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A Comprehensive Study of Zoonotic Viruses: SARS-CoV, MERS-CoV, and SARS-CoV-2

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ABSTRACT

Coronavirus infections are responsible for mild, moderate, and severe infections in birds and mammals. Coronavirus is a respiratory viral infection. It is a spreadable and human-to-human transmission disease. It consists of different types of Coronavirus MERS, SARS infection, and SARS-Cov-2. Severe acute respiratory syndrome coronavirus (SARS-CoV) and the Middle East respiratory syndrome coronavirus (MERS-CoV), a third, highly pathogenic coronavirus, the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) appearing at end of 2019 led to a pandemic, increased panic, and attracted global attention. The new pandemic virus SARS-CoV-2 emerged in China and spread around the world in <3 months, infecting millions of people, and causing countries to shut down public life and businesses. The major clinical features include persistent fever, chills/rigor, myalgia, malaise, dry cough, headache, and dyspnoea. The high transmission rate of the virus has resulted in the current need for a fast and effective approach either to prevent or treat the infection. There is a need to improve our understanding immunology of this disease to developing vaccines and medicine for the prevention and treatment of patients. Here we discuss the characteristics, transmission, diagnosis, and treatment, etc.

INTRODUCTION

Corona virus is a single-stranded, highly diverse RNA virus with high virulence and mortality capacity which causes severe respiratory syndrome and due to this nowadays global public health is in an emergency.[1] Difficult days are being witnessed as the COVID-19 pandemic conditions continue to evolve, generating uncertainty and stress throughout the world. The emergence of infectious diseases throughout history has been the cause of suffering for many human groups, in addition to economic instability and disruption of daily life.[2] There are 7 identified human pathogenic coronavirus strains worldwide, among which two strains, i.e. Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) and the Middle East Respiratory Syndrome Coronavirus (MERS-CoV) can cause severe diseases.[3] Four of these strains, including Human Coronavirus 229E (HCoV-229E), Human Coronavirus OC43 (HCoV-OC43), Human Coronavirus NL63 (HCoV-NL63), and Human Coronavirus HKU1 (HCoV-HKU1), only cause relatively mild and self-limiting respiratory symptoms.

The first lethal coronavirus SARS-CoV emerged in 2002 in Guangdong Province, China.[4] In 2012, MERS-CoV emerged in Saudi Arabia. It caused two outbreaks in South Korea in 2015 and Saudi Arabia in 2018.[5] In December 2019, a new type of CoV was found that causes severe respiratory illness and first emerged in Wuhan, China. The World Health Organization named this novel virus SARS-CoV-2 and the as disease COVID-19 or Coronavirus Disease 2019.[6] The Coronaviridae is recognized as a novel virus family of enveloped and single-strand RNA (ssRNA) viruses. The Coronaviridae includes two subfamilies -the Coronavirinae and the Torovirinae.[7]

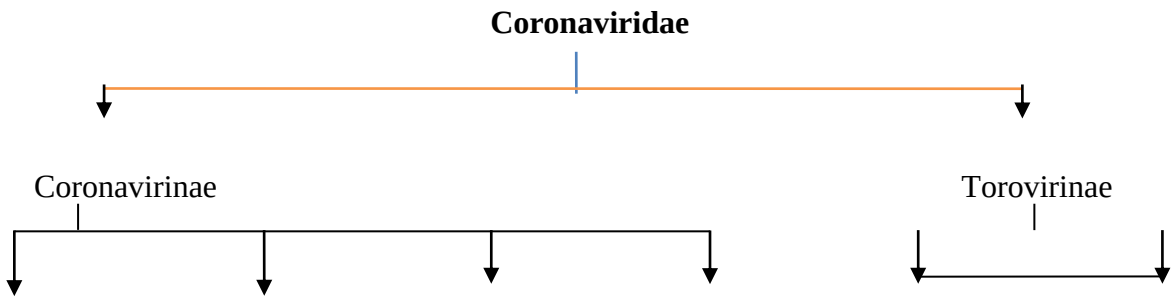

Table No. 1: Classification of Coronaviridae [7]

This review article aims to briefly summarize the knowledge and to provide an update of the major features of SARS-CoV, MERS-CoV, and SARS-CoV 2. We have included viral origin and evolution, cellular entry and viral transmission, incubation period and clinical manifestation, pathogenic response, diagnosis test, interpretation, and treatment.

General characteristics and virion structure:

1] SARS-CoV

The CoV family is named so because of the large spike protein molecules that are present on the virus surface and gives the virions a crown-like shape; coronavirus genomes are the largest among RNA viruses[8]. SARS-CoV has four main genera: Alpha-, Beta-, Gamma-, and Delta-coronavirus. SARS-CoV, SARS-CoV-2, and Middle East respiratory syndrome coronavirus are beta-coronavirus.[9] Within this, seven viruses are currently known to infect humans namely, NL63 and 229E from the alpha genus and OC43, HKU1, SARS-CoV, MERS-CoV, and SARS-CoV-2 from the beta genus. SARS-CoV is a positive-stranded RNA virus belonging to the family Coronaviridae.[10] It was characterized as a giant, enveloped, positive-stranded RNA virus with a genome comprising 29,727 nucleotides (~30 kb) and 41% of guanine or cytosine. The genomic body of this virus has the gene order of 5'-replicate (rep), which makes up approximately two-thirds of the genome and it consists of the large genes i.e. ORF1a and ORF1b. ORF1a and ORF1b of the rep gene encodes two large polyproteins known as pp1a (486 kDa) and pp1ab (790 kDa). Besides, the 3'structural spike(S), envelope(E), membrane(M), and nucleocapsid(N) proteins are encoded by four open reading frames (ORFs) downstream of the rep gene.[11]

