Immune Response to SARS-CoV-2 Infection

Fengji Cai*

School of Life and Health, Dalian University, Dalian City, Liaoning Province, 116000, China

Abstract: SARS-CoV-2 is a positive RNA virus which is responsible for the COVID-19 epidemic in 2020. This article provides an overview of this novel coronavirus, including its genome, proteins, receptor recognition, and synthesizes research on the immune responses and marker events triggered by the novel coronavirus. It emphasizes the critical roles of immune cells, specifically T cells and B cells, in the immune response. The article also highlights the virus's ability to replicate within lung-resident macrophages and variations in immune responses among different patients. The discussion regarding the persistence of immune protection is ongoing, but studies indicate that prior infection may offer protection for at least ten months, even though reinfection remains possible. These findings are of significant importance for understanding the immune mechanisms of COVID-19, assessing disease severity, and devising effective treatment strategies.

1. Introduction

The COVID-19 pandemic is caused by the beta-coronavirus SARS-CoV-2 (severe acute respiratory syndrome coronavirus-2), which has been ongoing since September 8, 2019 [1][2]. To date, the global reported cases of SARS-CoV-2 have surpassed 765 million, with cumulative deaths reaching 6.9 million [1]. As a positive-sense virus, SARS-CoV-2 carries genomic RNA that directly serves as messenger RNA (mRNA) for protein synthesis upon infecting host cells. Its key components consist of the surface spike (S) protein, membrane (M) protein, nucleocapsid (N) protein, and the RNA genome [3]. Droplet and touch transmission are the two primary ways that SARS-CoV-2 spreads. The incubation period lasts, on average, 3 to 7 days, but can last up to 14 days [4]. Its characterized respiratory lower and upper tract infection cause common symptoms include fever, cough, muscle pain, fatigue, and difficulty breathing, with occasional occurrences of sputum production, headache, and hemoptysis [4-6]. Also, according to a count in March 2023, one million people in the UK who have been chronically infected with the virus reported difficulty concentrating, with three quarters of them reporting memory loss or confusion. And these symptoms are often referred to as “brain fog” [7]. SARS-CoV-2 shares a considerable genetic homology for about 79.6% with SARS-CoV, both possessing regions that encode for the S protein, M protein, and other elements [2] [8]. However, a notable distinction lies in the length of their genomes, with SARS-CoV-2 having approximately 30,000 bp, slightly longer than SARS-CoV's 29,700 bp. Despite similar genome sizes, significant differences emerge in their coding and non-coding regions, including promoters and regulatory elements within their RNA sequences. These variations contribute to the unique characteristics and behavior of each virus.

A minimum of 29 proteins are encoded by SARS-CoV-2's (+) RNA genome. Among the four structural proteins, the spike protein is a large membrane protein comprising approximately 1,200 amino acids and forms spike-like protrusions on the virus's outer layer. The spike protein consists of two subparts, S1 and S2, where S1 is for binding to receptors on the surface of host cells, while S2 mediates the membrane fusion of virus and cell [9]. Despite the spike protein's protein sequence being quite different between SARS-CoV-2 and SARS-CoV, the three-dimensional structures of these two kinds are strikingly similar, displaying less than 75% sequence homology. The spike protein of SARS-CoV-2 has two conformations, RBD down and RBD up [4]. A noteworthy feature unique to the SARS-CoV-2 spike protein is the Furin cleavage site,which dose not exist in SARS-CoV. The Furin cleavage site is a specific sequence, "RRAR" (Arg-Arg-Ala-Arg), located between the S1 and S2 subunits. Human Furin protease thermodynamically shows high affinity(-37 kcal) that binds to the SARS-CoV-2 spike protein. The Furin enzyme can recognize and cleave this site, leading to the separation of the S1 and S2 subunits. It is believed that the presence of this cleavage site plays a key role in the entry of virus into host cells, potentially strengthening its efficiency and transmissibility. The Furin cleavage site in the SARS-CoV-2 spike protein highlights a significant difference between SARS-CoV-2 and its related virus, SARS-CoV, contributing to the distinct characteristics and behavior of the two viruses during infection [3][5].

