Life Science Informatics Publications

Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences

Journal Home page http://www.rjlbpcs.com/

RJLBPCS

ISSN 2454-6348

**Original Review Article** **DOI: 10.26479/2021.0701.04**

# IMMUNOPATHOGENESIS AND POSSIBLE THERAPEUTIC INTERVENTIONS FOR COVID-19: AN UPDATE

**S. Mateen[1], A. Mateen[2], A. Khalid[3], A. F. Faizy[4], S. Shahzad[4], S. Moin[4*]**

1.Department of Biochemistry, Faculty of Medicine, Jawaharlal Nehru Medical College, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India.

2. Department of Forensic Medicine, MRA Medical College, Saddarpur, Ambedkar Nagar, 224227, Uttar Pradesh, India.

3. Department of Biochemistry, Faculty of Life sciences, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India.

4. Department of Biochemistry, Faculty of Medicine, Jawaharlal Nehru Medical College, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India.

*** (S. Mateen, A. Mateen, A. Khalid) - Co-First Authors**

**ABSTRACT:** Coronavirus disease 2019 (covid-19) is a pandemic which is instigated by Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). So far around 9 million people are infected by SARS-CoV-2 across the world and this number is increasing at an alarming rate. SARS-CoV-2 uses ACE-2 receptor for entering into host cells and person-person transmission occurs via direct contact with infected individuals, contaminated objects/surface or respiratory droplets. Elderly people and those individuals with other complications are more prone to the development of this severe disease due to cytokine storm associated acute respiratory distress syndrome (ARDS). This review describes the virology of SARS-CoV-2, its transmission and immunopathagenesis. Further the potential anti-virals and immune-based drug therapies for covid-19 have also been discussed. Vaccines which are undergoing clinical trials for the management of covid-19 have also been highlighted.

**Keywords:** Covid-19; SARS-CoV-2; cytokine storm; acute respiratory distress syndrome; covid-19 vaccines.

**Article History: Received: December 25, 2020; Revised: January 06, 2020; Accepted: February 13, 2021.**

**Corresponding Author: Prof. Shagufta Moin*** Ph.D.
Department of Biochemistry, Jawaharlal Nehru Medical College, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India.
Email address: moinshagufta2@gmail.com

## 1.INTRODUCTION

A potentially fatal disease named as Coronavirus Disease 2019 (Covid-19) by World Health Organization (WHO) on 11 February, 2020 has been regarded as a major public health concern across the globe. Coronavirus (CoV) is regarded as the major pathogen which distresses humans lower respiratory tract and results in pneumonia. International virus classification commission named the novel coronavirus that caused covid-19 outbreak as Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Covid-19 is not the first coronavirus outbreak, rather other epidemic diseases such as Middle East respiratory syndrome (MERS)-CoV and Severe acute respiratory syndrome (SARS)-CoV have been reported in the past two decades [1]. In December 2019, a group of patients with pneumonia (fever, cough and dyspnea with acute respiratory distress syndrome) of unknown reason were observed in hospitals in Wuhan, Hubei province, China [2]. The seafood and wet animal wholesale market was identified as the source of infection thus leading to the shutting down of the area. However, during the spring festival the large influx of visitors resulted in the rapid spread of the virus to other parts of China. Later on, Covid-19 was declared as a Public Health Emergency of International Concern (PHEIC) and therefore a pandemic on 30 January, 2020 by WHO [3]. According to the latest reports (up to 22 December, 2020) the number of confirmed Covid-19 cases reached 77.3 million, out of which 43.6 million had recovered and 1.7 million had died.

### 1. Phylogeny and virology of SARS-CoV-2

CoV is RNA containing virus (single stranded and positive sense) which is enveloped in a lipid bilayer [4]. Four genera of CoV has been identified yet, with CoV infecting humans found in alpha (HCoV-229E and NL63) and beta (SARS-CoV. MARS-CoV, HCoV-HKU1 and HCoV-0C43) genera [5]. Genome sequencing of the virus obtained from five patients hospitalized in December, 2019 depicts the existence of the strain β of coronavirus in all of them [4]. The isolated genome of SARS-CoV-2 was found to be 96.2 % similar to the bat coronavirus isolate RaTG13 obtained from Yunnan province, China and 88% similar to 2 bat derived SARS-like CoV, SL-CoVZXC21 and SL-CoVZC45 collected from Zhoushan, China in 2018 [4, 6, 7]. Thus, bats are considered as most probable SARS-CoV-2's natural hosts owing to their close relationship with RaTG13. Furthermore,

SARS-CoV-2 showed seventy nine percent nucleotide sequence similarity to SARS-CoV and about fifty percent to MERS-CoV. It has been shown that SARS-CoV-2 genome is similar to a typical CoV and contains variable number of open reading frames (ORFs). Out of these ORFs, the first one (ORF1a/b) which comprises of the two-third viral RNA translates two huge polyproteins pp1a and pp1ab and encodes 16 non-structural proteins (NSP) [8]. The structural and accessory protein are encoded by rest of the ORFs. The ORFs in the other one-third of the genome encodes 4 structural proteins including spike (S) proteins, small envelope (E), nucleocapsid (N) and matrix (M) and other accessory proteins which interferes with the innate immune response of the host. The analysis of the SARS-CoV-2 genome obtained from several patients of different provinces in China revealed that the novel virus had been mutated in the patients [9]. Angeletti et al. showed that mutation in NSP2 and NSP3 plays an important part in the capability of infection and mechanism of differentiation of SARS-CoV-2 [10]. Tung Phan analysed the 86 whole or near complete SARS-CoV-2 genomes. Nucleotide sequence alignment showed 93 mutations in the whole genome of SARS-CoV-2 and 42 missense mutations in all the key structural and non-structural proteins (excluding envelope proteins). Three mutations ($F^{367}$, $D^{354}$ and $Y^{364}$) were found in receptor-binding domain of the spike protein [11]. The S glycoprotein plays an important role in viral binding to the receptor of host cell and is therefore the main target of neutralizing antibodies. Additionally, mutations in the S glycoprotein of SARS-CoV-2 may change its confirmation, thus allowing it to escape immune response by rapidly evolving and adapting to varying hosts in the future. The population genetic analysis of 103 genomes of SARS-CoV-2 revealed two broad types of SARS-CoV-2, one is L-type (70%) and the other S-type (30%) and the L-type strains were found to be more contagious and aggressive [12].