2] MERS-CoV

Although MERS-CoV belongs to the same family, order, and genus as SARS-CoV, it was the first beta coronavirus lineage C member which is identified as a "novel coronavirus" with a genome size of 30,119 nucleotides. The genome of MERS-CoV encodes 10 proteins in their structure. These 10 proteins comprised of two replicate polyproteins (ORF1ab and ORF1a), four structural proteins (E, N, S, and M), and four nonstructural proteins (ORFs 3, 4a, 4b, and 5).[12] In addition to the replicase and structural genes, there are accessory protein genes interspersed between the structural protein genes that may interfere with the host's innate immune response in infected animals.[13]

3] SARS-CoV-2

Although SARS-CoV-2 also belongs to the same family and genus as SARS-CoV and MERS-CoV, the genomic analysis revealed greater similarity between SARS-CoV-2 and SARS-CoV. Thus, researchers classified it as a member of lineage B from the International Committee on Taxonomy of Viruses. Initially, the Coronaviridae Study Group of the International Committee on Taxonomy of Viruses identified this virus as a sister clade to the prototype human and bat severe acute respiratory syndrome coronaviruses (SARS-CoVs) of the species Severe acute respiratory syndrome-related coronavirus. Later, it was labeled as SARS-CoV-2.[14] SARS-CoV-2 has an RNA genome size of 30,000 bases in length. Among other beta-coronaviruses, this virus is characterized by the unique combination of polybasic cleavage sites, a distinctive feature known to increase pathogenicity and transmissibility in other viruses.[15] Genomic analysis of SARS-CoV-2 revealed that the genome consists of six major ORFs and it shares less than 80% nucleotide sequence identity with SARS-CoV. Genomic analysis also revealed that the SARS-CoV-2 genome has a high similarity to that bat coronavirus (Bat CoV RaTG13), with a sequence identity of 96.2%. Furthermore, the receptor-binding spike protein shares a 93.1% similarity to Bat CoV RaTG13. [19]

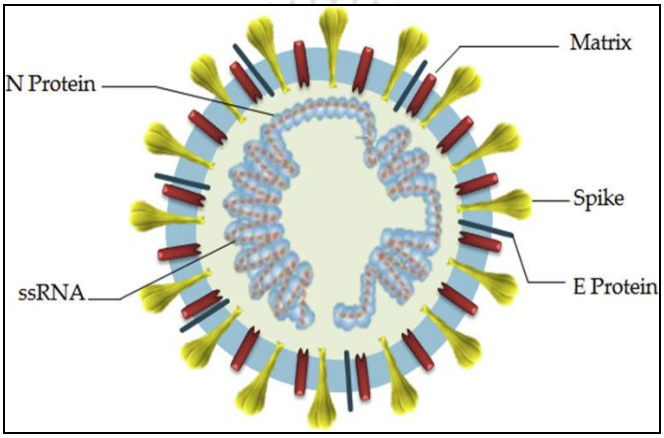

Fig No. 1: Structure of SARS-CoV [16, 17]

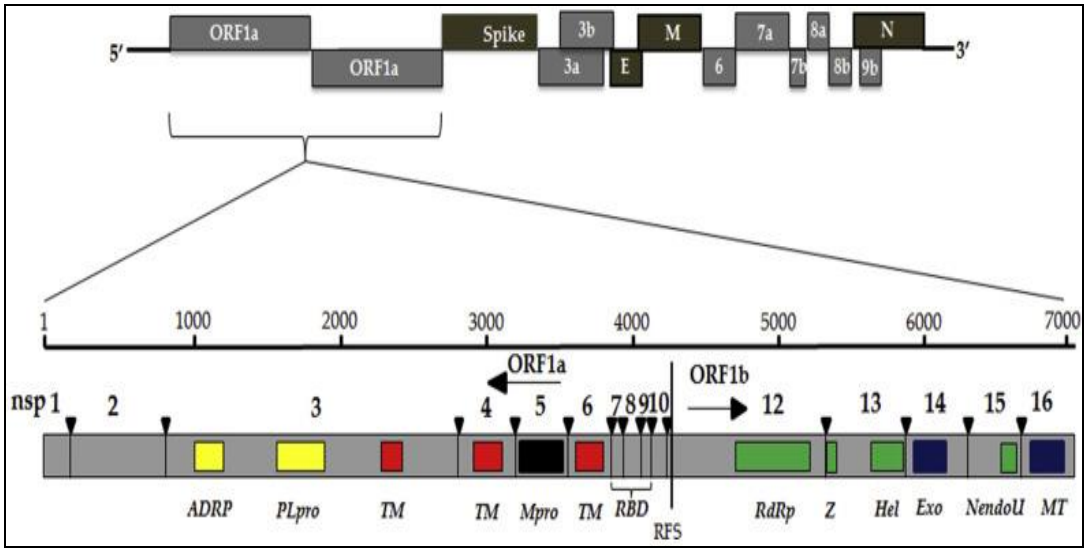

Fig. No. 2: Genome and nonstructural proteins of SARS-CoV [18]

Table No. 2: General Characteristics of SARS-CoV, MERS-CoV, and SARS-CoV 2 [20]

Characteristic	SARS-CoV	MERS-CoV	SARS-CoV 2
Year of the first reported case	2002	2012	2019
Country/Region of the first reported case	China	Middle east	China
Natural reservoir	Chinese horseshoe bats	Camels (possibly bats)	Unclear (possibly bats)
Intermediate host	Civet cats	Dromedary camels	Debatable (possibly pangolins)
Primary modes of transmission	Droplet, aerosol, and contact	Droplet, aerosol, and contact	Droplet, aerosol, and contact
Incubation period	2-7 days	2-14 days	2-14 days
Host receptor	ACE 2	DPP4	Unclear
Dominant cell entry pathway	Clathrin and caveolae-independent endocytic pathway	Cell membrane fusion	
Blood test results	Lymphopenia, thrombocytopenia, and leukopenia	Leucocytosis, monocytosis, and low CRP	Lymphopenia, thrombocytopenia, leukopenia, leukocytosis, monocytosis, and low CRP
Case fatality rate	≈15%	34.4%	1-3%

