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Chapter 5

Pathogenesis and mutagenesis of SARS-CoV-2: cellular attachment, entry, and infection

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5.1 Introduction

Coronavirus disease 2019 (COVID-19) is a highly contagious disease characterized by higher leukocyte counts, acute respiratory distress, and increased levels of plasma proinflammatory cytokines. In order to understand the pathogenic mechanism of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of COVID-19, it is important to consider the general replication cycle of a virus. Although some differences could exist in the replication cycle of some species, but generally, viruses are obligate intracellular pathogens that would require the host cell machinery to replicate, thereby causing infection. There are nine basic steps in viral replication (Fig. 5.1). They are listed as follows:

1. Attachment of the virus to the host cell receptor.
2. Fusion of the virus with cellular membranes and consequent release of the viral RNA into the cytoplasm.
3. Reverse transcription to make viral DNA in the cytoplasm.
4. Integration of the viral DNA into the host DNA in the nucleus.
5. Posttranscription in the nucleus to make viral mRNA outside the nucleus.
6. Translation of the coded information of the mRNA into viral protein.
7. Assembling and packaging of viral protein and genetic materials.
8. Budding out or lysis of the viral material at the membrane level.
9. Release of the virus and subsequent reinfection of other organs with similar receptors.

FIGURE 5.1 The general replication cycle of RNA viruses.
However, SARS-CoV-2 pathogenesis first step involves the binding of SARS-CoV-2 to the lungs epithelial angiotensin-converting enzyme 2 (ACE-2) receptor, which is inhaled from the droplets of an infected person or through other means [1]. Recently, a study showed that SARS-CoV-2 nucleic acid can be detected in the feces and urine of patients with COVID-19, an indication that SARS-CoV-2 may also be transmitted from the digestive tract through the fecal–oral route [2]. However, it is believed that the lung epithelial cells are the primary target of the virus [3,4].

The viral replication is presumed to start from the epithelium of the upper respiratory tract, and as the disease progresses, further multiplication occurs at the lower respiratory tract and in the gastrointestinal mucosa leading to mild level of viremia. At this point, the common symptoms of infections are fever and cough, though a few persons might be asymptomatic [5]. As the virus replicates, it moves to other organs that express ACE-2 such as the heart, kidneys, and gastrointestinal tract [6,7]. ACE-2 has been shown to be a co-receptor for viral entry for SARS-CoV-2 with a developing evidence that it has an extended role in the pathogenesis of COVID-19 [1]. The mechanism of the damage caused by SARS-CoV-2 infection has not been completely elucidated. However, the SARS model consists of three phases, namely, viral replication, immune hyperactivity, and pulmonary destruction [8].

The recent spike again in cases of COVID-19 after about a month of downturn, which led to ease in worldwide lockdown supports an earlier observation of mutation in SARS-CoV-2 [9]. One study found that infected samples of genomes collected from more than 7500 patients worldwide have identified mutations in the virus [10]. This development is scary because mutation could either make the virus more virulent (resistant to drugs) or less contagious. A comprehensive review on this and the pathogenic mechanisms of SARS-CoV-2 were discussed.

