Secondary Publication: SARS-CoV-2 and Immunological Response

TWMUJ 5: 1-9, 2021

Review

Secondary Publication: SARS-CoV-2 and Immunological Response

Hidehito Kato and **Naoko Yanagisawa**

Department of Microbiology and Immunology, Tokyo Women's Medical University, Tokyo, Japan

(Accepted April 12, 2021)

(Advance Publication by J-STAGE June 10, 2021)

Viruses require the host cellular machinery for protein translation and replication. Upon proliferation, virions damage cells and are released from the infected cells before infecting other cells. Acute inflammation occurs when host cells are damaged by infection. The cell receptors to which severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) binds, are widely distributed compared to those for other viruses, thereby resulting in various symptoms such as rhinitis, pneumonia, and enteritis. In general, RNA viruses, including SARS-CoV-2, exhibit a high frequency of gene mutations. Antigenic modulation due to genetic mutations of the spike protein causes cytokine storms because of strong activation of the innate immune system, similar to the phenomenon previously observed in highly pathogenic avian influenza. The proportion of severely ill patients due to coronavirus disease 2019 (COVID-19) varies from country to country, and factors that are responsible for the severity of the disease include antibody-dependent enhancement (ADE), Bacillus Calmette-Guérin (BCG) vaccination, and human leukocyte antigen (HLA) type. ADE and HLA types may also influence the protective effect of immunity, including its vaccine response against SARS-CoV-2.

This report is a secondary publication of our previous review report "J Tokyo Wom Med Univ 91: 2-10, 2021."

Keywords: spike protein, ACE2, ADE, BCG, HLA

Introduction

As viruses differ from bacteria and cannot self-replicate, they penetrate (infect) cells and use the replication enzymes of infected cells for replication. After replication, viruses damage cells, leave the infected cells, reinfect nearby cells, or transmit to other individuals through saliva, feces, blood, and other sources. If the virus is highly replicative, then the cell damage is worsened, leading to viral toxicity. In chronic infections, several cells do not cause acute injury. Viral genes are inserted into an infected cell's DNA (provirus), which is retained extrachromosomally as episomes. Until viruses are produced again by activation stimuli, the provirus synchronizes with the infected cell division. Presumably, at least 8% of human DNA comprises proviruses and their fragments. Because an antigenic shift occurred due to a mutation in its characteristic spike protein (the reason it is called corona), severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) is referred to as novel coronavirus; however, it is believed that the virus is not a unique divergence of the normal form, and the viral infection can be

Corresponding Author: Hidehito Kato, Department of Microbiology and Immunology, Tokyo Women's Medical University, 8-1 Kawada-cho, Shinjuku-ku, Tokyo 162-8666, Japan. kato.hidehito@twmu.ac.jp

doi: 10.24488/twmuj.2021001

treated by the host's normal immune response. In the present study, we explain the structure of SARS-CoV-2 as well as discuss factors that control the exacerbation of the disease, and the current state and challenges in vaccine development.

Characteristics of the Virus

Viruses are microscopic particles that cannot replicate. Until giant viruses were discovered.[1-3] It was believed that viruses could only be observed using electron microscopes. In general, the size of a virus ranges from 0.038 to 1 μm compared to 1-2 μm for a bacterium, and presumably, both bacteria and viruses can pass through the 5 μm mesh of a typical surgical mask. In a recent study, the virus uptake volume of the 0.1 μm SARS-CoV-2 was reduced by 20% to 40% by wearing cotton masks and surgical masks, and it was found that N95 masks could reduce uptake volume by 80%-90%.[4]

According to the International Committee on Taxonomy of Viruses (ICTV), a virus classification and naming institution, as of 2019, viruses were classified into 168 families, 103 subfamilies, 1,421 genera, 68 subgenera, and 6,590 species,[5] and moreover, multiple serotypes existed for each species. Among viruses that cause cold symptoms in humans, *rhinovirus* of the family *Picornaviridae* is the most common; however, other 110 types of viruses are also responsible for causing viral infections. Typically, *rhinovirus* and other viral infections exhibit minor symptoms. Nevertheless, viruses that cause fatal infections, such as *Ebolavirus*, *human immunodeficiency virus* (HIV), and *rabies Lyssavirus*, make up a negligible proportion of all viruses. Symptoms that arise from viral infections depend on the site of viral infection (the virus receptor expression site). For example, as *rabies Lyssavirus* receptors are nicotinic acetylcholine receptors, they cause neurological symptoms (impaired neurotransmission) by damaging the nerve cells. Moreover, HIV receptors are CD4 molecules, and thus, HIV produces immunodeficiencies by damaging helper T cells. Because the expression site and frequency of virus receptors differ depending on the species, viral infections are characterized by species and tissue specificity. Nevertheless, certain viruses use nonspecific receptors similar to those of bacteria. Due to these characteristics (species specificity), to create an experimental animal infection model, the target experimental animal must continue to be infected with a human virus for a period ranging from a few weeks to a few months (adaptation). SARS-CoV-2 infections have been confirmed *in vitro* in monkey kidney cell lines and *in vivo* in hamsters.[6] Animals such as rhesus macaques, cynomolgus macaques, and African green monkeys are known to present the same infection pathways and symptoms as humans.[7-9]