ACE 2: Angiotensin-converting enzyme; DPP4: Dipeptidyl peptidase-4 inhibitor; CRP: C-reactive protein

Viral origin and evolution

SARS-CoV

In the early stages of the SARS outbreak, mostly the new patient cases had an animal exposure before developing the disease. Investigators revealed that SARS-CoV strains were transmitted to palm civets from other animals. [21,22] Later, two studies reported that coronavirus related to human SARS-CoV or SARSr-CoV was found in horseshoe bat (genus Rhinolophus).[23,24] Bats are natural reservoirs for the virus and the palm civets are only the intermediate hosts.[25] SARS- CoV outbreak was produced by recombination within bats and then transmitted to palm civets or other mammals via fecal or oral transmission. When these virus-infected civets were transported to the Guangdong market, the virus spread among the civets and further mutation occurred before transmission to humans.[26]

MERS-CoV

Studies have shown that humans are infected through direct or indirect contact with infected dromedary camels. MERS-CoV strain isolated from camels and humans were almost identical.[27] MERS-CoV was highly prevalent in camels from the Middle East, Africa, and Asia.[28] MERS-CoV originated from bats and then transmitted to camels.[26]

SARS-CoV 2

The origin of SARS-CoV 2 and evolutionary crossover of the source between animals and humans are still obscure.[29] SARS-CoV 2 persists in the mystery of whether it was manmade in the lab or it was originated from a natural source.[30] The genomic and evolutionary analysis of pangolin-CoV with SARS-CoV 2 showed 91% and with bat CoV, RaTG13 showed 90.5% similarity, which suggested pangolin as an intermediate host of SARS-CoV 2.[31] This novel corona virus was first introduced in late December 2019 in Wuhan, the capital of Central China's Hubei Province, and then spread globally.[32]

Cellular entry and viral transmission

The spike glycoprotein not only acts as one of the requisite structure proteins of COVS but also plays an essential role in the interaction between COVS & host cells. This spike protein

consists of two subunits such as S1 and S2 subunit.[33] These subunits are responsible for viral fusion and exist in a non-covalent form.[34] The spike protein mediates viral entry into host cells by first binding to a host receptor through the receptor-binding domain (RBD) in the S1 subunit and then fusing the viral and host membranes through the S2 subunit.[35] SARS -COV, MERS-COV, & SARS-COV-2 these three viruses enter into the host cell by binding the receptor-binding domain to the functional receptors on the host cell surface.[36]

The dominant host receptor of SARS -COV is the Angiotensin-Converting Enzyme 2(ACE2).[37] The co-receptors involve in the entry of SARS-CoV are Neuropilins, heparan sulfate, sialic acids, or putative alternative receptors such as CD147 & GRP78.[38] Also, DC-SIGN (CD209) and L-SIGN (CD209L) are co-receptors of SARS-COV.[39] When the virus binds to DC-SIGN it does not cause SARS-CoV infection in dendritic cells but greatly enhances viral infection and dissemination. L-SIGN is also an alternative receptor for SARS-CoV because it can mediate cellular entry of SARS-COV by binding to its spike protein.[40] The cellular receptor of MERS -COV is dipeptidyl peptidase 4(DPP4, also termed as CD26).[41] Also, SARS-COV-2 enters into the host cell by binding its spike protein to ACE-2.[35,36] Compare to SARS-CoV, ACE -2 has a higher affinity to SARS-COV-2.[42] SARS -COV, MERS-COV, and SARS-COV-2 are employed by cellular serine protease TMPRSS2 and endosomal cysteine protease cathepsin B/L for spike protein priming, these are very important for them to enter into the host cells.[36] ACE-2 is vastly distributed in the respiratory tract, GIT, heart, Kidney & olfactory neuroepithelium. DPP4 expresses on the liver, thymus, prostate & bone marrow,[43] that results in broad cellular & tissue tropism of SARS -COV, MERS-COV &SARS-COV-2.[44]

