



## Review Article

## Insight on Novel-COVID-19- A review

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## ABSTRACT

In late December 2019, an outbreak of nCOVID-19 has posed a significant threat to mankind including the international health and economy. The epidemic has covered all over the world. It has created a global pandemic and there is an urge to find approach to control the spread of the disease. So, the purpose of the review is to focus primarily on the CoV its pathogenic features, genome orientations, phylogenetic variation and also comment on the epidemiology focusing on INDIA based on the current evidences. The effective treatment is also a criteria rising to control the outbreak, in this regard the study also focused on the recent development of drug/vaccine candidate and the complication associated against nCOVID-19.

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## 1. Introduction

In December 2019, Wuhan, china reported an unknown outbreak of pneumonia which spread rapidly through the country within a month. The etiological research was carried out by the Chinese government along with the researchers as they took a rapid measures to control the epidemics. As its clinical features resembles that of viral pneumonia, an analysis was carried out by Centre for disease control (CDC) experts and confirmed as Novel coronavirus pneumonia (NCP).<sup>1</sup> Later WHO officially renamed the disease as COVID-19 where the International Committee on Taxonomy of Viruses (ICTV) named as acute respiratory syndrome coronavirus (SARS-COV-2).<sup>2</sup> Previous outbreaks of coronaviruses (CoV's) reported as severe acute respiratory syndrome (SARS)-CoV and Middle East respiratory syndrome (MERS)-CoV which caused an eminent threat to the public health.<sup>3</sup>

Coronavirus are the group of enveloped, positive sense single stranded RNA virus responsible for wide variations of diseases which involves respiratory, enteric, hepatic and

neurological that varies from severity of person.<sup>3,4</sup> Likewise SARS-CoV-2 possess potential natural hosts similar to the other viruses- natural, intermediate and final hosts. This may acts as a great challenges to prevention and treatment of the virus infected patient.<sup>5</sup> While comparing to the SARS and MERS, the virus has a high transmissibility and infectivity despite carrying low mortality rate.<sup>1,5,6</sup>

The chronology and the transmissibility of the COVID-19 is as follows. The first case was reported in December-2019. From December 18, 2019 through December 29, 2019, five patients were hospitalized with acute respiratory distress syndrome and one patient died out of it. By January 2, 2020, 41 were admitted to hospital confirmed of COVID-19 where less than half of patients suffered from underlying disease includes diabetes, hypertension and cardiovascular diseases.<sup>3,7-9</sup> These patients were presumed to have been carrying due to the nosocomial infection. Prior to this reports established till January 30, 2020, the WHO declared the COVID-19 outbreaks as a sixth public health emergency of international concern. As of March 11, 2020 WHO has declared as a global pandemic. Regardless of age, sex distribution, incubation period, clinical features and optimal treatment the important factors associated with COVID-19

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remain uncertain from all geographic regions and across the all the disciplines.<sup>1,2,10,11</sup> However, the transmission of COVID-19 through asymptomatic carrier via person to person contact was observe in many cases. Therefore, herein, we performed a literature review, focusing on the epidemiological characteristics, symptoms, pathogenesis, transmission pattern even the complication associated with nCov along with the therapeutics options in this early safe of the outbreak.

## 2. Morphology and the characteristic features

Coronavirus are pleomorphic or spherical enveloped viruses, approx. 150 to 160 nm in size; observed through electron microscope. The virus with positive single stranded RNA, nucleoprotein, capsid, matrix and S-protein. Nucleocapsid protein (N), membrane glycoprotein (M) and spike glycoprotein (S) plays an important role in infection. It differs from the other coronavirus as they possess an additional glycoprotein that has acetyl esterase and hemmagglutinin (HE) properties. Reports suggest that the antibodies generated against the N protein of SARS-CoV may cross react with COVID-19, whereas a heterophilic antibodies of SARS-CoV may not provide cross protection to COVID-19.<sup>8</sup> But these hetrophilic antibodies can be used for diagnostic purposes. The potential role of N protein found to counteract the host immune responses as a viral suppressor protein of RNAi (VSR). A considerable variation in the length of the CoV genome was found where it showed high variability for ORF1a/ORF1b and four structural proteins which is associated with the number and size of accessory proteins. Alternatively, genomes of CoV contains 6 open reading frame (ORF's). The first ORF's (ORF1a/b), is about two-third of genome length which encodes 16 non-structural proteins (nsp1-16). This nsp protein has a specific role in replication of CoV. As it was found that the mutation rate in the RNA virus is much higher than that of DNA virus, so the genome of RNA virus are usually less than 10K nucleoides. From the study it was known that the genome size of COV is approx. 30 kB and is the largest among the RNA virus. RNA processing enzyme such as 3'-5' exo-ribonuclease of nsp14 helps in maintenance of the genome. Apart from the S protein, CoV also carry M and E protein. The M protein is responsible for maintaining the shape, promotes the membrane curvature and binds to nucleocapsid having three transmembrane domain where E protein plays an important role in virus assembly and releases during pathogenesis. The nsp3 protein bind to N protein helps to package the encapsidated genome into virions using replicase-transcriptase complex (RTC).<sup>2,5</sup> (Figure 1)

