspnea (shortness of breath), palpitations, diarrhea (Zhang et al., 2020), nausea, vomiting, ageusia (loss of taste), anosmia (loss of smell), new-onset confusion, cyanosis (bluish-discoloration) of the face and lips, and chest discomfort (Kissoon, 2019).

The case fatality rate (CFR) for COVID-19 is the ratio of individuals who died from the disease to the individuals who contracted the disease (Spychalski et al., 2020). CRF is based on various factors, including region, population demographics, and healthcare capabilities (Eastman et al., 2020). While some cases experience a severe illness that may require hospitalization, intensive care, and (or) mechanical ventilation, other cases may die (Centers for Disease Control and Prevention, 2021c,d). The risk for hospitalization is 40 times greater for the age group of 65–74, 65 times greater for the age group of 75–84, and 95 times greater for anyone over the age of 85 (Centers for Disease Control and Prevention, 2021c). The risk of death is 1300 times greater for the age group of 65–74, 3200 times greater for the age group of 75–84, and 8700 times greater for anyone greater than 85 years of age (Centers for Disease Control and Prevention, 2021c). The reference group used is between the age of 5–17 (Centers for Disease Control and Prevention, 2021c). Older adults and people with comorbidities are at an increased risk of severe disease (Centers for Disease Control and Prevention, 2021d). More than 8 out of 10 COVID-19 related deaths occur in adults over the age of 65 (Centers for Disease Control and Prevention, 2021c). Underlying health conditions include but are not limited to diabetes, chronic kidney disease, chronic lung disease, neurological conditions, cancer, dementia, down syndrome, cardiac conditions HIV infections, immunocompromised patients, liver disease, obesity, sickle cell anemia, thalassemia, transplants (organ or stem cell transplant), history of strokes, cerebrovascular disease, substance abuse disorders, current or former cigarette smoking, and

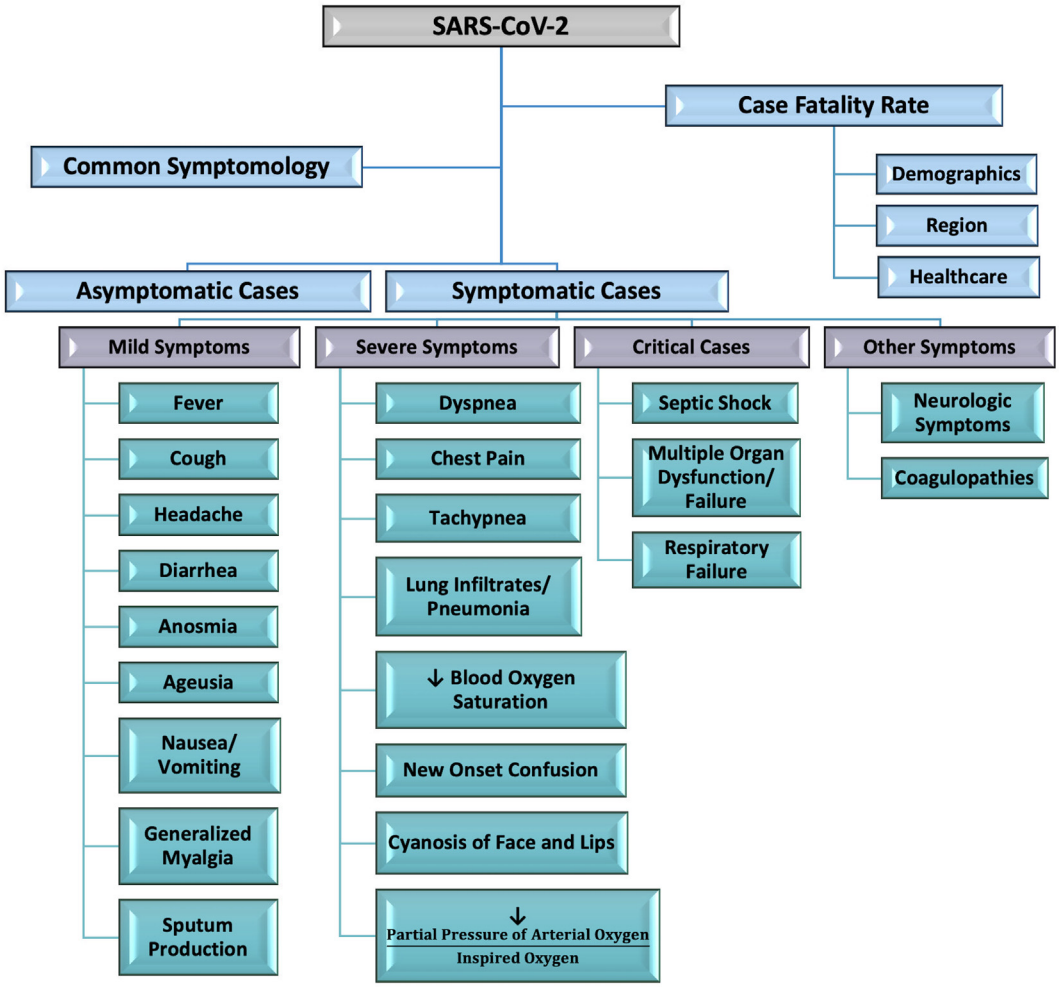

FIGURE 25.3 Clinical presentation of severe acute respiratory syndrome-Coronavirus-2 . Case fatality rate of SARS-CoV-2 is dependent on region or territory, demographics of the residents, and access to care. Infected individuals may be symptomatic or asymptomatic. Symptomatic individuals may experience a variety of symptoms. Mild symptoms include fever over 100.4°F, productive cough, nonproductive cough, generalized myalgia, headaches, diarrhea, anosmia, ageusia, nausea, and/ or vomiting. Severe symptoms include dyspnea, tachypnea, chest pain, lung infiltrates, decreased blood oxygen saturation levels, cyanosis of face and lips, new onset of confusion, diarrhea, and a decrease in the ratio of partial pressure arterial oxygen and a fraction of inspired oxygen. Additionally, critical cases may experience respiratory failure, multiple organ dysfunction or failure, and septic shock. Finally, other symptoms include neurologic symptoms and coagulopathies.

pregnancy (Centers for Disease Control and Prevention, 2021d). Fig. 25.3 depicts the clinical presentation of SARS-Co-V-2 in symptomatic and asymptomatic cases along with the factors that impact the CFR.