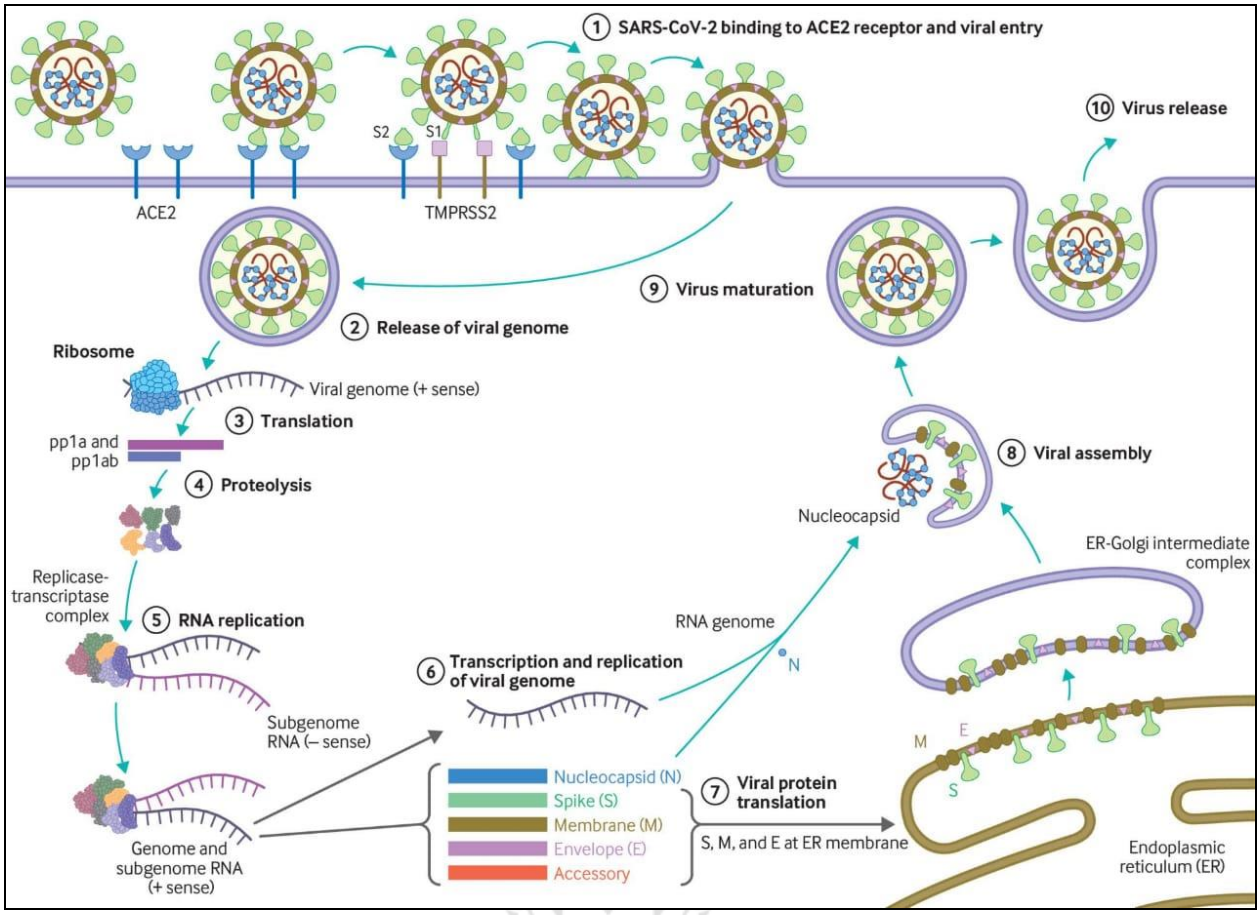

Fig. No. 3: Cellular entry and viral transmission of SARS-CoV 2

SARS -COV was transmitted by person-to-person close contact through inhaled air droplets and contaminated surfaces of devices.[45] The major irruption of SARS -COV is transmitted by airborne & another one was fecal contamination, feco-oral transmission.[46] The MERS-COV was transmitted by person-to-person but MERS-COV is not sustainably & frequently transmitted between humans. MERS -COV could be spread by contact with stool, vomitus, serum, urine & cerebrospinal fluid of patients.[43] Likewise, SARS-COV-2 is also transmitted from person to person.[47] SARS-COV-2 could be detected in saliva, sputum, urine, blood/serum, aerosol, etc.[48]

Incubation period and Clinical manifestation

The incubation period of the infectious disease varies from person to person. It is the period from the initial exposure to the infectious agent until the appearance of the signs and the symptoms of the disease.[49,50] Knowledge of the incubation period of viral infection is the key to investigate and control infectious disease. The incubation period of SARS-CoV is 4 days [51] and it could be longer with <10 days.[52] The incubation period of MERS-CoV is

5.2 days but in immunocompromised patients, the period could be longer.[53,54] The investigation showed that the incubation period of SARS-CoV 2 varies across different countries; the incubation period ranges from 1.8 to 12.8 days (mean) in China, 4 days (median) in Singapore, 3.6 days (median) in South Korea, and 4.9 days (median) globally.[55,56,57,58,59]

After the incubation period, many pathological abnormalities emerged due to weakened stabilization of endothelial cell to cell interactions, damaged integrity of vascular barrier and capillaries, diffused damage of alveolus, and multiple organ dysfunction[60], resulting in the onset of acute respiratory infection with the systemic disorder.[61,62] The clinical manifestations including sore throat, fever, dyspnea, cough, diarrhea, headache, and fatigue are pretty similar in SARS, MERS, and SARS-CoV 2.[63,64]

Immunopathology of Coronavirus

Immunopathology has been shown that SARS-CoV-2 disrupts normal immune responses, an impaired immune system, and uncontrolled inflammatory responses in severe and critical patients with COVID-19.[65] The hospitalized patients with severe COVID-19 indicated high levels of cytokines including IL-10, IL-7, IL-2, granulocyte colony-stimulating factor (G-CSF), C-X-C motif chemokine 10/interferon gamma-induced protein 10 (CXCL10/IP-10), monocyte chemoattractant protein-1 (MCP-1), macrophage inflammatory protein 1 alpha (MIP-1A), and tumor necrosis factor-alpha (TNF-α). These findings are in line with SARS and MERS in that presence of lymphopenia and “cytokine storm” may play a significant role in the pathogenesis of COVID-19. [66,67]

(A) When the SARS-CoV-2 virus invades the host, it is first recognized by the angiotensin-converting enzyme (ACE) 2 receptor present on respiratory epithelial cells allowing viral entry. Following viral replication within the cells, the virus is released where it is met by the host’s innate immune system. T lymphocytes and dendritic cells are activated through pattern recognition receptors (PRRs) including C-type lectin-like receptors, Toll-like receptors (TLR), NOD-like receptors (NLR), and RIG-I-like receptor (RLR). The virus induces the expression of numerous inflammatory factors, maturation of dendritic cells, and the synthesis of type I interferons (IFNs) which limits the viral spread and accelerates macrophage phagocytosis of viral antigens resulting in clinical recovery. However, the N protein of SARS-CoV can help the virus escape from the immune responses and overreaction of the immune system generates high levels of inflammatory mediators and free radicals. These

induce severe local damage to the lungs and other organs, and, in the worst scenario, multi-organ failure, and even death.

