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Review Article

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2019 Novel Human Coronavirus Sars-Cov-2 And Covid-19: A Brief Review

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Abstract: The emergence of the 2019 novel coronavirus SARS-CoV-2, causing a highly contagious disease COVID-19 poses a tremendous global public health concern. As the disease is quickly developing into a worldwide pandemic, a careful analysis of its origin, evolution, transmission, and cellular mechanism is urgently needed to combat the deadly virus. Successful isolation of the 2019-nCoV has promoted some bioinformatic studies to understand the viral origin and the feature of its infectivity. However, at this stage, much remains unclear about the origin of the novel virus and to be investigated to develop ways to control its spread. The mechanisms associated with the infectiousness of SARS-CoV-2 are not entirely understood. The current knowledge in 2019-nCoV pathogenicity and transmissibility along with several commonly known emerging viruses and information is very much important for better control of the disease. So far, the treatment is only supportive. Any antiviral agent is yet to emerge. The scientific community raced to understand the pathogenesis of the disease for developing treatment options. Extensive research is going on to understand the host response to the pandemic virus to develop the disease therapeutic. To fill the knowledge gap about the human immune response to SARS-CoV- 2 infections that may help in designing the appropriate immune intervention for treatment, diagnosis, and prophylactic/therapeutic vaccines against COVID-19, we present hereby a brief review on the genomic organization, origin, and evolution, transmission and pathogenesis of the novel virus, and clinical spectrum and possible preventive measures against COVID-19 which may be used for future references.

Keywords: CAI • Codon usage bias• GC content • gene expression • RSCU • SARS-CoV-2

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1. INTRODUCTION

Upon a viral outbreak of Pneumonia cases with unknown causes that emerged in Wuhan, China, in December 2019, the epidemic investigation and gene sequencing revealed that a novel coronavirus was the causative agent [1-3]. The virus was initially named as 2019-nCoV. Later, the virus was officially named Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) by the International Committee on Taxonomy of Viruses (ICTV). The disease caused by this virus, named COVID-19 by the World Health Organization, turns out to be a potential threat to global public health. Up-to-date information about COVID-19 is available at https://www.who.int/emergencies /diseases/novel-coronavirus-2019. Several thorough investigations on SARS-CoV-2 and COVID-19 have been done in the past few months to report on the evolutionary reservoir, possible intermediate host, and genomic sequence of SARS-CoV-2 and clinical characteristics COVID-19. The present review attempts to describe the various 2019-Novel Coronavirus features that cause disease COVID-19 and search for a feasible strategy to combat the disease.

2. *TAXONOMY*

Based on molecular characterization, the newly emerged novel virus is a member of the order *Nidovirales*, family *Coronaviridae*, sub-family *Orthocoronavirinae*[4]. Based on the difference in protein sequences, *Ortho corona viridae* is subdivided into four major genera: *Alphacoronavirus, Betacoronavirus*, *Gammacoronavirus*, and *Deltacoronavirus*[5], among which the beta-CoV(*Betacoronavirus*) genera contains most human CoVs (HCoVs) and is subdivided into four lineages (A, B, C, and D). According to the family classification and taxonomy, as developed by the *Coronaviridae* Study Group (CSG), a working group of the ICTV, SARS -CoV-2 belongs to the genus *Betacoronavirus*, subgenus *Sarbecovirus*[6]. CoVs are positive-stranded RNA viruses having a crown-like appearance and cause mostly respiratory and enteric diseases in different animals, including camels, cattle, cats, bats, and humans. SARS-CoV-2 has round or elliptic and often pleomorphic forms. Its diameter is approximately 60–140 nm[7]. It is susceptible to ultraviolet rays and heat like other CoVs[8]. Inactivation of these viruses can be achieved by lipid solvents, including ether (75%), ethanol, chlorine-containing disinfectant, per-oxyacetic acid, and chloroform[9]. The genome sequence of SARS-CoV-2 contains 29891 nucleotides, encoding for 9860 amino acids[10]. From the extensive genetic analysis, it has been observed that the genome of the novel coronavirus had 89% sequence similarity with bat SARS-like-CoVZXC21 and 82% with human SARS-CoV[11]. For this reason, the new virus was called SARS-CoV-2. Although its origins are not entirely understood, in-depth genomic analyses suggest that SARS-CoV-2 may have originated from bats[12]. The mutation in the virus's original strain could have directly triggered virulence towards humans, and it is not sure that any intermediate host exists between bats and humans.