Virus Classification

Viruses are classified by genotype as either DNA or RNA viruses and can be further separated into two types: single-stranded (ss) and double-stranded (ds). Furthermore, ssRNA viruses can be divided into those with positive polarity (+) and those with negative polarity (−) (**Table 1**).[5] Although (+) RNAs can be directly translated into viral proteins as mRNAs, (−) RNAs cannot be translated into viral proteins until they are transcribed into (+) RNAs. Furthermore, viruses can be categorized into those with an envelope derived from the infected cell (enveloped) and those without (nonenveloped). Because envelopes represent the plasma membranes of virus-infected cells, viruses do not comprise the genetic information of envelopes. Viral genes are protected by capsid proteins. Capsids can be classified as icosahedral or helical. The capsid of SARS-CoV-2 is helical.

SARS-CoV-2

SARS-CoV-2 is a (+) ssRNA virus belonging to the family Coronaviridae. The ICTV named the virus SARS-CoV-2 and the disease caused by it as coronavirus disease 2019 (COVID-19).[10] Coronaviruses are classified as alpha (α), beta (β), gamma (γ), and delta (δ). α- and β-coronaviruses trigger respiratory tract or intestinal infections in 10%-30% of infected individuals. They account for 15%-30% of viruses that trigger typical cold symptoms *rhinoviruses* are the most common cause of cold symptoms, followed by coronaviruses and then by influenza viruses.

Viruses in the coronavirus genera are (+) ssRNA viruses that comprise envelopes derived from infected cells containing three major glycoproteins (spike protein, en-

Table 1. Taxonomy of virus.[5]

Nucleic acid		Naked or enveloped	Capsid	Family and major diseases		
DNA	ss	Naked	Icosahedral	Parvoviridae Erythema infectiosum		
	ds	Naked	Icosahedral	Adenoviridae Gastroenteritis	Papillomaviridae Papilloma	
		Enveloped	Icosahedral	Herpesviridae Herpes	Hepadnaviridae HB	
		Enveloped	Complex	Poxviridae Variola		
RNA	ss (-)	Enveloped	Helical	Rhabdoviridae Rabies	Orthomyxoviridae Influenza	Arenaviridae Lassa fever
		Enveloped	Helical	Paramyxoviridae Measles/Mumps/RS	Filoviridae Ebola virus disease	Bunyaviridae CCHF/SFTS
	ss (+)	Enveloped	Helical	Coronaviridae SARS/MEAS/COVID-19		
		Enveloped	Polyhedron	Flaviviridae DF/JE/YF		
		Enveloped	Spherical	Togaviridae Rubella	Retroviridae AIDS/ATL	
		Naked	Icosahedral	Caliciviridae HE/IG (Noro)	Picornaviridae polio/HA/Rhino	
	ds	Naked	Icosahedral	Reoviridae IG (Rota)		

Virus-derived RNA dependant RNA polymerase is utilized for mRNA synthesis complementary to the genome in single-strand (-) RNA virus. Complex capsids include helical and icosahedral structures.

ss, single-stranded; ds, double-stranded; CCHF, Crimea-Congo Hemorrhagic Fever; SFTS, severe fever with thrombocytopenia syndrome; JE, Japanese encephalitis; YF, Yellow fever; RS, respiratory syncytial; IG, infectious gastroenteritis; HA, HB, HE, hepatitis A, B, E; AIDS, acquired immunodeficiency syndrome; COVID-19, coronavirus disease 2019; ATL, adult T-cell leukemialymphoma; SARS, severe acute respiratory syndrome; MERS, middle east respiratory syndrome; DF, dengue fever.

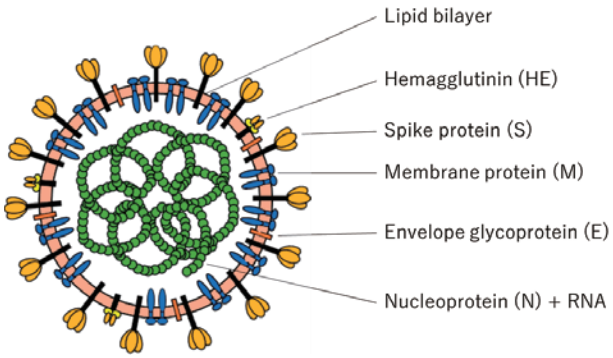

Figure 1. Structural diagram of SARS-CoV-2.
SARS-CoV-2 is a + ssRNA virus, in which the RNA is covered with a helical capsid. The envelope derived from the infected cell consists of three major glycoproteins, namely, spike protein (S), an envelope protein (E), and membrane protein (M), and a small number of haemagglutinin esterases (HE), similar to those observed in type C influenza virus.