The wide range of asymptomatic state to acute respiratory distress syndrome and multi-organ failure was observed in cases of COVID-19. The most common clinical symptoms includes fever, cough, sore throat, headache,

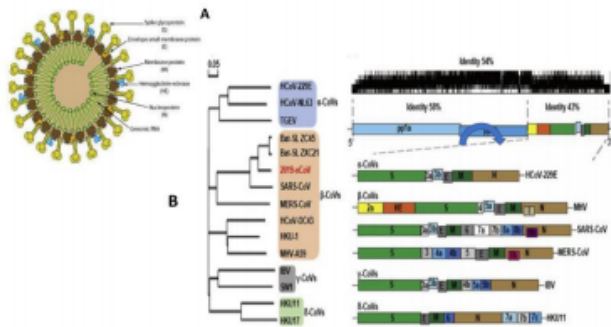
fatigue, headache, myalgia and breathlessness. The most common symptoms were found to be fever 30% and cough 38%.<sup>4,9,11</sup> Subsequently in some patients during the first week of the infection the diseases may progress to pneumonia, respiratory failure leading to death. Even the uncommon symptoms like sputum production, headache, hemoptysis and diarrhea was reported.

Based on the severity of symptoms, the patients may be classified as mild, severe and critical types. Mild patients may suffer from mild pneumonia to non-pneumonia. Apart form that the respiratory frequency was found to be  $\geq 30$ /min, blood oxygen saturation  $\leq 93\%$ , partial pressure of arterial oxygen to fraction of inspired oxygen ratio less than 300 and lung infiltrates that 50% within 24 to 48 hours.<sup>12,13</sup> Other clinical finding includes leucopenia and lymphopenia. Elevated levels of lactate dehydrogenase and creatinine kinase along with abnormal liver function with elevated level of alanine aminotransferase or aspartate aminotransferase was reported. C-reactive protein was responded high in the patients above the normal range. Study also investigated the rise in inflammatory cytokines including IL2, IL7, IL10, GCSF, IP10, MCP1, MIP1A AND TNF $\alpha$ .<sup>1,6,10,12,13</sup>

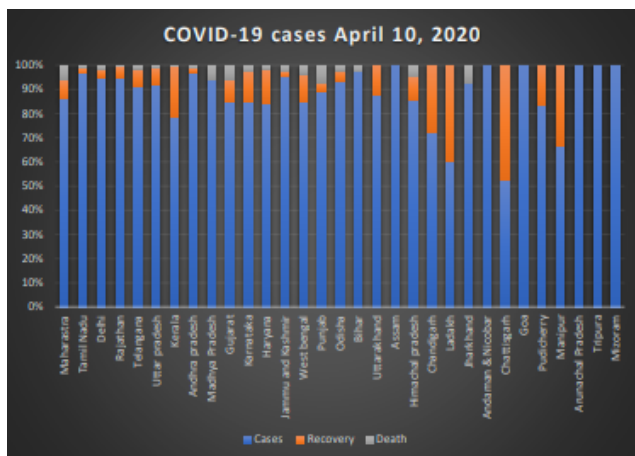
COVID-19 was classified as  $\beta$ -CoV of group 2B by World Health organization. A sequence similarity of 99.8% was obtained from the nine patients where 10 strains were isolated. Even the genetic similarity of 80% was found to be with SARS-CoV and 50% with MERS-CoV which was originated from bats. So from the phylogenetic point of view it was confirmed that COVID-19 belong to genus  $\beta$ -coronavirus under the subfamily orthocoronavirinae.<sup>14,15</sup> From the genetic sequence identity and phylogenetic reports, COVID-19 is sufficiently different form SARS-Cov and it can be considered as a new  $\beta$ -coronavirus that infects only the human. Further, an integrated sequence-based analysis was carried out from different geographical location in order to identify the unique pattern of infection which showed spike glycoprotein(S) plays a vital role in binding the virus to India showed variation from wuhan while Nepal does not showed any variation.<sup>16</sup> The comparison among the coronavirus with respect to the spike protein in 3° end is different ie depicted as 1273aa, 21493aa and 1270aa among the COVID-19, SARS-CoV and MERS-CoV. There occurs 79% similarity among SARS-CoV with COVID-19 and 50% with MERS-CoV. (Figure 2).