25.3 Coronavirus disease-2019 treatment options

25.3.1 Computational analysis of repurposed drugs

Traditional drugs and vaccines take several years to develop but now scientists can manufacture drugs rapidly due to computational analysis. One such example is RNA-based drugs, which are faster to manufacture and easy to manipulate. Computational in silico analysis can analyze drugs to ensure that they are effective in treating targeted diseases. Computational analysis for predicting bioactive compounds is rapid, methodical (Joshi et al., 2020), and cost-effective for the development of COVID-19 treatment drugs (Dhama et al., 2020; Mishra et al., 2020). With the urgent need, it was important to use creative methods to repurpose drugs rather than discover effective treatment mechanisms (Mishra et al., 2020). With no drugs available to treat COVID-19 (Mishra et al., 2020; Wu et al., 2020), drug repurposing was the only method to fight the pandemic (Mishra et al., 2020). Drug repurposing is a unique technique that involves using an existing drug and targeting a different disease (Lu et al., 2020; Mishra et al., 2020). Therefore because the drug targets of COVID-19 are known and the protein structure or its homologs are available, molecular docking can be used (Joshi et al., 2020; Mishra et al., 2020). Virtual screening approaches have also provided researchers with structures of SARS-CoV-2 proteins (Wu et al., 2020). These findings assist in the identification of numerous drugs that have potent antiinfective activity against the identified proteins (Mishra et al., 2020; Sun et al., 2018). The network approach method is an in silico approach that was utilized in drug repurposing (Mishra et al., 2020; Sun et al., 2018). This approach prioritizes potential drug targets and uses existing drugs that can possibly treat COVID-19 (Mishra et al., 2020; Sun et al., 2018). Some of the prospective repurposed drugs for SARS-CoV-2 include remdesivir, hydroxychloroquine, chloroquine, lopinavir, nelfinavir, streptomycin, and azithromycin (Li, Michelson et al., 2021; Mishra et al., 2020).

25.3.2 Pharmacological research

There are various studies of COVID-19 treatment drugs that are in their recruitment phase (Department of Health and Human Services). The list of global clinical trials shows that there are little to no studies in the Asian, Australian, and African subcontinents (Department of Health and Human Services, 2020). As of March 31, 2021, the Coronavirus Treatment Acceleration Program (CTAP), the brainchild of the United States's FDA, shows that there are over 600 drug development programs for the treatment of COVID-19 that are in the planning stages. The FDA has reviewed over 440 trials (The United States Food and Drug Administration, 2020a). These statistics do not include studies on vaccinations (The United States Food and Drug Administration, 2020a). The treatments that are being studied include but are not limited to: antivirals, neutralizing agents, immunomodulators, cell therapy, gene therapy, neutralizing antibodies, and combination therapies (The United States Food and Drug Administration, 2020a). Among the studies conducted in the United States, there are over 100 trials that are in the early stage trials testing for drug dosage and safety, while over 330 trials are in their late-stage trials testing for drug efficacy and safety (The United States

Food and Drug Administration, 2020a). Additionally, there are 10 drugs with emergency use authorization (EUA), including one approved for treatment (The United States Food and Drug Administration, 2020a).

On April 30, 2020, the first combination drugs, multiFiltrate PRO System and multiBic/multiPlus Solutions, were granted an EUA for treatment of severe COVID-19 (The United States Food & Drug Administration, 2021b). Other approved drugs for severe COVID-19 disease include Olumiant & Remdesivir with EUA issued on November 19, 2020; COVID-19 Convalescent Plasma with EUA issued on August 23, 2020; Fresenius Propoven 2% Emulsion with EUA issued on May 8, 2020; REGIOCIT renal replacement solution with EUA issued on August 13, 2020; and Propofool-Lipuro 1% with EUA issued on March 12, 2021 (The United States Food & Drug Administration, 2021b). Finally, COVID-19 convalescent plasma was authorized to be administered to hospitalized patients and was granted a EUA on August 23, 2020 (The United States Food & Drug Administration, 2021b).

One of the drugs currently authorized for emergency use for a mild-to-moderate disease is Bamlanivimab with EUA provided on November 11, 2020 (The United States Food & Drug Administration, 2021b). However, the EUA for Bamlanivimab to be administered alone was revoked on April 16, 2021 (Department of Health and Human Services, 2020; The United States Food and Drug Administration, 2021a). The drug was withdrawn because there was an increase in the frequency of resistant variants to the monoclonal antibodies (The United States Food and Drug Administration, 2021a). Other authorized drugs that were effective in combination therapy (The United States Food & Drug Administration, 2021b) are Bamlanivimab and Etesevimab with EUA issued on February 9, 2021, and Casirivimab and Imdevimab with EUA issued on November 21, 2020 (Ison et al., 2020).

25.3.3 Veklury—Remdesivir

Eastman et al. discuss a class of drugs, known as nucleoside analogs that serve as a therapeutic agents against various viruses (2020). This class of drugs can be further subdivided into three types. One type is the delayed chain terminator (Eastman et al., 2020). Remdesivir (GS-5734), an antiviral prodrug in this category, targets RdRp and blocks transcription (Eastman et al., 2020). It is a modified nucleotide with increased permeability and high potency towards viruses (Eastman et al., 2020). The goal of the primary assay was to develop an antiviral drug with no cytotoxic effects (Eastman et al., 2020). In vitro and in vivo models show that this antiviral is effective against different types of Coronaviruses (Eastman et al., 2020). These include but are not limited to: MERS, SARS, HCoV- OC43, HcoV- 229E, Ebola, RSV, and murine hepatitis virus (MHV) (Eastman et al., 2020). Remdesivir was a recommended repurposed drug of choice for the ongoing pandemic because it is efficient against various viruses and has a safety profile against Ebola (Eastman et al., 2020).

