Features of Pathobiology and Clinical Translation of Approved Treatments for Coronavirus Disease 2019

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Keywords

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Abstract

Background: Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) is currently the most important etiological agent of acute respiratory distress syndrome (ARDS) with millions of infections and deaths in the last 2 years worldwide. Several reasons and parameters are responsible for the difficult management of coronavirus disease-2019 (COVID-19) patients; the first is virus behavioral factors such as high transmission rate, and the different molecular and cellular mechanisms of pathogenesis remain a matter of controversy, which is another factor. **Summary:** In the present review, we attempted to explain about features of SARS-CoV-2, particularly focusing on the various aspects of pathogenesis and treatment strategies. **Key Messages:** We note evidence for the understanding of the precise molecular and

cellular mechanisms of SARS-CoV-2 pathogenesis, which can help design the appropriate drug or vaccine. Additionally, and importantly, we reported the updated issues associated with the history and development of treatment strategies such as, drugs, vaccines, and other medications that have been approved or under consideration in clinics and markets worldwide.

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Introduction

The novel coronavirus-infected persons from Wuhan city, Hubei Province, China, were described in December 2019 [1]. To date, the acute respiratory distress syndrome (ARDS) related to novel coronavirus affected

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>200 countries, with millions of confirmed cases and deaths [2]. The coronavirus that was named as the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), and other human coronaviruses including OC43 (HCoV-OC43), human coronavirus HKU1, severe acute respiratory syndrome coronavirus-1 (SARS-CoV-1), and Middle East respiratory syndrome-related coronavirus (MERS-CoV) belong to the genus Beta coronaviruses [3, 4]. SARS-CoV-2 has high homology to bat coronavirus (~89%), and SARS-CoV-1 (80%), which indicate have common ancestry with viruses found in bats, and recombination occurred in intermediated hosts including Pangolin. However, there is not yet strong evidence for an intermediate host. The reproduction number (R_0) of SARS-CoV-2 was estimated to be 2.24–3.58, which is higher than that of MERS ($R_0 = 1$) and lower than that of SARS-CoV-1 (R_0 value of 2–5). Besides, the case fatality rate of SARS-CoV-2 is lower (3.4%) than that of SARS-CoV-1 (9.6%) and MERS (35%), and the incubation period of these viruses was found to be 1–14 days, with an average of 5 days [5, 6]. Clinical manifestation of different coronavirus is variable from the common cold to severe respiratory diseases, with high fever, cough, and multiple system dysfunction. The disease caused by SARS-CoV-2 is known as coronavirus disease 19 (COVID-19) [7]. The mechanism of SARS-CoV-2-induced pathophysiology is a multifactorial process, and is not fully understood. Advances in the prevention and effective management of COVID-19 will require detailed knowledge about SARS-CoV-2 pathogenesis [8]. The present study was performed to identify and evaluate the available data on different molecular and cellular mechanisms involved in SARS-CoV-2 pathogenesis that may be useful in the design of appropriate drugs or vaccines.

SARS-CoV-2 Features (Morphology, Genome Organization, and Its Proteins)

SARS-CoV-2 includes pleomorphic spherical particles of 70–90 nm diameter with coronavirus-specific morphology that were derived from clinical samples and seen under a transmission electron microscope [9, 10]. Coronaviruses are enveloped viruses containing an unsegmented, single-stranded, positive-sense RNA genome of around 30 kb in length, which is enclosed by a 5'-cap and 3'-poly (A) tail [10, 11]. The genome organization of SARS-CoV-2 has similarities to that of other beta-coronaviruses. SARS-CoV-2 genome is demarcated by short

RNA breakpoint sequences that lead to recombination at specific nonrandom locations within the viral genome, suggesting the evolutionary pattern of coronaviruses over vast distances in time [12].

The genome and subgenome produce 6 open reading frames (ORFs). The majority of the 5' end is occupied by ORF1a/b, encoding sixteen nonstructural proteins (NSP1-NSP16) [11, 13]. One large polyprotein is initially produced from ORF1a/b and cleaved by the papain-like protease encoded within NSP3 and the 3C-like protease, to produce replication-transcription complex, which are necessary for viral transcription and replication. The remaining ORFs encode for 9 putative accessory proteins and 4 structural proteins (Spike-S, Envelope-E, Membrane-M, and Nucleocapsid-N) (Fig. 1) [14]. The specific role and function of each protein in the life cycle of the virus are shown in Table 1. Phylogenetic analysis of the SARS-CoV-2 S gene sequence illustrates that there are distinguished 27 amino acid substitutes in contrast to SARS-CoV-1/SARS-like coronaviruses. These substitutions are about higher infectivity and lower pathogenicity of SARS-CoV-2 than SARS-like coronaviruses [15]. SARS-CoV-2 evolved 2 major types L and S that differ in 2 SNPs. These are at positions of 8782 and 28114 that are located in ORF1ab (T8517C, synonymous) and ORF8 (C251T, S84L), respectively [16]. In addition, L type was the most prevalent, detected in 70% of the samples amplified, and S type was detected in 30% of the specimens. L and S types of SARS-CoV-2 have very small genetic differences and may not influence the immune response [4].