3. *HISTORY*

For thousands of years, CoVs have repeatedly crossed species barriers, and some have emerged as critical human pathogens. Animal CoVs have been known since the late 1930s[13]. Before the first isolation of HCoV-229E strain B814 from humans[14], different CoVs had been isolated in various infected animals e.g. turkey, mouse, cow, pig, cat, and dog[15]. To date, seven human CoVs (HCoVs) have been identified[7]. Among them, HCoV-229E and HCoV-NL63 are alpha-CoVs (*Alphacoronavirus*). The other five belong to beta- CoVs, which include HCoV-OC43(lineage-A), HCoV-HKU1(lineage-A), Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) [lineage-B], Middle East Respiratory Syndrome Coronavirus (MERS-CoV)[lineage-B], and SARS-CoV-2 [lineage-B][15]. HCoV-229E, HCoV-OC43, HCoV-HKU1 and HCoV-NL63 usually cause mild symptoms, like the common cold and/or diarrhea[16]. In contrast, SARS-CoV, MERS-CoV, and the newly emerged SARS-CoV-2 are highly pathogenic and may cause severe lower respiratory tract infection leading to a higher chance of developing acute respiratory distress syndrome (ARDS) and extra pulmonary manifestations[15]. The first HCoV-229E strain, B814, was isolated from the respiratory tract of patients with upper respiratory tract infection in 1966 and was subsequently adapted to grow in WI-38 lung cell lines. Only a few immune-compromised patients exhibited severe lower respiratory tract infection with fever and cold. Later in 1967, HCoV-OC43 was isolated from organ culture[17]. The clinical features of HCoV-OC43 disease appear to be similar to those caused by HCoV-229E. These symptoms are almost identical to infection with other respiratory tract pathogens such as influenza A viruses and rhinoviruses. Both HCoV-229E and HCoV-OC43 are distributed globally, and they tend to be predominantly transmitted during winter in temperate climates. Generally, these two viruses' incubation time is less than one week, followed by an approximately 2-week illness. Since then, more knowledge was accumulated through extensive studies on HCoV-229E and HCoV-OC43, both of which cause self-limiting symptoms. Indeed, the infection with HCoVs is generally thought to be harmless until the outbreak of SARS[15]. SARS-CoV was the first well-documented HCoV-caused pandemic in human history, and the etiological agent was SARS-CoV, the third HCoV discovered. The first case of SARS can be traced back to late 2002 in Guangdong Province of China. The SARS epidemic resulted in 8,096 reported cases with 774 deaths, spreading across many countries and continents. Patients infected with SARS-CoV initially present with myalgia, headache, fever, malaise, and chills, followed by dyspnea, cough, and respiratory distress as late symptoms. The incubation period is 4 to 7 days, and the peak of viral load appears on the 10th day of illness. Lymphopenia, deranged liver function tests, and elevated creatine kinase are common abnormalities in SARS[18,19]. Diffuse alveolar damage, epithelial cell proliferation, and an increase of macrophages are also observed in SARS patient[20]. Approximately 20-30% of patients subsequently require intensive care and mechanical ventilation. In addition to the lower respiratory tract, multiple organs, including the gastrointestinal tract, liver, and kidney, can also be infected in these severe cases, usually accompanied by a cytokine storm, which might be lethal, particularly in immune-compromised patients. Since then, tremendous efforts have been dedicated to HCoV research. HCoV-NL63 was isolated in the Netherlands during late 2004. It was initially prevalent in young children, the elderly, and immune-compromised patients with respiratory illnesses. Presentation of coryza, conjunctivitis, fever, and bronchiolitis is expected in the disease caused by HCoV-NL63[21,22]. Another independent study described the same virus's isolation from a nasal specimen from an 8-month-old boy suffering from Pneumonia in the Netherlands. Although it was identified in the Netherlands, it is distributed globally. It has been estimated that HCoV-NL63 accounts for

approximately 4.7% of common respiratory diseases, and its peak incidence occurs during early summer, spring and winter. HCoV-NL63 is associated with obstructive laryngitis, also known as croup. In the same year, HCoV-HKU1 was isolated from a patient hospitalized with pneumonia and bronchiolitis in Hong Kong. Besides community-acquired pneumonia and bronchiolitis, HCoV-HKU1 was reported to be associated with acute asthmatic exacerbation [23,24]. Similar to HCoV-NL63, HCoV-229E and HCoV-OC43, HCoV-HKU1 was found worldwide, causing mild respiratory diseases[15]. All these four community-acquired HCoVs have been well adapted to humans. They are generally less likely to mutate to cause highly pathogenic diseases. However, accidents did occur for unknown reasons as in the rare case of a more virulent subtype of HCoV-NL63, which has recently been reported to cause severe lower respiratory tract infection in China. Generally, when these HCoVs acquire the ability to transmit efficiently and maintain themselves continuously within humans, they also become less virulent or pathogenic. Ten years after the SARS epidemic in 2002, the Middle East respiratory syndrome (MERS) outbreak resulted in a persistent epidemic in the Arabian Peninsula with sporadic spreading to the rest of the world [25,26]. Clinical manifestations of MERS resemble those of SARS, characterized by progressive acute pneumonia. Unlike SARS, many patients with MERS also developed acute renal failure, which is thus far unique for MERS among HCoV-caused diseases. More than 30% of patients present with gastrointestinal symptoms, such as diarrhea and vomiting. So far, over 2500 laboratory-confirmed cases were reported with a high case fatality of 34.4%, making MERS-CoV one of the most devastating viruses known to humans. The 2019 novel HCoV (2019-nCoV), which has subsequently been renamed SARS-CoV-2, is the causative agent of the ongoing coronavirus disease 2019 (COVID-19), which has claimed more than 3,120 lives and infected more than 91,000 people as of March 3, 2020. The alarming devastation indicates that the world has to prepare for the deadly pandemic of SARS-CoV-2. During the middle to late December 2019, clusters of pneumonia patients retrospectively known to be associated with SARS-CoV-2 infection were detected in Wuhan, Hubei Province, China. The World Health Organization declared the ongoing outbreak of lower respiratory tract infection caused by SARS-CoV-2, a Public Health Emergency of International Concern, and named the disease COVID-19[27]. As of March 3, 2020, 90,053 cases have been confirmed worldwide, with a crude case fatality of 3.4%. SARS-CoV-2 causes severe respiratory infections like SARS-CoV and MERS-CoV presented as fever, cough, and dyspnea. Diarrhea is also seen in some patients. Pneumonia is one of the most severe symptoms and can progress rapidly to acute respiratory distress syndrome[15]. Although SARS-CoV and SARS-CoV-2 are very similar due to high nucleotide sequence homology of 82%, they cluster into different branches in the phylogenetic tree. So far, SARS-CoV-2 is thought to be less pathogenic but more transmissible compared to SARS-CoV and MERS-CoV[28]. Asymptomatic subjects infected with SARS-CoV-2 have been reported and might contribute to its rapid spreading around the world. Comparing and contrasting SARS-CoV-2 with the other six HCoVs reveal similarities and differences of great interest. First, the incubation period and the duration of the course of HCoV disease are very similar. In this regard, SARS-CoV-2 follows the general trend of the other six HCoVs. Second, the severity of symptoms of COVID-19 lies between SARS-CoV and the four community-acquired HCoVs (i.e., HCoV-229E, HCoV-OC43, HCoV-HKU1, and HCoV-NL63). SARS-CoV-2 infection exhibits more commonly seen features during infection with community-acquired HCoVs, including non-specific, mild, or even no symptoms. But, a small subset of severe cases of COVID-19 can also be seen as in the case of SARS-CoV infection, although the ratio is a bit lower. Third, the transmission of SARS-CoV-2 also shows interesting patterns characteristic of both community-acquired HCoVs and SARS-CoV. The transmissibility of SARS-CoV-2 is at least as high as that of community-acquired HCoVs[15]. But, it remains to be verified whether the transmissibility of SARS-CoV-2 decreases after passages in humans as SARS-CoV and MERS-CoV. Finally, same as the other HCoVs, SARS-CoV-2 can be detected in fecal samples. Whether fecal-oral transmission of SARS-CoV-2 plays an important role, as in the case of SARS-CoV, at least under some circumstances remains to be clarified by future studies. It is also a great interest to see whether SARS-CoV-2 might exhibit seasonality as in community-acquired HCoVs. The various features of SARS-CoV-2, such as its transmission, pathogenicity, and sustainable spreading after passages in humans, will be influential on the ultimate fate of the ongoing outbreak of COVID-19.