Since the initial draft of this article, multiple variants of SARS-CoV-2 have emerged, including Delta and Omicron, which have higher transmissibility and partial immune escape characteristics, and have had a significant impact on the global pandemic. Figure 1 shows that the discovery of a new variant B.1.1.529 (Omicron) in South

*Corresponding author: felixcai2001@foxmail.com

Africa has led to a significant increase in the number of infected cases, making it highly contagious.

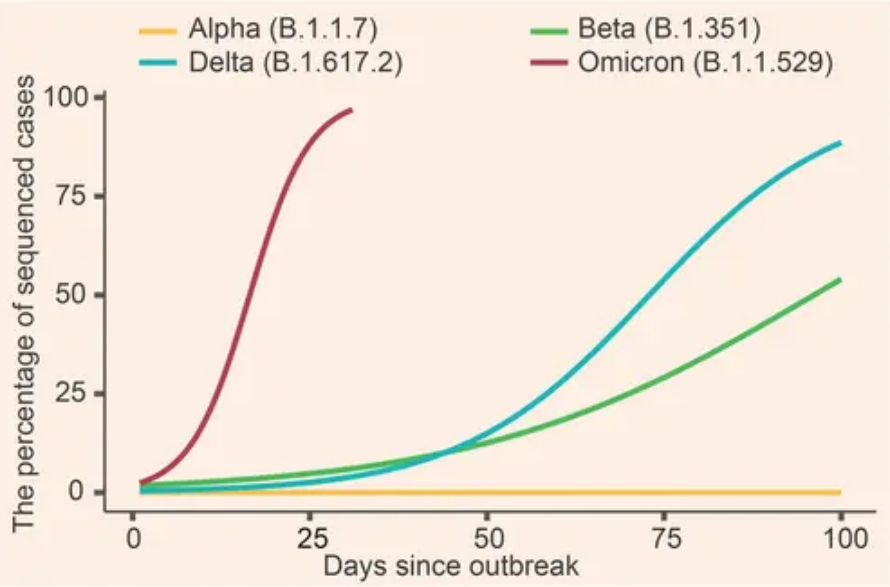

Figure 1. Omicron's ultra-high propagation rate

According to a research article published by Academician Huang Jianping's team at Lanzhou University in The Innovation, researchers used the Global Prediction System (GPCP) for the COVID-19 pandemic and used a modified model to predict the development of the COVID-19 pandemic. This model uses real epidemic data for relevant predictions. Therefore, the results can reflect the true development of the COVID-19 pandemic. According to the predicted results, by November 2023, the daily COVID-19 cases worldwide will decrease to about 3000. With more and more countries announcing the relaxation of restrictions and the restoration of social life, it can be said that the COVID-19 pandemic will end in November 2023. But by the end of the pandemic, the total number of confirmed cases will reach at least 750 million.

2. Mechanism of Sars-Cov-2 infection

Research on the beginning of SARS-CoV-2 can refer to the relevant contents of SARS. Studies have proved that the SARS coronavirus is transmitted by bat to civet to man, especially in 2013, the SARS coronavirus was traced to Rhinolepis sinensis [10]. It was previously believed that the Huanan Seafood market in Wuhan was the cause of the outbreak when a cluster of cases surfaced there. However, there is evidence that a total of 13 of the first 41 confirmed COVID-19 cases had no history of contact with this seafood market; three of the four people who were infected earlier had not been to the seafood market, so the seafood market was not the only source of exposure.

The invasion mechanism of SARS-CoV-2 is a multifaceted process encompassing viral attachment, binding, and entry into the host cell (known as cellular intrusion), followed by viral replication, assembly, and release. Among these steps, viral attachment stands out as a pivotal stage.