Gilead Sciences developed Remdesivir, a therapeutic agent, in collaboration with other organizations to treat RNA-dependent viruses that could cause a global pandemic (Eastman et al., 2020). Remdesivir (GS-5734), registered under the name Veklury (Gilead Sciences, 2020a, 2020b), was issued a EUA on May 1, 2020 (The United States Food & Drug Administration, 2021b). As of October 22, 2020, the EUA was reissued for COVID-19

hospitalized patients over the age of 12 and with a minimum weight of 40 kg (The United States Food and Drug Administration, 2020b). It is the first and only FDA-approved drug currently available in the market for the treatment of COVID-19 (The United States Food and Drug Administration, 2020b). There is various clinical research supporting its potency in the treatment of COVID-19 (Eastman et al., 2020).

Just like any other drug, Remdesivir (Veklury) has various side effects; the most common one being nausea (Gilead Sciences, 2020a, 2021). Lab abnormalities such as increased aspartate aminotransferase (AST) and increased alanine transaminase (ALT) were identified (Gilead Sciences, 2020a, 2021). Hypersensitivity reactions include infusion-related reactions, anaphylactic reactions, fever, rash, shivering, wheezing, diaphoresis, angioedema, dyspnea, blood pressure changes such as hypertension, or hypotension, and heart rate abnormalities including bradycardia or tachycardia (Gilead Sciences, 2020a, 2021). Additionally, Remdesivir can also cause kidney impairment and liver damage (Gilead Sciences, 2020a, 2021).

Another thing to consider regarding drug administration is that it is intravenously (IV) administered daily for up to 10 days (Eastman et al., 2020; The United States Food and Drug Administration, 2020b). Since the drug has to be administered through IV, it will require close monitoring during the treatment phase. It is also important to ensure that the recommended dose is administered for the prescribed duration. Gilead Sciences provides an ideal infusion time based on dosage and the rate of infusion. Before the drug can be administered, the patient's renal clearance must be checked to ensure that the estimated glomerular filtration rate is less than 30 mL/min (Gilead Sciences, 2020a, 2020b). However, it is important to note that data is scarce on the drug effects on pregnant women and their fetuses (Gilead Sciences, 2020a, 2020b). Finally, it is also unknown whether Remdesivir can pass through breast milk (Gilead Sciences, 2020a, 2020b).

25.4 Coronavirus disease-2019 vaccinations

25.4.1 Computational analysis of Coronavirus disease-2019 vaccines

Per the CDC, the United States ensures its vaccine safety by approving all vaccines before they are made available for administration (Centers for Disease Control and Prevention, 2011). The FDA has strict guidelines for ensuring the safety, availability, and effectiveness of the vaccines that are provided by EUA's (Centers for Disease Control and Prevention, 2011). This is ensured by three phases of clinical trials (Centers for Disease Control and Prevention, 2011).

On February 4, 2020, the Department of Health and Human Services (HHS), determined SARS-CoV-2 causing COVID-19 as a public health emergency affecting the US (Department of Health and Human Services, 2020). This declaration took effect on March 27, 2020, to provide EUA for drugs and other biological products (Department of Health and Human Services, 2020). Due to this emergency, health scientists have focused on the identification of ideal and efficient vaccine candidates (Mishra et al., 2020). With the urgent need to develop multiepitope vaccines and the high pathogenicity of SARS-CoV-2, a lucrative in silico analysis was implemented for predicting active antigenic epitopes (Mishra et al., 2020; Sanami et al., 2021). One of the methods for vaccine development

used was immunoinformatics (Mishra et al., 2020) and the reverse vaccinology method. These methods were used to develop a vaccine using SARS-CoV-2 genetic data and computer algorithms (Sanami et al., 2021). The target antigens for epitope prediction for this method include SARS-CoV-2 S, M, N, and E proteins. Also, their cytotoxic T lymphocytes (CTLs) and helper T lymphocytes (HTLs) epitopes were predicted and linked (Sanami et al., 2021). Epitope selection of the vaccine candidates was evaluated for "physicochemical properties, antigenicity, allergenicity," toxicity, secondary and three-dimensional structure prediction of the vaccine, linear B-cell epitope prediction, molecular docking, molecular dynamics simulation (to confirm stability), immune simulation (to confirm vaccine efficacy), reverse translation, codon optimization, and in silico cloning of the vaccine construct (Sanami et al., 2021).

While the FDA has granted EUAs to three vaccine candidates, many other researchers are using immunoinformatics approaches to develop other vaccines (Sanami et al., 2021). Reverse vaccination development utilize computational tools to virtually design a vaccine before further experimentation (Behmard et al., 2020; Hwang et al., 2021; Moxon et al., 2019). Some of the computational tools that have been used for reverse vaccination development include antigen selection, epitope prediction, immunogenicity prediction, and toxicology and allergenicity prediction.

25.4.1.1 Antigen selection

Phylogenetic analysis of SARS-CoV and SARS-CoV-2 revealed SARS-CoV has 76% of similarity in the spike glycoprotein S and 90.6% with protein N. Additionally, low levels of similarity was identified with MERS. Due to this genetic similarity, SARS-CoV information could aid in vaccine development. Based on the genetic similarity, the S2 subunit of the spike glycoprotein S was proposed to be studied further to potentially create a strong antibody response (Ahmed et al., 2020). Spike glycoprotein S was the first protein candidate because it is involved in the binding of the SARS-CoV-2 with the host cell (Fig. 25.2A) (Hwang et al., 2021). ANTIGENpro is a computational tool used for the prediction of protective antigens (Donald Bren School of Information and Computer Sciences, 2021; Sunita et al., 2020).