4. GENOMIC ORGANIZATION

The Yongzhen Zhang team in China was the first to determine the full-length genomic sequence of the SARS-CoV-2 virus (GenBank accession number AY274119)[29]. This novel coronavirus's genome is 29.891 kb long, with a G + C content of 38%. It is an unsegmented, single-stranded (ss) positive-sense RNA genome, enclosed by a 5′-cap and 3′-poly-A tail. These viruses encircled within an envelope containing viral nucleocapsid arranged in helical symmetry revealed a divulging spherical outline with virion diameter varying from 60 to 140 nm and distinct spikes of 9 to 12 nm, giving the virus an appearance of a solar corona [Fig.1]. The genomic structure, typical of other beta coronaviruses, contains 14 open reading frames (ORFs), encoding for 27 proteins. The genome is arranged in the order of a 5′-untranslated region(UTR)-replicase complex (orf1ab)-structural proteins (Spike(S)-Envelope(E)- Membrane (M)-Nucleocapsid (N))−3′UTR and non-structural open reading frames (ORFs)[Fig.2]. The 5′ and 3′ terminal sequences have 265 nt at the 5′ terminal end and 229 nt at the 3′ terminal end. The predicted replicase ORF1ab is 21,291 nt in length and contains 15 predicted non-structural proteins(nsps), which are essential for virus replication, followed by 13 downstream ORFs. The synthesis of polyprotein 1a/1ab (pp1a/pp1ab) in the host is realized from viral RNA[30]. The transcription works through the replication-transcription complex (RCT) organized in double-membrane vesicles and via the synthesis of sub-genomic RNAs (sgRNAs) sequences. Of note, transcription termination occurs at transcription regulatory sequences, located between the so-called open reading frames (ORFs) that work as templates for subgenomic mRNAs production. The two poly-proteins, pp1a and pp1ab, are initially produced from ORF1a/b by a -1 frameshift between ORF1a and ORF1b. The viral encoded proteases [Main protease (Mpro), chymotrypsin-like protease (3CLpro), and papain-like protease (PLPs)] cleave these poly-proteins into individual nsps. They together comprise 15 nsps, including nsp1 to nsp10 and nsp12 to nsp16. Apart from ORF1a and ORF1b, other ORFs encode for structural proteins and accessory proteins. The 3′-terminal region of the genome encodes for structural proteins, namely spike (S),

envelope protein (E), membrane protein (M), and nucleocapsid (N), plus eight accessory proteins(3a, 3b, p6, 7a, 7b, 8b, 9b, and ORF14). The predicted S, E, M, and N genes are 3,822, 228, 669, and 1,260 nt in length. Accessory genes are seen intermingled within the structural genes, and it is found that 2019-nCoV lacks the hemagglutinin-esterase gene (HE), which is the characteristic of some Betacorona virus [10]. Pathophysiology and virulence mechanisms of SARS-CoV-2 have links to the function of the nsps and structural proteins. For instance, research underlined that nsps could block the host innate immune response[31]. Among structural proteins functions, the envelope has a crucial role in virus pathogenicity as it promotes viral assembly and release. However, many of these features (e.g., those of nsp 2, and 11) have not yet been described. S protein is a large multifunctional class I viral transmembrane protein, which lies as a trimer on the virion surface, giving the virion a 'corona' or crown-like appearance. Functionally it is required to enter the infectious virion particles inside the cell through interaction with various host cellular receptors. Notably, Spike glycoproteins are one of the vital immunodominant proteins of coronaviruses. They can induce a host immune response[32]. The spike glycoproteins are composed of two subunits (S1 and S2). The first one, S1, helps host receptor binding while the latter one, S2, is responsible for the fusion. The former (S1) is further divided into two subdomains, namely the N- terminal domain (NTD) and C- terminal domain (CTD). Both these subdomains act as the receptor-binding domains interacting efficiently with various host receptors. The S1 CTD contains the receptor-binding motif (RBM). The trimeric S1 locates itself on top of the trimeric S2 stalk. The S2 subunit containing a fusion peptide, a transmembrane domain, and cytoplasmic domain — is highly conserved. Thus, it could be a target for antiviral (anti-S2) compounds. The M protein is the most abundant viral protein present in the virion particle, giving a definite shape to the viral envelope. It binds to the nucleocapsid and acts as a central organizer of the coronavirus assembly. The M protein containing three transmembrane domains, flanked by a short amino-terminal outside the virion, and a long carboxy-terminal inside the virion has a highly diverse amino acid composition. The E protein consisting of three domains, namely short hydrophilic amino-terminal, a large hydrophobic transmembrane domain, and a C terminal domain, is the smallest among the major structural proteins. It plays a multifunctional role in the pathogenesis, assembly, and release of the virus. The envelope protein acts as viroporin (ion-channel). Its absence or inactivation is related to altered virulence due to morphological changes and tropism. The N protein is multifunctional and plays a role in complex formation with the viral genome. It facilitates M protein interaction needed during virion assembly and enhances the virus's transcription efficiency. It contains three highly conserved and distinct domains, namely an N-terminal domain (NTD), RNA-binding domain or linker region (LKR), and a C-terminal domain (CTD). The NTD binds with the 3' end of the viral genome and is highly diverged both in length and sequence. The receptor-binding domain (RBD), rich in serine and arginine, can directly interact with RNA and is responsible for cell signalling[33]. It also modulates the host's antiviral response by working as an antagonist for interferon 302 and RNA interference[34]. On the contrary, the spike receptor-binding domain presents only a 40% amino acid identity with other SARS-CoVs. Other structural elements on which research must necessarily focus are the ORF3b with no homology with SARS-CoVs and a secreted protein (encoded by ORF8), which is structurally different from those of SARS-CoV[10].