Previous studies have illuminated that the receptor for the SARS virus is angiotensin-converting enzyme 2 (ACE2). In this intricate dance between the virus and host cells, the spike protein of SARS-CoV-2 assumes a critical role. This spike protein acts as the primary mediator, binding to and entering host cells. Further investigations have confirmed that SARS-CoV-2 also relies on ACE2 for its entry. The process of infection by SARS-CoV-2 additionally requires the involvement of cell protease TMPRSS2. The main surface glycoprotein spike of SARS-CoV-2 is proteolytically cleaved by TMPRSS2. Research has demonstrated that inhibitors of TMPRSS2 have broad preventive and therapeutic effects against various strains of SARS-CoV-2.

Crucially, the SARS-CoV-2 spike protein's receptor-binding domain (RBD) has a more compact shape and is changing in certain regions. These modifications greatly increase ACE2's binding affinity for SARS-CoV-2 by stabilizing two virus-binding hotspots at the RBD-ACE2 interface. This heightened binding affinity enhances the virus's infectiousness. These structural modifications in the spike protein contribute to the increased transmissibility of SARS-CoV-2. Recognizing the pivotal role of the spike protein in viral infection, it has become a focal point for the development of COVID-19 vaccines.

Presently, numerous COVID-19 vaccines are engineered using the spike protein or its fragments. These vaccines are designed to stimulate the human immune system, inducing the production of antibodies targeting this protein. The presence of these antibodies effectively prevents the virus from binding to host cells, thwarting infection and providing robust protection against COVID-19 [5].

Indeed, NRP1 (Neuropilin-1) stands out as a significant player in the infection process of SARS-CoV-2 too. This protein has the ability to bind furin-cleaved substrates, specifically S1(Arg-Arg-Ala-Arg), substantially enhancing the infectivity of SARS-CoV-2

[3]. Moreover, a monoclonal antibody that targets NRP1 can reverse this impact. An in-depth pathological examination of the olfactory epithelium from corpses of patients who died with COVID-19 showed that NRP1-positive cells in close proximity to the nasal cavity were infected with SARS-CoV-2.

On another front, CD147, which is sometimes referred to as basigin or EMMPRIN, is a notable transmembrane glycoprotein that belongs to the immunoglobulin superfamily. CD147 has been shown to play roles in the growth of tumors, the invasion of Plasmodium, and the vulnerability to viral and bacterial infections. [10]. In 2020, a crucial discovery was made by researchers who found that the SARS-CoV-2 spike protein interacts with the host cell receptor CD147. SARS-CoV-2 replication was inhibited by using the CD147 antibody Meplazumab, reducing CD147 expression, or inhibiting CD147 in Vero-E6 and BEAS-2B cell lines. Additionally, CD147-antagonistic peptide-9 exhibited inhibitory effects on SARS-CoV.

SARS-CoV-2 can effectively replicate in resident macrophages of the lungs, which has a significant impact on the progression and severity of the disease. The infection of macrophages not only increases the efficiency of virus transmission, but may also exacerbate the inflammatory process by activating or inhibiting immune responses, leading to damage and dysfunction of lung tissue. Research has shown that this replication in resident macrophages in the lungs may be associated with severe clinical manifestations of COVID-19, especially in patients with weaker immune systems. Therefore, a deep understanding of this mechanism is of great significance for developing effective treatment strategies and managing clinical care for COVID-19 patients.

These discoveries underscore the intricate ways in which SARS-CoV-2 interacts with host cells, shedding light on potential avenues for therapeutic interventions and further deepening our understanding of the virus's mechanisms of infection.

3. Life cycles of the virus

Following the virus invasion, a series of intricate events unfold within the host cell. Initially, receptor recognition and binding take place, facilitated by TMPRRS2 and other proteases. After the S protein's second cleavage, the cellular and viral membranes fuse together to reveal a fusion peptide that inserts into the membrane. The antiparallel six-helix bundle formed by two heptad repeats in S2 permits membrane mixing and the release of the viral DNA into the cell.