5.2 Pathogenesis of SARS-CoV-2

SARS-CoV-2 is the causative agent of COVID-19, a novel fatal disease with great global and public health concerns. The symptoms associated with COVID-19 patients include elevated levels of leukocytes, abnormal respiratory functions, and increased levels of proinflammatory cytokines in the plasma [11]. The leading pathogenesis of SARS-CoV-2 infection is known for targeting the respiratory system with pneumonia, RNAemia, in addition to the incidence of ground-glass opacities, and acute cardiac injury [12]. High blood levels of cytokines and chemokines were recorded in COVID-19 patients which included interleukin (IL) 1 beta (IL-1β), IL-1RA, IL-7, IL-8, IL-9, IL-10, basic FGF2, GCSF, GMCSF, IFNγ, IP10, MCP1, MIP1Aα, MIP1β, PDGFB, TNF-α, and VEGFA [12]. However, some of the severe cases that were admitted into the intensive care unit of hospitals had elevated levels of proinflammatory cytokines including IL-2, IL-7, IL-10, GCSF, IP10, MCP1, MIP1α, and TNF-α thought to promote the severity of the disease [12].
SARS-CoV-2 utilizes the ribosomes of the host cell to translate mRNA into polyproteins. These polyproteins are the structural framework of the viral molecule [13,14]. The synthesized polyproteins in turn will utilize the host RNA-dependent RNA polymerase to replicate itself, increasing the viral load within the host cell. The polyproteins use the host enzymes such as proteinases, which will degrade the polyproteins to create the individual spike proteins, E protein, nucleocapsid (Figs. 5.1 and 5.2) [15]. These structural components, together with the replicated ssRNA, give rise to a mature SARS-CoV-2 that would bud off the type II pneumocyte, back into the alveolus [16]. Type II pneumocytes are damaged in the process of SARS-CoV-2 budding off of the host cell. The destruction of type II pneumocytes will release specific inflammatory mediators which will stimulate macrophages. Stimulated macrophages trigger the release of certain cytokines such as IL-1, IL-6, and TNF-α [17]. In cases of acute inflammation, IL-1 and IL-6 are the major cytokines released leading to fever [11]. Furthermore, IL-1, IL-6, and TNF-α enter the blood circulation, causing the dilation of smooth muscles along with contraction of blood vessel endothelial cells resulting to increased capillary permeability. Increased capillary permeability causes plasma from the bloodstream to leak into the interstitial spaces, leading to alveolar edema.

Damage to type II pneumocytes within the alveoli occasioned by SARS-CoV-2 causes a decrease in the production of surfactants since the primary function of type II pneumocyte is the production of surfactant. Decreased surfactant within the alveolus will cause alveolar collapse due to increased surface tension as well as alveolar edema [15]. This collapse of the alveoli will impair gaseous exchange causing refractory hypoxemia and increased work associated with breathing in an attempt to breathe in as much air as possible to reopen the collapsed alveolar and interstitial edema. This is the mechanism
that causes acute respiratory distress syndrome (ARDS) in SARS-CoV-2 infection. Another feared complication alongside ARDS has been disseminated intravascular coagulation which is a manifestation of coagulation failure and an intermediate link in multiorgan failure [16].

Inflammatory mediators released by the destruction of type II pneumocytes will increase the influx of neutrophils into the alveolus, releasing reactive oxygen species and proteases in which neutrophils will attempt to destroy the virus. In an attempt to destroy the invading virus, neutrophils will cause mass destruction of all alveolar cells: type I and type II pneumocytes. Damaged type I pneumocytes will cause impaired gaseous exchange as they play integral role in gas exchange. Type II pneumocytes are also destroyed by the neutrophils causing increased surface tension, leading to alveolar collapse. Damaged cells will collect at the basement of the alveolar membrane and into the center of the alveolus creating a collection of fluid, along with cellular debris comprising type I pneumocytes, type II pneumocytes, neutrophils, and macrophages, leading to consolidation. The consolidation will also hinder gas exchange, causing hypoxemia [11]. On the other hand, hypoxemia will trigger the release of peripheral chemoreceptors causing the sympathetic nervous system to increase respiration rate and heart rate to compensate for a reduced partial pressure of oxygen. Consolidation will lead to cough induction and may also cause the presentation of dyspnea due to decreased gas exchange. During the infection, inflammation of the lungs can advance to systemic inflammatory response syndrome as cytokines circulate in the vascular system, causing increased capillary permeability [18]. Capillary permeability uses the same mechanism as in the lungs to deposit plasma within tissue spaces, decreasing the blood volume. The vasodilation of blood vessels will decrease the total peripheral resistance, causing hypotension. The hypotension will cause decreased perfusion, leading to multisystem organ failure [17]. Also, decreased perfusion to the kidneys causes an increase in blood urea nitrogen and creatinine levels. Kidneys are unable to filter the blood urea nitrogen and creatinine from the blood, causing acute renal injury. Consistently, this causes IL-1 and IL-6 to circulate the vascular system and enter the central nervous system, targeting the hypothalamus, known for its role in maintaining body temperature. The elevated levels of IL-1 and IL-6 within the hypothalamus will cause it to release prostaglandins that will help reset the core body temperature to be higher than normal resulting in fever. The role of IL-1 and IL-6 in increasing the core body temperature is important as the most common initial symptom of COVID-19 [14].