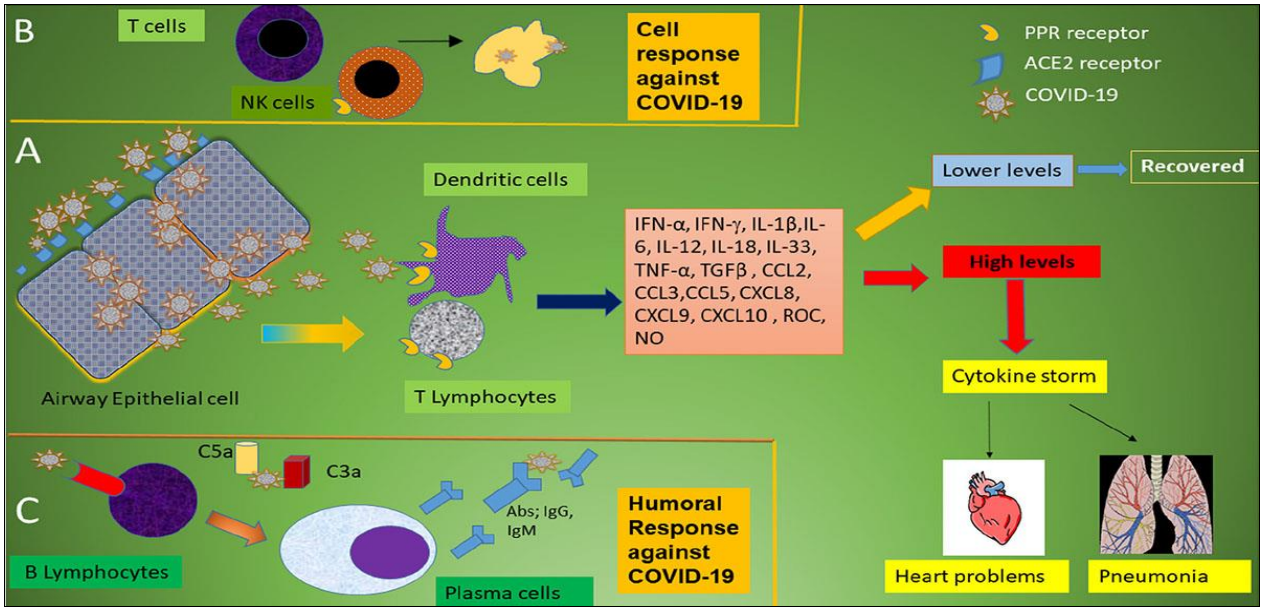

Fig. No. 4: Schematic immune response to CoVs [68]

(B) The adaptive immune response joins the fight against the virus. T lymphocytes including CD4 + and CD8 + T cells play an important role in this defense. CD4 + T cells stimulate B cells to produce virus-specific antibodies whilst CD8 + T cells can directly kill virus-infected cells. T helper cells produce pro-inflammatory cytokines to help the defending cells. However, SARS-CoV-2 can inhibit T cells by inducing programmed cell death (apoptosis).

(C) Humoral immunity including complement factors such as C3a and C5a and specific B cell-derived antibodies are also essential in combating SARS-CoV-2 infection.

In most cases, the immune response is capable of eliminating the virus; however, deficient immune response or respiratory hyper inflammation can lead to severe respiratory failure in severe COVID-19 cases.[69]

Adaptive Immune Response in SARS and MERS Infection

Antibodies, CD4+ T cells, and CD8+ T cells can each have protective roles in controlling viral infections, but the importance of each component of adaptive immunity varies depending on the viral infection.[70] SARS-CoV infection induces seroconversion as early as day 4 after the onset of disease and in MERS-CoV infection, seroconversion is seen at the second or third week of disease onset. For both types of coronavirus infections, delayed and

weak antibody response are associated with severe outcomes.[71] Depletion of CD4+ T cells is reduced pulmonary recruitment of lymphocytes and neutralizing antibody and cytokine production, resulting in a strong immune-mediated interstitial pneumonitis and delayed clearance of SARS-CoV from the lung.[72] Dendritic cells presented the viral antigens to T lymphocytes. T cells differentiate into different subtypes under the influence of secreted cytokines. Th1 helps CD8+ T cells to eradicate infected cells by secreting interferon-gamma. Th17 cytokines attract inflammatory cells to the infection site, and IL-4 produced by Th2 cells activates B cells to secrete neutralizing antibodies.[66]

Adaptive immune responses to SARS-CoV-2 infection:

It involves the aspects of innate immune response and T- and B-cell immunity and antiviral neutralizing antibody response.[69] SARS-CoV-2 infects human T-cell lines via a novel route through CD147 spike protein, present on the surface of T lymphocytes.[69] CD4+ memory T cells, upon re-stimulation, trigger B cells and other immune cells by cytokine production, while cytotoxic memory T cells help in destroying the infected cells during subsequent infection.[73] The SARS-CoV-2 infection leads to a robust antibody response within 7-14 days as IgM, IgG and IgA have been detected in almost all infected individuals and that IgG antibodies persist in the weeks following recovery. The antibodies most commonly detected are against the SARS-CoV-2 N and S proteins. [74]