## 2. Transmission of SARC-CoV-2

SARC-CoV-2 is considered to be of zoonotic origin due to the exposure of large number of infected people to animal market in Wuhan where live animals are regularly sold. Earlier reports have identified two snake species which could act as reservoir of human covid-19. Apart from birds and mammals, no other reservoir of covid-19 has been reported [13]. Many studies have suggested person-person transmission to be the likely route of covid-19 spread. Person-person transmission takes place mostly by direct contact or via droplets (particles >5-10 μm in diameter) which spread by either sneezing or coughing of an infected person [14]. About 80% transmission is suggested to be contributed by pre- and asymptomatic individuals. This transmission is mostly limited to close contacts such as family members and healthcare professionals (6 feet). Owing to the virus mediated contamination of surfaces and objects, a recent study showed that this virus can be found on stainless steel for up to two-three days, plastic for up to two-three days, copper for up to four hours and on cardboard for up to one day. Covid-19 can be found on computers, trashcans, sickbed handrails and floors as well as in air up to four meters from patients in hospitals [15]. The mean incubation time

has been reported to be three-seven days (with median of 5.1 days and up to two weeks). This implies that SARS-CoV-2 doubled itself after around seven days and the basic reproduction number was found to be 2.2, that is, each patient transmits this virus to additional 2.2 people [16]. About 18% of the cases have been reported to be asymptomatic. Numerous studies have shown the potential of asymptomatic individuals to transmit infection to other people. 10 days have been reported for the resolution of symptoms. However viral shedding occurs even after disappearance of symptoms. SARS-CoV-2 viral shedding continues for eighteen days (nasopharyngeal swab) or nineteen days (feces). Viral shedding for asymptomatic and mild cases continues for 10 days (between 8-15 days) after disappearance of symptoms [14].

**3. Clinical features and diagnosis of covid-19**

The symptoms of covid-19 arise after an incubation period of 5.1 days. The time period between appearance of symptoms and death have been reported to be between six to forty one days (median of fourteen days) [17]. This period is dependent on the immune system of the infected person and his/her age. At the onset of covid-19, common symptoms includes fever, cough, fatigue, myalgia, dyspnea, sore throat, anorexia, productive sputum. However a smaller percentage of patients also display nausea, dizziness, diarrhea, headache and abdominal pain. The elderly and individuals with other complications (diabetes, hypertension, cardiovascular complication and chronic obstructive pulmonary disease) may rapidly develop ARDS, metabolic acidosis, septic shock which is hard to revert back and coagulation dysfunction eventually causing death [14]. Laboratory features include decreased or normal white blood cell count, lymphocytopenia and eosinopenia. An increase in alanine aminotransferase (ALT), prothrombin time (PT), lactate dehydrogenase (LDH), D-dimer, neutrophils, C-reactive protein (CRP), troponin, blood urea and creatinine have also been reported. The most common laboratory features are eosinopenia (78.8%) and lymphopenia (68.7%) [18, 19]. An increase in pro-inflammatory mediators including cytokines and chemokines have also been shown [19]. X-ray and chest CT-scans have shown ground-glass opacity and bilateral patchy shadowing in the lungs of moderate and severe patients [20]. On the basis of severity of symptoms, covid-19 can be approximately divided into four phases: mild, moderate, severe and critical. Infected individuals with only mild symptoms and without radiographic changes comes under mild phase. Patients with fever, radiographic features and respiratory symptoms comes under moderate phase. One of the three criteria should be fulfilled for a patient to be said in severe phase: RR>30 times per minutes; oxygen saturation< 93% in ambient air; $PaO_2/ FiO_2$<300 mm Hg. For critical phase one of the following feature is required: septic shock, respiratory failure, multiple organ failure [21]. ARDS is the most common complication of covid-19. About 48.2% of covid-19 cases develop ARDS. High mortality rate has been found in moderate and severe ARDS cases [22]. Age plays an important role in worsening ARDS as patients with age more than 65 years have been shown to have more severe ARDS and hence there is higher probability of mortality [23]. Elevated

LDH, blood urea nitrogen and reduced albumin level predicts the mortality of covid-19 ARDS individuals [23]. Some other complications associated with covid-19 includes diabetes mellitus, cardiovascular disease, hypertension and chronic kidney disease [22]. Since signs and symptoms of covids-19 patients are atypical therefore auxiliary examination is also done. Current diagnostic tests for covid-19 include auxiliary examinations such as detection of nucleic acid in samples taken from throat and nasal passage or in other respiratory tract samplings by real-time PCR, CT-scan, ELISA and point-of-care testing of IgM/IgG [24].