Four major structural proteins in SARS-CoV-2 are mentioned in brief below:

(1) S is a large multifunctional transmembrane glycoprotein, and cleaved into S1 and S2 units. (2) Matrix glycoprotein (M) is the most abundant viral protein, which gave a definite shape to the viral envelope, and is essential for virus morphogenesis and assembly. (3) E is the smallest of the major structural proteins. It acts as a viroporin (ion channel) and is essential for various stages of the virus cycle, such as pathogenesis, assembly, and release of the virus. (4) N is the only structural protein that binds to the genomic RNA, and facilitates virion assembly, and enhances the transcription efficiency of the virus [17–20].

SARS-CoV-2 Pathobiology and Treatment Options

Binding to ACE2 and Entry

The first SARS-CoV-2 targets human cells, such as nasal and bronchial epithelial cells and pneumocystis,

Fig. 1. Genome properties of SARS-CoV-2. **A** The large replica polyproteins encoded by ORF1a/b are cleaved by the PL^{pro} and the 3CL^{pro}, to produce nonstructural proteins that are highly conserved throughout coronaviruses. **B** The S protein mainly contains

the S1 and S2 subunits. The S1/S2 cleavage sites are highlighted. This scheme is a mixed conclusion from a previous study [15–20]. PL^{pro}, papain-like protease; 3CL^{pro}, 3C-like protease; S, spike; ORF1a/b, open reading frame 1a/b; SARS-CoV-2, .

through the binding of viral structural S glycoprotein to the angiotensin-converting enzyme 2 (ACE2), as a zinc-containing metalloenzyme, which is widely expressed in many cells [21]. The attachment of receptor-binding domain (RBD) located on the surface of S glycoprotein to ACE2 prompts endocytosis of the virus [22]. The S1 subunit binds to the ACE2 via its RBD, and the S2 subunit is responsible for membrane fusion (Fig. 2) [3]. Additionally, the priming of the virus S protein is mediated by different co-receptors and activators, including transmembrane serine protease 2 and endosomal/lysosomal cysteine proteases such as cathepsin B and L. Taken together, these events can cause downregulation of ACE2, through internalization and degradation of the protein, which in turn results in the loss of cilia and squamous metaplasia, which contribute to severe lung injury [23, 24]. In addition to the ACE2 receptor, SARS-CoV-2 could bind the putative alternative receptor CD147 to enter target cells. Research has shown that when CD147 protein expression is inhibited, cell infection with the new coronavirus is reduced by 50% [5, 25].

According to the WHO and COVID-19 treatment guidelines, many antiviral agents are known today as “effective compounds” against the SARS-CoV-2, but here we investigated the NIH- or WHO-recommended antiviral agents that are available at <https://www.covid19treatmentguidelines.nih.gov> [26] for a better understanding of their antiviral properties on SARS-CoV-2. These drugs can target viral replication machinery, RNA polymerase, and viral protease, or modulate inflammatory responses against SARS-CoV-2 [27, 28]. Characteristics of approved and under development therapeutics options such as medication class, product name, clinical phase, manufacturing, mechanisms of action, dosage, and limitation are shown in Table 2 and Figure 3.

Reduction of viral loads in COVID-19 patients treated with some antiviral agents that can inhibit the binding of SARS-CoV-2 to host cells was found in various phases of clinical trials, indicating the inhibitory effect of these molecules on viral envelope proteins and their host cell receptors/co-receptors [31, 32]. These are including antiviral drugs (ivermectin), neutralizing monoclonal anti-