## 3. Epidemiology

As of April 9, 2020, data from the WHO revealed 1,596,496 cases of COVID-19. 184 countries have been affected with the confirmed cases including including mainland China, Japan, Singapore, Hong Kong Special Administrative Region (SAR), Thailand, South Korea, Taiwan, Australia, Malaysia, Germany, Vietnam, the United States, Macao SAR, the United Arab Emirates, Canada,

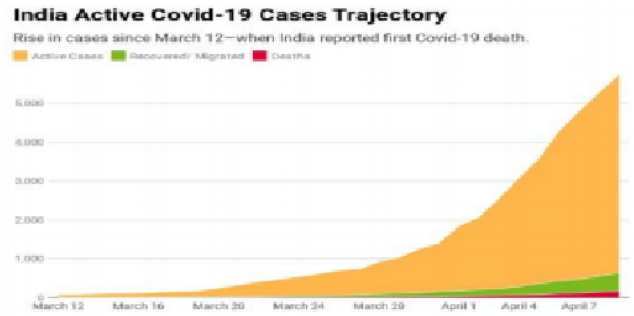


**Fig. 1:** Schematic Picture of coronavirus, B: phylogenetic tree of new COVID-19 where the genome structure of four of coronaviruses: two long polypeptides 16 nonstructural proteins have proceeded from Pp1a and pp1b represent. S, E, M, and N, are represented of the four structural proteins spike, envelope, membrane and nucleocapsid. COVID-19; CoVs, coronavirus; HE, hemagglutinin-esterase. Viral names: HKU, coronavirus identified by Hong Kong University; HCoV, coronavirus; IBV, infections bronchitis virus; MHV, murine hepatitis virus; TGEV, transmissible gastroenteritis virus.

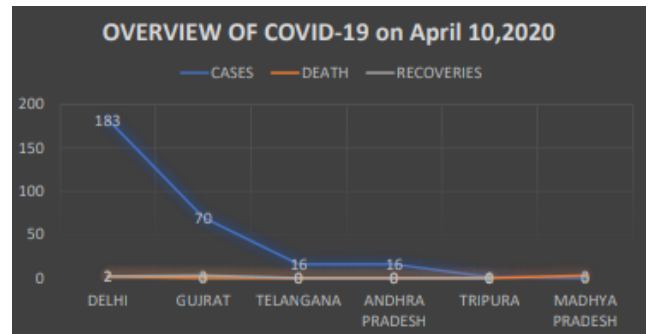


**Fig. 2:** Showing the rate of positive cases, recovery rate and death rate in the affected states of India till April 10, 2020. The highest number of suffered cases were observed in Maharashtra followed by Tamil Nadu where the highest recovery rate were seen in Kerala. The death cases were highest in Maharashtra followed by Punjab.

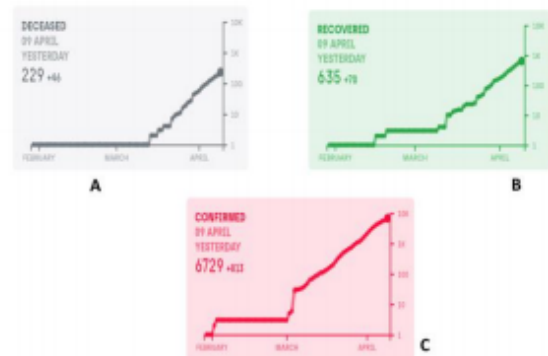
France, the Philippines, the United Kingdom, Italy, India, Russia, Finland, Sweden, Sri Lanka, Cambodia, Nepal, Spain, Belgium, Iran, Egypt, Israel, Lebanon and India. The number of deaths 94, 624 is reported to date all over the affected countries.<sup>3,9,12,13</sup> However, asymptomatic patients or patients with mild COVID-19 symptoms may not seek health care which leads to underestimation of the burden of COVID-19. Etiological investigations have been performed in the patients in hospitals due to similar viral pneumonia findings. On January 22, 2020, novel CoV has been declared to be originated from wild bats and belonged