**4. Pathogenesis of covid-19**

Covid-19 patients show symptoms which are comparable to those reported in SARS-CoV and MARS-CoV infections. Since the pathogenesis of SARS-CoV-2 is not well understood, the similar mechanism of action of SARS-CoV and MARS-CoV might be informative for understanding the biochemical interactions and SARS-CoV pathogenesis. The S protein (glycoprotein expressed as homotrimer on the envelope of virus) of coronavirus plays an imperative role in defining the entry of virus into host cells [25]. There are two subunits of S protein. S1 consists of receptor binding domain, hence it determines virus-host range while S2 regulates viral fusion with the host cell membrane. The S protein binds with angiotensin-converting enzyme-2 (ACE-2) which is abundantly present in lungs, heart, kidney and adipose tissue [7, 26]. Structural analysis by cryo-electron microscopy showed that SARS-CoV-2 has 10-20 fold greater affinity for ACE-2 as compared to SARS-CoV, thus suggesting SARS-CoV-2 to be more contagious than SARS-CoV [27]. Direct membrane fusion between plasma membrane of the host and viral envelope has been reported for the entry of SARS-CoV into host cells (Figure 1). However, SARS-CoV entry into the host cell by clathrin dependent and independent endocytosis has also been reported [28]. After virus entry into cells, viral genome is released into the cytoplasm. The released RNA is translated into two polyproteins pp1a and 1ab which are then proteolytically cleaved into 16 non-structural proteins including RNA-dependent RNA polymerase, helicase, exonuclease, 3C-like protease (3CLpro) and papain-like protease (PLpro). After that, genomic replication and sub-genomic transcription occurs. Subgenomic RNAs serves as the mRNAs for structural and accessory proteins including S, E and M proteins. Nucelocapsid proteins and genomic RNA associate to form viral nucleocapsid and the genomic RNA and viral proteins are assembled into mature virions in Endoplasmic reticulum-Golgi intermediate compartment (ERGIC). Finally the mature virions are transported to the cell surface in vesicles and are released outside by exocytosis. [5]. The early onset of rapidly replicating virus results in the endothelial and epithelial cell apoptosis, vascular leakage leading to the release of pro-inflammatory mediators [29]. Furthermore SARS-CoV-2 may also cause pyroptosis in lymphocytes and macrophages which might be responsible for the observed peripheral blood lymphopenia in 82.1% patients [20]. After the entry of virus into host cells, ACE-2 downregulation occurs which in turn has been reported to induce shedding of ACE-2 ectodomain. Downregulation of ACE-2 results

in compensatory ACE mediated formation of angiotensin II which in turn stimulates its type 1a receptor leading to enhanced vascular permeability and lung pathology [1]. Loss of pulmonary ACE-2 has also been reported to be associated with acute lung injury, renin-angiotensin system (RAS) dysfunction and increased inflammation [30]. Furthermore SARS-CoV S protein mediated shedding of ACE-2 has been linked with the production of TNF-α in cell culture conditions [31]. For the initiation of immune response pathogen recognition receptor (PRR) detects the pathogen associated molecular pattern (PAMP) on viral RNA. In case of coronavirus, key PRRs are toll like receptors 7 (recognize RNA in endosomes), nucleotidyl transferase cyclic GMP-AMP synthase (cGAS) (recognition DNA in cytoplasm), retinoic-acid inducible gene I (RIG-I) and melanoma differentiation-associated gene 5 (MDA5) (recognize cytosolic ds RNA). [32] These signalling molecules recruit adaptors such as TIR-domain-containing adaptor protein including IFN-β (TRIF), stimulator of interferon genes protein (STING) and mitochondrial antiviral-signalling protein (MAVS). This is followed by the activation of transcription factor nuclear factor-κB (NF-κB) via adaptor molecule MyD88 which in turn results in the production of pro-inflammatory cytokines [32]. A significantly higher level of cytokines and chemokines such as IFN-γ, TNF-α, IL-1, IL-2, IL- 4, IL-7, IL-10, IL-12, IL-13, IL-17, hepatocyte growth factor (HGF), macrophage colony-stimulating factor (MCSF), GCSF, , IP-10, MIP-1α and MCP-1 have been found in Covid-19 patients [33]. The uncontrolled systemic inflammation caused by pro-inflammatory cytokines and chemokines (cytokine storm) by immune effector cells have been shown to cause ARDS which is considered as the leading cause of death in SARS-CoV and MERS-CoV patients. The main pathological change in ARDS is the infiltration of non-specific cell which results in pulmonary and interstitial tissue damage. Cytokine storm also determines the clinical course of extra pulmonary multiple-organ failure [34]. Therefore in covid-19 patients cytokine storm might also be responsible for damage to extra pulmonary organs and tissues (elevated creatinine and liver enzymes). It was found that the levels of TNF-*α,* GCSF, MCP-1, IP-10 and macrophage inflammatory protein-1A were higher in covid-19 patients in ICU as compared to those in general ward, thus suggesting the role of cytokine storm in enhancing the severity of disease [19]. IFN has an important antiviral role in the early stages of infection as the formation of IFN-I or IFN- *α*/ *β* is considered as the natural host defence response against viral infection. Upon activation of PRRs by either cellular damage or release of mitochondrial DNA by viral infection, initiation of signalling cascades leading to the production of protective type I IFN occurs [35]. However, in SARS-CoV and MERS-CoV, antiviral response is hindered due to the delayed release of IFN. Then, the cytokine and chemokine mediated attraction of inflammatory cells (monocytes and neutrophils) occurs which in turn is followed by the lung injury due to severe infiltration of inflammatory cells in lung tissue [36]. Activation of the cells of the myeloid lineage by PRRs or damage associated inflammatory stimuli, presentation of antigens to antigen presenting cells (APCs) via major histocompatibility complex (MHC) occurs. Both

CD8+T (recognize peptide presented by MHC I) and CD4+T (recognize peptide presented by MHC II) cells are required for viral clearance in SARS-CoV-1 infection animal model [37]. HLA molecules such as HLA-B∗4601, HLA-B∗0703, HLA-DR B1∗1202 and HLA-Cw∗0801 have been correlated with the susceptibility to SARS-CoV whereas HLA-DR0301, HLA-Cw1502 and HLA-A∗0201 alleles have been shown to impart protection from SARS-CoV infection. On the other hand, MHC II molecules, such as HLA-DRB1∗11:01 and HLA-DQB1∗02:0, impart susceptibility to MERS-CoV infection [24]. After the presentation of antigen, the humoral and cell mediated response of the body is stimulated by B and T lymphocytes respectively. Virally infected cells are killed by the activated CD8+T cells. On the other hand CD4+T cells are involved in the production of factors required for the help of B cells and antibody responses as well as enhancement of CD8+ T cell function. Studies suggests a reduction in the number of CD4+T cells and CD8+T cells in the peripheral blood of individuals infected with SARS-CoV-2. A typical pattern of IgM and IgG antibodies is observed for SARS-CoV infection [38]. The SARS-CoV specific IgM antibodies vanish after around 12 weeks while IgG antibodies lasts for much longer time [39]. These antiviral antibodies plays a vital role in the clearance of virus. However in case of SARS-CoV infection, the generation of neutralizing antibodies for S protein (anti-S-IgG) results in severe lung injury by changing inflammatory responses [40]. A greater chance of death was observed in patients who developed anti-S-IgG faster as the time required for neutralizing antibodies to reach their peak in deceased patients was 14.7 days while it was 20 days in recovered patients [41]. The appearance of antiviral IgG is related to the development of ARDS in 80% of SARS-CoV infected patients [42]. It has been shown that in lungs anti-S-IgG promotes inflammatory monocyte/macrophage accumulation and IL-8 and MCP-1 production. This inflammatory response is mediated by binding of virus-anti-S-IgG complex to the Fc receptors (FcR) present on monocytes/macrophages and FcR blocking resulted in diminished formation of inflammatory cytokines [40].