Table 1. Function of nonstructural and structural proteins of SARS-CoV-2

Protein name	Function
NSP1 (N-terminal product of the viral replicase)	Inhibition of host translation machinery and innate immune response (virulent factor)
NSP2 (N-terminal product)	Binds to PHBs 1, 2 (prohibitin), supposed role in apoptosis induction
NSP3 (papain-like proteinase)	Release NSPs 1, 2, and 3 from the N-terminal region of pp1a and 1ab
NSP4 (double-membrane vesicle maker)	Viral RTC and membrane rearrangement
NSP5 (main proteinase or 3CLpro)	Cleaves at multiple distinct sites of NSP polypeptide
NSP6 (putative transmembrane domain)	Induces the formation of autophagosomes
NSP7 (RNA-dependent RNA polymerase)	Part of the RTC, and forms complex with NSP8 and 12
NSP8 (multimeric RNA polymerase; replicase)	Part of the RTC, and forms heterodimer with NSP8 and 12
NSP9 (RNA-binding protein)	May bind to helicase
NSP10 (growth-factor-like protein possessing 2 zinc binding motifs)	Modulates NSP16, as a methyltransferase stimulator
NSP11	Unknown (consists of 13 amino acids and identical to the first segment of Nsp12)
NSP12 (RNA-dependent RNA polymerase)	Part of the RTC, and copies viral RNA and methylation (guanine)
NSP13 (RNA helicase)	Unwinds duplex RNA (helicase), part of the RNA polymerase complex, involved in virus replication
NSP14 (proofreading exonuclease)	Proofreading of the viral genome, which prevents lethal mutagenesis and functions as a methyltransferase for mRNA capping
NSP15 (RNA endonuclease)	Degrade RNA to hide from host defense
NSP16 (2'-O-ribose methyltransferase)	5'-cap RNA methylation
ORF3a	Interactions with some structural proteins and involved in virus release, apoptosis, and pathogenesis
ORF3b	Apoptosis stimulator, and inhibits the antiviral innate immune response
ORF6	Effective in viral pathogenesis, and inhibition of IFN induction
ORF7a	Apoptosis induction
ORF7b	Unknown (an integral membrane protein, expressed in viral-infected cells)
ORF8	May enhance replication and shows interaction with some structural proteins
ORF9b	Shows interaction with some NSPs and interferon antagonist
ORF10	Its function is not clearly understood but may have an immune modulatory role
ORF14	Unknown (consists of 73 amino acid residues)
S protein	Mediates attachment and viral entry into the host cell
E protein	It acts as a viroporin and is essential for stages of the virus cycle, such as pathogenesis, assembly, and release of the virus
M protein	It is essential for virus morphogenesis and assembly
N protein	It facilitates virion assembly and enhances the transcription efficiency of the virus

SARS-CoV-2, severe acute respiratory syndrome coronavirus-2; N, Nucleocapsid; M, Membrane; S, Spike; E, Envelope; ORF, open reading frame; NSPs, nonstructural proteins; RTC, replication-transcription complex. *Concluded from previous studies [17–20].

bodies (bamlanivimab and etesevimab), recombinant human monoclonal antibodies (casirivimab, imdevimab, and sotrovimab), and convalescence plasma [33]. At the beginning of the pandemic, several studies reported data on the antiviral activity of ivermectin, hydroxychloroquine alone, or in combination with azithromycin against SARS-CoV-2 [34–36]. The updates obtained from different trials with thousands of COVID-19 patients indicated these drugs do not reduce mortality or the duration of mechanical ventilation, and even cause adverse drug reactions [37]. Convalescent plasma or serum from a patient who recovered from COVID-19 could be another option for prophylaxis of infection and treatment of COVID-19 patients, particularly after the onset of symptoms

[38]. The antibody binds the S protein which prevents the entry of SARS-CoV-2 into the host cell and viral neutralization. In addition, the antibody modulates the inflammatory response, which is also more easily achieved during the initial immune response, a stage that may be asymptomatic [39]. There are reports that convalescent serum was used for the therapy of patients with COVID-19 in China during the current outbreak [40]. Recently, the connection of the SARS-CoV-specific human MAb CR3022 to SARS-CoV-2 RBD showed its potential as a remedial factor in the management of SARS-CoV-2. Indeed, it can be applied alone or in combination with other impressive treatments [41].

Fig. 2. The S protein of coronaviruses facilitates viral entry into target cells. The S protein of SARS-CoV-2 binds to ACE2 as the entry receptor, through its S2 subunit for viral attachment. The S protein is cleaved by the cellular serine protease that called TMPRSSs at the S1/S2 boundary or within the S1 subunit, which removes the structural constraint of S1 on S2, and releases the internal fusion peptide combined with the S TM domain for the viral fusion. This scheme is a mixed conclusion from a previous study [24]. S, spike; SARS-CoV-2, severe acute respiratory syndrome coronavirus-2; ACE2, angiotensin-converting enzyme 2; TMPRSSs, transmembrane serine proteases.