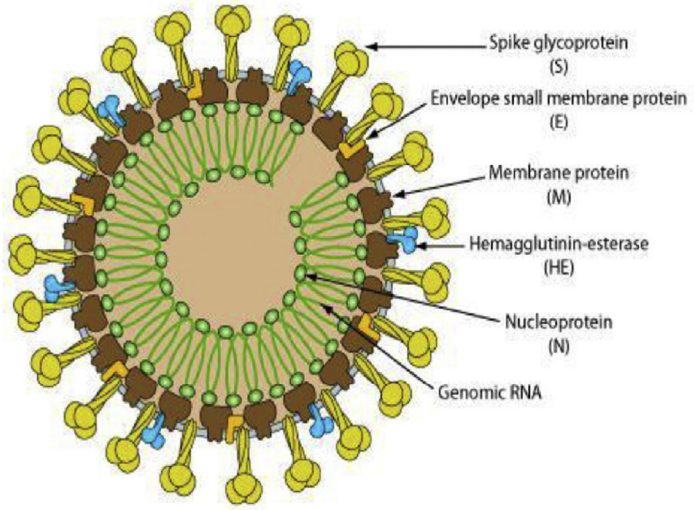

Fig.1: Schematic of a coronavirus
(http://ruleof6ix.fieldofscience.com/2012/09/a-new-coronavirus-should-youcare.html).

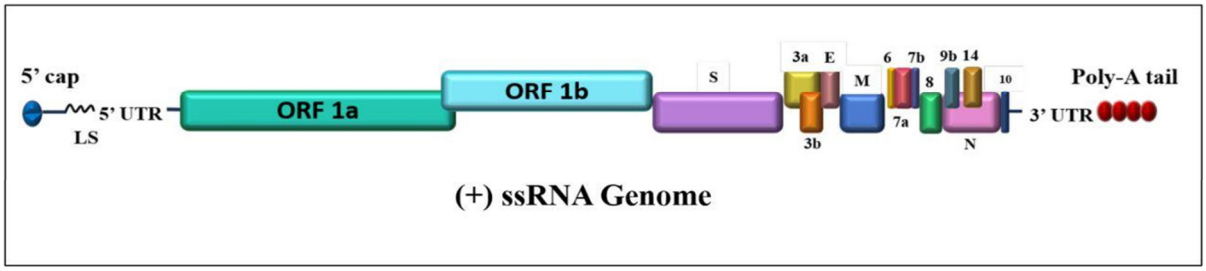

Fig. 2:The SARSCoV-2 genome is arranged in the order of 5′-replicase (ORF1a/b)–structural proteins [spike (S)–envelope (E)–membrane (M)–nucleocapsid (N)] −3′

5. THE ORIGIN AND EVOLUTION OF SARS-COV-2

Previous investigations revealed that the genera alpha coronavirus and beta coronavirus originated from bats[35]. The gamma coronavirus and delta coronavirus evolved from birds and swine[36]. Gamma and delta coronaviruses generally infect birds, although some can cause infection in mammals. Alpha and beta coronaviruses are known to harm humans and animals. SARS-CoV, HCoV-HKU1,HCoV-OC43 , MERS-CoV, and HCoV-229E and HCoV-NL63 can cause infections in humans. SARS-CoV, HCoV-HKU1, HCoV-OC43, and MERS-CoV belong to the genera beta coronavirus. HCoV-229E and HCoV-NL63 are alpha coronaviruses. The beta coronaviruses are the most important group because they comprise the most highly pathogenic viruses against humans, including SARS-CoV-2, MERS-CoV, and SARS-CoV[37]. The highly pathogenic MERS and SARS coronaviruses originated in bats[38]. However, the origin of the newly emerged SARS-CoV-2 remains debatable. The association of initially confirmed SARS-CoV-2 cases with the Huanan Seafood market suggested that the marketplace has played a role in the early spreading. However, whether it is the origin of the outbreak and the native host(s) of SARS-CoV-2 remains uncertain. All HCoVs are thought to have originated from bats, mice, or domestic animals[15]. Several studies supported an evolutionary origin of all HCoVs from bats. In the host, these viruses become well adapted but show great genetic diversity. Although parental viruses of HCoVs are typically non-pathogenic in their natural reservoir hosts but become pathogenic after interspecies transmission to a new host. Tracing the zoonotic origin of HCoVs provides a framework to understand the natural history, driving force, and restriction factors of species jumping. It might help the reservoir's search, intermediate and amplifying animal host(s) of SARS-CoV-2, with important implications in preventing future spillover. All four community-acquired HCoVs causing mild symptoms have been well adapted to humans. From another perspective, it might also be true that humans have been well adapted to these four HCoVs. In other words, both could be the survivors of ancient HCoV pandemics. HCoVs that cause severe diseases in humans and humans who developed severe HCoV diseases have been eliminated. For this to happen, HCoVs have to replicate in humans to a sufficient extent to allow the accumulation of adaptive mutations that counteract host restriction factors. In this sense, the longer the SARS-CoV-2 outbreak persists and the more people it infects, the greater the chance that it will fully adapt to humans. If it adapts well, its transmission in humans would be difficult to stop by quarantine or other infection control measures. For many years, the four community-acquired CoVs circulate in human populations, triggering common cold in immunocompetent subjects. These viruses do not need an animal reservoir. In contrast, highly pathogenic SARS-CoV and MERS-CoV have not adapted to humans well, and their transmission cannot be sustained. They need to maintain and propagate in their zoonotic reservoirs and seek the chance to spillover to susceptible human targets, possibly via one or more intermediate and amplifying hosts. SARS-CoV-2 has features that are similar to both SARS-CoV/MERS-CoV and the four community-acquired HCoVs. It is highly transmissible, like community-acquired HCoVs, at least for the time being. However, it is more pathogenic than community-acquired HCoVs and less pathogenic than SARS-CoV or MERS-CoV. Whether it will adapt fully to humans and circulate within humans without a reservoir or intermediate animal host remains to be seen.