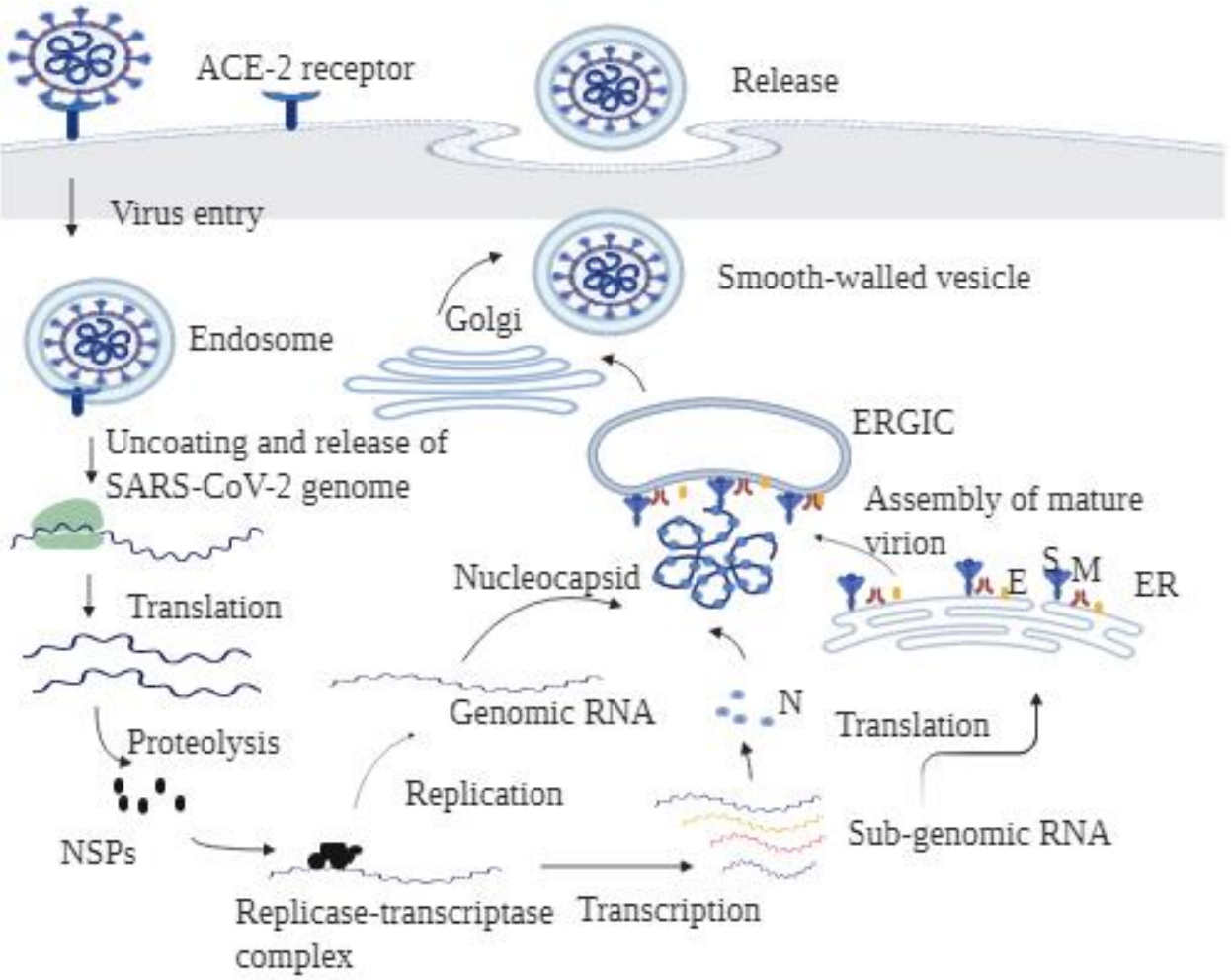


**Fig I.** Replication cycle of SARS-Cov-2.

## 5. Possible therapeutic interventions

High virus titre and subsequent inflammation due to heightened cytokine and chemokine levels leads to higher morbidity and mortality in SARS-CoV-2 infected individuals. Currently we don't have any clinically approved anti-viral drug or vaccine for the SARS-CoV-2 infection. The management of SARS-CoV-2 infection includes the use of broad-spectrum antibiotics for preventing secondary bacterial infection, fluid management and oxygen therapy. The genomic structure of SARS-CoV-2 and its molecular mechanism of infection enables the repurposing of existing anti-viral drugs or other therapeutic interventions against covid-19 (Table I).

### 5.1 Antiviral therapeutics

Remdesivir is an adenosine analogue which can block viral RNA replication by targeting RNA dependent RNA polymerase. It has shown promising results in many RNA virus infections (SARS, MERS) in cultured cells and animal models [43, 44]. It has also been shown to treat the first case of covid 19 in US [45]. Chloroquine has been used to treat malaria and hydroxychloroquine is a less toxic analogue of chloroquine which has been shown to be more effective for SARS-CoV infected