The replicase produces the polyproteins pp1a and pp1ab prior to replication by translating two open reading frames (ORFs) from the virion genomic RNA, rep1a and rep1b. These polyproteins contain specific non-structural proteins (nsps), which are later cleaved into individual nsps. A significant portion of these nsps forms the replicase-transcriptase complex (RTC), crucial for both replicating viral RNA and transcribing sub-genomic RNAs.

Once the viral replication machinery is active, it begins generating copies of the virus's genetic material, including both the complete genome and smaller RNA segments. Coronaviruses are known for their ability to undergo genetic recombination, which contributes significantly to their diversity and evolution.

Translation produces the viral structural proteins (Spike, Envelope, and Membrane) subsequent to replication and sub-genomic RNA synthesis. These proteins enter the endoplasmic reticulum (ER) and proceed through the secretory pathway to the Golgi intermediate compartment (ERGIC) of the ER. Mature virions are assembled inside the ERGIC when viral genomes, which are encapsidated by the Nucleocapsid (N) protein, combine with membranes made of structural proteins. Despite its low abundance, the E protein assists the M protein in the assembly of virion through a number of proposed processes.

Once constructed, virions are delivered by vesicles to the cell surface, where they are exocytosed and freed from the cell. This marks the completion of one viral lifecycle, setting the stage for the repetition of these intricate processes in subsequent infections.

4. Immune response after infection

Although the durability of the immune protection is debated, both animals and human studies show that this protection is likely. Reinfection with SARS-CoV-2 is possible, but prior infection offers a consistent level of protection of approximately 87% for up to at least 10 months.

T cell responses during the immune response arise early and are correlated with protection; however, in cases of severe disease, these responses are substantially weakened and are linked to strong activation and lymphopenia. The impact of T cell immunity in controlling SARS-CoV-2 may have been understated up until this point. Research on MERS and SARS-CoV-1 indicates that T cells may be important in managing the illness, and increased antibody levels in SARS-CoV-1 infections have been connected to worsening clinical outcomes and increased inflammation. Early cytotoxic CD8+ T cell response initiation, usually shown 7 days after symptom onset and peaking at 14 days, is linked to both successful viral clearance and modest disease progression. Serious lymphocytopenia is observed in the blood of many acutely infected people, and it is also linked to a catastrophic clinical outcome. This results in a pattern of "coexisting suppression and activation," with peripheral T cell loss of up to 80%. A type 1 CD4+ profile is linked to effective viral management, but a type 2 profile is frequently observed in patients with severe illness. Nonetheless, an overactive immune response can have negative consequences, and excessively elevated levels of T cell activation have been linked to unfavorable clinical outcomes. A balanced generation of IL-10 and inflammatory cytokines characterizes virus-specific T cell responses in asymptomatic infection, whereas the severe pattern of T cell activation is completely different in acute SARS-CoV-2 infection. Conversely, the generation of

inflammatory mediators is more polarized in symptomatic disease. Certain post-mortem studies have suggested the presence of lymphocytic infiltration in tissue, although the patterns observed are quite diverse. High viral load levels are also frequently found in the afflicted tissue. The CD4+ and CD8+ memory T cell responses that are unique to SARS-CoV-2 normally represent 0.5% and 0.2% of the T cell repertoire, respectively. However, it's important to note that there is significant heterogeneity in these responses among different individuals.