5.2.1 SARS-CoV-2 cellular attachment

SARS-CoV-2 attaches itself to the host through virus—host interaction occasioned by the attachment of the viral structural protein with a specific receptor on the host cell. Once the virus attaches to the host via receptors,
it enters the host cell through the process of endocytosis pathway triggered by the spike protein [19] followed by incorporation of the virus, release of the viral genome, transcription and translation, and assembly of the new viral particles as well as the final release of the virus from the host cell [20]. As previously discussed, once the virus has access to the mucosal membranes of the host, it travels through the respiratory tract and enters the lung alveoli. In the alveoli, the SARS-CoV-2 binds specifically to the ACE-2 receptor, present on the type II pneumocyte. There is a spike glycoprotein present on the outer membrane of the SARS-CoV-2 which has a high affinity for the ACE-2 receptor [21]. Once the viral particle binds to the ACE-2 receptor, the virus is endocytosed into the cytoplasm of the type II pneumocyte where the lysosomal enzymes of the host cell will break down the lipid bilayer of the virus.

5.2.2 SARS-CoV-2 entry

Human ACE-2 is a functional receptor for the attachment of SARS-CoV-2 for cell entry, similar to SARS-CoV (Fig. 5.4) [22–24]. ACE-2 is a type I membrane protein which is expressed in the lung, heart, kidney, and intestine but mainly associated with cardiovascular diseases [25]. The ACE-2 protein is made up of an N-terminal peptidase domain and a C terminal collectrin-like domain that ends with a single transmembrane helix and a ~40-residue intracellular segment [25]. In addition to cleavage of angiotensin I to produce angiotensin (1–9), ACE-2 also provides a direct binding site for the spike proteins of coronaviruses [25]. The spike protein of coronaviruses exists in a metastable prefusion conformation that goes through a structural rearrangement to fuse the viral membrane with the host cell membrane [26]. The above process is initiated by the S1 subunit and a host—cell receptor binding, which destabilizes the prefusion trimer, resulting in the S1 subunit shedding and the S2 subunit transition to a highly stable postfusion conformation [26]. To engage a host—cell receptor (Fig. 5.3), the receptor-binding domain of

FIGURE 5.3 Spike receptor binding mechanism of SARS-CoV-2.
S1 undergoes hinge-like conformational movements that transiently hide or expose the determinants of receptor binding [27]. The biophysical and structural evidence of the receptor-binding domain of its spike protein in contact with ACE-2 suggests that SARS-CoV-2 the protein likely binds to human ACE-2 with 10–20 fold higher affinity than SARS-CoV [28]. According to Zhou et al. [24], another structural evidence suggests that the ACE-2–B0AT1 complex can bind two spike proteins simultaneously.

5.2.3 SARS-CoV-2 infection

Severely infected COVID-19 patients are currently the major source of transmission. Although some infected persons are asymptomatic, they could still spread the disease [29]. Additionally, it was found the samples taken from patients after recovery continuously show a positive RT-PCR test (Fig. 5.5) [30], which is something that has never been seen in the history of human infectious diseases. This trend may pose serious challenges for disease prevention and control. As reported by Epidemiology Working Group [31], COVID-19 has been considered as a type of self-limiting infectious disease, and most cases with mild symptoms can recover in 1–2 weeks. SARS-CoV-2 infection can cause five different outcomes: asymptomatically infected persons (1.2%); mild to medium cases (80.9%); severe cases (13.8%); critical case (4.7%); and death (2.3% in all reported cases). In a study conducted by Lu et al. [31], the proportion of asymptomatic infection in children under 10 years old was as high as 15.8%.
FIGURE 5.5 Coronavirus disease 2019 test through RT-PCR.
5.3 Mutagenesis of SARS-CoV-2