vero cells than chloroquine [46]. Both chloroquine and hydroxychloroquine have been shown to inhibit the pH dependent steps of viral replication. They also inhibit the glycosylation of ACE receptor, thus interfering with binding of the virus with host receptor [47]. They have immunomodulatory effects and hence they might inhibit the cytokine storm observed in covid 19 patients. A combination of remdesivir and chloroquine has been successfully shown to inhibit SARS-CoV in vitro [32]. Azithromycin has been used for the treatment of various bacterial infections. It imparts anti-viral effects by induction of IFN stimulated genes thereby inhibiting viral replication. It has also been shown to possess immunomodulatory effects by increasing neutrophil activation and decreasing the levels of pro-inflammatory cytokines. The combination of hydroxychloroquine and azithromycin seems to be potent for covid 19 [48]. Ribavirin is a guanosine analogue used for the treatment of numerous viral infections including respiratory syncytial virus and hepatitis C virus. By molecular docking, ribavirin was found to target the RNA dependent RNA polymerase of SARS-CoV and hence this may increase its antiviral potential against SARS-CoV-2 [49]. Faripiravir, a RNA polymerase inhibitor has been associated with enhanced viral clearance and more frequent radiographic improvement than ritonavir-lopinavir [50]. Protease inhibitors (ritonavir and lopinavir) which are used for the treatment of human immune deficiency virus (HIV) could be used for treating SARS-CoV and MERS-CoV infected patients [32]. Molecular docking showed that ritonavir and lopinarvir might inhibit protease 3CLpro, thus inhibiting replication of SARS-CoV-2 [51]. In Korea, the viral load of covid-19 patients were significantly decreased by ritonavir and lopinavir treatment [52]. Triple combination therapy consisting of interferon beta-1β, lopinavir–ritonavir and ribavirin was found to be safe and effective than lopinavir–ritonavir alone in reducing viral shedding duration and symptoms in patients with mild to moderate covid-19 infection [53].

**6.2 Immune-based therapy**

**Anti-cytokine therapy**

Anakinra (IL-1β antagonist) is an anti-cytokine inhibitor which inhibits binding of IL-1β with its receptor IL-1 type I receptor. During cytokine storm, most important cytokines of IL-1 family are IL- $\beta$, IL-18, and IL-33. Anakinra was found to produce good clinical outcomes as assessed by oxygen flow, reduction in CRP, no progression in infiltrates on serial CT scans in 9 hospitalized covid-19 patients. However, 3 patients showed elevated liver transaminase level [54]. Tocilizumab and sarilumab are IL-6 receptor antagonists while siltuximab is IL-6 antagonist. These drugs are now a days studied for the management of individuals suffering from covid-19. Increased serum levels of IL-6 have been found in covid-19 patients [55]. Therefore inhibition of IL-6 signalling might also supress the observed cytokine storm in covid-19 patients. A clinical trial (ChiCTR2000029765) with tocilizumab reported quick fever reduction and improvement of respiratory function in 21 covid-19 patients in China. Siltuximab was found to reduce CRP level

and improved clinical conditions in covid-19 patients who developed ARDS [56]. In a Phase 2/3 clinical trial, SARS-CoV-2 infected persons were grouped into three; first group received sarilumab 400 mg, second group received sarilumab 200 mg and third group received placebo. It was found that the fraction of patients who were on ventilator or who died was lowest in patients receiving sarilumab 400 mg followed by those receiving sarilumab 200 mg and placebo (*ClinicalTrials.gov* Identifier NCT04315298). For the control of coronavirus infection, repurposing of IFNs is another potential option for the development of effective drug. IFN-α and IFN-β have been shown to possess antiviral, antiproliferative and immune modulatory effects on several cell types [57, 58]. However no clinical data is available for SARS-CoV-2 infection.

**Convalescent plasma and antibody treatment**

Covid-19 convalescent plasma is the plasma obtained from the individuals who have recovered from SARS-CoV-2 infection and it includes antibodies against SARS-CoV-2. The use of convalescent plasma is limited to small case series, small retrospective cohort study, and case reports. Generation of monoclonal antibodies against SARS-CoV represents one effective way of treating covid-19 patients. Monoclonal antibodies neutralizing SARS-CoV such as CR3022, CR3014, m396 could prove to be an effective covid-19 treatment [59, 60].

**Corticosteroids**

Corticosteroids are steroid hormones which have anti-inflammatory function. Timely administration of corticosteroids have shown clinical improvements in SARS infection [55]. However dosage of glucocorticoid and its timing of administration are critical for severely ill patients. Initiation of body's immune response is inhibited by the too early administration of glucocorticoid thus increasing viral load and consequent adverse effects. Therefore, glucocorticoids are mostly given to severely ill patients experiencing cytokine storm. The timely administration of glucocorticoid in early stages of cytokine storm may prevent the inflammation induced ARDS and subsequent multiple organ failure [53]. Recently dexamethasone has been shown to decrease mortality in patients with COVID-19 requiring oxygen or ventilator support (ClinicalTrials.gov Identifier: NCT04381936).

**Signalling pathway inhibition**

Baricitinib is an inhibitor of janus kinase (JAK) and used for the treatment of rheumatoid arthritis. It is predicted to impede receptor mediated endocytosis of SARS-CoV-2 in AT2 alveolar epithelial cells of lungs [61]. However no clinical data is available for SARS, MERS, and covid-19.

**Blood purification treatment**

Blood purification system including blood/plasma filtration, plasma exchange, perfusion, adsorption etc., can remove inflammatory mediators and suppress the cytokine storm for the reduction of inflammation induced body damage. This therapy can be used in the early and middle stages of the disease of severe and critically ill patients. The artificial liver technology which was used for

reducing cytokine storm in H7N9 patients seems to be potent for covid-19 patients as well [62].

**Stem cell therapy**

Mesenchymal stem cells (MSCs) have been shown to possess anti-inflammatory and immune regulatory effects. They can hinder the hyper activation of macrophages and T lymphocytes and promote their differentiation into inhibitory macrophages and T regulatory cells, respectively. MSCs have been shown to produce anti-inflammatory factors which can suppress ARDS, resist fibrosis, regenerate and repair damaged lung tissue. Hence, many immune modulatory effects of MSCs can be expected in the treatment of covid-19 patients [63].