25.4.1.2 Epitope prediction

There are different computational and in-silico methods to identify cell epitopes, like B cell and T cell epitopes. The Immune Epitope Database (IEDB) is a prediction tool that helped to identify epitopes that were further considered for vaccine design (Behmard et al., 2020). Furthermore, VaxiJen v2.0 is a computational tool where the epitopes are tested for antigenic potential (Naz et al., 2020). Moreover, EpiJen v1.0 is an algorithm for T cell epitope prediction, which uses a multistep epitope selection (Doytchinova et al., 2006; Research TEJI, 2021). Also, HADDOCK is a biomolecular modeling platform that helps to check the binding affinity of the epitopes (Computational Structural Biology Group, 2021; Naz et al., 2020).

25.4.1.3 Immunogenicity prediction

There are various tools that can assist in the prediction of Immunogenicity. NEC Immune profiler software is a bioinformatic tool used to predict immunogenic neoantigens (Malone et al., 2020; NEC, 2020). Additionally, Monte Carlo random sampling procedure utilizing Phyton is a probability technique that can be used. For example, it can be used to

identify regions that are most likely to carry viable T-cell targets for vaccines or identify the selected epitope hotspots. Furthermore, EvoDesign is a protein design program in which mutations can be introduced to the spike glycoprotein S with the goal to achieve a greater immune response (Ong et al., 2021; Pearce et al., 2019).

25.4.1.4 *Toxicology and allergenicity prediction*

Allertop v2.0 is a computational bioinformatics tool used in vaccine designs to check the probability of compounds causing allergic reactions (Medical University of Sofia, 2021; Naz et al., 2020). Additionally, the University of Southern California created DeepVacPred as an AI framework that provides vaccine subunits that are low in allergenicity. This tool can be also used for epitope prediction and antigenicity (Sherwood, 2021; Yang et al., 2021). Furthermore, ToxCast and Tox21 are both US research agencies, which use high-throughput screening and computational methods to evaluate the safety of the proposed vaccine (Keshavarzi Arshadi et al., 2020; Agency, 2021; U.S. Environmental Protection Agency, 2021). The use of these computational tools aid in proposing a vaccine design, however, in vivo studies are required to confirm the predictions and analyze the effectiveness of the vaccine candidates (Hwang et al., 2021). Once potential is identified with the in silico design, the investigational vaccine is tested in animals (Centers for Disease Control and Prevention, 2011).

Under normal circumstances, Phase 1 clinical trial proceed Phase 2 clinical trial. Due to the pandemic, for the SARS-CoV-2 investigational vaccine, Phase 1 and Phase 2 clinical trials were combined (Levin, 2020). These are known as Phase 1/2a clinical trials. Their focus is to identify safety and the correct dosage before the clinical trial moved on the Phase 2b or Phase 3, obtain licensure, and finally move on to Phase 4, with the overall outcome to ensure vaccine safety (Levin, 2020) (Table 25.1).

25.4.2 Coronavirus disease-2019 vaccines

As of April 2021, Pfizer (BNT162b2); Moderna (mRNA 1273); and Johnson & Johnson (JNJ7843673) have received the FDA's EUA for the prevention of COVID-19 (The United States Food & Drug Administration, 2021b). AstraZeneca, on the other hand, is awaiting a EUA application in the United States, but it is already approved in the United Kingdom (Shimabukuro and Oliver 2021). After receiving the EUA, per the CDC, as of April 3, 2021, the total number of vaccine doses delivered was 207,866,645 with 161,688,422 vaccines administered (Centers for Disease Control and Prevention, 2021a). The population that received at least one dose included 31.4% of the total population in the United States, with 40.1% of the population over the age of 18, and 75% over the age of 65 (Centers for Disease Control and Prevention, 2021a). On the other hand, fully vaccinated people included 18% of the total population, with 23.1% of the population over the age of 18, and 54.7% over the age of 65 (Centers for Disease Control and Prevention, 2021a). Out of these, 29,855,663 received the two doses of Pfizer-BioNTech, 26,153,607 received the two doses of Moderna, 3,815,015 received the single dose of Johnson & Johnson, and 33,861 received two doses of an unknown vaccine (Centers for Disease Control and Prevention, 2021a). With these numbers, the country is heading towards normalization.

TABLE 25.1 Vaccine development during severe acute respiratory syndrome-Coronavirus-2 pandemic.

Vaccine development during SARS-CoV-2 pandemic		
Computational preclinical research	Antigen selection	Genetic similarity test
		ANTIGENpro
	Epitdpe selection	Immune epitope database (IEDB)
		VAXIJen v2.0
		EPIJen v1.0
		HADDOCK
	Immunogenicity prediction	NEC immune profiles
		Monte Carlo random sampling test
		EVODesign
	Toxicology and allergenicity prediction	Allertop v2.0
		DeepVacPred
		ToxCast
		Tox21
	Phase 1	
Simultaneous	Phase 2	
	Phase 3	
Emergency use authorization		

25.4.2.1 *Pfizer—BioNTech BNT162 RNA-based vaccine*

Pfizer—BioNTech is BNT162b2 vaccine was the first vaccine to receive a EUA for the prevention of SARS-CoV-2 (The United States Food & Drug Administration, 2021b). It is a double-dose vaccine administered 21 days apart as an intramuscular (IM) injection (Pfizer-BioNTech, 2020) for individuals over the age of 12 (The United States Food & Drug Administration, 2021b). It is the only vaccine currently available in the market that is considered safe for the age group of 12–18. A EUA request was submitted to the FDA on November 20, 2020 (The United States Food and Drug Administration, 2020c). This is when the Advisory Committee scheduled a meeting for December 10, 2020. FDA granted EUA on December 11, 2020 (The United States Food & Drug Administration, 2021b), which was amended on May 10, 2021, to include minors (The United States Food & Drug Administration, 2021a).

Pfizer—BioNTech is a lipid nanoparticle (LNP) vaccine based on the "nucleoside-messenger RNA" encoding for the antigen P2 mutant for prefusion of full-length spike glycoprotein S (Fig. 25.4A1) (Pfizer-BioNTech, 2020). The genetically modified mRNA antigen P2 locks the spike protein in its prefusion conformation, which blocks the spike protein in the conformation before it binds with the ACE2 receptors (Pfizer-BioNTech, 2020). The vaccine stimulates the creation of antibodies that would attack the spike protein of SARS-CoV-2 before it can fuse with the host cell and change its conformation (Fig. 25.4B) (Pfizer-BioNTech, 2020).