velope protein, and membrane protein) and possess limited amount of hemagglutinin-esterase (HE) similar to those of the type C influenza virus (**Figure 1**). Influenza virus structures differ in polarity and segmentation (segmented / unsegmented) of ssRNA (influenza viruses: negative strand, segmented; coronaviruses: positive strand, unsegmented) and types of constituent proteins (infected cells are different). Presumably, RNA viruses may undergo frequent base substitutions by mutation, and their genetic information changes continually. Moreover, unsegmented ssRNA has no mutations due to antigen shift (reassortment of segments) compared to segmented ssRNA, and these viruses are therefore less prone to mutations. An antigenic shift occurred due to a mutation to SARS-CoV-2's characteristic spike protein.[11, 12] Furthermore, because SARS-CoV-2 has potent interferon (IFN) inhibitor genes, it is likely to replicate.[13] As with the Spanish flu of 1918 influenza and highly pathogenic avian influenza infection, it tends to induce a cytokine storm and triggers an overactive innate immune response.[14-18] After its spike protein binds to the angiotensin-converting enzyme (ACE) 2 expressed on host cells, SARS-CoV-2 enters the cell by the action of transmembrane protease/serine (TMPRSS) 2 on the sur-

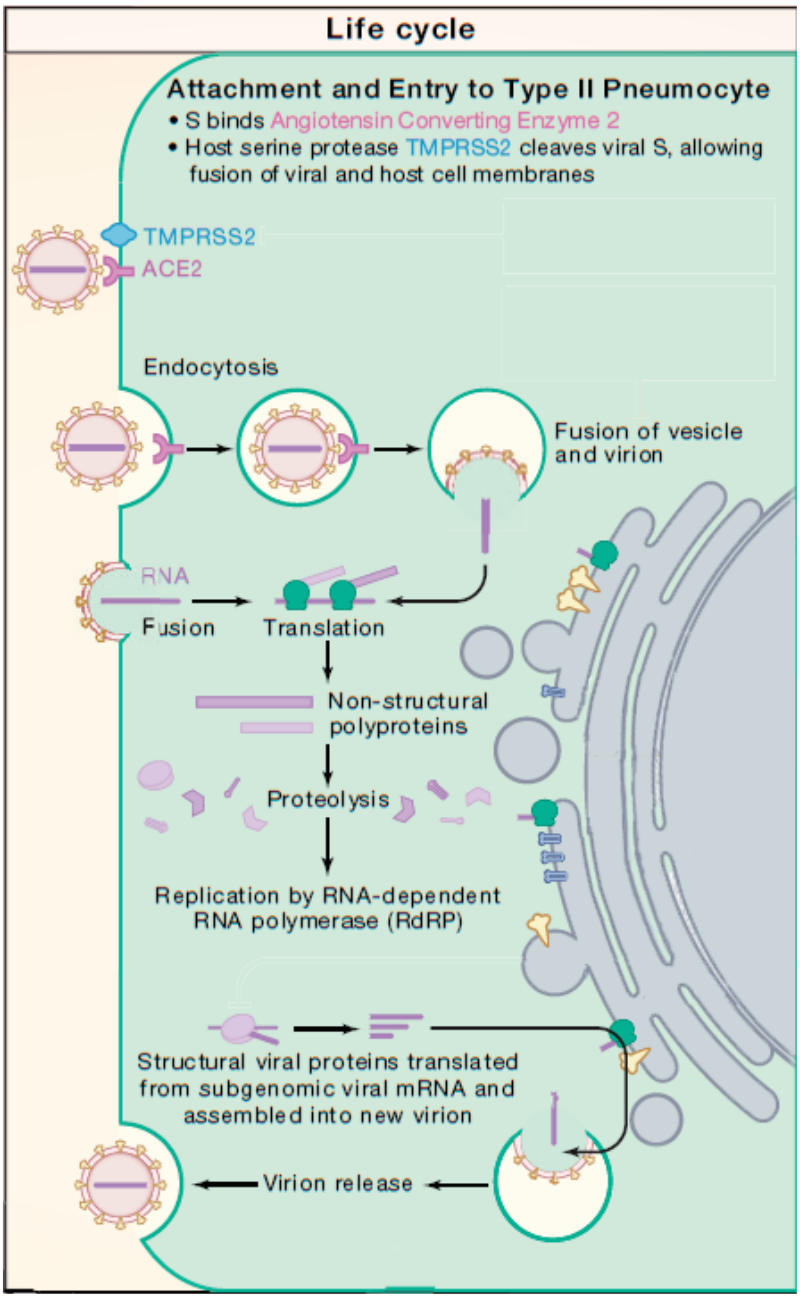

Figure 2. Life cycle of the coronavirus.
The spike protein binds to the ACE2 enzyme on the plasma membrane of type 2 pneumocytes and intestinal epithelial cells. After binding, the spike protein is cleaved by a host membrane serine protease, TMPRSS2, facilitating viral entry. Modified from reference 19.

face of the infected cell (**Figure 2**).[19,20] Thus, the simultaneous expression of ACE2 and TMPRSS2 are necessary for coronavirus infections.[21] Although the expression of human influenza virus receptors and proteases is limited to the upper respiratory epithelial cells, these molecules exhibited various expressions in the the nasal passage, bronchi, lung parenchyma, esophagus, ileum, colon, prostate, and cornea.[22] Therefore, SARS-CoV-2 is similar to influenza considering the onset of fever that accompanies the activation of innate immunity; however, its infections induce various symptoms such as rhinitis, pneumonia, and intestinal inflammation.