Translation and Replication

After the attachment, the ACE2/S SARS-CoV-2 complex is internalized into the cytoplasm by receptor-mediated endocytosis and prompts uncoating of virion in the acidic endosomal vesicles to release of the single-stranded viral RNA [42]. The positive single-stranded viral RNA translated into replicase polyproteins pp1a/pp1b and other products such as nsp1-16 collectively constitute the functional replication-transcription complexes by the host cell machinery [43]. Ribosomal frame shifting during the translation process has been seen in the replication of SARS-CoV-2, which produces genomic and multiple copies of subgenomic RNA species [23, 44]. The assembly of viral particles takes place via the interaction of genomic RNA and viral envelope proteins (S, E, and M) at the endoplasmic reticulum and Golgi complex [45]. Finally, these virions are subsequently released out of the cells via exocytosis [46]. It has been shown that several antiviral drugs influence the viral replication machinery in different ways: (i) directly targeting the viral proteins, such as RdRp and viral protease, and (ii) interruption of viral replication machinery through modulating cellular factors [47, 48]. Remdesivir, favipiravir, ribavirin, sofosbuvir, and tenofovir revealed the interaction and inhibition of RdRp, resulting in the reduced viral RNA synthesis and mRNA capping [49]. Remdesivir is the best example of a novel nucleotide analog with strong therapeutic applications against a diverse range of human viruses such as Ebola virus disease, SARS-CoV-1 and MERS, and SARS-like coro-

naviruses that inhibit viral RNA synthesis [50]. In addition, other inhibitors including lopinavir, ritonavir (Kaletra), and darunavir have been tested in clinical trials in the treatment of COVID-19 patients. This class of drugs interferes with the processing of the viral polyprotein by blocking the function of viral protease. Among these drugs, remdesivir is the only FDA-approved antiviral agent for the treatment of COVID-19 [51].

Inflammatory Responses

Virus replication (Viral phase) in pneumocytes leads to the inflammatory response, including macrophages, natural killer cells, CD4+T cells, cytotoxic T lymphocytes/CTLs, and antibody responses [52]. In later stages of infection, epithelial-endothelial barrier integrity is compromised, which potentially mediates lung injury, as well as extrapulmonary systemic involvement caused by SARS-CoV-2 [53]. Viral replication and pathobiology of SARS-CoV-2 virus are shown in Figure 4A–D. Several therapeutics plans modulate inflammatory responses against SARS-CoV-2 by different mechanisms. Approved and under evaluation plans include (1) immunomodulatory (colchicine, corticosteroids, interleukin inhibitors [IL-1 and IL-6], and interferons) and (2) cell-based therapy (mesenchymal stem cell) [54–56].

Pathology

Pathophysiology of SARS-CoV-2-induced ARDS is a multifactorial process and is very similar to SARS-CoV-1

Table 2. Drug options, which can be a candidate to treat COVID-19

Medication class	Product name	Examples for developer	Examples for clinical phases	Dosage	MOA	Limitation/side effects
Antiviral	Remdesivir	Gilead sciences	(NCT04280705) (different phases)	100 mg/day for 10 days	It is an adenosine analog that binds to the viral RNA-dependent RNA polymerase and inhibits viral replication	Gastrointestinal symptoms, ALT and AST elevations, hypersensitivity, increases in prothrombin time Liver function tests and prothrombin time should be obtained in all patients before remdesivir is administered and during treatment
	Ivermectin	Various	Phase 3 (NCT04834115) (different phases)	0.2 mg/kg single dose, maximum dose 18 mg	Interfering with intracellular transport process and attachment of the spike protein to the cell membrane	Generally well tolerated Dizziness, pruritis, nausea or diarrhea, and neurological adverse effects
	Anti-SARS-CoV-2 monoclonal antibodies	Bamlanivimab plus etesevimab	Phase 3 (NCT04497987) (different phases)	BAM 700 mg and ETE 1,400 mg IV administered together as a single dose	Neutralizing monoclonal antibody that binds to the RBD of the S protein (blocking viral attachment and cell entry)	Nausea, dizziness, rash, pruritis, pyrexia, hypersensitivity, including anaphylaxis and infusion-related reactions
		Casirivimab plus imdevimab	Phase 3 (NCT04452318) (different phases)	CAS 1,200 mg and IMD 1,200 mg IV administered together as a single dose	Recombinant human monoclonal antibodies that bind to the S protein RBD (blocking viral attachment and cell entry)	Hypersensitivity, including anaphylaxis and infusion-related reactions
	Convalescent plasma	Plasma from donors who have recovered from COVID-19	Phase 3 (NCT04361253) (different phases)	High-titer COVID-19 CP unit (about 200 mL) and based on the prescribing provider's medical judgment and the patient's clinical response	May contain antibodies that suppress the virus and modify the inflammatory response	TRALI, TACO, allergic reactions, anaphylactic reactions, febrile nonhemolytic reactions, hemolytic reactions, hypothermia, metabolic complications, transfusion-transmitted infections, thrombotic events, theoretical risk of antibody-mediated enhancement of infection and suppressed long-term immunity
Cell-based therapy	AdMSCs	Celltex Therapeutics Corporation	Phase 2 (NCT04428801)	Each subject receives 3 doses of 200 million autologous adipose-derived mesenchymal stem cells via intravenous infusion every 3 days	May reduce the acute lung injury and inhibit the cell-mediated inflammatory response induced by SARS-CoV-2	Uncommon Multiply or change into inappropriate cell types, tumor genesis, infection, and thrombus formation
Immuno-modulators	Colchicine	NHLBI; Bill and Melinda Gates Foundation; Government of Quebec	Phase 3 (NCT04322682) (different phases)	0.5 mg twice daily for 3 days then once daily for 27 days	Anti-inflammatory with reducing of the chemotaxis of neutrophils, inhibit inflammasome signaling and decrease the production of cytokines	Diarrhea, nausea, vomiting, cramping, abdominal pain, bloating, loss of appetite, neuromyotoxicity (rare), and blood dyscrasias (rare)
Corticosteroids	Dexamethasone (prednisone, methylprednisolone, hydrocortisone)	Various	Phase 3 (NCT04327401) (different phases)	Dexamethasone: 6 mg IV or PO once daily, for up to 10 days	Anti-inflammatory effects of corticosteroids might prevent or mitigate systemic inflammatory response that can lead to lung injury and multisystem organ dysfunction	Hyperglycemia, secondary infections, reactivation of latent infections, psychiatric disturbances, avascular necrosis, adrenal insufficiency, increased blood pressure, peripheral edema, and myopathy
	Fluvoxamine	Various	Phase 3 (NCT04668950)	Various dosing regimens used	Probably reduction in the production of inflammatory cytokines and expression of inflammatory genes	Nausea, diarrhea, dyspepsia, asthenia, insomnia, somnolence, and sweating