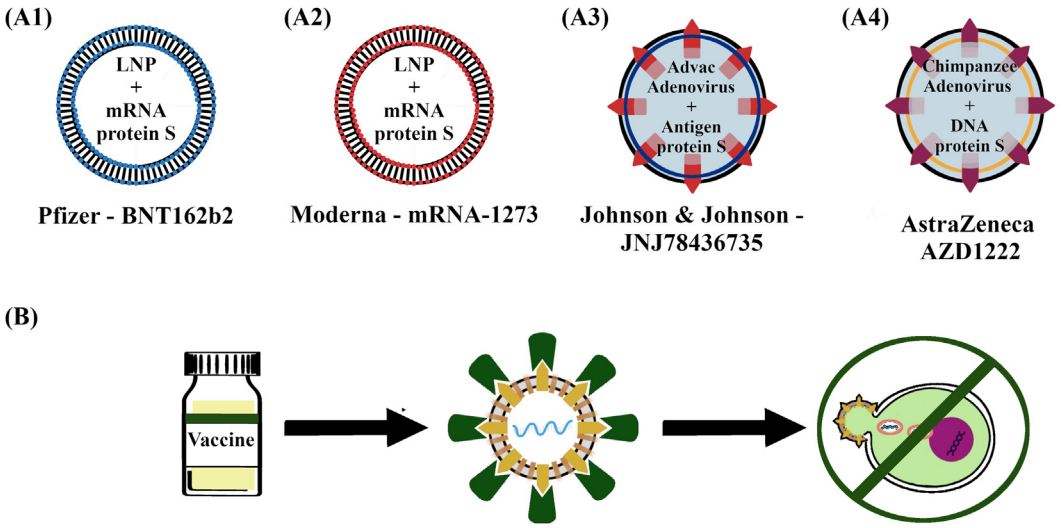

FIGURE 25.4 Severe acute respiratory syndrome-Coronavirus-2vaccines. (A1) Pfizer vaccine BNT162b2: nucleoside messenger RNA platform encoding antigen for spike glycoprotein S, inside a lipid nanoparticle (LNP). (A2) Moderna vaccine mRNA-1273: nucleoside messenger RNA platform encoding antigen for spike glycoprotein S, inside a lipid nanoparticle (LNP). (A3) Johnson & Johnson vaccine JNJ78436735: genetic material vector encoded for spike protein S loaded in Advac adenovirus. (A4) AstraZeneca vaccine AZD1222: double-stranded DNA with prefusion conformation for spike protein S loaded in a Chimpanzee Adenovirus. (B) Vaccine mechanism of action: Vaccine administration in host leads to the production of antibodies and T lymphocytes that would bind with the spike glycoprotein S, preventing the coupling of the SARS-CoV-2 virus with the angiotensin-converting enzyme 2 receptors of the host cells.

Without LNPs, mRNA will degrade in the human body and will trigger an unwanted immune response (McCoy, 2021). To ensure that the genetic materials reach the target cells, a mixture of lipids known as LNPs is used (McCoy, 2021). The LNPs capsule consists of four lipids: (Jackson et al., 2020) cationic lipid, PEGylated lipid, distearoylphosphatidylcholine (DSPC), and cholesterol (McCoy, 2021). In the capsule, the lipids are in a specific ratio with the mRNA (Jackson et al., 2020). The cationic lipid is ionizable and the most important out of the four lipids because it encapsulates the negatively charged mRNA (McCoy, 2021). Additionally, PEGylated lipid assists in controlling the size and life of the particles (McCoy, 2021). Furthermore, DSPC (a phospholipid) and cholesterol contribute to the LNP structure (McCoy, 2021).

25.4.2.2 Moderna—mRNA 1273 vaccine

Moderna—mRNA1273 was the second vaccine to receive a EUA for the prevention of SARS-CoV-2. It is a double-dose vaccine administered one month apart as an IM injection for individuals who are over the age of 18 (Moderna, 2021b). A EUA request was submitted to the FDA on November 30, 2020. This was when the Advisory Committee scheduled a meeting for December 17, 2020. FDA granted the EUA on December 11, 2020 (The United States Food & Drug Administration, 2021b).

Just like Pfizer—BioNTech, Moderna—mRNA-1273 is also a nucleoside-modified mRNA vaccine that is formulated in LNP (Jackson et al., 2020). It encodes for stabilized SARS-CoV-2 spike glycoprotein S in a prefusion conformation (Jackson et al., 2020). This includes a transmembrane anchor and the S1-S2 cleavage site, which in combination is known as the S-2P antigen (Fig. 25.4A2) (Jackson et al., 2020). The stabilization is achieved with two proline substitutions at 986 and 987 positions of the amino acid, which is located on the top of the S2 subunit (Jackson et al., 2020). These enable the modified mRNA delivery into the host cells, which then expresses the SARS-CoV-2 spike antigen (Jackson et al., 2020), causing an immune response that provides protection against COVID-19 (Fig. 25.4B) (Moderna, 2021a).

25.4.2.3 Johnson & Johnson Janssen—JNJ7843673 vaccine

Johnson & Johnson Janssen COVID-19 vaccine is also known as the JNJ7843673 vaccine (Janssen Biotech Inc, 2021a, 2021b). It was the third vaccine to receive a EUA for the prevention of COVID-19. It is a single-dose replication-incompetent human adenovirus type 26 (Ad26) vector vaccine (Janssen Biotech Inc, 2021a; Shimabukuro and Oliver 2021) that can be administered as an IM injection for individuals who are over the age of 18 (Shimabukuro and Oliver 2021). A EUA request was submitted to the FDA on February 4, 2021, and the Advisory Committee meeting was scheduled on February 26, 2021 (Janssen Biotech Inc, 2021a). EUA was granted on February 27, 2021 (Janssen Biotech Inc, 2021a).