Viral Interference

The phenomenon of viral interference (where infection by one type of virus hinders the infection by another type of virus) has long been known. Previous studies reported

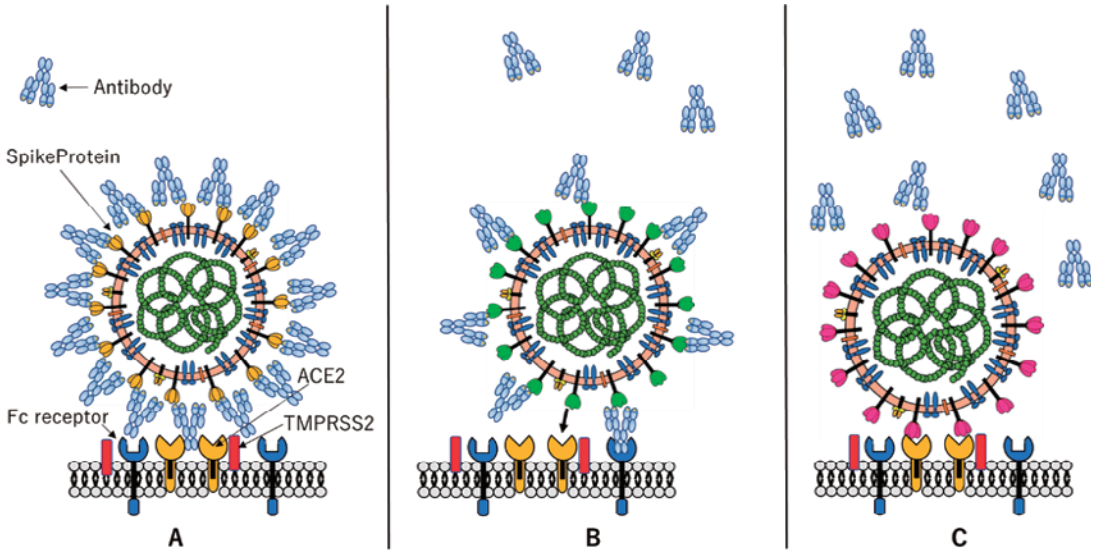

Figure 3. Fc-dependent antibody-dependent enhancement (ADE) hypothesis in SARS-CoV-2 infection.

A. Specific antibodies targeting viral surface proteins (yellow) bind to and neutralize the virus. This promotes viral entry into host phagocytes and blocks their attachment to the cell surface viral receptor.

B. Interplay of the cell surface Fc receptor with the viral epitope-bound antibodies promotes viral infection of the target cells (ADE). Incomplete binding of the mutated viral ligand (green) to antibodies promotes exposure of the viral epitopes to the cell surface receptors, leading to enhanced invasion of the virus to the target cells.

C. Viral infection is mediated by host cell surface receptors. Mismatched antibodies are incapable of blocking viral epitopes (pink), thereby accelerating subsequent infection.

a substance that inhibits the replication of other viruses, namely IFN (currently, Type I IFN).[23] Presumably, one reason for viral interference is that the Type I IFN produced by the activation of innate immunity from the initial virus infection inhibits the replication of the other virus infection. Another possible reason is the competition for replication enzymes because the initial virus expends the replication/transcription enzymes of the infected cell. On comparing influenza virus and coronavirus, it is assumed that a coronavirus would prevail (since it has (+) RNA that can directly assemble proteins) due to the difference between (+) RNA and (−) RNA. Additionally, the virus that prevails also differs depending on whether it has an IFN inhibitor gene. In recent years, interference between influenza virus and rhinovirus infections has been observed.[24] As this is also a (+) ssRNA virus akin to the *rhinovirus*, presumably similar interference will occur with the SARS-CoV-2 outbreak. In particular, influenza did not spread in Japan during the winter of 2019; however, COVID-19 was widely spread.