B cells are essential for developing protective immunity against SARS-CoV-2 because they also produce antibodies, which are essential for the humoral immune response. B cells have a complex and dynamic function in viral infection, presenting antigens, secreting antibodies, and producing cytokines. Research on severe COVID-19 cases has revealed a more robust memory B cell response to the S protein and a higher antibody response to the N and S proteins. This suggests that severe infections may offer better protection against SARS-CoV-2 reinfection. Humoral immune responses are essential for efficiently clearing and preventing reinfection with SARS-CoV-2. They play a vital role in individuals infected with the virus. In cases of immune deficiency, where humoral responses are limited, the clearance of SARS-CoV-2 is ineffective, potentially leading to recurrent infections. In germinal centers, naïve B cells interact with antigens and CD4+ T cells to form memory B cells (MBCs) that produce high-affinity antibodies and long-lived plasma cells (LLPCs). Some B cells undergo extrafollicular responses, rapidly turning into short-lived plasma cells (SLPCs) that secrete low-affinity antibodies. T follicular helper cells (Tfh) in germinal centers activate and regulate B cell activity. Long-lived plasma cells (LLPCs) move and settle in the bone marrow as part of the initial immune response to SARS-CoV-2 infection. There, they constantly manufacture neutralizing antibodies that offer protection against reinfection. This process occurs after the germinal center (GC) response. Research has shown that SARS-CoV-2-specific LLPCs can persist in the bone marrow for 7 to 11 months, offering effective protection, particularly in cases of mild COVID-19 infections. Upon activation, B cells undergo differentiation into cells that produce antibodies (Abs), which serve as a powerful defense against infections that may be primary or secondary. Serum IgM antibody production usually outweighs that of other antigen-specific antibodies such as IgA and IgG in viral infections. However, serum antibody formation against SARS-CoV-2 S and N proteins, including IgM, IgA, and IgG, happens almost simultaneously in COVID-19 patients. Research also suggests that IgA plays a more significant role in neutralizing the virus compared to IgG and dominates the early-stage humoral response to SARS-CoV-2 infection in mucosal tissues. Age, gender, antigen load, levels of matching neutralizing antibodies (nAb), and ethnicity are among the characteristics that are linked to the variability in disease severity, humoral immune response, B cell reaction, and antibody titers among patients. These outcomes are also seen in cases of SARS infection. B cells are essential for controlling the release of antibodies as well as for controlling inflammatory cytokines. However, the exact mechanisms underlying the formation and amplification of this cytokine storm remain unknown. Then comes to the secondary response. In cases of COVID-19 reinfection, memory B cells (MBCs) that circulate throughout the body are swiftly activated and differentiate into antibody-secreting cells. However, the MBCs are driven to form new germinal centers (GCs) in order to manufacture higher-affinity antibodies if these circulating antibodies are absent or if they are activated by a variation of the original pathogen. Unfortunately, most patients infected with SARS-CoV lose their antigen-specific antibodies, implying that the immune memory's protection against reinfection by coronaviruses only lasts for a limited duration. Presently, our understanding of mucosal immunity against SARS-CoV-2 lags behind our knowledge of systemic immunity. Those with severe illnesses had larger IgG responses in nasal samples taken from COVID-19 patients, while those with milder or moderate symptoms had higher IgA responses. Memorization B cells (MBCs) are essential during reinfection because they produce a local response that quickly increases the production of antigen-specific antibodies. This is in addition to the fact that preexisting antibodies in mucosal tissues can provide protection against pathogen infections such as COVID-19. MBCs are found in various human tissues, including the lungs, spleen, lymph nodes, and gut.

Cytotoxic T cells recognize the viral peptide MHC I complex on the surface of infected cells, release perforin and granzymes, and directly kill infected cells. Helper T cells promote immune response by secreting cytokines such as interferon gamma and interleukin. After recognizing viral antigens, B cells differentiate into plasma cells and memory B cells. Plasma cells produce a large amount of antibodies to neutralize the virus and prevent it from binding to host cells, while memory B cells respond quickly upon reinfection, providing long-term immune protection.

The infection and replication of SARS-CoV-2 in human macrophages residing in the lung is another important cause of illness. A research study employed a reporter strain, SARS-CoV-2-mNG (encoding the fluorescent protein mNG), to track infected cells in MISTRG6-hACE2 mice, ultimately elucidating the replication of SARS-CoV-2 within human macrophages. Research has shown that individuals previously infected with SARS-CoV-2 may receive a certain degree of immune protection for at least ten months. For example, Cohen et al. (2021) tracked blood samples from infected individuals for up to 8 months and found that neutralizing antibody levels and memory B and T cell responses remained relatively stable during this period, supporting the possibility of long-term immune protection after infection.