The Janssen Advac technology uses adenovirus as vectors and encodes for a stabilized variant in the prefusion conformation of SARS-CoV-2 spike glycoprotein S (Fig. 25.4A3) (Janssen Biotech Inc, 2021b). Adenoviruses are a group of double-stranded, linear DNA viruses (Custers et al., 2020) that cause diseases such as the common cold (Janssen Biotech Inc, 2021b). Janssen has genetically modified the Ad26 viral vector to ensure that the vector is unable to replicate or cause ailments in the host (Janssen Biotech Inc, 2021b). Ad26 viral vector will carry the antigen's genetic code to mimic the SARS-CoV-2 viral pathogen (thus giving the Janssen vaccine the name of Ad26.CoV2.S) (Janssen Biotech Inc, 2021b). When the antigen is encountered by the host cell, an immune response is mediated by the fusion of the antigen to the host cells, producing immune cells (cell-mediated immunity) and antibodies (humoral immunity) (Janssen Biotech Inc, 2021b). After vaccination, when the person encounters SARS-CoV-2, the body will produce immune cells and antibodies to prevent severe disease (Fig. 25.4B) (Janssen Biotech Inc, 2021b).

25.4.2.4 AstraZeneca—AZD1222

AstraZeneca—AZD1222 is awaiting EUA application in the United States but is already approved in the United Kingdom (Shimabukuro and Oliver 2021). It is a double-dose vaccine administered with an interdose interval of 4–12 weeks (Voysey et al., 2021) as an IM injection for individuals who are over the age of 18 (Villafana, 2021; Voysey et al., 2021). On August 31, 2020, AstraZeneca—AZD1222 began Phase III trials in the United States (AstraZeneca, 2021a, 2021b; Villafana, 2021). On September 8, 2020, the clinical trials were temporarily paused (Villafana, 2021). On December 29, 2020, the United Kingdom was granted a EUA for AstraZeneca (Medicines & Healthcare Products Regulatory Agency, 2021). As of January 25, 2021, AZD1222 had received EUA or full approval in 18 countries (Villafana, 2021).

AstraZeneca was developed by the University of Oxford and Vaccitech (AstraZeneca, 2021a, 2021b). Just like the Janssen COVID-19 vaccine, AstraZeneca utilizes a chimpanzee

adenovirus [ChAdOx1] vector (Fig. 25.4A4) (Medicines & Healthcare Products Regulatory Agency, 2021; Villafana, 2021; Voysey et al., 2021). The active substance on this monovalent SARS-CoV-2 vaccine is a single recombinant, replication-deficient ChAdOx1 that codes for the replication-deficient S glycoprotein (ChAdOx1-S [recombinant]) with E1 and E3 deletion (Medicines & Healthcare Products Regulatory Agency, 2021). Postvaccination, the ChAdOx1-S (recombinant) enters the host cell and produces SARS-CoV-2 glycoproteins, thus inducing cell-mediated and humoral immunity (Medicines & Healthcare Products Regulatory Agency, 2021). With future infections, the immune system can recognize and prevent SARS-CoV-2 infection (Medicines & Healthcare Products Regulatory Agency, 2021).

25.4.3 Vaccine adverse events

Currently, in the United States there have been no reports of adverse events for Pfizer–BioNTech COVID-19 Vaccine after 97.9 million doses had been administered. On the other hand, Moderna- mRNA 1273 has three reports of cerebral venous sinus thrombosis (CVST) after 84.7 million doses were administered. Those individuals had normal platelet counts, 150–450K/mm^3. Their onset of the adverse reaction was 2, 6, and 12 days after receiving the Moderna vaccination. Additionally, Johnson & Johnson and AstraZeneca COVID-19 vaccines both have various reports of adverse events and both contain "replication-incompetent adenoviral vectors" (Shimabukuro and Oliver 2021).

Johnson & Johnson—JNJ7843673 has six reports of adverse events among 6.86 million doses administered (Shimabukuro, 2021; Shimabukuro and Oliver 2021). The reporting rate of 0.87 cases per million doses of vaccinations (Shimabukuro and Oliver 2021) that were administered led the CDC and FDA to issue a statement recommending a pause on the vaccine as a precautionary measure (Centers for Disease Control and Prevention, 2021b). The pause was lifted on April 23, 2021 (The United States Food and Drug Administration, 2021b). The adverse events noted included CVST in combination with thrombocytopenia (Centers for Disease Control and Prevention, 2021b). CVST is a type of blood clot and thrombocytopenia. Thrombocytopenia is defined as a low blood platelet count that is less than 150 K/mm^3 (Centers for Disease Control and Prevention, 2021b; Shimabukuro, 2021; Shimabukuro and Oliver 2021). These side effects were seen in white females between the ages of 18–48 with a mean age of 33 years (Shimabukuro, 2021; Shimabukuro and Oliver 2021). Their symptom onset was between 6 and 13 days with the median onset being 8 days (Shimabukuro, 2021; Shimabukuro and Oliver 2021). None of the women were postpartum or pregnant and 1 of them was using estrogen/progesterone. Additionally, none of them were diagnosed with coagulation disorders (Shimabukuro, 2021; Shimabukuro and Oliver 2021). A few of the underlying health conditions include obesity, hypertension, asthma, and hypothyroidism (Shimabukuro, 2021; Shimabukuro and Oliver 2021). Furthermore, it is important to note that thrombosis usually does not occur in combination with low platelet count and these atypical cases are consistent with the adverse reactions after the AstraZeneca COVID-19 vaccine (Shimabukuro and Oliver 2021).

On April 7, 2021, the European Medicines Agency's (EMA) safety committee reported a strong association along with a probable causal link between the AstraZeneca COVID-19 vaccine and a rare blood clotting disorder (European Medicines Agency, 2021). These

include CVST, splanchnic vein thrombosis, and arterial thrombosis in combination with thrombocytopenia in women under the age of 60 (European Medicines Agency, 2021; Shimabukuro and oliver 2021). In the European Union alone there have been 62 cases of CVST and 24 cases of splanchnic vein thrombosis with thrombocytopenia out of which 18 were fatal (European Medicines Agency, 2021). Most of these were identified in females (European Medicines Agency, 2021; Shimabukuro and oliver 2021).