Before discussing the animal origins of HCoVs, it will serve us well to discuss the definitions and characteristics of the evolutionary, natural, reservoir, intermediate, and amplifying hosts of HCoVs. An animal serves as the evolutionary host of an HCoV if it harbours a closely related ancestor sharing high homology at the nucleotide sequence level. The ancestral virus is usually well adapted and non-pathogenic in this host. Likewise, a reservoir host harbours HCoV continuously and for the long term. In both cases, the hosts are naturally infected, and animals are the natural hosts of HCoV or its parental virus. In contrast, if the HCoV is newly introduced to an intermediate host right before or around its introduction to humans, it is not well adapted to the new host and is often pathogenic. This intermediate host can serve as the zoonotic source of human infection and play the role of an amplifying host by allowing the virus to replicate transiently and then transmitting it to humans to amplify the scale of human disease. An HCoV can undergo a dead-end infection if it cannot sustain its transmission within the intermediate host. On the contrary, HCoVs can also adapt to the intermediate host and even establish long-term endemicity. In this case, the intermediate host becomes a natural reservoir host. The analysis of SARS-CoV-2 origin was performed based on the complete genome sequence. Investigations have revealed that the SARS-CoV strains detected in market civets were transmitted from horseshoe bats. These viruses were phylogenetically related to SARS-CoV in bats from China, Europe, Southeast Asia, and Africa. Besides, the genome sequences of SARS-CoV strains isolated from humans were highly similar to those in bats. The origin of SARS-CoV has been extensively investigated. SARS-CoV-2 had 96.2% overall genome sequence identity throughout the genome to BatCoV RaTG13, a bat coronavirus detected in Rhinolophus affinis from Yunnan province[39]. Furthermore, the phylogenetic analysis of the full-length genome, the receptor-binding protein spike (S) gene, and RNA-dependent RNA polymerase (RdRp) gene demonstrated that RaTG13 was the closest relative of the SARS-CoV-2. Although the zoonotic source of SARS-CoV-2 is not confirmed, its genome sequence exhibits close relatedness with two other bat-derived SARS-like coronaviruses,bat-SL-CoVZC45 and bat-SL-CoVZXC21[40]. Comparing the genome of SARS-CoV-2 with that of the closely related SARS/SARS-like CoV revealed that the sequence coding for the spike protein with a total length of 1,273 amino acids showed 27 amino acid substitutions. The sequence identity between SARS-CoV-2 and bat-SL-CoVZC45 or bat-SL-CoVZXC21, the closest relatives in their analyses, is lower than 90%[40,41]. Furthermore, the genome sequencing-based studies have revealed that MERS-CoV strains from humans are phylogenetically related to bats[41,42]. The strains have identical genomic and protein structures except for the S proteins. Also, recombination analysis of genes encoding orf1ab and S revealed that MERS-CoV originated from the exchange of genetic elements between coronaviruses in camels and bats [43,44]. Phylogenetic analysis reveals that SARS-CoV-2 is genetically distinct from SARS-CoV (79% similarity) and MERS-CoV (50%). However, homology modeling shows that both SARS-CoV and SARS-CoV-2 have similar receptor-binding domain structures, despite amino acid variation at some key residues, including the absence of the 8a protein and the fluctuation in the number of amino acids in the 8b and 3c proteins in SARS-CoV2[38,45]. In contrast, the primary protease is highly conserved between SARS-CoV-2 and SARS-CoV, with a 96% overall identity. Thus, it is reasonable to suspect that the bat is the natural host of SARS-CoV-2,

considering its similarity with SARS-CoV. Despite some evidence pointing out that bats are a potential reservoir of SARS-CoV-2, there remains doubt whether other hosts of 2019-nCoVs exist and that transmitted the virus to humans. Because most bat species in Wuhan were hibernating in late December, and no bats were found or sold in the Huanan Seafood market in late December. So, it may be speculated that an animal sold at the Wuhan seafood market might represent an intermediate host facilitating the virus's emergence in humans. Previous reports suggest that other human- infecting coronaviruses that originate from bat have intermediate hosts. For example, masked palm civet and dromedary camels are the intermediate hosts for SARS-CoV and MERS-CoV, respectively. A study of the relative synonymous codon usage (RSCU) found that SARS-CoV-2, bat-SL-CoVZC45, and snakes had similar synonymous codon usage bias, and speculated that snakes might be the intermediate host. However, no SARS-CoV-2 has been isolated from the snake yet. Interestingly, the coronavirus strains isolated from pangolins were found to have 85.5% to 92.4% similarity in nucleotide sequence and 97.4% similarity in amino acid sequences to SARS-CoV-2[46]. The sequence similarity of SARS-CoV-2 to these identified coronaviruses from pangolins is lower than the bat coronavirus RaTG13 (96.2%). The receptor-binding domain of S protein from one sub-lineage of the pangolin coronaviruses has97.4% similarity in amino acid sequences to that of SARS-CoV-2, higher than that of RaTG13 (89.2%). In other studies, coronaviruses isolated from Malayan pangolins showed 100%, 98.2%, 96.7%, and 90.4% amino acid identity with 2019-nCoV in the E, M, N, and S genes, respectively[47]. Moreover, the pangolin coronavirus and SARS-CoV-2 share identical amino acids at the five critical residues of RBD of S protein, while RaTG13 only possesses one. The discovery of coronavirus from pangolin and its closeness to SARS-CoV-2 in sequence similarity suggests that pangolin may be a potential intermediate host. The receptor-binding domain of the S protein of the Pangolin-CoV is virtually identical to that of 2019-nCoV, with one amino acid difference. A comparison of available genomes suggests 2019-nCoV might have originated from the recombination of a Pangolin-CoV-like virus with a Bat-CoV-RaTG13-like virus. However, bat and pangolin roles as the respective natural reservoir and intermediate host still need further investigation.