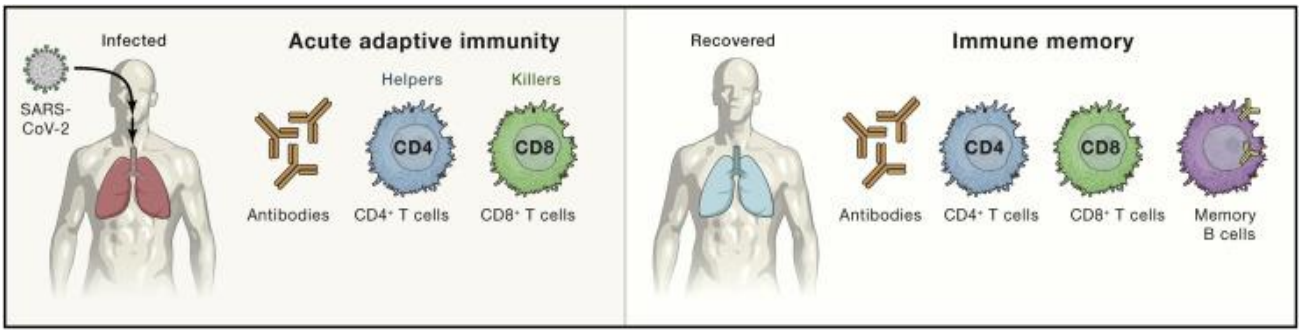

Fig. No. 5: The major components of adaptive immunity in viral immune

Innate Immune Response in SARS, MERS infection:

Immunological memory in the innate immune system is called “trained immunity” and may affect the spread and intensity of certain infections.[75] The recognition of virus infection begins with the identification of viral single standard RNA, double standard RNA by host cell pathogen recognition receptors (PRRs), which signal downstream via recruited adaptor

proteins, ubiquitin ligases, and kinases, culminating in transcription factors and the ultimate expression of immune genes, including IFNs, cytokines, and chemokines.[76] High serum levels of proinflammatory cytokines were observed in MERS-CoV and SARS-CoV infection, indicating a potential similar cytokine storm-mediated illness severity.[66] Several studies have been performed on cytokines secreted in SARS-CoV infection, and the result has shown that IP-10, IL-1, TNF-α, IL-6, IL-8, and MCP-1 were increased in the blood of the patients infected with SARS-CoV.[66] The innate immune cells express pathogen-recognition receptors (PRRs) to sense pathogen-associated patterns (PAMP) that include C-type lectin receptors, NOD-like receptors (NLRs), RIG-I-like receptors (RLRs), and Toll-like receptors (TLRs).[68] Both MERS-CoV and SARS-CoV, in human epithelial cell and fibroblast culture, show a delay (24–30 h post-infection) in the induction of proinflammatory cytokines with slightly different cytokine/chemokine profiles. This delay in cytokine induction was confirmed in another study using the same epithelial cell lines as well as in human alveolar type II cells. In both cell lines and primary alveolar type II cells, SARS-CoV induced IFN-β, IFN-λ, CXCL10, CXCL11, IL-6, IP-10, and TNF-α but MERS-CoV did not induce IFN-β but induced a higher level of IL-8 transcript in cell culture.[77]

Innate immune Response in SARS COVID-2 infection

ACE-2 is the primary receptor site of SARS-CoV-2 and is expressed on type 2 alveolar cells of the lungs.[78] SARS-CoV-2 could be detected by RLR and TLR pathways, and the cGAS pathway might also be involved, resulting in the activation of innate immune responses.[79] The innate immune system senses foreign material that is possibly pathogenic, and this triggers downstream signaling to ultimately induce transcription factors in the nucleus which in turn stimulate expression of types I and III interferons (IFNs) and other proinflammatory cytokines.[80] CoVs entry into human host cells, viral RNAs are released and act as pathogen-associated molecular patterns (PAMPs), which are recognized by pattern receptors (PRRs) as toll-like receptors (TLR3, TLR7, and TLR9) and retinoic acid-inducible (RIG-I) type I receptors.[81]