The complete knowledge of the pathogenicity of SARS-CoV-2 might take years to be fully elucidated. This includes the evolutionary mechanism, its emergence, and mutation rate. Characterizing the mutation rate will open doors to understanding the mechanism of action, immune invasion, and possible resistance of this deadly virus to drugs or vaccines. It has been reported that there is slow mutation rate in genome of SARS-CoV-2 which can modulate the viral transmission in different parts of the globe [9]. This mutation rate in turn depends upon the replicating enzymes, host enzymes, abrupt nucleic acid damage due to different physiochemical processes, and recombination [32,33]. Usually, the mutation rate in RNA viruses is on a higher side than that of the host which could increase the virulence and adaptability of the pathogen [34]. Recently, 13 variation sites in SARS-CoV-2 have been characterized, ORF1ab, S, ORF3a, ORF8, and N regions, among which positions 28144 in ORF8 and 8782 in ORF1a had showed a mutation rate of 30.53% and 29.47%, respectively [21]. This mutation rate might affect the disease severity and transmission rate. There have been eight novel recurrent mutations characterized of SARS-CoV-2, located at positions 1397, 2891, 14408, 17746, 17857, 18060, 23403, and 28881. Mutations in 2891, 3036, 14408, 23403, and 28881 positions are predominantly observed in Europe, whereas others located at positions 17746, 17857, and 18060 were found in North America; other silent mutation was detected in RNA-dependent RNA polymerase (RdRp) gene in England (UK) on February 9, 2020 while a different mutation in RdRp changing its amino acid composition emerged on February 20, 2020 in Italy [9]. Another study reported that SARS-CoV-2 is rapidly evolving across different regions across the globe and new mutations are evolving [35]. Lately, it has been found that among 103 sequenced strains of SARS-CoV-2, there have been 149 sites of mutations in genome of the virus [36]. These mutations showed differences in geographical distribution, transmission ability, and severity of the disease, causing more difficulty in drug and vaccine development. Therefore, tracking such mutations is very pivotal in order to find a strong cure against SARS-CoV-2 [37].

Koyama et al. [35] conducted a study to examine the rate of variation in the genome of SARS-CoV-2. Their study was conducted between February 1 and May 1, 2020. They downloaded 10,022 SARS-CoV-2 infected genomes from 68 countries of four databases and compared the genomes using the reference genome, NC_045512 with EMBOSS needle. Clad studies were conducted using Bayesian evolutionary analysis by sampling trees, version 2.5. In their findings, authors identified six major clads (basal, D614G, L84S, L3606F, D448del, and G392D) (Table 5.1), out of which D614G (a G-to-A base change at position 23403 in the Wuhan reference strain) was found to be the most
| Clade/sublevel 1/sublevel 2              | Date                | First observation of strain                  | Country       | No. of samples |
|----------------------------------------|---------------------|---------------------------------------------|---------------|---------------|
| Basal<sup>a</sup>                      | December 2019       | MN90894                                     | China         | 670           |
| D614G//                                | January 24, 2020    | EPI_ISL_422425                              | China         | 1889          |
| D614G/Q57H/                            | February 26, 2020   | EPI_ISL_418219                              | France        | 469           |
| D614G/Q57H/T265I                       | February 21, 2020   | EPI_ISL_418218                              | France        | 2391          |
| D614G/203_204delinsKR/                 | February 25, 2020   | EPI_ISL_412912                              | Germany       | 1330          |
| D614G/203_204delinsKR/T175M            | March 1, 2020       | EPI_ISL_413647 and EPI_ISL_417688          | Portugal and Iceland | 215 |
|                                        |                     | Total                                       |               | 6294          |
| L84S/                                  | December 30, 2019   | MT291826                                    | China         | 525           |
| L84S/P5828L                            | February 20, 2020   | EPI_ISL_413456                              | The United States | 1137 |
|                                        |                     | Total                                       |               | 1662          |
| L3606F/                                | January 18, 2020    | EPI_ISL_408481                              | China         | 182           |
| L3606F/V378I                           | January 18, 2020    | EPI_ISL_412981                              | China         | 127           |
| L3606F/G251V                           | January 29, 2020    | EPI_ISL_412974                              | Italy         | 419           |
| L3606F/G251V/P7655                     | February 20, 2020   | EPI_ISL_415128                              | Brazil        | 260           |
|                                        |                     | Total                                       |               | 988           |
| D448del/                               | February 8, 2020    | EPI_ISL_410486                              | France        | 248           |
| G392D/                                 | February 25, 2020   | EPI_ISL_414497                              | Germany       | 160           |

<sup>del</sup>, deletion; <sup>delins</sup>, deletion–insertion.