**Table I. Possible therapeutic intervention of covid-19 treatment**

| Drug | Mechanism of Action | Reference |
|---|---|---|
| Remdesivir | Interferes with viral replication (RNA dependent RNA polymerase) | 42, 43 |
| Chloroquine | Interferes with viral replication (glycosylation of cellular receptors, binding of virus to cell surface), immunomodulatory effects (reduction in pro-infammatory cytokines) | 46 |
| Hydroxychloroquine | Interferes with viral replication (virus-cell fusion, glycosylation of cellular receptors,), immunomodulatory effects (reduction in pro-inflammtory cytokines) | 46 |
| Azithromycin | Interferes with viral replication (induction of IFN stimulated genes), immunomodulatory effects (neutrophil activation, reduction in pro-inflammatory cytokines) | 47 |
| Ribavirin | Interferes with viral replication(RNA dependent RNA polymerase) | 48 |
| Faripiravir | Interferes with viral replication (3CLpro inhibitor) | 49 |
| Anakinra | Competitively inhibits IL-1 binding to the IL-1 type I receptor | 53 |
| Tocilizumab | Human recombinant monoclonal antibody (IL-6 receptor antagonist) | ChiCTR2000029765 |

| | | |
|---|---|---|
| Sarilumab | Human recombinant monoclonal antibody (IL-6 receptor antagonist) | NCT04315298 |
| Siltuximab | Human-mouse chimeric monoclonal antibody (IL-6 antagonist) | 55 |
| IFN-α and IFN-β | Antiviral, anti-inflammatory and immunomodulatory effects | 56, 57 |
| Convalescent plasma and antibody treatment | Modification of immune response | 58, 59 |
| Dexamethasone | Corticosteroid (modification of immune response) | NCT04381936 |
| Baricitinib | JAK inhibitor (interfere with receptor mediated endocytosis) | 60 |
| Blood purification treatment | Cytokine storm suppression | 61 |
| Mesenchymal stem cells | Anti-inflammatory and immune regulatory effects | 62 |

**Vaccines**

The development of effective SARS-CoV-2 vaccine is the need of hour for reducing severity of disease, viral shedding and transmission for controlling coronavirus outbreak. Many vaccination strategies such as live attenuated virus, inactivated virus, viral vectors, protein vaccines, subunit vaccines and recombinant DNA vaccines have been tested in animals against SARS-CoV and MERS-CoV infection [64]. Currently vaccines are not available for covid-19 treatment. Experimental vaccines which are being tested in humans for covid-19 are listed in table II.

**Table II: Most advanced covid-19 vaccine candidates.**

| **Candidate** | **Trial phase** | **Technology** | **Clinical Trial** |
|---|---|---|---|
| VPM1002 | Phase 3 | Other | NCT04435379 |
| MMR vaccine | Phase 3 | Other | NCT04357028 |
| Bacillus Calmette-Guerin (BCG) live-attenuated vaccine | Phase 2/3 | Other | NCT04328441 |
| AZD1222 | Phase 1/2/3 | Non-replicating viral vector | NCT04324606, NCT04335032 |
| mRNA-1273 | Phase 2 | RNA based vaccine | NCT04405076 |

| | | | |
|---|---|---|---|
| BNT162 | Phase 1/2 | RNA-based vaccine | NCT04380701 |
| Inactivated vaccine | Phase 1/2 | Inactivated virus | ChiCTR2000031809 |
| BBIBP-CorV | Phase 1/2 | Inactivated virus | ChiCTR2000032459 |
| CoronaVac | Phase 1/2 | Inactivated virus | NCT04352608 |
| V-SARS | Phase 1/2 | Inactivated virus | NCT04380532 |
| AV-COVID-19 | Phase 1/2 | Modified APC | NCT04386252 |
| Unnamed Inactivated SARS-CoV-2 Vaccine | Phase 1/2 | Inactivated virus | NCT04412538 |
| LV-SMENP-DC | Phase 1/2 | Modified APC | NCT04276896 |
| Gam-COVID-Vac Lyo | Phase 1/2 | Non-replicating viral vector | NCT04437875 |
| Ad5-nCoV | Phase 1 | Non-replicating viral vector | NCT04313127 |
| INO-4800 | Phase 1 | DNA-based vaccine | NCT04336410 |
| bacTRL-Spike | Phase 1 | DNA-based vaccine | NCT04334980 |
| NVX-CoV2373 | Phase 1 | Protein subunit | NCT04368988 |
| Covid-19/aAPC Vaccine | Phase 1 | Modified APC | NCT04299724 |
| SCB-2019 | Phase 1 | Protein subunit | NCT04405908 |
| COVAX-19 | Phase 1 | Protein subunit | NCT04428073 |

## 2. CONCLUSION

The occurrence and development of covid-19 is dependent on the interaction between SARS-CoV-2 and immune system of the host which in turn is dependent on the genetics, age, gender, physical and nutritional status and neuroendocrine system. These factors will decide the severity, duration and reinfection of covid-19. Scientists across the globe have made tremendous efforts for the characterization of novel SARS-CoV. Diagnosis of individuals suffering from SARS-CoV infection in the initial stages is of utmost importance for preventing the spread of this virus. Due to the non-availability of effective anti-viral and vaccine, self-isolation and quarantine are necessary. Since covid-19 is a pandemic, therefore coordinated international efforts are required for the development of potent therapy and vaccine against this deadly virus.

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

**HUMAN AND ANIMAL RIGHTS**

No Animals/Humans were used for studies that are base of this research.

**CONSENT FOR PUBLICATION**

Not applicable.

**AVAILABILITY OF DATA AND MATERIALS**

The author confirms that the data supporting the findings of this research are available within the article.

**FUNDING**

None

**ACKNOWLEDGEMENT**

None

**CONFLICT OF INTEREST**

The authors have declared that no competing interests exist.