Table 2 (continued)

Medication class	Product name	Examples for developer	Examples for clinical phases	Dosage	MOA	Limitation/side effects
Interferons	Interferons Alfa	Cadila Healthcare Limited	Phase 2 (NCT04480138) (different phases)	Nebulized IFN alfa-2b 5 million international units twice daily	Stimulate the expression of several genes that contribute to shifting the host cells toward an antiviral activity	Flu-like symptoms(e.g., fever, fatigue, myalgia), injection site reactions, liver function abnormalities, decreased blood counts, worsening depression, insomnia, irritability, nausea, vomiting, and induction of autoimmunity
	Interferons beta	ShahidBeheshti University of Medical Sciences	Phase 4 (NCT04350671) (different phases)	IFN beta-1b 8 million international units subcutaneous every other day, up to 7 days total	Stimulate the expression of several genes that contribute to shifting the host cells toward an antiviral activity	Flu-like symptoms (e.g., fever, fatigue, myalgia), leukopenia, neutropenia, thrombocytopenia, lymphopenia, liver function abnormalities, injection site reactions, headache, hypertension, pain, rash, worsening depression, and induction of autoimmunity
Interleukin-1 inhibitors	Anakinra	Hellenic Institute for the Study of Sepsis	Phase 3 (NCT04680949) (different phases)	Dose and duration vary by study	Human IL-1 receptor antagonist	Neutropenia, anaphylaxis, headache, nausea, diarrhea, sinusitis, arthralgia, flu-like symptoms, abdominal pain, injection site reactions, and liver enzyme elevations
Interleukin-6 inhibitors	Sarilumab	Sanofi; Regeneron	Phase 2/3 (NCT04315298) (different phases)	400 mg IV (single dose)	Anti-interleukin-6 receptor monoclonal antibodies	Neutropenia, thrombocytopenia, GI perforation, HSR, increased liver enzymes, HBV reactivation, and infusion-related reaction
	Tocilizumab	Hoffmann-La Roche	Phase 3 (NCT04409262) (different phases)	A single dose of tocilizumab 8 mg/kg actual body weight IV The dose should not exceed tocilizumab 800 mg	Anti-interleukin-6 receptor monoclonal antibodies	Infusion-related reaction, HSR, GI perforation, hepatotoxicity, treatment-related changes on laboratory tests for neutrophils, platelets, lipids, and liver enzymes; and HBV reactivation
	Siltuximab	JuditPichMartínez	Phase 2 (NCT04329650) (different phases)	Dose and duration unknown	Anti-interleukin-6 monoclonal antibody	Infusion-related reaction, HSR, GI perforation, neutropenia, HTN, dizziness, rash, pruritus, and hyperuricemia
Kinase inhibitors	Acalabrutinib	AstraZeneca	Phase 2 (NCT04380688) (different phases)	Dose and duration unknown	Bruton's tyrosine kinase inhibitor that leads to immune and inflammation suppressing	-Hemorrhage, cytopenias, atrial fibrillation, and flutter, infection, headache, diarrhea, fatigue, and myalgia
	Baricitinib	Eli Lilly and Company	Phase 3 (NCT04421027) (different phases)	4 mg PO once daily for 14 days or until hospital discharge (for adults)	Janus kinase inhibitor that leads to immune and inflammation suppressing	-Lymphoma and other malignancies, thrombosis, GI perforation, liver enzymes, HSV reactivation, and changes in lymphocytes, neutrophils, Hgb, and liver enzymes

