Neutralizing Antibodies against SARS-CoV-2 and Other Human Coronaviruses

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Coronavirus (CoV) disease 2019 (COVID-19) caused by severe acute respiratory syndrome (SARS)-CoV-2 (also known as 2019-nCoV) is threatening global public health, social stability, and economic development. To meet this challenge, this article discusses advances in the research and development of neutralizing antibodies (nAbs) for the prevention and treatment of infection by SARS-CoV-2 and other human CoVs.

Current Situation with SARS-CoV-2 and Other Human CoVs

Three emerging, highly pathogenic human CoVs are SARS-CoV, Middle East respiratory syndrome (MERS)-CoV, and COVID-19 virus, which was previously named 2019-nCoV by the World Health Organization (WHO), and is also known as hCoV-19 or SARS-CoV-2 [1]. Atypical pneumonia (SARS) was first reported from Guangdong Province, China in late 2002. SARS caused a global pandemic in 2003 with approximately 10% (774/8098) case fatality rate (CFR) [2]. SARS-CoV has not circulated in humans since 2004. MERS-CoV was first reported from Saudi Arabia in 2012 and has continued to infect humans with limited human-to-human transmission, leading to a CFR of approximately 34.4% (858/2494) in 27 countries, according to the most recent WHO report [1]. Both SARS-CoV and MERS-CoV are zoonotic viruses. They use bats as their natural reservoirs and transmit from bats to intermediate hosts (e.g., palm civets for SARS-CoV, dromedary camels for MERS-CoV), leading to infection in humans [2,3].

Different from SARS-CoV and MERS-CoV, SARS-CoV-2 was first reported in Wuhan, China in December 2019 and is characterized by its rapid spread and virulent human-to-human transmission [4], resulting in 125 048 confirmed cases including 4613 deaths (CFR 3.7%), particularly in Wuhan, China and in at least 117 other countries, territories, or areas as of March 12, 2020. With no vaccines or treatments on the horizon, researchers are exploring various medical interventions, including nAbs, to control the continuous spread of SARS-CoV-2 and the global COVID-19 pandemic [5]. SARS-CoV-2 is also a zoonotic virus with bats as its natural reservoir [4], but its intermediate hosts have not been identified.

Pathogenesis and Key Proteins of SARS-CoV-2 and Other Human CoVs

SARS-CoV-2 infection mainly results in pneumonia and upper/lower respiratory tract infection. Fever and cough are two major clinical symptoms, but others include shortness of breath, muscle pain (myalgias)/fatigue, confusion, headache, sore throat, and even acute respiratory distress syndrome, leading to respiratory or multiorgan failure [6]. For elderly people with underlying comorbidities such as diabetes, hypertension, or cardiovascular disease, SARS-CoV-2 infection may result in severe and fatal respiratory diseases. So far, its effects on children have been generally mild. The virus can be transmitted through respiratory droplets or close contact with infected surfaces or objects and is detectable in multiple samples, including saliva, stool, and blood [7]. To develop vaccines and therapeutics, we must understand the behavior of key proteins in SARS-CoV-2.

Similar to SARS-CoV and MERS-CoV, SARS-CoV-2 is an enveloped, single-stranded, and positive (+)-sense RNA virus, belonging to the beta-CoV genera in the family Coronaviridae [4]. The genome of this and other emerging pathogenic human CoVs encodes four major structural proteins [spike (S), envelope (E), membrane (M), and nucleocapsid (N)], approximately 16 nonstructural proteins (nsps1–16), and five to eight accessory proteins. Among them, the S protein plays an essential role in viral attachment, fusion, entry, and transmission. It comprises an N-terminal S1 subunit responsible for virus–receptor binding and a C-terminal S2 subunit responsible for virus–cell membrane fusion [2,3]. S1 is further divided into an N-terminal domain (NTD) and a receptor-binding domain (RBD). SARS-CoV-2 and SARS-CoV bind angiotensin-converting enzyme 2 (ACE2) while MERS-CoV binds dipeptidyl peptidase 4 (DPP4), as receptors on the host cell expressing ACE2 (e.g., pneumocytes, enterocytes) or DPP4 (e.g., liver or lung cells including Huh-7, MRC-5, and Calu-3) [2,3,8]. Phylogenetically, SARS-CoV-2 is closely related to SARS-CoV, sharing approximately 79.6% genomic sequence identity [4]. During infection, CoV first binds the host cell through interaction between its S1-RBD and the cell membrane receptor, triggering conformational changes in the S2 subunit that result in virus fusion and entry into the target cell (see human CoV life cycle in Figure 1A) [2,3].