6. PATHOGENESIS

The ability of coronaviruses to infect humans is invariably associated with their binding strengths to human receptor proteins. Given the emergency, the characterization of the biology of SARS-CoV-2 and features of virus-host interactions demands identification of the candidate molecules to inhibit viral functions. Perhaps the most well-known human protein related to beta-coronavirus infection is ACE2 (Angiotensin Converting Enzyme 2), a zinc metalloprotease able to hydrolyze angiotensin I, angiotensin II, apelin-13, dynorphin A 1-13, and 7 additional small peptides. The SARS-related coronaviruses are covered by spike proteins that contain a variable receptor-binding domain (RBD). This RBD binds to the angiotensin-converting enzyme-2 (ACE2) receptor found in the heart, lungs, kidneys, and gastrointestinal tract, thus facilitating viral entry into target cells. SARS-CoV exploits ACE2 as its cellular entrance based on the initial interaction between the viral S (spike) protein and ACE2[48]. The SARS-CoV infects ciliated bronchial epithelial cells and type-II pneumocytes through angiotensin-converting enzyme 2 (ACE2) as a receptor. Based on biophysical and structural evidence and the sequence similarity, many investigations reported that both SARS‑CoV ‑2 and severe acute respiratory syndrome coronavirus (SARS‑CoV) use the ACE2 receptor to facilitate viral entry in target cells. The mechanisms associated with the infectiousness of SARS-CoV-2 are not entirely understood; however, several recent studies have shown that SARS-CoV-2 access in human cells also depends on the interaction between the S protein and ACE2. The SARS-CoV-2 spike protein may directly bind with the host cell surface ACE2 receptor facilitating virus entry and replication. The lung appears to be the most vulnerable target organ as its vast surface area makes the lung highly susceptible to inhaled viruses. The biophysical and structural evidence that the SARS-CoV-2 spike protein was predicted to have a strong binding affinity to human ACE2[49]. The wide distribution and similarity of this receptor in the animal kingdom may account for cross-species transmission. Simultaneously, the pattern of expression of ACE2 in human respiratory epithelia and oral mucosa may explain the fast human-human transmission. The down-regulation of ACE2 could be a SARS-CoV-2-induced mechanism from which the virus gains the capability to spread faster by damaging the host lung tissue. Further analysis even suggested that SARS-CoV-2 recognizes human ACE2 more efficiently than SARS-CoV, increasing the ability of SARS-CoV-2 to transmit from person to person. Since SARS-CoV and SARS-CoV-2 are so similar, the biochemical interactions and the pathogenesis are likely similar. Binding of the SARS-CoV to the angiotensin-converting enzyme 2 (ACE-2) receptors in the type II pneumocytes in the lungs triggers a cascade of inflammation in the lower respiratory tract. It has been demonstrated that when the SARS spike protein binds to the ACE-2 receptor, the complex is proteolytically processed by type 2 transmembrane protease TMPRSS2[50]. It leads to cleavage of ACE-2 and activation of the spike protein, similar to the mechanism employed by influenza and human metapneumovirus, thus facilitating viral entry into the target cell. It has been suggested that cells in which ACE-2 and TMPRSS2 are simultaneously present are most susceptible to access by SARS-CoV. Early indications are that the SARS-CoV-2 virus also requires ACE-2 and TMPRSS2 to enter cells. The host's immune response system is activated when the cell is infected by viral entry, and then, the inflammatory cascade is initiated by antigen-presenting cells (APC). The APC performs its functions by presenting the foreign antigen to CD4+-T-helper (Th1) cells, and releasing interleukin-12 to stimulate the Th1 cell further. The Th1 cells stimulate CD8+-T-killer (Tk) cells, which target any cells containing the foreign antigen and stimulate B-cells to produce antigen-specific antibodies[51]. The clinical spectrum of COVID-19 varies from asymptomatic or paucisymptomatic forms to clinical conditions. The critical cases are characterized by respiratory failure that necessitates mechanical ventilation and support in an intensive care unit (ICU) to multiorgan and systemic manifestations in sepsis, septic shock, and multiple organ dysfunction syndromes (MODS).In 81% of cases, the patients suffered from uncomplicated illness disease with non-pneumonia, mild Pneumonia, or moderate Pneumonia. In most of the issues, these patients usually have an upper respiratory tract viral infection, including mild fever, cough (dry), sore throat, nasal congestion, malaise, headache, muscle pain, or malaise. In some moderate Pneumonia cases, fever is associated with respiratory symptoms such as cough and shortness of breath (or tachypnea in children) without

signs of severe pneumonia. The disease manifested as severe pneumonia in 14% of cases. In this case, fever is associated with severe dyspnea, respiratory distress, tachypnea (> 30 breaths/min), and hypoxia (SpO2 < 90% on room air). However, the fever symptom may fluctuate even in severe forms of the disease. Cyanosis can occur in the case of children. In 5% of cases, the condition becomes critical with respiratory failure, septic shock, and/or multiple organ dysfunction (MOD) or failure (MOF).

7. TREATMENT

There is no specific antiviral treatment recommended for COVID-19, and no vaccine is currently available. The treatment is symptomatic, and oxygen therapy represents the primary treatment intervention for patients with severe infection. Mechanical ventilation may be necessary in respiratory failure cases refractory to oxygen therapy, whereas hemodynamic support is essential for managing septic shock. No definite therapeutic strategies have been developed to date. Systemic corticosteroids for treating viral pneumonia or acute respiratory distress syndrome (ARDS) are not recommended in this case. Although unselective or inappropriate administration of antibiotics should be avoided, several approaches[52] have been adopted.

1. The lopinavir/ritonavir, chloroquine, and hydroxychloroquine are used in the proper dosage. Alpha-interferon (e.g., 5 million units by aerosol inhalation twice per day) is also used.
2. Remdesivir (GS5734) — an inhibitor of RNA polymerase with in vitro activity against multiple RNA viruses, including Ebola — could be useful for both prophylaxis and therapy of HCoVs infections. This drug was positively tested in a rhesus macaque model of MERS-CoV infection.
3. Tolicizumab, a humanized IgG1 monoclonal antibody, directed against the IL-6 receptor and commonly used to treat rheumatoid arthritis is also used.