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BAM, bamlanivimab; ETE, etesevimab; CAS, casirivimab; CP, convalescent Plasma; TRAIL, transfusion-related acute lung injury; TACO, transfusion-associated circulatory overload; IV, intravenous; PO, by mouth; FLU, influenza; GI, gastrointestinal; HSR, hypersensitivity reaction; HBV, hepatitis B virus; HTN, hypertension; HSV, herpes simplex virus; Hgb, hemoglobin; MOA, mechanisms of action; COVID-19, coronavirus disease-2019; NIH, National Institutes of Health. *Concluded from different studies in the NIH (<https://www.covid19treatmentguidelines.nih.gov>) [29] <clear-delete> </clear-delete>

Fig. 3. Featured and critical data for approved and under development of therapeutics plans. These plans can treat COVID-19 patients in various stages such as attachment, entry, replication and hyper inflammation phase [80]. COVID-19, coronavirus disease 19; RBD, receptor-binding domain; SARS-CoV-2, severe acute re-

spiratory syndrome coronavirus-2; S, spike; ACE2, angiotensin-converting enzyme 2; TMPRSS2, transmembrane serine protease 2; 3CLpro, 3C-like protease; PLpro, papain-like protease; M, Membrane; N, Nucleocapsid; E, Envelope.

Fig. 4. Molecular and cellular mechanisms of SARS-CoV-2 pathogenesis, from air to the blood: SARS-CoV-2 adjusted to alveoli epithelial cell (A); the infection cycle of the SARS-CoV-2 starts with the binding of the virion to the receptor ACE2 via receptor-mediated endocytosis and its proliferation (B); immune responses to SARS-CoV-2 including (C) (1) macrophages that efficiently capture and kill viruses, and produce NO and cytokines; (2) NK cells that secrete cytokines and kill infected host cells that fail to express sufficient peptide-MHC class I and infected DCs; (3) CD4+ T cells, which reciprocally license DCs for T-cell activation; (4) CTLs that kill virus-infected host cells by death ligands (FAS/FASL) and by cytokines or perforin/granzyme; (5) neutralizing an-

tibody production that bounds to the virus and engaged FcRs on an NK cell, macrophage, or neutrophil that triggers the ADCC. On the other hand, these antibodies can bind to the complement component C1, resulting in the activation of MAC and destruction of the infected cell. Another complement component such as free C3b binds to the virus surface and mediated phagocytosis by neutrophil CR1 receptors. D The viral spread to the cardiovascular system. NK cells, natural killer cells; DCs, dendritic cells; CTLs, cytotoxic T lymphocytes; ADCC, antibody-dependent cellular cytotoxicity; MAC, membrane attack complex; SARS-CoV-2, severe acute respiratory syndrome coronavirus-2; ACE2, angiotensin-converting enzyme 2. (For figure see next page.)

and MERS infectious patients, with less severe pathogenesis [57, 58]. The clinical symptoms of SARS-CoV-2 can be asymptomatic, symptomatic, mild, and also lead to severe disease with multi-organ failure. In the symptomatic phase or Viral phase, in which the clinical manifestations of the disease usually start 5 days after exposure, patients may experience symptoms such as fever, dyspnea, sore throat, chest pain, expectoration, cough, and myalgia, but fever, cough, and fatigue are common symptoms of COVID-19 [59]. At the present, there are different confirming diagnostic methods for the detection of SARS-CoV-2 in patients, around the time of symptom onset, in laboratories, as follows: (1) nucleic acid tests like real-time RT-PCR or next-generation sequencing; (2) antibody or antigen detection tests, including enzyme-linked immunosorbent assay; (3) chest computed tomography and spectroscopic techniques [60–63]. Real-time RT-PCR on nasopharyngeal and oropharyngeal swabs is considered the “gold standard” for confirming the diagnosis in clinical cases of COVID-19 [64, 65]. Besides clinical symptoms, the blood biochemistry indexes such as the total white blood cell, lymphocyte, platelet, and thromboplastin time decline, while C-reactive protein, lactate dehydrogenase, aspartate transaminase, alanine aminotransferase, cytokine level, and bilirubin increase in most patients [66]. ARDS is a prevalent phenomenon in patients, followed by anemia, acute heart injury, and secondary infections [67]. Reports illustrate that middle-aged and older people with chronic and underlying diseases, especially high blood pressure and diabetes, are susceptible to respiratory failure and have poorer prognoses, but it does not mean that children are lesser than old people susceptible to SARS-CoV-2 [68–70]. In the later stages of infection or the thrombo-inflammatory phase, ARDS is a common complication, and resulted from the occurrence of cytokine storms and immune regulatory network imbalance, which is finally followed by anemia, acute heart damage, multiple organ failure, and secondary bacterial infections [71]. Bilateral severe interstitial inflammation of the lungs is found in the chest computed tomography pictures or chest X-ray, which is named ground-glass opacity and involves a local lobe but later expands to multiple lung lobes [67].