Factors that Control Exacerbation of Disease

Antibody-dependent enhancement (ADE)

ADE explains the exacerbation of disease after vaccination and after a secondary viral infection by a different type (serotype) of virus.[25-27] For example, for viruses with two or more serotypes (antigen types), exacerbation of disease occurs when there is an infection with a type 2 virus after the initial infection with a type 1 virus establishes immunity. After infection with the type 1 virus and production of neutralizing antibodies, even if a second infection with the type 1 virus occurs, the infection would not be successful because the viral antigens would be completely neutralized by the neutralizing antibodies (**Figure 3-A**); however, if the virus in the second infection was a type 2 virus, the neutralizing antibodies could not completely neutralize the virus, but the viral antigens would remain uncovered because the antigenicity of type 1 slightly differs from that of type 2 (**Figure 3-B**). As antibodies that incompletely bind to the virus also bind to the Fc receptors of infected cells and promote exposure

of the cells to the virus, the frequency of binding between the viral antigens and the cell's viral receptors is higher than in typical infections (**Figure 3-B**). As a result, after establishing immunity, infection with a virus of different antigenicity causes exacerbation of the disease. With SARS-CoV-2 infections, fusion of the viral and cellular membranes (uncoating) occurs by TMPRSS2 acting on the viral envelope. When neutralizing antibodies firmly bind to the spike protein (**Figure 3-A**), and if phagocytosis of the virus occurs, TMPRSS2 is unable to act on the spike protein, and uncoating does not occur. In contrast, with a mutated spike protein, ACE2 and TMPRSS2 both act on the spike protein, and infection (uncoating) is successfully established by the aforementioned mechanisms (**Figure 3-B**). The principle of the current mainstream ADE hypothesis is as follows: antiviral efficacy is inhibited, and viral replication accelerates because expression signals for Type I IFN inhibitory signals, which have antiviral effects and interleukin (IL)-10, which is an inhibitory cytokine are transmitted from nonspecific molecules (leukocyte immunoglobulin [Ig] like receptor [LILR]) that recognize viruses and Fc receptors. On analyzing the antigenicity of 80 variants of natural mutants and artificial mutants of the spike protein, it was found that neutralizing antibody responsiveness of SARS-CoV-2 was altered.[28] Furthermore, as the variants differ country wise, it can be inferred that enhancement by ADE also differed.[29, 30]

Bacille de Calmette et Guerin (BCG): Inhibition by activation of innate immunity

In Africa in 1990, BCG vaccination was carried out to reduce the mortality caused by tuberculosis in children; however, in addition to tuberculosis, lower mortality from viral pneumonia was observed after vaccination.[31] This gave rise to the Trained Immunity hypothesis in 2011,[32] and immense research was conducted. As a result, in 2016, it was found that BCG vaccination enabled the innate immune system to swiftly respond to the invading pathogens via epigenetics (changes in gene expression or cell phenotype that are not accompanied by changes in the DNA sequences).[33] In 2018, it was demonstrated that BCG vaccination could inhibit experimental infection from a yellow fever viral vaccine strain,[34] thereby suggesting that chromatin that encodes IL-1β unravels due to BCG's repeated stimulation and its production becomes easier (trained immunity).[35] Since IL-1β induces the production of Type I IFNs that have an antiviral effect,[36] it can be assumed that it inhibits the replication of unspecified viruses. As these studies were conducted before the SARS-CoV-2 pandemic, they did not cover SARS-CoV-2; however, if considered along with epidemiologic studies in recent years, presumably, the enhancement of SARS-CoV-2 infections can be suppressed by trained immunity. As BCG is a live attenuated vaccine, it continues to replicate to maintain a trained immunity state within the body until approximately 15 years of age; however, it is unclear whether this trained immunity state persists afterward. Several severe symptoms in elderly population may result from age-related inactivation of trained immunity by BCG vaccination. Epidemiologic studies have reported that the number of deaths from SARS-CoV-2 per million population reduced in countries that use the Japan and Russia BCG strains for vaccines including higher viable counts.[37-39] Furthermore, in a "retrospective observational study," it was reported that when BCG vaccination rates increased by 10%, mortality from coronavirus infections reduced by 10.4%.[40] In a "prospective clinical study" that excluded immunocompromised patients and patients taking steroids, the safety of vaccinating those aged 65 years or older with BCG (strain 1331) and protection against viral respiratory infections has been confirmed.[41] In contrast, Israel implemented BCG vaccination between 1955 and 1982 and has continued administering BCG vaccination to immigrants from regions with a high prevalence of tuberculosis since 1982. Although it did not detail whether the BCG used was the Japanese, Russian, or Danish strain, there was a report from Israel that the efficacy of the BCG vaccine was not supported after comparing the SARS-CoV-2 positive rates of 3,064 subjects born from 1979 to 1981 (aged 39-41 years) with 2,809 subjects born from 1983 to 1985 (aged 35-37 years).[42] WHO stated that there is no evidence that BCG prevents severe COVID-19, and the clinical relevance is unknown. Thus, BCG vaccination is not recommended for preventing severe COVID-19.[43]

Table 2. Draft landscape of COVID-19 candidate vaccines in phase 3 compiled by WHO (October 19, 2020).[49]