<sup>a</sup>The reference genome (NC_045512) used belongs to the basal clade.

Reproduced from Koyama T, Platt D, Parida L. Variant analysis of SARS-CoV-2 genomes. Bull World Health Organ 2020;98(7):495–504. https://doi.org/10.2471/BLT.20.253591. Licence: Creative Commons BY 3.0 IGO.
re-occurring clad. Authors also identified 14 subclads (Table 5.1). Variants was also found in the open reading frame 1ab of SARS-CoV-2 genomes (Table 5.2). In order to examine variants in nucleotides, the nucleotides within the coding areas were converted to amino acid residues. The findings suggest that the C>T mutation was the most common with 1670 distinct variants. In summary, the most common variants are the synonymous 3037C>T in 6334 samples (Table 5.3), P4715L in the open reading frame 1ab of 6319 samples (Table 5.3), and D614G in the spike protein in 6294 samples (Table 5.1).

According to BBC News of July 19, 2020, her health reporter, Rachel Schraer, was quoted as saying that the most notable mutation—named D614G (situated within the protein making up the virus’s “spike” it uses to break into the cells)—is now seen in as many as 97% of samples around the world. She further stated that the D614G variant is so dominant that it is now the pandemic [38]. This correlates with the findings of Koyama et al. [35] and Korber et al. [39].

5.4 Future perspectives

There are serious concerns about rising cases and more people getting infected by the virus that can lead to an increase in the fatality rate. On the other hand, the process of treatment gets disrupted as there is a spike in number of cases. It becomes more interesting for those who are immunocompromised. At present, only a little is known about this virus, there are large number of questions which remain unanswered. The most important among which is response of host toward the virus involving cellular and humoral response. All these queries must be addressed which would help in successful vaccine development. As virus has been evolved only some months before, less data and information is available on the mechanism of pathogenesis and mutagenesis.

As the cases of COVID-19 keep soaring, authors support social distancing, wearing of face mask, and the observation of all sanitary measures such as frequent hand washing. Authors also recommend the consumption of food rich in micronutrients such as vitamins and minerals for immune system boosting. Chapters 16, 18–21 of this book presents the various food sources of these nutrients. Also, the Editor of this book and his collaborating team has worked on many related projects especially as it regards drug discovery from natural products [40–51].

As SARS-CoV-2 mutates, it is wise to sustain continuous, evidence-based analysis of evolutionary changes so that public health measures can be adjusted in response to substantive changes in the infectivity or severity of COVID-19 [52].
| Final protein | Missense mutation | Synonymous mutation | Noncoding region | In-frame | Frameshift deletion | Stop gained | Total |
|---------------|------------------|---------------------|------------------|---------|--------------------|-------------|-------|
|               |                  |                     | Mutation | Deletion | Insertion          | Deletion | Insertion |            |         |
| NSP1          | 64               | 45                  | 0         | 0        | 0                  | 13        | 0        | 1           | 0       | 123     |
| NSP2          | 237              | 130                 | 0         | 0        | 0                  | 5         | 0        | 0           | 0       | 372     |
| NSP3          | 547              | 349                 | 0         | 0        | 0                  | 16        | 0        | 2           | 3       | 917     |
| NSP4          | 116              | 113                 | 0         | 0        | 0                  | 1         | 0        | 0           | 1       | 232     |
| 3CLPro        | 67               | 54                  | 0         | 0        | 0                  | 0         | 0        | 0           | 0       | 121     |
| NSP6          | 82               | 67                  | 0         | 0        | 0                  | 4         | 1        | 2           | 0       | 156     |
| NSP7          | 27               | 21                  | 0         | 0        | 0                  | 0         | 0        | 0           | 0       | 48      |
| NSP8          | 60               | 25                  | 0         | 0        | 0                  | 1         | 0        | 0           | 1       | 87      |
| NSP9          | 29               | 22                  | 0         | 0        | 0                  | 0         | 0        | 0           | 1       | 52      |
| NSP10         | 25               | 25                  | 0         | 0        | 0                  | 0         | 0        | 0           | 2       | 52      |
| RdRp          | 194              | 157                 | 0         | 0        | 0                  | 2         | 0        | 1           | 3       | 357     |
| Helicase      | 148              | 101                 | 0         | 0        | 0                  | 0         | 0        | 0           | 0       | 249     |
| ExoN          | 141              | 118                 | 0         | 0        | 0                  | 11        | 0        | 1           | 2       | 273     |
| EndoRNase     | 92               | 67                  | 0         | 0        | 0                  | 3         | 0        | 0           | 0       | 162     |
| OMT           | 76               | 50                  | 0         | 0        | 0                  | 1         | 1        | 0           | 0       | 128     |
| Total         | 1905             | 1344                | 0         | 0        | 0                  | 57        | 2        | 7           | 13      | 3329    |