Although these adverse effects are rare, it is important to treat the reaction appropriately as standard measures for blood clots (Centers for Disease Control and Prevention, 2021b). It is important to note that if anyone has developed symptoms such as severe headache, dyspnea, abdominal or leg pain within 3 weeks after receiving the Johnson & Johnson single-dose vaccination, they must seek medical attention (Centers for Disease Control and Prevention, 2021b). Also, if anyone had received the AstraZeneca COVID-19 vaccine and has experienced dyspnea, chest pain, neurological symptoms, swelling in their legs, or under the skin blood spots beyond the injection administration site, they should seek medical attention (European Medicines Agency, 2021).

25.5 Severe acute respiratory syndrome-Coronavirus-2 variants

SARS-CoV-2 currently has several variants. They can cause severe disease, increase transmissibility, hamper the effectiveness of treatments, vaccines, and lead to diagnostic failures (Centers for Disease Control and Prevention, 2021e). As of July 2021, the variants of concern being monitored by the United States are Alpha, Beta, Delta, and Gamma. Other variants include B.1.427, and B.1.429 (Centers for Disease Control and Prevention, 2021e).

Variants can impact the efficacy of vaccines, therefore to combat the situation and create optimal prevention and treatment mechanisms, it is important to identify and explore the dynamics of the SARS-CoV-2 variants (Dey et al., 2021). In silico analysis enables various sequences of different SARS-CoV-2 proteins to be identified (Dey et al., 2021). Various mutations were noted in the spike glycoprotein S (S1 subunit and S2 subunit). S1 subunit includes the receptor-binding domain (RBD) (Huang et al., 2020). One of the mutational hotspots was identified at site 614. Additional mutations were seen at sites 300 and 500 (Dey et al., 2021). A further study of this mutation will be important for the development of the COVID-19 vaccine (Isabel et al., 2020).

25.5.1 Alpha variant—B.1.1.7 lineage

Alpha variant also known as B.1.1.7 lineage, 20I/501Y.V1, Kent variant (Wise, 2021), or N501Y (Centers for Disease Control and Prevention, 2021f; Erol, 2021), was first detected in the United Kingdom in September 2020 (Centers for Disease Control and Prevention, 2021f). The identified attributes include increased transmission, increased severity of hospitalization, and increased CFRs. N501Y carries 14 mutations at RBD of the spike protein (Erol, 2021). At site 501 of the spike protein, where asparagine is replaced with tyrosine, there is an increased affinity for the ACE2 receptor, which thereby causes increased transmission of the virus (Fig. 25.5) (Erol, 2021; Xie et al., 2021).

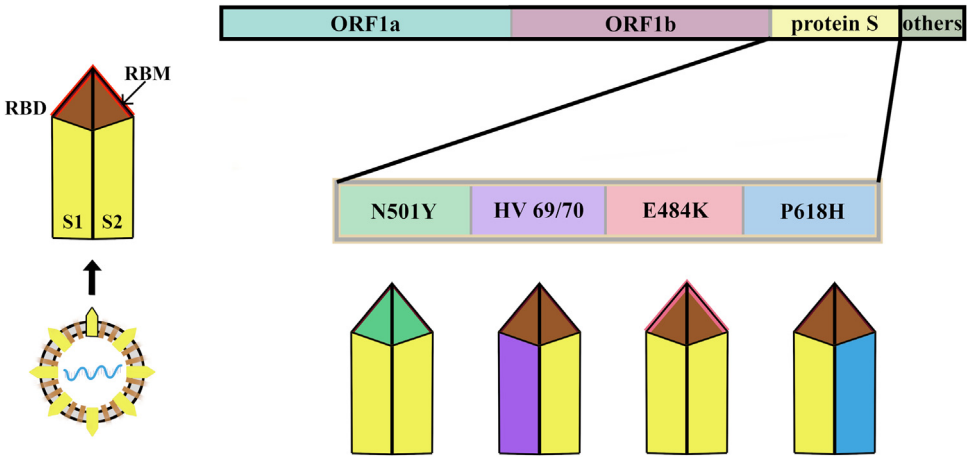

FIGURE 25.5 Spike protein S gene mutations. For the purpose of this chapter, SARS-CoV-2 genome is divided into: ORF1a, ORF1b, spike glycoprotein S, and other SARS-CoV-2 proteins (Tan et al., 2005). Mutations of the spike glycoprotein's (S) genetic code are responsible for the SARS-CoV-2 strand variations. N501Y (green) mutation of the receptor-binding domain. HV69/70 (purple) mutation of the S1 subunit (S1). E484K (pink) mutation of the receptor-binding motif. P618H (blue) mutation of the S2 subunit (S2).

P681H mutation has the amino acid in the 681st position of the spike protein S substituted from Proline to Histidine (SignalChem Biotech Inc, 2021). This leads to a conformation change of the S2 subunit (Fig. 25.5) (SignalChem Biotech Inc, 2021). The adjacent proximity of the subunits to the furin cleavage site amplifies viral fusion by stimulating TMPRSS-2 hydrolysis (SignalChem Biotech Inc, 2021) and promotes viral entry into lung and primary epithelial cells leading to increased transmission of SARS-CoV-2 (Frampton et al., 2021).

In January 2020, D614G was the spike protein predominant from B.1.1.7 lineage and was spreading worldwide (Isabel et al., 2020). It was one of the first mutations of SARS-CoV-2 and was initially identified in Thailand (Dey et al., 2021). Being one of the most studied genomes, this mutation of spike glycoprotein S has polar aspartic acid replaced with nonpolar glycine on the 614th position (Dey et al., 2021). This mutation also led to a change in between the sites 615 and 616 as the development of Elastase 2 or Neutrophil Elastase protease site was noted (Dey et al., 2021).