**REFERENCES**

1. De Wit E, Van Doremalen N, Falzarano D, Munster VJ. SARS and MERS: recent insights into emerging coronaviruses. Nature Reviews Microbiology. 2016;14(8):523.
2. Lu H, Stratton CW, Tang YW. Outbreak of pneumonia of unknown etiology in Wuhan, China: The mystery and the miracle. Journal of medical virology. 2020;92(4):401-2.
3. Harapan H, Itoh N, Yufika A, Winardi W, Keam S, Te H, et al. Coronavirus disease 2019 (COVID-19): A literature review. Journal of Infection and Public Health. 2020.
4. Lu R, Zhao X, Li J, Niu P, Yang B, Wu H, et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. The Lancet. 2020;395(10224):565-74.
5. Perlman S, Netland J. Coronaviruses post-SARS: update on replication and pathogenesis. Nature reviews microbiology. 2009;7(6):439-50.
6. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A novel coronavirus from patients with pneumonia in China, 2019. New England Journal of Medicine. 2020.
7. Zhou P, Yang X-L, Wang X-G, Hu B, Zhang L, Zhang W, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. nature. 2020;579(7798):270-3.
8. Fehr AR, Perlman S. Coronaviruses: an overview of their replication and pathogenesis. Coronaviruses: Springer; 2015. p. 1-23.
9. Zhang L, Shen F-m, Chen F, Lin Z. Origin and evolution of the 2019 novel coronavirus. Clinical Infectious Diseases. 2020.
10. Angeletti S, Benvenuto D, Bianchi M, Giovanetti M, Pascarella S, Ciccozzi M. COVID-2019: the role of the nsp2 and nsp3 in its pathogenesis. Journal of medical virology. 2020;92(6):584-8.

11. Phan T. Genetic diversity and evolution of SARS-CoV-2. Infection, genetics and evolution. 2020;81:104260.
12. Tang X, Wu C, Li X, Song Y, Yao X, Wu X. On the origin and continuing evolution of SARS-CoV-2. Natl Sci Rev. 2020.
13. Bassetti M, Vena A, Giacobbe DR. The novel Chinese coronavirus (2019-nCoV) infections: Challenges for fighting the storm. European journal of clinical investigation. 2020;50(3):e13209.
14. Siordia Jr JA. Epidemiology and clinical features of COVID-19: A review of current literature. Journal of Clinical Virology. 2020:104357.
15. Guo Z-D, Wang Z-Y, Zhang S-F, Li X, Li L, Li C, et al. Aerosol and surface distribution of severe acute respiratory syndrome coronavirus 2 in hospital wards, Wuhan, China, 2020. Emerg Infect Dis. 2020;26(7):10.3201.
16. Cascella M, Rajnik M, Cuomo A, Dulebohn SC, Di Napoli R. Features, evaluation and treatment coronavirus (COVID-19). Statpearls [internet]: StatPearls Publishing; 2020.
17. Wang W, Tang J, Wei F. Updated understanding of the outbreak of 2019 novel coronavirus (2019-nCoV) in Wuhan, China. Journal of medical virology. 2020;92(4):441-7.
18. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, et al. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus–infected pneumonia in Wuhan, China. Jama. 2020;323(11):1061-9.
19. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The lancet. 2020;395(10223):497-506.
20. Guan W-j, Ni Z-y, Hu Y, Liang W-h, Ou C-q, He J-x, et al. Clinical characteristics of coronavirus disease 2019 in China. New England journal of medicine. 2020;382(18):1708-20.
21. Wang Y, Wang Y, Chen Y, Qin Q. Unique epidemiological and clinical features of the emerging 2019 novel coronavirus pneumonia (COVID-19) implicate special control measures. Journal of medical virology. 2020;92(6):568-76.
22. Liu Y, Sun W, Li J, Chen L, Wang Y, Zhang L, et al. Clinical features and progression of acute respiratory distress syndrome in coronavirus disease 2019. MedRxiv. 2020.
23. Wu C, Chen X, Cai Y, Zhou X, Xu S, Huang H, et al. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. JAMA internal medicine. 2020.
24. Li X, Geng M, Peng Y, Meng L, Lu S. Molecular immune pathogenesis and diagnosis of COVID-19. Journal of Pharmaceutical Analysis. 2020.
25. Li F. Structure, function, and evolution of coronavirus spike proteins. Annual review of virology. 2016;3:237-61.
26. Turner AJ, Hiscox JA, Hooper NM. ACE2: from vasopeptidase to SARS virus receptor. Trends in pharmacological sciences. 2004;25(6):291-4.

27. Song W, Gui M, Wang X, Xiang Y. Cryo-EM structure of the SARS coronavirus spike glycoprotein in complex with its host cell receptor ACE2. PLoS pathogens. 2018;14(8):e1007236.
28. Wang H, Yang P, Liu K, Guo F, Zhang Y, Zhang G, et al. SARS coronavirus entry into host cells through a novel clathrin-and caveolae-independent endocytic pathway. Cell research. 2008;18(2):290-301.
29. Yang M. Cell pyroptosis, a potential pathogenic mechanism of 2019-nCoV infection. Available at SSRN 3527420. 2020.
30. Imai Y, Kuba K, Penninger JM. The discovery of angiotensin-converting enzyme 2 and its role in acute lung injury in mice. Experimental physiology. 2008;93(5):543-8.
31. Haga S, Yamamoto N, Nakai-Murakami C, Osawa Y, Tokunaga K, Sata T, et al. Modulation of TNF-α-converting enzyme by the spike protein of SARS-CoV and ACE2 induces TNF-α production and facilitates viral entry. Proceedings of the National Academy of Sciences. 2008;105(22):7809-14.
32. Guo Y-R, Cao Q-D, Hong Z-S, Tan Y-Y, Chen S-D, Jin H-J, et al. The origin, transmission and clinical therapies on coronavirus disease 2019 (COVID-19) outbreak–an update on the status. Military Medical Research. 2020;7(1):1-10.
33. Chen C, Zhang X, Ju Z, He W. Advances in the research of cytokine storm mechanism induced by Corona Virus Disease 2019 and the corresponding immunotherapies. Zhonghua shao shang za zhi= Zhonghua shaoshang zazhi= Chinese journal of burns. 2020;36:E005-E.
34. Wang H, Ma S. The cytokine storm and factors determining the sequence and severity of organ dysfunction in multiple organ dysfunction syndrome. The American journal of emergency medicine. 2008;26(6):711-5.
35. Schneider WM, Chevillotte MD, Rice CM. Interferon-stimulated genes: a complex web of host defenses. Annual review of immunology. 2014;32:513-45.
36. Channappanavar R, Fehr AR, Zheng J, Wohlford-Lenane C, Abrahante JE, Mack M, et al. IFN-I response timing relative to virus replication determines MERS coronavirus infection outcomes. The Journal of clinical investigation. 2019;129(9).
37. Zhao J, Zhao J, Perlman S. T cell responses are required for protection from clinical disease and for virus clearance in severe acute respiratory syndrome coronavirus-infected mice. Journal of virology. 2010;84(18):9318-25.
38. Xu Z, Shi L, Wang Y, Zhang J, Huang L, Zhang C, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. The Lancet respiratory medicine. 2020;8(4):420-2.
39. Li G, Chen X, Xu A. Profile of specific antibodies to the SARS-associated coronavirus. New England Journal of Medicine. 2003;349(5):508-9.