**Fig. 3:** Active Cases reported in India from March 12, 2020 to April 7, 2020.



**Fig. 4:** A Demographic review of Covid-19 on April 10, 2020.



**Fig. 5:** Graph predicting the deceased person (A), recovery rate (B) and positive cases (C) in India from February to April (till date)

to Group 2 of beta-coronavirus that contains SARS-CoV. Although the reports suggested that the SARS-CoV and COVID-19 belongs to the same beta-coronavirus subgroup, with 70% genome similarity but has been found to show genetic differences from SARS-CoV which varies from locations. Often the transmissibility rate of COVID-19 infection is through the exposure to the virus and both the immune-compromised and normal population appears to be susceptible. According to the experts, the age distribution plays a vital role in susceptibility and recovery, where

predicted age distribution between 25 to 89 years old has been reported so far. The adult patients between 35 and 55 years old and fewer identified cases were found among the children and infants. A study on the early transmission dynamic pattern predicted that the virus reported at a median age of patients was 59 years, where majority being the male (59%).<sup>11,17,18</sup> The reports also suggested that the poor immunity in old person and those suffering from renal and hepatic dysfunctions are at the high risk and the survival rate is less as compared to the others. The average incubation duration of COVID-19 was estimated to be  $4.8 \pm 2.6$ , which may range from 2 to 11 days. According to the latest guidelines set by WHO the average incubation duration was found to be 7 days ranging from 2 to 14 days respectively. Concerning about India, 5,865 cases has been confirmed and 169 deaths has been reported so far among 32 states as of 10 April, 2020. Even though INDIA reported the first confirms case of COVID-19 on 30<sup>th</sup> January, 2020 in the state Kerala who had a history of travel from Wuhan, China. Till then highest has been reported in Maharashtra with 1,364 active cases carrying 97 as number of death. The first death due to coronavirus in INDIA was reported from Karnataka on 12<sup>th</sup> March, 2020 and thereby on 10 April, 2020, 25 death was reported from the state with a bulk of 229 positive cases being outnumbered. The biggest concern among the states was the slum with 8 lakh residents where 13 cases were reported earlier. Further, it was followed by Tamil Nadu carrying 738 active cases with 8 deaths as on 10 April, 2020. The regions in Kerala worst affected includes Kottayam, Kararagod, Pathnamthitta and Ernakulam. Among these regions Kararagod I one of the worst-affected regions recording 90 cases in the country. Bhilwara is one of the areas in Rajasthan where a high number of cases have been recorded. Two person with a travel history from Italy to delhi has been tested positive on 2<sup>th</sup> March, 2020. Till, 10 the April, 2020 Delhi, became the worst affected state in the list of COVID-19. Concerning about the situation and rising cases in INDIA, the government announced 21- days lock down to curb the spread of coronavirus. Even though, the case fatality rate among the SARS-CoV-2 infected patients in this study were 56% among the diabetic individual, almost 47% had hypertension and 16% carrying the cardiac diseases.<sup>19–22</sup> The three co-morbidities was found to be 17% with 40% cases examined to have pre-existing diseases condition. According to the ICMR survey, morbidity rate was 12% of urban and nearly 8% of rural population had a prevalence of diabetes and hypertension. While a combination of diabetes and hypertension is bad enough as a risk factor making as worse and increases the risk with the age. A nationwide manhunt happened after the religious gathering at Delhi Nizamuddin which has emerged as a hotspot and linked 7 COVID-19 deaths. Nearly 2,100 people has been evacuated where 50 were tested positive in Tamil Nadu, 18 in Andra Pradesh, 10 Andaman & Nicobar

Islands. Combining, the death rate of the confirmed COVID-19 ceases was found to be 1.38% (Figure II & III). There occurring alarming increasing rate of the number of cases in INDIA in a day (Figures 4 and 5).