Vaccines

With the threat of millions of people being infected and health-care systems becoming overwhelmed, the race is on to develop a vaccine that will protect individuals and

slow the spread of the disease [72]. S protein plays a significant role in the induction of protective immunity against SARS-CoV-2 by mediating T-cell responses and neutralizing antibody production [73]. In the past few decades, scientists would develop vaccines that induce the body to produce antibodies that recognize and block human coronaviruses with the use of S protein as the target [74]. Nonetheless, the expanded vaccines have minimal usage, even between strains close together of the virus, owing to an absence of cross-conservation [75]. Recently, researchers identified the at least target domain of the virus's S protein that is critical for docking with ACE2 receptor and this region or RBD located in the S1 subunit of the S protein [76–78]. Furthermore, several studies strongly reported that viral structural proteins such as N, M, and E proteins have the potential for inclusion within future vaccine candidates to stimulate T-cell responses, and may significantly contribute to the recovery from COVID-19 [1].

In addition, inactivated and live attenuated vaccine platforms can induce broad and strong immune responses, in comparison to the other platforms, because they have the whole virion including structural and nonstructural proteins [1, 79, 80]. According to the vaccine tracker reported by the World Health Organization, October 2021, nearly 300 vaccine candidates are currently under various phases of development. In total candidate vaccines, 194 and 123 are in clinical and preclinical phases, respectively, that are available at <https://www.who.int/publications/m/item/draft-landscape-of-covid-19-candidate-vaccines>. There are 10 platforms for COVID-19 vaccines including, PS, nonreplicating viral vector (VVnr), replicating viral vector (VVr), VVnr in combination with an antigen-presenting cell (VVnr + APC), VVr in combination with an antigen-presenting cell (VVr + APC), virus-like particle, inactivated virus, live attenuated virus, mRNA vaccine (RNA), and DNA [30, 81, 82]. Around 11 vaccine candidates have been authorized/approved up to now in clinics and markets worldwide. Additionally, their platforms, clinical phase, manufacturing, and dosage are shown in Table 3 and Figure 5.

Conclusion

The pandemic of the newly identified coronavirus that is also known as COVID-19 is the third highly pathogenic human coronavirus. SARS-CoV-2 has less mortality than SARS-CoV-1 and MERS, but it has spread fast all over the world and has been declared a public health

