

The Microbiology of Coronaviruses

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Abstract

The end of 2019 marked the start of coronavirus disease (COVID-19) pandemic from China, which went on to envelope more than 190 countries and territories across the globe. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), from a group of betacoronaviruses, is responsible for COVID-19. The virulent factors include the presence of envelope and spike proteins having receptor bonding domains (RBD). Clinical manifestations can range from mild respiratory infections to fatal outcomes. The viability of virus ranges from 3 to 72 hours. Polymerase chain reaction (PCR) is the diagnostic test of choice in this pandemic situation. Due to the absence of specific antivirals and vaccine, adoption of preventive option can help to combat the specific life-threatening outcomes.

Keywords: Coronaviruses, SARS-CoV-2, Respiratory Infections, Receptor Binding Domains, Angiotensin Converting Enzyme 2, Pandemic.

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History of Coronaviruses

The discovery of coronaviruses (CoVs) can be traced back to 1960s. At that time, they were labelled as pathogens causing upper respiratory tract infections (URTI) in children.¹ In 1965, Tyrrell and Bynoe ascertained its presence from the tracheal aspirates of an adult patient with common cold. At that point, they were given the name B-814 group of virus. After initial unsuccessful results of viral cultures, they inoculated the infectious agent via intranasal route in healthy volunteers. The presence of common cold symptoms were considered to label these viruses as being the significant aetiology for said state. Afterwards they were categorised as organ culture viruses "OC", which were later given a name of coronavirus. The word corona means 'crown-like appearance having surface projections'.²

Group Characteristics

The virus prefers moderate climates. Therefore the

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frequency of respiratory tract infections is higher in winter and spring compared to summer and fall.²

Morphology

Coronaviruses belongs to a group of enveloped positive-sense RNA viruses. The RNA genome is considered to be the largest (26-33kb) amongst the entire family of RNA viruses. When viewed via electron microscope, the size ranges from 60nm to 140nm. The club-shaped spikes project on the surface, giving a crown-like appearance. Besides, the inimitable replication strategy makes it distinctive.^{3,4}

Serotypes

The coronaviruses are from a subfamily of Othocoronavirinae, in the family of Coronaviridae (Nidovirales).⁵ The Coronaviridae family comprises two subfamilies i.e. Coronavirinae and Torovirinae.⁴ Based upon the taxonomy communicated by International Committee on Taxonomy of Viruses (ICTV), and phylogenetic clustering, Othocoronavirinae are further divided into four genera i.e. the alphacoronavirus (alpha-CoV), betacoronavirus (beta-CoV), gammacoronavirus (gamma-CoV), and deltacoronavirus (delta-CoV). Amongst these, alpha and beta-CoVs can infect humans, mammals, bats, cats, mice, and pigs. Gamma- and delta-CoVs commonly infects birds, but occasionally mammals can be infected.⁵

Besides many serotypes, seven are commonly involved in human infections. Two of them i.e. human coronavirus-229E (HCoV-229E), and human coronavirus NL-63 (HCoV-NL63) belongs to alpha-CoVs. Human coronavirus OC-43 (HCoV-OC43), human coronavirus HKU1, severe acute respiratory syndrome (SARS-CoV-1), Middle East respiratory syndrome coronavirus (MERS-CoV), and novel coronavirus (SARS-CoV-2, nCoV-2019 or Wuhan coronavirus) belongs to beta-CoVs.⁵

Mild respiratory infections are usually seen with HCoV-229E, HCoV-NL63, HCoV-OC43, and HCoV-HKU1. However SARS-CoV-1, MERS-CoV, and SARS-CoV-2, can cause infection ranging from mild intensity to fatal outcomes. Amongst all these, SARS-CoV-2 is the microbe responsible for the current coronavirus disease (COVID -19).^{6,7}