COVID-19 Vaccine developer/manufacturer	Vaccine platform	Type of candidate vaccine	Number of doses	Timing of doses	Route of Administration
Sinovac	Inactivated	Inactivated	2	0, 14 days	IM
Wuhan Institute of Biological Products/ Sinopharm	Inactivated	Inactivated	2	0, 21 days	IM
Beijing Institute of Biological Products/ Sinopharm	Inactivated	Inactivated	2	0, 21 days	IM
University of Oxford/AstraZeneca	Non-Replicating Viral Vector	ChAdOx1-S	1		IM
CanSino Biological Inc./Beijing Institute of Biotechnology	Non-Replicating Viral Vector	Adenovirus Type 5 Vector	1		IM
Gamaleya Research Institute	Non-Replicating Viral Vector	Adeno-based (rAd26-S+rAd5-S)	2	0, 21 days	IM
Janssen Pharmaceutical Companies	Non-Replicating Viral Vector	Ad26COVS1	2	0, 56 days	IM
Novavax	Protein Subunit	Full length recombinant SARS CoV-2 glycoprotein nanoparticle vaccine adjuvanted with Matrix M	2	0, 21 days	IM
Moderna/NIAID	RNA	LNP-encapsulated mRNA	2	0, 28 days	IM
BioNTech/Fosun Pharma/Pfizer	RNA	3 LNP-mRNAs	2	0, 28 days	IM

Human leukocyte antigen (HLA): Inhibition by antigen-presenting capacity

When a virus infects a cell, its antigens are broken down into peptides, and the major histocompatibility complex (MHC) of the antigen-presenting cell is expressed (HLA in humans). Cytolytic T cells (Tc) specific to virus-antigens are activated and damage cells expressing antigens on their self-MHC (MHC restriction). Because Tc activation strength toward viral antigens depends on the MHC type, differing MHC types may influence disease exacerbation upon viral infection. Moreover, the onset of acquired immunodeficiency syndrome (AIDS) in HIV-infected individuals possessing HLA-B*35 is accelerated because they cannot induce HLA-B*35-restricted Tcs, which indicates poor interaction between HLA-B*35 and HIV antigen peptides (the level of HLA-B*35 antigen presentation capability is low).[44] Conversely, in HIV patients, slower progression to AIDS is correlated with HLA-B*57 and HLA-B*27.[45,46] In SARS-CoV infections, numerous HLA polymorphisms are correlated with susceptibility: HLA-B*4601, HLA-B*0703, HLA-DRB1*1202,[47] and HLA-Cw*0801.[48] The correlation between SARS-CoV-2 infections and HLA susceptibility is presently being investigated.

Current State and Challenges of SARS-CoV-2 Vaccine Development

According to WHO, as of October 19, 2020, 42 COVID-19 candidate vaccines were in the clinical phase. Among these, 10 candidates were in the final stage, that is, phase 3 (**Table 2**).[49] The types of vaccines include viral vector, RNA, DNA, recombinant protein, and inactivated vaccines. Of these, viral vectors, RNA, and DNA vaccines imitate viral infections to simultaneously stimulate innate immunity and induce type 1 helper T cells (Th1, which activates cellular immunity) and follicular helper T cells (Tfh, which activates humoral immunity). In contrast, recombinant proteins and inactivated vaccines can induce Tfh, but they need to be administered with adjuvants because it is difficult to induce innate immunity and Th1. Of these, viral vector vaccines are of replication-gene defective strain, and cell death of infected cells by viral replication does not occur. Nevertheless, recombinant proteins are synthesized, and they can also induce Th1, which is the state closest to viral infection. Therefore, their efficacy was presumed. AstraZeneca's vaccine uses chimpanzee-derived adenovirus antibodies because anti-chimpanzee adenovirus antibodies do not exist in human sera.[50] CanSino Biologics Inc.'s viral vector vaccine uses human adenovirus type-5 vector (a typical virus vector used in gene therapy) that is less likely eliminated by an-

tibodies *in vivo*.[51] This vector was initially used as an Ebola vaccine in 2014, and its safety and immunogenicity have been verified by China and Sierra Leone. The results of experiments in both countries are excellent.[50, 51]

Even after concluding clinical trials and selling vaccines, we should be prepared for the possibility that these vaccines may only be effective on certain individuals and not on others; furthermore, in few individuals, these vaccines may cause adverse reactions due to interactions between HLA and antigens, and because ADE is triggered by differences in viral antigenicity, as aforementioned.

Conclusion

No structural gap was observed between SARS-CoV-2 and other coronaviruses, and it has also been verified that SARS-CoV-2 evokes a normal immune response.[52] Detailed research is available into the structure and properties of the virus, and expectations for the practical use of these vaccines are increasing.

Remarks: This report is a secondary publication of our previous review report "J Tokyo Wom Med Univ 91: 2-10, 2021."

Conflicts of Interest: The authors declare no conflicting interests.

Author Contributions: HK designed the overall framework of the review and wrote the manuscript. NY edited the draft and contributed to the final version of the manuscript.

