Analysis of the Characteristics of SARS-Cov-2 Spike Compared to SARS-Cov Spike and Its Role in the Pathogenesis of Infection– A Literature Review

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Abstract

The spread of COVID-19 that caused by SARS-CoV-2 happened very fast & has infected many people. WHO reported 1,051,635 cases of COVID-19 and 56,985 confirmed deaths caused by COVID-19 as of April 4, 2020. SARS-CoV-2 is a novel coronavirus that originates from *betacoronavirus* together with SARS-CoV. This study aims to find and understand the differences between the SARS-CoV-2 and SARS-CoV spikes in the infection and the spread of the resulting disease. This is a literature studies obtained from national journal articles indexed by SINTA and international journals indexed by Scimago or Scopus and published in 2015-2020. We concluded that there is a difference between the SARS-CoV-2 spike and the SARS-CoV spike, so that it can affect the spread of the disease. SARS-CoV-2 has a furin cleavage site that absent in SARS-CoV. The existence of this furin cleavage site can help the activation and the efficiency of the infection of the SARS-CoV-2 and increases its transmissibility to organs and populations when compared to viruses without furin cleavage sites such as SARS-CoV.

Keywords: *SARS-CoV-2, SARS-CoV, COVID-19 infection, coronavirus spike*

Introduction

The virus that caused COVID-19 has been identified as the novel coronavirus[1]. Coronavirus (CoV) is included in the coronaviridae family and classified into four genera : Alphacoronavirus, Betacoronavirus, Deltacoronavirus and Gammacoronavirus. The virus that causes COVID-19 itself is a betacoronavirus[2]. Other examples of viruses from betacoronavirus are Human Severe Acute Respiratory Syndrome (SARS)[1].

This viral pneumonia disorder was initially detected in patients who came from Wuhan (China) since December 12, 2019. Some infected patients had been exposed to or had contact with Huanan Seafood wholesale Market at the beginning of its spread[3]. World Health Organization officially named the disease as a coronavirus disease 2019 (COVID-19). The Coronavirus Study Group gave the name Severe Acute Respiratory Syndrome coronavirus 2 (SARS-CoV-2) for the virus that caused COVID-19 on February 11, 2020[4].

The spread of this viral infection occurs very quickly and has infected many people. As of April 4, 2020, WHO has reported, as many as 1,051,635 positive cases of COVID-19 and 56,985 deaths due to COVID-19 infection have been confirmed. Most cases of COVID-19 infection in Asia occurred in China with a total of 82,875 confirmed cases with a total of 3,335 deaths. In Indonesia, there have been 1,986 confirmed cases with 196 deaths(WHO, 2020).

COVID-19 has spread rapidly in more than 200 countries. The speed of spread and the high mortality rate due to COVID-19 infection have become a threat to society so that on January 30 2020 WHO officially announced that the COVID-19 epidemic was a public health emergency of international concern[4].

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Coronaviruses has its own characteristic, which is a spike that forms a crown which is also the origin of the 'coronavirus' name. The spike of the coronavirus is considered very important because it has a function to mediate the entry of viruses in host cells so that they can replicate[6].

From the description above, it is necessary to do further research on the pathogenesis of the coronavirus, especially COVID-19. It is very interesting to understand the morphology of this virus, especially the coronavirus spike, because the coronavirus spike is thought to play an important role in mediating the entry of the virus in the host cells[7]. Based on the data and description above, we conducted this research on the differences in the SARS-CoV-2 spike and the SARS-CoV spike which can affect the spread of viral infections in various organs and populations.

Methodology

This research is a literature Review with samples taken from at least 15 national journal articles indexed by SINTA and international journals indexed by Scimago or Scopus published in 2015-2020. The inclusion criteria are indexed journal articles with the theme COVID-19, SARS-CoV-2, SARS-CoV and Spike Coronavirus. The exclusion criteria were journal articles whose full text was inaccessible, not indexed by Scopus, Thomson Reuters, Web Science, Scimago, and SINTA, and published before 2015.

Similarities between SARS-CoV-2 and SARS-CoV

There's a large similarity between SARS-CoV and SARS-CoV-2. An experimental study was conducted by by comparing the genome sequences between the two viruses using multiple sequence alignment and phylogenetic analysis. The viral genome was obtained from samples of one of the patients who traveled to Wuhan between 29 December 2019 and 4 January as well as other βCoVs genomes from humans or mammals, including SARS-CoV which was used as a comparison. It was found that there was an 82% similarity between SARS-CoV-2 and SARS-CoV. The results of this study indicate that SARS-CoV-2 is included in the betacoronavirus, along with SARS-CoV[8].