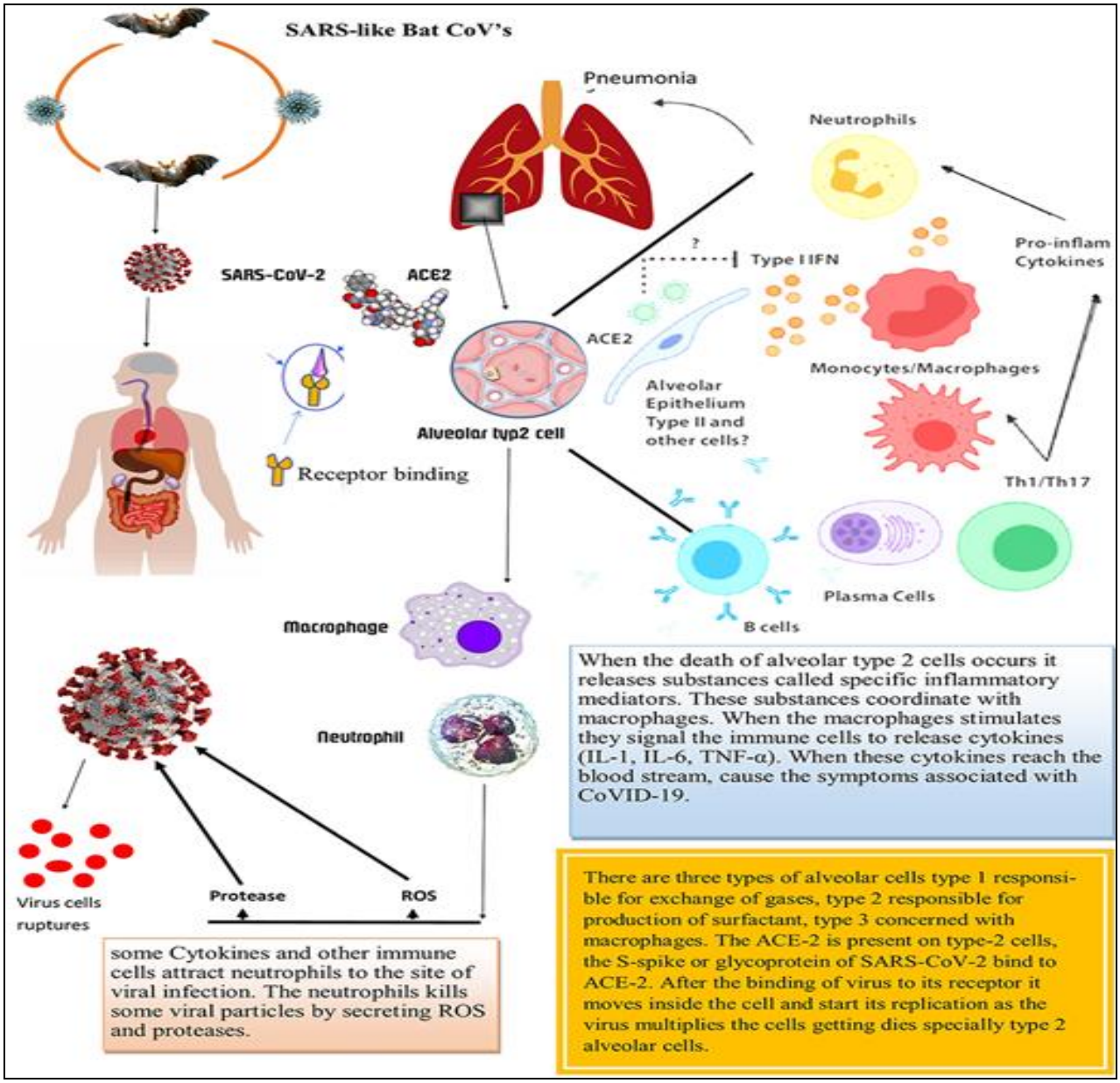

Fig. No. 6: Innate immune response to SARS-CoV-2[11]

Diagnosis

All over the world, emerging and reemerging infectious diseases are becoming very easily transmissible. The very important and first key to respond to an outbreak is early diagnosis. [82] Identification of virus mainly includes virus isolation and viral nucleic acid detection. According to Koch's postulate, virus isolation is the "gold standard" for the virus diagnosis in the laboratory. [83]

1] **Physical examination**

If the patient has mild symptoms, then positive signs may not be present. But if the patient is in severe condition may have moist rales in the lungs, shortness of breath, dullness in percussion increased or decreased tactile speech tremor and weakened breath sounds, etc.[84]

2] **RT-PCR**

This method is the most widely used nucleic acid test (NAT).[82] The real-time reverse transcription-polymerase chain reaction has become the primary and standard diagnostic test of SARS-COV, MERS-COV, and Covid 19. It has high sensitivity, specificity, and simplicity.[82,85] For rapid detection of virus kit was developed using fluorescent RT-PCR method and a hydrolysis probe. The kit consists of the probe, primers, and positive and negative control for detection. Three fragments of reverse transcription of segments of the virus were used as positive control and complementary human DNA were used as a negative control.[86] This method is used to detect and quantify multiple species from the sample.[87] Viral antigens, viral RNA, DNA, and biomarkers can be detected using this method.[88] For this test, the sample can be collected from the upper respiratory tract (oropharyngeal and nasopharyngeal) and lower respiratory tract (expectorated sputum, bronchoalveolar lavage, or endotracheal aspirate) of a patient suspected of viral infection.[89] In the early stage of infectious disease, the total number of leukocytes decreases or remains constant with decreased lymphocyte count or increased or normal monocytes also indicating the diagnosis of Covid-19.[90] RT-PCR has been recognized with a disadvantage to its relatively high cost.[91] In small-scale studies, it has been revealed that in patients with initial negative RT-PCR results, chest CT may reveal pulmonary abnormalities consistent with COVID-19.[92]

3] **CT scan**

Chest CT (Computed Tomography) is a non-invasive, conventional imaging modality with high accuracy and speed.[93] CT scan showed pulmonary lesions very clearly including ground-glass opacity and segmental consolidation in bilateral lungs, especially in the lung periphery.[83] Pleural effusion is common in MERS cases, but it is rare or only occurs in severe SARS and Covid-19 patients. In the Covid-19 disease, both lungs (multiple lobes, especially the lower lobes are involved) are simultaneously infected by SARS-COV-2. While in the initial period of SARS and MERS, lungs are more commonly involved in unilateral or unifocal than multifocal involvement.[94] To ensure accurate detection of cases and to

facilitate infection prevention measures, diagnostic algorithms based on a combination of RT-PCR and CT may prove to be necessary.[95]

4] **Pulmonary pathology**

SARS, MERS, and SARS-CoV 2 have non-specific pulmonary histopathological abnormalities. These changes are due to direct viral cytotoxic and immunopathogenic effects.[96] They are characterized by diffused alveolar damage (DAD), which includes two categories, acute exudative DAD and proliferative DAD. According to several SARS autopsy research, it is shown that SARS-CoV could damage multiple tissues and the major histopathology involves lungs.[97,98] The exudative traits of DAD were rarely seen while proliferative features of DAD became obvious. Specifically, in the early period of SARS infection exudative DAD is the predominant pulmonary pathology finding and proliferative DAD was additionally observed in the progress period.[99] The autopsy investigation of MERS has shown that DAD is the predominant pathological feature of MERS.[100] The major pathological feature of COVID-19 includes bilateral DAD as well as interstitial inflammation and fibrosis. The combination of DAD and fibrosis leads to the rapid deterioration of clinical conditions in severe COVID-19 cases.[101,102]

Interpretation of COVID-19 tests

The interpretation of a test for SARS-CoV-2 will depend upon a combination of the accuracy of the test to be performed and the estimated risk of COVID-19 before performing the test.[103] Test sensitivity is the first major property needed to properly interpret test results., which refers to the proportion of patients with the disease that the test correctly classifies as positive.[104] There are many reasons why an infection may not be detected by RT-PCR, ranging from whether the nasopharyngeal swab was taken properly, to the abundance of SARS-CoV-2 in the tested anatomical location, to when in the course of infection the sample is obtained.[105] False negatives are occurred due to clinical laboratory errors involving steps such as sample preparation, machine or operator error, and reporting errors.[106,107] For example, In the United State during the early stage of COVID-19a labeling error resulted in a patient receiving a false-negative test result, which in turn led to the patient being discharged from a hospital that might enhance the community disease transmission.[108]

Therapeutics and treatment options

No specific antiviral treatment or vaccine is available for the treatment of COVID-19. Molecular interaction between virus cell receptors and host cells with surface spike glycoprotein(s) is mainly important for the development of antiviral treatment. Treatment of severe influenza still presents multiple challenges.