8. TRANSMISSION

Because the first cases of the CoVID-19 disease were linked to direct exposure to the Huanan Seafood Wholesale Market of Wuhan, the animal-to-human transmission was presumed the primary mechanism. Nevertheless, subsequent cases were not associated with this exposure mechanism. Therefore, it was concluded that the virus could also be transmitted from human-to-human, and symptomatic people are the most frequent COVID-19 spread source. The possibility of transmission before symptoms develop seems to be infrequent, although it cannot be excluded. Moreover, there are suggestions that individuals who remain asymptomatic could transmit the virus. This data suggests that the use of isolation is the best way to contain this epidemic. As with other respiratory pathogens, including flu and rhinovirus, the transmission is believed to occur through respiratory droplets from coughing and sneezing. Aerosol transmission is also possible in case of prolonged exposure to elevated aerosol concentrations in closed spaces. Analysis of data related to the spread of SARS-CoV-2 seems to indicate that close contact between individuals is necessary to transmit the disease[53]. It may be reasonably hypothesized that the virus might pass through the mucous membranes, especially nasal and larynx mucosa, then enter the lungs through the respiratory tract, causing common infection symptoms are fever and cough. Then the virus would attack the targeting organs that express ACE2, such as the lungs, heart, renal, gastrointestinal tract. The SARS-CoV-2 detected in the faecal samples is more likely because it enters the blood from the lungs and then travels from the blood to the intestines[54]. During the infection process, the white blood cell count in peripheral blood in the early stage of the disease is normal or slightly low, and lymphopenia is observed in patients. B lymphocyte reduction may occur early in the disease, which may affect antibody production in the patient. In severe type patients, lymphocytes were significantly reduced. In that case, lymphocytes in patients with COVID-19 might gradually decrease as the disease progresses. But the mechanism of significant lymphocyte reduction in severe type patients remains unclear. The inflammatory factors associated with diseases mainly containing IL-6 were significantly increased, which also contributed to the disease's aggravation. Non-survivors had higher neutrophils, D-Dimer, blood urea nitrogen, and creatinine than the survivors. If patients' immune function in the acute phase (pneumonia phase) is sufficient, and no more basic diseases, the virus can be effectively suppressed. In that case, the patients enter the recovery phase. Suppose the patient is older or in an immune impaired state, combined with other primary diseases such as hypertension and diabetes. In that case, the immune system cannot effectively control the virus in the acute phase (pneumonia phase). The patient will become a severe or critical type. In those cases, T cells, B cells were further reduced, while inflammatory cytokines and D-Dimer continued to increase in severe type patients. To enhance patients' immune function and inhibit the formation of inflammatory factor storms, only a few therapeutic measures[52] have been suggested because COVID-19 does not currently have a specific antiviral drug treatment. Based on data from published epidemiology and virologic studies provide evidence that COVID-19 is primarily transmitted from symptomatic people to others who are in close contact through respiratory droplets, by direct contact with infected persons, or by contact with contaminated objects and surfaces. The incubation period for COVID-19, which is the time between exposure to the virus (becoming infected) and symptom onset, is 5-6 days. However, it can be up to 14 days. During this period, also known as the "presymptomatic" period, some infected persons can be contagious. In a small number of case reports and studies, a presymptomatic transmission has been documented. It has been observed that some people can test positive for COVID-19 from 1-3 days before they develop symptoms. Thus, people infected with COVID-19 could transmit the virus before significant symptoms developed. It is essential to recognize that presymptomatic transmission still requires the virus to be spread via infectious droplets or through touching contaminated surfaces. Therefore, transmission from a presymptomatic case can occur before symptom onset. There are few reports[55] of laboratory-confirmed cases that are genuinely asymptomatic(person infected with COVID-19 who does not develop symptoms). To date, there has been no documented asymptomatic transmission(refers to transmission of the virus from a person who does not develop symptoms). However, this does not exclude the possibility that asymptomatic transmission may not occur. So, the use of isolation may be the best way to contain this epidemic.

9. TRANSMISSION RATE, INCUBATION PERIOD, AND MODE OF TRANSMISSION

The reproductive number denoted R_0 (pronounced as 'R-naught') is a mathematical term that defines contagiousness i.e., the average or expected number of people that an infected person could spread the virus to in the infected period. The average reproductive number of SARS-CoV-2 is estimated to be 3.28, with a median value of 2.79, although the World Health Organization estimates it between 2 and 2.5[56]. For comparison, It may be mentioned that the mean R_0 for seasonal influenza is between 1.1 and 2.3, whereas for SARS, it was between 1.7 and 1.9 and for MERS less than 1, suggesting that SARS-CoV-2 has higher pandemic potential. Since the estimation of R_0 depends on the estimation method used and the measure of R_0 may be biased by insufficient data and short onset time of the diseases. If R_0 is greater than 1, then the disease outbreak will lead to an epidemic, and if R_0 is less than 1, then the spell will become extinct. If R_0 is equal to 1, one infected person will infect precisely one other person, so the number of infected persons will remain constant over time. So, there is reason to believe that estimates of R_0 play a significant role to chalk out strategies to contain the spread of the virus. Understanding incubation periods is significant as it allows health authorities to introduce more effective measures to control R_0. The best current estimates of the SARS-CoV-2 infection range from 2 to 14 days. Coronaviruses are generally thought to be spread by respiratory droplets, not confused with the airborne transmission[57]. Droplets are more massive and tend to fall to the ground close to the infected host and only infect others if a susceptible host intercepts the droplet before landing. Droplet transmission is typically limited to short distances, generally less than 2 m. However, the airborne route involves much smaller droplets floating and moving long distances with air currents. Under specific humidity and temperature environments, airborne droplets can remain in flight for hours. Generally, pathogens transmissible via the airborne route have higher R_0 because infected particles can stay in the air long after the infected individual has left the premises. For example, airborne transmission happens in measles (R_0 between 12 and 18 and chickenpox(R_0s between 3.7 and 5.0. Once infected droplets have landed on surfaces, their survivability on those surfaces determines if contact transmission is possible. The viruses can survive and remain infectious from 2 h up to 9 days on surfaces as metal, glass, or plastic, with increased survival in colder and dryer environments. Repeated cleaning of surfaces with common biocidal substances such as ethanol and sodium hypochlorite may be very effective for the inactivation of the coronaviruses. To find out the strategies to contain the disease is very much important. Further studies are needed to understand the transmission mechanisms, incubation times and clinical course, and infectivity duration.