nAbs against SARS-CoV, MERS-CoV, and SARS-CoV-2

Virus nAbs induced by vaccines or infected virus play crucial roles in controlling viral infection. Currently developed SARS-CoV- and MERS-CoV-specific nAbs include monoclonal antibodies (mAbs), their functional antigen-binding fragment (Fab), the single-chain variable region fragment (scFv), or single-domain antibodies [nanobodies (Nbs)] [8]. They target S1-RBD, S1-NTD, or the S2 region, blocking
Figure 1. Life Cycle of Highly Pathogenic Human Coronaviruses (CoVs) and Specific Neutralizing Antibodies (nAbs) against These Coronaviruses. (A) Life cycle of highly pathogenic human CoVs. These CoVs enter host cells by first binding to their respective cellular receptors [angiotensin-converting enzyme 2 (ACE2) for severe acute respiratory syndrome (SARS)-CoV-2 or SARS-CoV and dipeptidyl peptidase 4 (DPP4) for Middle East respiratory syndrome (MERS)-CoV] on the membranes of host cells expressing ACE2 (e.g., pneumocytes, enterocytes) or DPP4 (e.g., liver or lung cells including Huh-7, MRC-5, and Calu-3) via the surface spike (S) protein, which mediates virus–cell membrane fusion and viral entry. Viral genomic RNA is released and translated into viral polymerase proteins. The negative (−)-sense genomic RNA is synthesized and used as a template to form subgenomic or genomic positive (+)-sense RNA. Viral RNA and nucleocapsid (N) structural protein are replicated, transcribed, or synthesized in the cytoplasm, whereas other viral structural proteins, including S, membrane (M), and envelope (E), are synthesized in the endoplasmic reticulum (ER) and transported to the Golgi. The viral RNA–N complex and S, M, and E proteins are further assembled in the ER–Golgi intermediate compartment (ERGIC) to form a mature virion, then released from host cells. (B) Potential targets of nAbs against SARS-CoV-2 and other pathogenic human coronaviruses. (Figure legend continued at the bottom of the next page.)
CoVs. (a) Human CoV receptor binding and membrane fusion process. The CoV first binds a viral receptor (ACE2 or DPP4) through the receptor-binding domain (RBD) in the S protein, followed by fusion of the virus with cell membranes via the formation of a six-helix bundle (6-HB) fusion core. NTD, N-terminal domain. (b) Potential targets of nAbs on the S protein of human CoVs. Monoclonal antibody (mAb), antigen-binding fragment (Fab), single-chain variable region fragment (scFv), or single-domain antibody (vH-H) derived from cameld heavy chain antibody (HcAb) binds to the RBD, S1 subunit (non-RBD, including NTD), or S2 of the viral S protein, blocking binding between the RBD and the respective receptor (for RBD-targeting nAbs), interfering with the conformational change of S for S1-targeting nAbs, or hindering S2-mediated membrane fusion (for S2-targeting nAbs), leading to the inhibition of infection with pathogenic human CoVs in the host cells. This figure was created using BioRender (https://biorender.com/).
CoV RBD-based vaccines that might eventually prevent SARS-CoV-2 and SARS-CoV infection [15]. It is also possible that SARS-CoV RBD-targeting nAbs might be applied for prophylaxis and treatment of SARS-CoV-2 infection in the current absence of SARS-CoV-2-specific vaccines and antibodies. However, robust testing lies ahead.

**Concluding Remarks and Future Perspectives**

SARS-CoV-2 continues to infect people globally with the concomitant urgency to develop effective nAbs as prophylactic and therapeutic agents to prevent and treat its infection and control its spread. Studies from SARS-CoV and MERS-CoV have demonstrated that many fragments (S1-NTD, RBD, S2) in S proteins can be used as targets to develop nAbs. Still,
RBD-specific antibodies have greater potency to neutralize infection with divergent virus strains, suggesting that the RBD of SARS-CoV-2 can also serve as an important target for the development of potent and specific nAbs. Cocktails comprising antibodies specific for RBD and other regions in the S protein may further improve the breadth and potency of nAbs against SARS-CoV-2 and its escape-mutant strains. Human sera from convalescent patients have been used to treat COVID-19, but lessons learned from SARS show that some non-nAbs targeting the non-RBD regions in the S protein may cause an antibody-dependent enhancement (ADE) effect on viral infectivity and disease, as well as other harmful immune responses [2]. On a positive note, some anti-SARS-CoV nAbs have shown cross-reactivity or cross-neutralizing activity against SARS-CoV-2 infection in vitro. Thus, overall, research on SARS-CoV- and MERS-CoV-specific nAbs should provide important guidelines for the rapid design and development of SARS-CoV-2-specific nAbs.

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References

Resources
1. www.who.int/emergencies/mers-cov/en/
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