1. Medicine therapy

Traditional Chinese medicine (TCM)

TCM is responsible for improving symptoms and decreasing the deterioration, mortality, and recurrence rates of COVID-19. Chinese scholars have proposed that TCM can control the dysfunction of ACE2 caused by a viral infection in multiple pathways. It can inhibit ribosomal proteins to obstruct viral replication, conferring a protective effect in humans.[109]

Chloroquine and Hydroxychloroquine

CQ is an amine anisotropic form of quinine and it has been used worldwide as a front-line drug for the treatment and prophylaxis of malaria [110]. HCQ is a 4-aminoquiniline analogue of CQ. The pharmacokinetics of HCQ is similar to CQ. [111] During long-term therapy, the clinical safety of HCQ is better as compared to CQ. Clinical responses and broad-spectrum antiviral effects of CQ and HCQ in SARS-CoV warrant particular attention for repurposing this drug for use in the treatment of COVID-19. [111, 112].

Remdesivir

Remdesivir is an adenosine nucleotide analog prodrug having broad-spectrum antiviral activity, is expected to become a potent drug for COVID-19. [113] It was developed by Gilead Sciences, Inc. in response to the Ebola outbreak in West Africa from 2014 to 2016. In its active triphosphate nucleoside form, remdesivir binds to ribonucleic acid (RNA)-dependent RNA polymerase and it acts as an RNA-chain terminator. It is having potent *in vitro* activity against SARS-CoV-2 with an EC50 at 48 hours of 0.77 μM in Vero E6 cells. [114] It has also had similar activity against other zoonotic coronaviruses with EC50 values of 0.07 μM demonstrated for both SARS-CoV-1 and MERS-CoV. [114-117]

Ivermectin

Ivermectin is a synthetic derivative of macrocyclic lactones commonly known as avermectins and it's having broad-spectrum antiparasitic activity. It is approved by the US Food and Drug Administration as an antiparasitic agent. [118] It inhibits the interaction between the HIV-1 integrase protein (IN) and the importin (IMP) α/β heterodimer which is responsible for importing the integrase protein. [119,120]. It also inhibits IN nuclear import and HIV-1 and DENV replication.[121]

Lopinavir/ritonavir (protease inhibitors) and antiviral drugs

Lopinavir/ritonavir are antiretroviral drugs having protease inhibitor action, widely used for the treatment of HIV, and recently suggested as potential candidates for the treatment of COVID-19. [122,123] Lopinavir/Ritonavir is a kind of viral replication inhibitor, was used for SARS patients [124] and it may be effective for SARS-CoV-2 infection.[125]

Nitazoxanide

Nitazoxanide has potent *in-vitro* activity against SARS CoV-2, with an EC50 at 48 hours of 2.12 µM in Vero E6 cells.[126] This potent activity is consistent with EC50 values for nitazoxanide and its active metabolite like tizoxanide acts against MERS-CoV in LLC-MK2 cells in which EC50 values of 0.92 and 0.83 µM, respectively.[127] It possesses broad-spectrum *in vitro* antiviral activity against influenza, rotavirus, respiratory syncytial virus, parainfluenza, and norovirus among others in addition to coronaviruses.[127] Due to its broad-spectrum antiviral activity, Nitazoxanide is being investigated for the management of influenza and other acute respiratory infections due to its broad-spectrum activity.[128]

2. Immunomodulatory therapy

There is much attention on the use of dexamethasone, tocilizumab, and anakinra for COVID-19.[129] Dexamethasone is a cheap and widely available steroid and it has such a large effect on reducing mortality of COVID-19 patients and it is expected to be an effective and affordable drug for treatment. Tocilizumab is the first IL-6 receptor inhibitor that has a significant effect on the treatment of COVID-19 patients.[130] A retrospective study with 29 COVID-19 patients in that respiratory function was improved among 72% COVID-19 patients after using high-dose anakinra.[131,132]

3. Specific immunotherapy

Vaccination

COVID-19 vaccine incorporating nucleic acid vaccine (including mRNA vaccine, DNA vaccine), recombinant genetic engineering (protein recombinant) vaccine, inactivated vaccine, attenuated influenza virus vector vaccine, and adenovirus vector vaccine are yet to be explored.[133,134] Cytotoxic T-lymphocyte cell epitopes and B cell epitopes on the surface of SARS-CoV-2 are prospective targets for the SARS-CoV-2 vaccine.[135] Some researchers think that the complete S protein or the S1 protein holding the RBD is an antigen that can be used for vaccine development[136]; however, some studies have pointed out that vaccines targeting antibodies against S2 linear epitopes may be more effective, because of less genetic mismatches providing SARS-CoV-derived antibodies ineffective compared with S1 subunit.[137]

CONCLUSION

SARS-CoV, MERS-CoV, and SARS-CoV these three highly pathogenic corona viruses that cause severe respiratory infection. The economic burden and health threats caused by coronaviruses are extremely dreadful. The emergence of a new coronavirus (SARS-CoV-2) outbreak represents a challenge for all healthcare professionals to control the source of infection, prevent the spread of the disease, and treat patients. The rapid and accurate detection of coronavirus is useful in preventing the spread of disease. Nowadays, RT-PCR and CT have become the first-line diagnosis for COVID-19 and it plays a very important role in managing the outbreak of SARS-CoV-2.

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