Epidemiology

About 15% of common cold in adult population is due to coronavirus infection. With the passage of time, it was discovered that an individual of any age can be infected. It was also observed that it is responsible for 35% of the cases of respiratory infection epidemics.² WHO report for the year 2020 showed that older age group individuals along with the presence of comorbidities are at high risk for serious outcome. The highlighted comorbidities include malignancy, cardiovascular disease, chronic respiratory disease, and diabetes.^{2,8}

The COVID-19 pandemic is the result of SARS-CoV-2 infection. Like the previously known pandemic of influenza, the same Pandemic Intervals Framework (PIF) was observed for COVID-19. This progression outlines an investigation phase, case recognition, initiation, and acceleration and deceleration phases. The case peak usually appears at the end of the acceleration phase. It is then followed by deceleration phase in which the number of cases starts to decline. Depending on all the said phases, variation of PIF can be seen amongst various regions across the globe. The estimation of PIF can be done by identifying the transmissibility speed amongst specific community, availability of vaccines and other medical resources.²

Spread of infection

The mode of transmission is via respiratory droplets in most cases. However, other modes include contact with inanimate objects like surfaces, plastic, copper, stainless steel, and cardboards etc.⁷

Transmissibility

Coronaviruses harbour the error-prone RNA-dependent RNA polymerases (RdRP), mutations and recombination events. These sequences are the basis for acquiring evolution.⁵ The basic viral reproduction number i.e. R_0 (pronounced "R naught") is used to detect the viral transmissibility. The description of R_0 implies the approximate figure of persons acquiring infection in a community. This is especially true for the infection-free areas and those in which the persons are not vaccinated. Depending upon specific R_0 values, there can be three possibilities. First of all, if the value is <1 , it suggests every single infection can cause <1 new infection. In such situations the disease will disappear soon. If the R_0 equals 1, the disease will be transmitted and remain in active state, but with minimum chances of epidemic. In the third case, if R_0 is >1 , it suggests that cases will grow exponentially,

ultimately causing an epidemic or even a pandemic situation.⁹

Surface Stability

van Doremalen et al evaluated the surface stability for aerosols, stainless steel, plastic, cardboard, and copper.⁷ It was estimated that the viability of virus in aerosols was 3 hours. A reduction in infectious titre i.e. $10^{3.5}$ to $10^{2.7}$ for median tissue culture infectious dose (TCID₅₀) was observed per litre of air. The same TCID₅₀ per millilitre was observed in case of SARS-CoV-1, i.e. from $10^{4.3}$ to $10^{3.5}$. The stability on plastic and stainless steel was observed to be till 72 hours. Upon comparison with SARS-CoV-1, similar stability kinetics was observed. But the stability of both was more on plastic and stainless steel, as compared to copper and cardboard. For plastics, the reduction of TCID₅₀ per millilitre was $10^{3.7}$ to $10^{0.6}$ after 72 hours. For stainless steel it reduced from $10^{3.7}$ to $10^{0.6}$ per millilitre after 48 hours. The viability of SARS-CoV-2, on copper was 4 hours, while it was 8 hours for SARS-CoV-1. On cardboards, the viability of SARS-CoV-2 was 24 hours, while it was 8 hours for SARS-CoV-1.

Virulent Factors and Pathogenesis

Most of the coronaviruses are covered by an envelope having spike protein, which contains a specific variable receptor-binding domain (RBD). The RBD has affinity with angiotensin-converting enzyme-2 (ACE-2) receptor, especially those located in kidneys, heart, lungs, and gastrointestinal tract. This attachment facilitates the entry of the virus into target cells. Literature review supports the evidence that the RBD of SARS-CoV-2 was comparable to a mutated version of a similar virus i.e. RaTG13, upon genomic sequencing. It was believed to be sampled from bats (*Rhinolophus affinis*). This supports the theory that the origin of SARS-CoV-2 is from bats, which due to mutation can infect other animals and humans.¹⁰