Note: Koyama et al. [36] compared 10,022 genomes to the NC_045512 genome sequence. Reproduced from Koyama T, Platt D, Parida L. Variant analysis of SARS-CoV-2 genomes. Bull World Health Organ 2020;98(7):495–504. https://doi.org/10.2471/BLT.20.253591. Licence: Creative Commons BY 3.0 IGO.
| Genomic change | Type of mutation | Gene/protein          | Amino acid change   | No. of samples |
|---------------|------------------|-----------------------|---------------------|---------------|
| 3037C > T     | Synonymous       | ORF1ab/NSP3           | F924F/F106F         | 6334          |
| 14408C > T    | Missense         | ORF1ab/RdRp           | P4715L/P323L        | 6319          |
| 23403A > G    | Missense         | S                     | D614G               | 6294          |
| 241C > T      | Noncoding        | 5’-UTR                | NA                  | 5928          |
| 25563G > T    | Missense         | ORF3a                 | Q57H                | 2893          |
| 1059C > T     | Missense         | ORF1ab/NSP2           | T2651/T851          | 2442          |
| 28144T > C    | Missense         | ORF8                  | L84S                | 1669          |
| 8782C > T     | Synonymous       | ORF1ab/NSP4           | S2839S/S76S         | 1598          |
| 28881_28883delinsAAC | Missense     | N                     | 203_204delinsKR     | 1573          |
| 18060C > T    | Synonymous       | ORF1ab/ExoN           | L5932L/L7L          | 1178          |
| 17858A > G    | Missense         | ORF1ab/helicase       | Y5865C/Y541C        | 1166          |
| 17747C > T    | Missense         | ORF1ab/helicase       | P5828L/P504L        | 1147          |
| 11083G > T    | Missense         | ORF1ab/NSP6           | L3606F/L37F         | 1070          |
| 14805C > T    | Synonymous       | ORF1ab/RdRp           | Y4847Y/Y455Y        | 844           |
| 26144G > T    | Missense         | ORF3a                 | G251V               | 769           |
| 20268A > G    | Synonymous       | ORF1ab/endoRNase      | L6668L/L216L        | 452           |