40. Liu L, Wei Q, Lin Q, Fang J, Wang H, Kwok H, et al. Anti–spike IgG causes severe acute lung injury by skewing macrophage responses during acute SARS-CoV infection. JCI insight. 2019;4(4).
41. Zhang L, Zhang F, Yu W, He T, Yu J, Yi CE, et al. Antibody responses against SARS coronavirus are correlated with disease outcome of infected individuals. Journal of medical virology. 2006;78(1):1-8.
42. Peiris JSM, Chu C-M, Cheng VC-C, Chan K, Hung I, Poon LL, et al. Clinical progression and viral load in a community outbreak of coronavirus-associated SARS pneumonia: a prospective study. The Lancet. 2003;361(9371):1767-72.
43. Lo MK, Jordan R, Arvey A, Sudhamsu J, Shrivastava-Ranjan P, Hotard AL, et al. GS-5734 and its parent nucleoside analog inhibit Filo-, Pneumo-, and Paramyxoviruses. Scientific reports. 2017;7:43395.
44. Sheahan TP, Sims AC, Leist SR, Schäfer A, Won J, Brown AJ, et al. Comparative therapeutic efficacy of remdesivir and combination lopinavir, ritonavir, and interferon beta against MERS-CoV. Nature communications. 2020;11(1):1-14.
45. Holshue ML, DeBolt C, Lindquist S, Lofy KH, Wiesman J, Bruce H, et al. First case of 2019 novel coronavirus in the United States. New England Journal of Medicine. 2020.
46. Yao X, Ye F, Zhang M, Cui C, Huang B, Niu P, et al. In vitro antiviral activity and projection of optimized dosing design of hydroxychloroquine for the treatment of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Clinical Infectious Diseases. 2020.
47. Savarino A, Boelaert JR, Cassone A, Majori G, Cauda R. Effects of chloroquine on viral infections: an old drug against today's diseases. The Lancet infectious diseases. 2003;3(11):722-7.
48. Gautret P, Lagier J-C, Parola P, Meddeb L, Mailhe M, Doudier B, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. International journal of antimicrobial agents. 2020:105949.
49. Elfiky AA. Anti-HCV, nucleotide inhibitors, repurposing against COVID-19. Life sciences. 2020:117477.
50. Cai Q, Yang M, Liu D, Chen J, Shu D, Xia J, et al. Experimental treatment with favipiravir for COVID-19: an open-label control study. Engineering. 2020.
51. Nukoolkarn V, Lee VS, Malaisree M, Aruksakulwong O, Hannongbua S. Molecular dynamic simulations analysis of ritronavir and lopinavir as SARS-CoV 3CLpro inhibitors. Journal of theoretical biology. 2008;254(4):861-7.
52. Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. The Lancet. 2020;395(10223):507-13.

53. Hung IF-N, Lung K-C, Tso EY-K, Liu R, Chung TW-H, Chu M-Y, et al. Triple combination of interferon beta-1b, lopinavir–ritonavir, and ribavirin in the treatment of patients admitted to hospital with COVID-19: an open-label, randomised, phase 2 trial. The Lancet. 2020;395(10238):1695-704.
54. Aouba A, Baldolli A, Geffray L, Verdon R, Bergot E, Martin-Silva N, et al. Targeting the inflammatory cascade with anakinra in moderate to severe COVID-19 pneumonia: case series. Annals of the Rheumatic Diseases. 2020.
55. Ye Q, Wang B, Mao J. The pathogenesis and treatment of theCytokine Storm'in COVID-19. Journal of infection. 2020;80(6):607-13.
56. Gritti G, Raimondi F, Ripamonti D, Riva I, Landi F, Alborghetti L, et al. Use of siltuximab in patients with COVID-19 pneumonia requiring ventilatory support. MedRxiv. 2020.
57. Spiegel M, Pichlmair A, Mühlberger E, Haller O, Weber F. The antiviral effect of interferon-beta against SARS-coronavirus is not mediated by MxA protein. Journal of clinical virology. 2004;30(3):211-3.
58. Abdolvahab MH, Mofrad M, Schellekens H. Interferon beta: from molecular level to therapeutic effects. International review of cell and molecular biology. 326: Elsevier; 2016. p. 343-72.
59. Tian X, Li C, Huang A, Xia S, Lu S, Shi Z, et al. Potent binding of 2019 novel coronavirus spike protein by a SARS coronavirus-specific human monoclonal antibody. Emerging microbes & infections. 2020;9(1):382-5.
60. Zhang L, Liu Y. Potential interventions for novel coronavirus in China: A systematic review. Journal of medical virology. 2020;92(5):479-90.
61. Richardson P, Griffin I, Tucker C, Smith D, Oechsle O, Phelan A, et al. Baricitinib as potential treatment for 2019-nCoV acute respiratory disease. Lancet (London, England). 2020;395(10223):e30.
62. Xu K, Cai H, Shen Y, Ni Q, Chen Y, Hu S, et al. Management of corona virus disease-19 (COVID-19): the Zhejiang experience. Journal of Zhejiang University (medical science). 2020;49(1):0-.
63. Uccelli A, de Rosbo NK. The immunomodulatory function of mesenchymal stem cells: mode of action and pathways. Annals of the New York Academy of Sciences. 2015;1351(1):114-26.
64. Graham RL, Donaldson EF, Baric RS. A decade after SARS: strategies for controlling emerging coronaviruses. Nature Reviews Microbiology. 2013;11(12):836-48.