References

1. La Scola B, Audic S, Robert C, et al. A giant virus in amoebae. Science. 2003;299(5615):2033.
2. Philippe N, Legendre M, Doutre G, et al. Pandoraviruses: amoeba viruses with genomes up to 2.5 Mb reaching that of parasitic eukaryotes. Science. 2013;341(6143):281−6.
3. Takemura M. Morphological and Taxonomic Properties of Tokyovirus, the First Marseilleviridae Member Isolated from Japan. Microbes Environ. 2016;31(4):442−8.
4. Ueki H, Furusawa Y, Iwatsuki-Horimoto K, et al. Effectiveness of Face Masks in Preventing Airborne Transmission of SARS-CoV-2. mSphere. 2020;5(5):e00637-20.
5. Virus Taxonomy: 2019 Release [Internet]. Berlin: International Committee on Taxonomy of Viruses; 2019 [cited 2020 Oct 10]. Available from: https://talk.ictvonline.org/taxonomy/.
6. Imai M, Iwatsuki-Horimoto K, Hatta M, et al. Syrian hamsters as a small animal model for SARS-CoV-2 infection and countermeasure development. Proc Natl Acad Sci U S A. 2020;117(28):16587−95.
7. Munster VJ, Feldmann F, Williamson BN, et al. Respiratory disease in rhesus macaques inoculated with SARS-CoV-2. Nature. 2020;585(7824):268−72.
8. Muñoz-Fontela C, Dowling WE, Funnell SGP, et al. Animal models for COVID-19. Nature. 2020;586(7830):509−15.
9. Rockx B, Kuiken T, Herfst S, et al. Comparative pathogenesis of COVID-19, MERS, and SARS in a nonhuman primate model. Science. 2020;368(6494):1012−5.
10. Coronaviridae Study Group of the International Committee on Taxonomy of Viruses. The species Severe acute respiratory syndrome-related coronavirus : classifying 2019-nCoV and naming it SARS-CoV-2. Nat Microbiol. 2020;5(4):536−44.
11. Konno Y, Kimura I, Uriu K, et al. SARS-CoV-2 ORF3b is a potent interferon antagonist whose activity is further increased by a naturally occurring elongation variant. Cell Rep. 2020;32(12):108185.
12. Kobasa D, Jones SM, Shinya K, et al. Aberrant innate immune response in lethal infection of macaques with the 1918 influenza virus. Nature. 2007;445(7125):319−23.
13. Wrapp D, Wang N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science. 2020;367(6483):1260−3.
14. Walls AC, Park Y-J, Tortorici MA, et al. Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein. Cell. 2020;181(2):281−92.
15. Ishikawa T. Clinical Preparedness for Cytokine Storm Induced By the Highly Pathogenic H5N1 Influenza Virus. J Pharmacogenom Pharmacoproteomics. 2012;3(6):1000e131.
16. Beigel JH, Farrar J, Han AM, et al. Avian influenza A (H5N1) infection in humans. N Engl J Med. 2005;353(13):1374−85.
17. Tumpey TM, Basler CF, Aguilar PV, et al. Characterization of the reconstructed 1918 Spanish influenza pandemic virus. Science. 2005;310(5745):77−80.
18. Imai Y, Kuba K, Neely GG, et al. Identification of oxidative stress and Toll-like receptor 4 signaling as a key pathway of acute lung injury. Cell. 2008;133(2):235−49.
19. Oberfeld B, Achanta A, Carpenter K, et al. SnapShot: COVID-19. Cell. 2020;181(4):954.
20. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. Cell. 2020;181(2):271−80.
21. Matsuyama S, Nao N, Shirato K, et al. Enhanced isolation of SARS-CoV-2 by TMPRSS2-expressing cells. Proc Natl Acad Sci U S A. 2020;117(13):7001−3.
22. Sungnak W, Huang N, Bécavin C, et al. SARS-CoV-2 entry factors are highly expressed in nasal epithelial cells together with innate immune genes. Nat Med. 2020;26(5):681−7.
23. Isaacs A, Lindenmann J. Virus interference. I. The interferon. Proc R Soc Lond B Biol Sci. 1957;147(927):