Individual differences in immune response may be caused by various factors, including age, comorbidity, and genetic factors. For example, the immune system of elderly people may not be as effective as that of young people, which may explain why they often have more severe clinical manifestations after being infected with SARS-CoV-2. Comorbidity, such as cardiovascular disease or diabetes, may also affect the function of

immune cells and inflammatory response. In addition, genetic factors play a crucial role in determining an individual's sensitivity to viruses and the intensity of their immune response. The interaction of these factors may explain why different patients have such diverse responses to SARS-CoV-2 infection.

The global imbalance in vaccination has led to the sustained spread and mutation of the virus in certain regions, which not only exacerbates local epidemics but also threatens global public health security. In order to effectively combat emerging variants, the international community must strengthen cooperation, improve vaccine accessibility and vaccination rates, especially in countries and regions with limited resources.

5. Conclusion

Here are the key points from the provided information regarding immune responses and markers in COVID-19: (1) T-cell responses: When levels drop below 20%, severe patients frequently show lymphopenia. Severe illness may be predicted by initial lymphopenia. (2) CD8+ T cells: A significant decrease in lymphopenia, falling below 5%, is seen in CD8+ T cells and may indicate a serious illness. (3) Th1-Th2 responses: Both CD4 and CD8 Th1 responses are required for normal antiviral immunity. A cytokine storm, primarily including Th1 and inflammatory responses, is indicative of a systemic inflammatory response linked to severe illness. Additionally, this may aid in the activation of inflammasomes. (4) Eosinophils: A considerable proportion of hospitalized patients (between 50% and 80%) have reduced levels of circulating eosinophils. (5) Particular antibody levels: During the acute phase, there is an increase in IgM that is specific to the virus, and during the convalescent phase, there is an increase in IgG that is particular to the virus. (6) Cytokine storm: Severe cases release high amounts of both innate and adaptive cytokines, which are linked to the severity of the disease. (7) Acute-phase reactants: Severe cases have high amounts of these reactants, and initial high values are predictive of severe illness.

The findings of this study have important implications for current and future treatment strategies. Identifying the key molecular mechanisms of virus replication and immune escape provides targets for the development of new therapeutic drugs. Its in-depth understanding of immune responses can help improve the design of existing vaccines to stimulate more effective immune protection. With further understanding of the virus lifecycle, we can develop new treatment strategies to address virus mutations and improve treatment targeting. These strategies will provide new weapons for the fight against COVID-19 and prepare for similar outbreaks in the future.

References

1. https://covid19.who.int
2. Kooraki S, ,et al. Coronavirus (COVID-19) Outbreak: What the Department of Radiology Should Know. J Am Coll Radiol. 2020 Apr;17(4):447-451.
3. Li F. Structure, function, and evolution of coronavirus spike proteins. Annu Rev Virol, 2016, 3 (1):237-261.
4. General office of the national health commission. Pneumonia diagnosis and treatment plan for new coronavirus infection (trial version 5). Issued on February 4, 2020.
5. Lai, , et al. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19): The epidemic and the challenges. Int. J. Antimicrob. Agents 2020, 55, 105924.
6. Cheetham NJ, et al. The effects of COVID-19 on cognitive performance in a community-based cohort: a COVID symptom study biobank prospective cohort study. eClinicalMedicine. Published online July 21, 2023:102086.
7. Kavanagh E. Long Covid brain fog: a neuroinflammation phenomenon? Oxf Open Immunol. 2022 Sep 27;3(1):iqac007.
8. Walls AC, et al. Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein. Cell. 2020;181(2):281-292.e6.
9. Yao H, et al. Molecular Architecture of the SARS-CoV-2 Virus. Cell. 2020;183(3):730-738.e13.
10. Lu, et al. Basolateral CD147 induces hepatocyte polarity loss by E-cadherin ubiquitination and degradation in hepatocellular carcinoma progress. Hepatology 1, 317–332 (2018).