Continued
| Genomic change | Type of mutation | Gene/protein | Amino acid change | No. of samples |
|----------------|------------------|--------------|-------------------|---------------|
| 17247T > C     | Synonymous       | ORF1ab/helicase | R5661R/R337R      | 325           |
| 2558C > T      | Missense         | ORF1ab/NSP2   | P765S/P585S       | 274           |
| 15324C > T     | Synonymous       | ORF1ab/RdRp   | N5020N/N628N      | 267           |
| 1605_1607del   | In-frame deletion| ORF1ab/NSP2   | D448del/D268del   | 250           |
| 18877C > T     | Synonymous       | ORF1ab/ExoN   | L6205L/L280L      | 234           |
| 2480A > G      | Missense         | ORF1ab/NSP2   | I739V/I559V       | 232           |
| 27046C > T     | Missense         | M             | T175M             | 221           |
| 11916C > T     | Missense         | ORF1ab/NSP7   | S3884L/S25L       | 185           |
| 2416C > T      | Synonymous       | ORF1ab/NSP2   | Y717Y/Y537Y       | 170           |
| 1440G > A      | Missense         | ORF1ab/NSP2   | G392D/G212D       | 164           |
| 27964C > T     | Missense         | ORF8          | S24L              | 164           |
| 36C > T        | Noncoding        | 5′-UTR        | NA                | 163           |
| 2891G > A      | Missense         | ORF1ab/NSP3   | A876T/A58T        | 159           |
| 28854C > T     | Missense         | N             | S194L             | 155           |
| 1397G > A      | Missense         | ORF1ab/NSP2   | V378I/V198I       | 139           |
| 28657C > T     | Synonymous       | N             | D128D             | 139           |
| SNV   | Type         | ORF/NSP | Position | Mutation   |
|-------|--------------|---------|----------|------------|
| 28688T > C | Synonymous   | N       | L139L    | 138        |
| 18998C > T | Missense     | ORF1ab/ExoN | A6245V/A320V | 137       |
| 28311C > T | Missense     | N       | P13L     | 136        |
| 28863C > T | Missense     | N       | S197L    | 136        |
| 9477T > A  | Missense     | ORF1ab/NSP4 | F3071Y/F308Y | 136       |
| 25979G > T | Missense     | ORF3a   | G196V    | 132        |
| 29742G > T | Noncoding    | 3'-UTR  | NA       | 131        |
| 25429G > T | Missense     | ORF3a   | V13L     | 128        |
| 24034C > T | Synonymous   | S       | N824N    | 118        |
| 29870C > A | Noncoding    | 3'-UTR  | NA       | 115        |
| 28077G > C | Missense     | ORF8    | V62L     | 113        |
| 26729T > C | Synonymous   | M       | A69A     | 106        |
| 27_37del  | Noncoding deletion | 5'-UTR  | NA       | 106        |
| 19_24del  | Noncoding deletion | 5'-UTR  | NA       | 105        |
| 514T > C  | Synonymous   | ORF1ab/NSP1 | H83H/H83H | 105       |
| 23731C > T | Synonymous   | S       | T723T    | 102        |
| 3177C > T | Missense     | ORF1ab/NSP3 | P971L/T1198K | 101       |

del, deletion; delins, deletion-insertion; NA, not applicable. 10,022 genomes were compared to the NC_045512 genome sequence. Reproduced from Koyama T, Platt D, Parida L. Variant analysis of SARS-CoV-2 genomes. Bull World Health Organ 2020;98(7):495–504. https://doi.org/10.2471/BLT.20.253591. Licence: Creative Commons BY 3.0 IGO.
5.5 Conclusion

At present, COVID-19 is the biggest challenge faced by people across the globe and remains a top concern. The experience from previous disease outbreaks due to SARS-CoV and MERS-CoV helps in understanding the pathogenesis and spread of this viral infection. Interaction between host and viral particles plays an important role in viral pathogenesis, thus by using receptor analog and inhibitors can prevent this binding. Therefore, controlling this binding can disable the viral entry into host cells and this could play an essential role in combating the spread of COVID-19. Continuous research and sequencing might play a vital role in understanding the mutagenic rate of reported strains throughout the globe, which in turn will help in successful drug and vaccine development.

List of abbreviations

3CLPro  3C-like protease  
ACE-2  Angiotensin-converting enzyme 2  
ARDS  Acute respiratory distress syndrome  
CLD  Collectrin-like domain  
COVID-19  Coronavirus disease 2019  
ExoN  3′-5′ exonuclease  
IL  Interleukin  
M  Membrane glycoprotein  
mRNA  Messenger RNA  
N  Nucleocapsid phosphoprotein  
NSP  Nonstructural protein  
OMT  O-methyltransferase  
ORF  Open reading frame  
RdRp  RNA-dependent RNA polymerase  
S  Spike glycoprotein  
SARS-CoV-2  Severe acute respiratory syndrome coronavirus-2  
SNS  Sympathetic nervous system  
ssRNA  Single-stranded RNA  
TNF-α  Tumor necrosis factor-alpha  
UTR  Untranslated region

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