258−67.
24. Wu A, Mihaylova VT, Landry ML, et al. Interference between rhinovirus and influenza A virus: a clinical data analysis and experimental infection study. Lancet Microbe. 2020;1(6):e254−62.
25. Takada A, Kawaoka Y. Antibody-dependent enhancement of viral infection: molecular mechanisms and in vivo implications. Rev Med Virol. 2003;13(6):387−98.
26. Taylor A, Foo S-S, Bruzzone R, et al. Fc receptors in antibody-dependent enhancement of viral infections. Immunol Rev. 2015;268(1):340−64.
27. Smatti MK, Al Thani AA, Yassine HM. Viral-Induced Enhanced Disease Illness. Front Microbiol. 2018;9:2991.
28. Li Q, Wu J, Nie J, et al. The Impact of Mutations in SARS-CoV-2 Spike on Viral Infectivity and Antigenicity. Cell. 2020;182(5):1284−94.
29. Forster P, Forster L, Renfrew C, et al. Phylogenetic network analysis of SARS-CoV-2 genomes. Proc Natl Acad Sci U S A. 2020;117(17):9241−3.
30. Genomic epidemiology of novel coronavirus - Global subsampling [Internet]. Nextstrain; 2020 [cited 2020 Oct 10]. Available from: https://nextstrain.org/ncov/global.
31. Kristensen I, Aaby P, Jensen H. Routine vaccinations and child survival: follow up study in Guinea-Bissau, West Africa. BMJ. 2000;321(7274):1435−8.
32. Netea MG, Quintin J, van der Meer JWM. Trained immunity: a memory for innate host defense. Cell Host Microbe. 2011;9(5):355−61.
33. Netea MG, Quintin J, van der Meer JW, et al. Trained immunity: A program of innate immune memory in health and disease. Science. 2016;352(6284):aaf1098.
34. Arts RJW, Moorlag SJCFM, Novakovic B, et al. BCG Vaccination Protects against Experimental Viral Infection in Humans through the Induction of Cytokines Associated with Trained Immunity. Cell Host Microbe. 2018;23(1):89−100.
35. Khader SA, Divangahi M, Hanekom W, et al. Targeting innate immunity for tuberculosis vaccination. J Clin Invest. 2019;129(9):3482−91.
36. Aarreberg LD, Wilkins C, Ramos HJ, et al. Interleukin-1β Signaling in Dendritic Cells Induces Antiviral Interferon Responses. mBio. 2018;9(2):e00342-18.
37. Miyasaka M. Is BCG vaccination causally related to reduced COVID-19 mortality? EMBO Mol Med. 2020;12(6):e12661.
38. Miller A, Reandelar MJ, Fasciglione K, et al. Correlation between universal BCG vaccination policy and reduced mortality for COVID-19. medRxiv. doi: 10.1101/2020.03.24.20042937.
39. Akiyama Y, Ishida T. Relationship between COVID-19 death toll doubling time and national BCG vaccination policy. medRxiv. doi: 10.1101/2020.04.06.20055251.
40. Escobar LE, Molina-Cruz A, Barillas-Mury C. BCG vaccine protection from severe coronavirus disease 2019 (COVID-19). Proc Natl Acad Sci U S A. 2020;117(30):17720−6.
41. Giamarellos-Bourboulis EJ, Tsilika M, Moorlag S, et al. Activate: Randomized Clinical Trial of BCG Vaccination against Infection in the Elderly. Cell. 2020;183(2):315−23.
42. Hamiel U, Kozer E, Youngster I. SARS-CoV-2 Rates in BCG-Vaccinated and Unvaccinated Young Adults. JAMA. 2020;323(22):2340−1.
43. Bacille Calmette-Guérin (BCG) vaccination and COVID-19 [Internet]. Geneva: World Health Organization; 2020 [cited 2020 Oct 10]. Available from: https://www.who.int/news-room/commentaries/detail/bacille-calmette-gu%C3%A9rin-(bcg)-vaccination-and-covid-19.
44. Tomiyama H, Miwa K, Shiga H, et al. Evidence of presentation of multiple HIV-1 cytotoxic T lymphocyte epitopes by HLA-B*3501 molecules that are associated with the accelerated progression of AIDS. J Immunol. 1997;158(10):5026−34.
45. Kawashima Y, Pfafferott K, Frater J, et al. Adaptation of HIV-1 to human leukocyte antigen class I. Nature. 2009;458(7238):641−5.
46. Murakoshi H, Akahoshi T, Koyanagi M, et al. Clinical Control of HIV-1 by Cytotoxic T Cells Specific for Multiple Conserved Epitopes. J Virol. 2015;89(10):5330−9.
47. Keicho N, Itoyama S, Kashiwase K, et al. Association of human leukocyte antigen class II alleles with severe acute respiratory syndrome in the Vietnamese population. Hum Immunol. 2009;70(7):527−31.
48. Chen YM, Liang SY, Shih YP, et al. Epidemiological and genetic correlates of severe acute respiratory syndrome coronavirus infection in the hospital with the highest nosocomial infection rate in Taiwan in 2003. J Clin Microbiol. 2006;44(2):359−65.
49. Draft landscape and tracker of COVID-19 candidate vaccines [Internet]. Geneva: World Health Organization; 2020 [cited 2020 Oct 10]. Available from: https://www.who.int/publications/m/item/draft-landscape-of-covid-19-candidate-vaccines.
50. Folegatti PM, Ewer KJ, Aley PK, et al. Safety and immunogenicity of the ChAdOx1 nCoV-19 vaccine against SARS-CoV-2: a preliminary report of a phase 1/2, single-blind, randomised controlled trial. Lancet. 2020;396(10249):467−78.
51. Zhu F-C, Li Y-H, Guan X-H, et al. Safety, tolerability, and immunogenicity of a recombinant adenovirus type-5 vectored COVID-19 vaccine: a dose-escalation, open-label, non-randomised, first-in-human trial. Lancet. 2020;395(10240):1845−54.
52. Grifoni A, Weiskopf D, Ramirez SI, et al. Targets of T Cell Responses to SARS-CoV-2 Coronavirus in Humans with COVID-19 Disease and Unexposed Individuals. Cell. 2020;181(7):1489−501.