Platform	Developer	Phase	Candidate vaccine	Doses	ID	Refs
Virus based LAV IV	Codagenix/Serum Institute of India	Phase 1	COVI-VAC	1-2	NCT04619628	[82]
	Bharat Biotech international Limited	Phase 3	Whole-virion inactivated SARS-CoV-2 vaccine (BBV152)	2	NCT04641481*	[83]
	Sinopharm + China National Biotech Group Co + Beijing Institute of Biological products	Phase 4	Inactivated SARS-CoV-2 vaccine (vero cell), vaccine name BBIBP-CoV	2	NCT04863638*	[84]
	Organization of Defensive Innovation and Research	Phase 1	Inactivated SARS-CoV-2 vaccine FAKHRAVAC (MIVAC)	2	IRCT20210206050259N1	[85]
Nucleic-acid based DNA RNA	Sinovac Research and development Co., Ltd	Phase 4	CoronaVac; inactivated SARS-CoV-2 vaccine (vero cell)	2	NCT04756830*	[86]
	Research Institute for Biological Safety Problems, Rep of Kazakhstan	Phase 3	QazCovid-in® – COVID-19 inactivated vaccine	2	NCT04691908	[87]
	Shifa Pharmed Industrial Co	Phase 2/3	COVID-19 inactivated vaccine	2	IRCT20201202049567N3	[88]
	AnGes + Takara Bio + Osaka University	Phase 2/3	AG0301-COVID19	2	NCT04655625	[89]
Vector-based Vvr Vnr	Pfizer/BioNTech + FosunPharma	Phase 4	BNT1 62b2 (3 LNP-mRNAs), also known as “comirnaty”	2	NCT04760132*	[90]
	CureVac AG	Phase 3	CVnCoV vaccine	2	NCT04674189	[91]
	Moderna + National Institute of Allergy and Infectious diseases (NIAID)	Phase 4	mRNA-1273	2	EUCTR2021-000930-32*	[92]
	University of Hong Kong, Xiamen University and, Beijing Wantai Biological Pharmacy	Phase 2	DeIN51-2019-nCoV-RBD-OPT1 (intranasal flu-based-RBD)	2	ChiCTR2000039715	[93]
Protein and peptide-based	Gamaleya Research Institute; Health Ministry of the Russian Federation	Phase 3	Gam-COVID-VacAdeno-based (rAd26-S+rAd5-S)	2	NCT04530396*	[90]
	AstraZeneca + University of Oxford	Phase 4	ChAdOx1-S – (AZD1222)	1-2	NCT04760132*	[94]
	CanSino Biological Inc./Beijing Institute of Biotechnology	Phase 4	Recombinant novel coronavirus vaccine (adenovirus type 5 vector)	1	NCT04892459*	[95]
	Janssen Pharmaceutical	Phase 4	Ad26.COV2.S	1-2	EUCTR2021-002327-38-NL*	[96]
VLP	Federal Budgetary Research Institution State Research Center of virology and Biotechnology “vector”	Phase 3	EpiVacCorona (EpiVacCorona vaccine based on peptide antigens for the prevention of COVID-19)	2	NCT04780035*	[97]
	Anhui ZhifeiLongcom Biopharmaceutical + Institute of Microbiology, Chinese Academy of Sciences	Phase 3	Recombinant SARS-CoV-2 vaccine (CHO cell)	2-3	NCT04466085*	[98]
	Novavax	Phase 3	SARS-CoV-2 rS/Matrix M1-Adjuvant (full-length recombinant SARS CoV-2 glycoprotein nanoparticle vaccine adjuvanted with matrix M)	2	NCT04611802	[99]
	Razi Vaccine and Serum Research Institute	Phase 3	CoV-2 glycoprotein nanoparticle vaccine adjuvanted with matrix M)	3	IRCT20210206050259N3	[100]
VLP	Medicago Inc	Phase 3	RaziCov pars, recombinant spike protein	2	NCT05040789	[101]
		Phase 3	Coronavirus-like particle COVID-19 (CoVLP)	2		

COVID-19, coronavirus disease-2019; VLPs, virus-like particles; SARS-CoV-2, severe acute respiratory syndrome coronavirus-2; Wnr, nonreplicating viral vector; LAV, live attenuated virus; Wvr, replicating viral vector; IV, inactivated virus. *Authorized/approved.

Fig. 5. Vaccines platforms against COVID-19. **A** Different vaccine platforms such as PS, VVnr, VVR, VLP, IV, LAV, mRNA (RNA), and DNA (DNA) vaccines can be considered to protect individuals, resulting in the reduction of the disease spread. **B** Distribution

of approved and ongoing platforms in different clinical phases. COVID-19, coronavirus disease-2019; VLP, virus like particle; PS, protein subunit; IV, inactivated virus; VVnr, nonreplicating viral vector; LAV, live attenuated virus; VVR, replicating viral vector.

emergency of international concern by the WHO. Despite extensive research and a flood of articles published daily on SARS-CoV-2, and advances in effective management of COVID-19, we will require in-depth studies about SARS-CoV-2 pathogenesis. For the discovery of an effective drugs and vaccines against SARS-CoV-2, identification and evaluation of the available data on different molecular and cellular mechanisms involved in SARS-CoV-2 pathogenesis is very promising.

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Conflict of Interest Statement

The authors declare that no conflict of interest exists.

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Author Contributions

Ayyoob Khosravi designed and supervised the study with the help of Abasalt Hosseinzadeh Colagar. Ali Fallah wrote the first draft of the manuscript with support from Azadeh Mohammad-Hasani. Parts of the manuscript were also written by Hadi Razavi Nikoo and Hamidreza Abbasi. Hamidreza Abbasi designed the figures and tables with the help of Ali Fallah. Ayyoob Khosravi, Hadi Razavi Nikoo, and Abasalt Hosseinzadeh Colagar participated in the final editing the manuscript. Hamidreza Abbasi played the main role in the submission of the manuscript. All authors give final approval of the manuscript to be submitted.

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