The mutation is believed to increase the affinity of RBD to ACE-2 receptors in humans. But for pangolin, rodents and civets, the affinity for RBD to ACE-2 was decreased. Pangolins are alleged to be the intermediate host of SARS-CoV-2. There are many genomic similarities amongst SARS-CoV-1 and SARS-CoV-2. Therefore, biochemical interactions and pathogenesis are much similar as well. The binding of ACE-2 receptors with type II pneumocytes initiates a cascade of inflammation in lower respiratory tract. The complex formed by binding of SARS-CoV-2 spike proteins to ACE-2 receptors gets proteolytically processed with type 2 transmembrane protease

(TMPRSS2). This sequence of events leads to cleavage of ACE-2 receptors and activation of the spike proteins facilitating viral entry into the target cell. Furthermore, there could be direct viral invasion, antibody-dependent enhancement by IgG that were primed by previous human CoV, and hyper-activation of TH1 cells. Besides these, secondary bacterial infection because of destruction of epithelial receptors, can also be seen as a part of pathogenesis.¹⁰

Diseases

Humans and animals both can be infected with coronaviruses. In humans they can cause infection ranging from common cold or mild upper respiratory tract infection or even severe respiratory illnesses with high mortality rate. Mild gastrointestinal and genitourinary symptoms like diarrhoea and urinary tract infection can also be seen in some cases.³ Coronaviruses have been identified to infect mammals and birds including bat, mouse, alpacas, swine, dog, cattle, chicken and horses.⁴ As per description of According to the CDC guidelines for the year 2020, it is rare that animal coronaviruses can infect humans. But if such a situation arises, then transmission amongst the humans is easy. Common serotypes for such condition include MERS-CoV, SARS-CoV-1, and SARS-CoV-2. It was also identified that bats were the reservoir for these three viruses. Once the community gets infected, the condition is inclined towards epidemics and pandemics, like in current scenario of COVID -19.¹¹

Diagnostic Options for Coronaviruses

The knowledge regarding laboratory diagnostic options for coronavirus infections have progressed markedly. The advanced tests harbour high sensitivity and specificity that they are now comparable with conventional gold standard. Amongst all these, molecular tests by polymerization chain reaction (PCR) found a great place. Specimens like broncho-alveolar lavage, sputum and nasopharyngeal secretions are preferred ones for various tests. Blood, urine, stool and rectal specimens can also be used for PCR but have less significance. Antibody detection from serum is another test with a success rate of 15%. To date, the supportive evidence for false-positive and false-negative is limited. Therefore, there was emphasis on repeating the test twice, at least one week apart, either for PCR or serological identification. It is also suggested that the specimens should be stored and frozen at -70°C or below, maximally up to 72 hours, in order to maintain the viability of virus isolation.¹²

Treatment Option

To date, no treatment options are available for coronaviruses especially for SARS-CoV-2. However one anti Ebola drug, remdesivir, which is a nucleotide analogue, had shown good outcomes for both SARS-CoV-2 and MERS-CoV. However, the efficacy was found to be directly related to disease severity, viral load and extent of lung damage.¹³ Further studies and their results are awaited.

Vaccine

The absence of vaccine is the main reason to fight this group of viruses. Though inactivated vaccine was developed for SARS-CoV- 1 and MERS-CoV, they are yet to be approved for humans. Moreover, the earlier described presence of mutations for RBD to ACE-2 receptors is the biggest hindrance for the creation of vaccine for SARS-CoV-2.⁵

Prevention

In the absence of specific vaccine and antivirals, adoption of preventive option is the only way to combat this infection. The preventive options include maintenance of good hygiene, frequent hand washings, use of alcohol-based hand sanitizers, avoid touching of the face, use of bleach as surface disinfectant and avoiding visits to public places and social gatherings.²

The only possible way to combat the current pandemic is adoption of preventive measures. The result of all this will be reduction in morbidity and mortality rates from said infection.

Conclusion

SARS-CoV-2 belongs to a group of beta coronaviruses. The viability of the virus ranges from 3 to 72 hours on different surfaces. PCR is the test of choice. Due to absence of specific antivirals and vaccine, adoption of preventive option can help to combat the specific life-threatening outcomes.

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