#### 4. Transmission pattern

Many domestic and wild animals such as cats, dogs, bats etc. can acts as a reservoir for the coronavirus transmission. According to the reports, it was considered that the animal corona virus do not spread among the human. However, there are exception regarding the SARS and MERS which spread rapidly while in close contact with the infected person via respiratory droplets. According to the latest guidelines, the three main transmission route was observed in cases of COVID-19: droplets transmission, contact transmission and aerosol transmission<sup>22–24</sup>. Droplets transmission occurs through the droplets produced by an infected person when cough or sneezes. The contact transmission occurs when a person touches a surface contaminated with virus and hereby subsequently touching the mouth, eyes or nose. In case of aerosol transmission, when the droplets gets mixed with air and form aerosols which may cause infection once inhaled and gets deposited in lungs. Additionally research has also inked the SARS-CoV-2 and detected in the sample of stool, gastro-intestinal tract, saliva and urine.<sup>4,10,12,17,22,23</sup> Bioinformatics approach have predicted that the digestive tract might be a potential route of infection. However from the retrospective study it was also found that the pregnant women with COVID-19 had indicated the possibility of intrauterine vertical transmission between mother and infants in late pregnancy was found to be inadequate so, the vertical transmissibility was yet to be predicted.<sup>25</sup> Typically, the most respiratory virus were found to be contagious when people are mostly symptomatic but the asymptomatic carriers were also been reported.<sup>20</sup>

#### 5. Therapeutics/treatment option

Reports suggested some potential drug candidates against COVID-19 but the clinical effectiveness of these drugs has not been investigated. These includes Lopinavir, nucleoside analogs, neuaraamidase inhibitors, remdesivir, umifenovir, DNA synthesis inhibitors (Tenofovir, disoproxil and lamivudine), chloroquinine, ACE2-based peptides, 3C-like protease inhibitors, novel vinylsulfone protease inhibitor, teicoplanin etc. Although the effectiveness of remdesivir is evident and has been found as a promising candidate for COVID-19 invitro.<sup>14</sup> The contains an indole derivative molecule “Arbidol” which was found to block viral fusion against influenza A and B virus and also confirmed an antiviral effect on SARS-CoV in cell experiment<sup>7</sup>. Based on clinical trials, chloroquine was proved to be an effective against COVID-19.<sup>18,26</sup> Reports also suggested that the

hydroxychloroquine is effective in clearing the viral load in three to six days in most of the patients but the antagonistic effect of hydroxychloroquine was found to be more effect when combined with azithromycin.<sup>12,18</sup> In this regard, INDIA has supplied Hydroxychloroquine in 55 corona hit counties. Further, in the drug trials it has been found that the RNA-dependant RNA polymerase (RdRp) sequence of SARS-CoV-2 has showed 96% similarity with SARS-CoV, which can be a critical findings. Even the S protein was considered as a target molecule for designing anti-CoV. All such strategies adopted has shown promising result in-vitro and in-vivo. Even though the efficacy is not sufficient to support due to randomized animal or human trials. As there is no sufficient vaccines and specific drugs, the convalescent plasma therapy was found to be an effective way.<sup>6,24,27</sup> The patients recovered from COVID-19 would produce specific antibodies against the SARS-CoV-2 and their serum could be an meditative way to prevent the infection.<sup>7</sup> The future direction is a gateway toward the convalescent sera from the recovered patients. A study reports showed that the patient possess a specific IgM at day 9 after disease onset and switching to IgG by week 2.25. This showed a cross-reactivity with SARS-CoV and able to neutralize the virus by mounting humoral immunity. Eventually the T-cell response was found to be productive as the neutralizing antibodies high CD4 and CD8 and more levels of Th2 cytokines as compared to the infective person.<sup>28,29</sup> A single dose of 200 ml convalescent plasma with the neutralizing antibodies above 1:640 can provide supportive care and can acts as antiviral agents. An efficacy and adequately assessing such approach is essential but the adjustment should be made necessary by taking the account of ABO groups but the convalescent plasma can be administered directly to the patients at high risk of deterioration<sup>4,9,12,30</sup> (ie at the age 70 & dependence on oxygen with a baseline oxygen saturation of less than 94%r). The ICMR has approved the technique convalescent plasma therapy where Kerala may be the first state to start the clinical trials among its patients, Even though, closed monitoring should be maintained as regular intervals to detect a further side effects of the transfer.