HV69/70 mutation is the loss of two amino acids, histidine on position 69 and valine on position 70, which are located in the N-terminal domain of the S1 subunit (Kemp et al., 2021). This deletion of these specific amino acids changes the conformation of the S subunits (Fig. 25.5) (Frampton et al., 2021; Kemp et al., 2021). This can interfere with antibody binding (Kemp et al., 2021) potentially impacting the efficacy of the vaccine and also leading to increased transmission in the community (Frampton et al., 2021; Kemp et al., 2021).

25.5.2 Beta variant—B.1.351 lineage

Beta variant also known as B.1.351 lineage or 20H/501.V2 was first detected in South Africa (Centers for Disease Control and Prevention, 2021e). This variant includes N501Y

(Centers for Disease Control and Prevention, 2021e), which was discussed earlier in the chapter. The E484K mutation, first identified in the South African variant and now also identified as the UK variant, is a mutation of the spike protein which changes the receptor-binding motif (Fig. 25.5) (Lasek-Nesselquist et al., 2021; Wise, 2021). It increases its affinity to the ACE receptors (Wang et al., 2021; Wise, 2021) and assists in monoclonal and neutralizing antibodies immune escape (Lasek-Nesselquist et al., 2021). It is not a new variant, but it occurs in different variants such as Beta and Gamma (Wise, 2021). It is a spike protein mutation impacting the body's immune response, perhaps affecting vaccine efficacy (Wise, 2021). Alpha together with the E484K variant increases the serum antibody necessary to prevent SARS-CoV-2 infection. This combination may have also increased its efficiency to reinfect people with the original viral strain (Wise, 2021), possibly achieved by weakening an immune response (Wise, 2021).

25.5.3 Delta variant—B.1.617.2 lineage

Delta variant is also known as B.1.351 lineage and was first detected in India (Centers for Disease Control and Prevention, 2021e; Cherian et al., 2021). The spike protein mutations in the RBD include P681R, L452R, D614G, T478K, and E484Q (Cherian et al., 2021). Analysis of L452R, E484Q, and T478K mutations have revealed increased ACE2 binding. Additionally, L452R and E484Q mutations may diminish the binding of certain monoclonal antibodies and may impact neutralization potential (Cherian et al., 2021). On the other hand, P681R analysis has identified increased rates of S1−S2 cleavage at the furin cleavage site, leading to an increase in transmissibility (Cherian et al., 2021).

25.5.4 Other variants

There are various other variants and mutations of SARS-CoV-2. One of them is Gamma variant also known as B.1.1.28 lineage, P.1 lineage, or 20J/501Y.V3 was first detected in either Japan or Brazil (Centers for Disease Control and Prevention, 2021e). Gamma variant includes E484K, D614G, and N501Y mutations, which was discussed earlier in this chapter (Centers for Disease Control and Prevention, 2021e). Other lineages of concern include SARS-CoV-2 B.1.427 and SARS-CoV-2 B.1.429 (Centers for Disease Control and Prevention, 2021e). They are also referred to as 20C/S:452R (Centers for Disease Control and Prevention, 2021e). These were first detected in California, United States (Centers for Disease Control and Prevention, 2021e). These variants can cause increased transmissibility, reduce neutralization in convalescent and postvaccination sera (Deng et al., 2021). A decrease in susceptibility was noted when these were treated with EUA monoclonal antibody agents (Centers for Disease Control and Prevention, 2021e).

25.6 Current challenges and future perspective

The current pandemic of COVID-19 has been an ongoing global challenge and still requires mastery. SARS-CoV-19 novel mutations have led to the formation of various

variant strains which will continue to emerge. Over time additional in vitro/in vivo studies would help confirm phenotypic changes of these new strains. It would also be extremely important to assess vaccine effectiveness.

Large-scale global COVID-19 vaccine utilization will impact the ongoing phases 2 and 3 randomized placebo-controlled trials (Dal-Ré et al., 2021). Rouw et al. state that vaccine disparities have been identified where high-income countries have purchased more than half of the vaccines. To ensure that we can control the pandemic, it is a critical challenge to ensure global access to these vaccines. A good strategy to create a balance would be for COVID-19 vaccine candidate sponsors to conduct additional clinical countries with limited affordability and availability for these vaccines. Clinical research could focus on "placebo-controlled, double-blind, crossover trials" to study potential vaccine candidates (Rouw et al., 2021). Once adequate information regarding immunogenicity response to COVID-19 vaccines is obtained, future studies would include benefits from noninferiority immunogenicity trials (Rouw et al., 2021).

25.7 Summary

SARS-CoV-2, a novel betacoronavirus, is the causative agent of COVID-19, contagious disease with varying clinical presentations (Pollock & Lancaster, 2020). SARS-CoV-2 is constantly changing and has undergone numerous mutations over a short period of time (Centers for Disease Control and Prevention, 2021f; Lasek-Nesselquist et al., 2021; Xie et al., 2021). Depending on the severity of the disease, treatment may be warranted. Though there are several EUA drugs for its treatment, remdesivir is the only FDA-approved drug for patient care and had advantages amidst the pandemic (The United States Food & Drug Administration, 2021b; The United States Food and Drug Administration, 2020b). Furthermore, Pfizer, Moderna, Johnson & Johnson vaccines are available as prevention measures (The United States Food & Drug Administration, 2021b), which give the public a ray of hope, especially for high-risk patients with comorbidities. Once the ongoing clinical trials end, we will have a better understanding of the side effects of treatment and prevention methods on our health. Currently, various vaccine trials are being conducted in the United States (The United States Food and Drug Administration, 2020a). Several computational approaches offer alternative screening methods and provide network-based pharmacology for identifying repurposed drugs, combination therapies, or vaccines for the treatment and prevention of COVID-19 (Dhama et al., 2020; Kissoon, 2019; Mishra et al., 2020; Sanami et al., 2021; Wu et al., 2020). Therefore ongoing research on SARS-CoV-2, its treatment, prevention measures, variants, and the overall health and long-term effects on mankind, will guide us into a brighter future and help us reach our ultimate goal of eradicating SARS-CoV-2.

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