10. PREVENTION

Preventive strategies are focused on patients' isolation, and careful preventive measures are the current strategy to limit the spread of cases. Because an epidemic will increase as long as R0 is greater than 1 (COVID-19 is 2.2), control measures must focus on reducing the value to less than 1. The WHO and other organizations have issued the following general recommendations:

- Avoiding close contact with subjects suffering from acute respiratory infections.
- Washing hands frequently, especially after contact with infected people or their environment.
- Avoiding unprotected contact with farm or wild animals.
- People with acute airway infection symptoms should keep their distance, cover coughs or sneezes with disposable tissues or clothes, and wash their hands.
- Immune-compromised individuals should avoid public gatherings.

The most crucial strategy for the populace to undertake is to frequently wash their hands and use portable hand sanitizer and avoid contact with their face and mouth after interacting with a possibly contaminated environment.

11. VACCINE

Meanwhile, scientific research is growing to develop a coronavirus vaccine towards novel coronavirus immunization. Several potential therapeutic approaches[58] are currently being extensively investigated to combat COVID-19 disease.

11.1. SPIKE PROTEIN-BASED VACCINE

The development of a spike protein-based vaccine may rely on the fact that ACE2 is the SARS-CoV-2 receptor. So, cell lines that facilitate viral replication in the presence of ACE2 may be most efficient in large-scale vaccine production.

11.2. INHIBITION OF TRANSMEMBRANE PROTEASE SERINE 2(TMPRSS2) ACTIVITY.

Transmembrane protease serine 2 (TMPRSS2) is essential for the entry and viral spread of SARS-CoV-2 through interaction with the ACE2 receptor. The serine protease inhibitor camostat mesylate has been shown to block TMPRSS2 activity[59] and is thus an exciting candidate.

11.3. BLOCKING THE ACE2 RECEPTOR.

Identifying interaction sites between ACE2 and SARS-CoV-2 at the atomic level, one could target this interaction site with antibodies or small molecules.

11.4. DELIVERING EXCESSIVE SOLUBLE FORM OF ACE2.

Suppose an excessive soluble form of ACE2 is administered. In that case, it may bind with SARS-CoV-2 not only to neutralize the virus but also to rescue cellular ACE activity, which negatively regulates the renin-angiotensin system(RAS) to protect the lung from injury[60]. Thus, a soluble form of ACE2 may be useful in drug design to combat the disease.

11.5. RNA-BASED VACCINE

Traditional vaccines that stimulate the production of antibodies via challenges with purified proteins from the pathogens, or by using whole cells (live, attenuated vaccines), generally take a long time to assess the clinical benefit. Alternatively, RNA-based vaccines use mRNA that, upon entering cells, are translated to antigenic molecules[61] that, in turn, stimulate the immune system and are more rapid and less expensive than traditional vaccines, which can be a major advantage in pandemic situations. So far, only a little

information is available about the targets of immune responses to SARS-CoV-2. The use of available information related to SARS-CoV epitopes in conjunction with bioinformatic predictions points to specific regions of SARS-CoV-2 that are likely being recognized by human immune responses. The observation that many B and T cell epitopes are highly conserved between SARS-CoV-2 and SARS-CoV is important. Vaccination strategies designed to target the immune response toward these conserved epitope regions could generate immunity[62]. Several *in silico* analysis [63] of SARS-CoV-2 genomic sequence along with other members of the coronaviridae family of viruses, the overall human genome, and the transcriptome of specific human tissues such as lung, which are primarily targeted by the virus can facilitate effective vaccine design against this novel virus. Predicting targets through computational methods is appealing because it circumvents expensive and difficult experiments.

12. CONCLUSION

At the moment, the therapeutic strategies to contain the infection are only supportive, and prevention aimed at reducing transmission in the community is our best weapon. Therefore, the aim is to collect information and scientific evidence and provide an overview of the topic that needs continuous updating. Many uncertainties remain regarding both the virus-host interaction and the evolution of the epidemic. Scientists worldwide work tirelessly, and information about the transmission mechanisms, the clinical spectrum of disease, new diagnostics, and prevention and therapeutic strategies are rapidly developing. Lack of sufficient experimental and clinical data and the urgency to understand the deadly coronaviruses' infectivity, we have been increasingly relying on computational analyses. To study the 2019-nCoV virus in terms of protein structures, functions, phylogeny, and interactions at molecular and organismal levels, computational analyses of the genome sequence are significant. It is well discussed in the literature that codon usage is biased across all life domains, i.e., synonymous codons occur at different frequencies in different organisms. So, some codons are preferred over others. The preferred codons correspond to more abundant tRNAs, and therefore, are translated more efficiently. Similarly, there are biases in dinucleotide usage or codon pair usage, with specific nucleotide pairs or codon pairs occurring at a much different frequency than expected based on the codon usage. The codon pair usage also appears to affect translation efficiency. Considering that viruses rely on the host-cell machinery for proper expression of their genes, a thorough characterization of codons, codon pair, and dinucleotide usage of SARS-CoV-2 can provide useful information regarding the expression potential of the viral genes and the fitness of the virus in its human or other hosts. Furthermore, viral attenuation can be achieved through extensive changes in codons, codon pairs, or dinucleotide usage of viral genes. Thus, in-depth analysis may guide the development of new vaccines against COVID-19.

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14. AUTHOR'S CONTRIBUTION

The manuscript was written by Dr. Satyabrata Sahoo. Miss Ria Rakshit collected the materials, the data and the references with regard to this work. All authors contributed to the final manuscript.

15. CONFLICTS OF INTEREST

Conflict of interest declared none

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