## 6. Complications and challenges associated with COVID-19

Passive immunization becomes an important factor to reduce the virus replication and disease severity. But there exists some key challenges associated like viral kinetics, host interaction etc. even though it has been found that the spike protein plays a principle antigenic role inducing immune response in the host. Further, Angiotension-converting enzyme 2 (ACE2) utilize as a host receptor for its attachment and entry. But this ACE2 receptor was highly expressed in heart and lungs is also involved in the development of hypertension and diabetes mellitus.<sup>31</sup>

So, the treatment of patients with hypertension and suspected of coronavirus remains controversial. Even reports suggested that patients recovered from SARS-CoV-2 carries 44% of cardiovascular disease abnormalities with 60% glucose metabolism disorders. Therefore, a large proportion of death has been accounted for the patients infected with SARS-CoV-2 carrying cardiovascular disease. Acute coronary syndrome also remains one of the prime concern in SARS-cov-2.<sup>32,33</sup>

A reports suggested of blood purification techniques (plasma exchange and absorption) form critically ill-patients with COVID-19 showed a minimum managing level of cytokine generation; the patients with hypertension expressed multiple complication, even though patients without any underlying disease was found to be critically ill even after treating with Lopanvir, Ritonavir and other supportive therapies.<sup>34</sup> The risk found to be associated with cardiovascular disease goes to 40% when associated with COVID-19. 16.7% patients suffered from arrhythmias. One can expect high mortality rate with the patients associated with acute myocarditis confected with SARS-COV-2.<sup>33</sup> The co-morbidities is often found to be associated with COVID-19. Several clinical features like hypertension, diabetes, cardio-vascular disease and cerebrovascular disease remains a prime concern.<sup>35</sup> Neurological complications 'encephalopathy' also remains a pivotal part along with COVID-19. But, this has been associated only with the elderly patients. A case history of 74 years raised with atrial fibrillation, chronic obstructive pulmonary disease suffered an altered mental disturbance. The encephalomalacia was found as an abnormality with poor prognosis when the patients was treated with hydroxychloroquine.<sup>36</sup> Higher risk of complication was found to be associated with the patients carrying lung cancer with a difficulties to differentials the patients with COVID-19 in terms of clinical symptoms. So, this creates a dilemma to treat the patients and provide a cocktail therapy with cefaselis, oseltamivir, meropenim etc. there occurs a confounding factor and a great challenge for the prompt diagnosis and management among the lung cancer patients.<sup>37</sup> For the severe cases, intensive care should be supplemented but still no recommendation has been provided. Further, Artificial liver system (ART) becomes an effective way for treating the lung failure associated with COVID-19. Cytokine storm has been found as a propounding factor associated as it activated the T-cell resulting in multi-organ failure. So once GCSF, Ip10, MCP1, MIPIA and TNF $\alpha$  were found to be elevated, the ALS was recommended. Alternatively, detection of several cases of COVID-19 with clinical manifestation also remains a concern.<sup>38</sup> As false positive and false negative test results are likely to obtain. Hence, positive IgG and negative IgM has been considered as a recovery criteria epically among COVID-19 cases. According to WHO, Laboratory diagnosis of COVID-19

will be based on RT-PCR. The target genes obtained are ORF1ab, N, RdRp, E, N has been considered as essential. As RT-PCR is an expensive test, chest CT scan remains one of the diagnostic approach. Further, the sensitivity and specificity of a test depends on the severity of cases which may vary according to the different population. Moreover, false negative results in early epidemics and low quality of specimen remains as a main challenge.<sup>39</sup> In INDIA, ICMR (autonomous body) has adopted the E gene assay for screening and RdRp, N and ORF gene as a confirmation. As screening requires a positive controls so, in low number of samples a positive control has to be made so, In-vitro transcribed E gene was found to be limited. Apart from that weak signal from RdRp was also found. Hence, invitro transcribed RNA for the entire N gene was found to show a promising result.<sup>40</sup>

So, combating COVID-19 is equally important as cancer prevention. Even though a major progress has been made in terms of treatment still a significant promising result has to be obtained to understand this virus associated with other concerned disease.

## 7. Conclusion

The virus outbreak has challenged the medical, economic and public health care infrastructure facility all over the world. So, in future it will be likely to continue such outbreaks and can occurs through zoonotic origin, so the efforts should be made to prevent such outbreak by sticking to the measure like social distancing etc.

## 8. Source of Funding

None.

## 9. Conflict of Interest

None.

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