A SARS-CoV-2 virus was detected in *balf* samples from 3 patients from Wuhan Jinyintan Hospital on December 30, 2019 by using RT-PCR. Then the virus genome sequence was carried out for comparison with another coronavirus. Genome sequence comparisons were performed using multiple sequence alignment, phylogenetic analysis was performed to determine the origin of the virus and the shape of the virus by means of transmission election microscopy. This detected virus has genetic similarities with viruses from the genus betacoronavirus. Seen from an electron microscope, the virus has a spherical shape with pleomorphism and has a solar corona appearance that matches the morphology of the coronaviridae family. It also concluded that SARS-CoV-2 that was detected belongs to the genera betacoronavirus, together with SARS-CoV[9].

An experimental study conducted on viruses taken from *balf* and cultures isolated samples from nine 2019-nCoV patients in Wuhan, China from 30 December - 7 January 2020. The viral genome was obtained through sequence analysis. Another viral genome used for comparison was obtained from GenBank. Then phylogenetic analysis was carried out to determine the genera of the virus. Through RT-PCR, the SARS-CoV-2 virus was found in the patient. Then it was also found that there was a genetic similarity between SARS-CoV-2 with SARS-CoV by 79% and with MERS-CoV by 50%. It was also concluded that SARS-CoV-2 is a novel betacoronavirus. SARS-CoV-2 also uses the angiotensin-converting enzyme 2 (ACE2) receptor to infect host cells[10].

A viruses obtained from samples taken from seven patients with severe pneumonia (six of whom had a history of exposure to the seafood market in Wuhan) at the ICU at the Wuhan Jin Yin Hospital at the beginning of the outbreak by RT-PCR was compared with other viral genomes obtained from GenBank. viral genomes were obtained through sequence analysis, cell receptors from SARS-CoV-2 were tested with hela cells that express ACE2, mock-transfected cells were used as control, phylogenetic analysis was performed to determine the genera of the virus and virus morphology was seen using an electron microscope. This study shows several results, namely 5 out of 7 patient samples showed positive PCR results. Then it was found that the similarity of the genome arrangement between SARS-

CoV-2 and SARS-CoV was 79.6%. Viral morphology shows the typical characteristics of coronavirus from an electron microscope. It was also concluded that SARS-CoV-2 was included in the SARSr-CoV (severe acute respiratory syndrome-related coronavirus) species. In addition, SARS-CoV-2 uses the same cell receptor as SARS-CoV for host cell infections, namely the angiotensin converting enzyme 2 (ACE2) receptor[11].

An experimental study on SARS-CoV-2 has been done by examined the virus obtained from a patient at the central hospital of Wuhan. A bronchoalveolar-lavage fluid (*balf*) sample from the patient was taken and the virus was confirmed by RT-PCR. Then the genomic sequence analysis was carried out on the viruses and phylogenetic analysis was done to determine the relationship with other viruses. Then the 2019-nCoV (now SARS-CoV-2) virus was found in the patient. The virus found (SARS-CoV-2) belongs to the subgenus sarbeCoVirus, genera betacoronavirus, just like SARS-CoV. The genomic sequence of the receptor binding domain (RBD) on the SARS-CoV-2 Spike protein shows a 73.3-74.9% similarity with RBD in the SARS-CoV spike. Then SARS-CoV-2 may also use the same cell receptor as SARS-CoV to infect, namely angiotensin converting enzyme 2 (ACE2)[12].

The five studies mentioned above[8-12] are discussing about the similiarity of SARS-CoV-2 and SARS-CoV, it is found that there is a similarity in genetic sequence between SARS-CoV-2 and SARS-CoV. The existence of this genetic sequence can be used as a evidence that SARS-CoV-2 may come from the same family as other viruses, especially SARS-CoV. The five studies[8-12] are also show that SARS-CoV-2 and SARS-CoV share a number of similarities, from genetic sequence to the common receptor that the two viruses use. In addition, the two viruses that have caused an outbreak are both included in the betacoronavirus genera[8-10,12]. These findings can be the basis for starting further studies on the differences between the two viruses.

The difference between the SARS-CoV-2 Spike and the SARS-CoV Spike

Although there are many similarities between SARS-CoV-2 and SARS-CoV, there are also differences in the Spike protein of the two viruses. This difference in spikes may affect the viral cycle, pathogenicity, antiviral development or also the spread of the virus. By using multiple-sequence alignment to compare the genome of the spike coronavirus (SARS-CoV-2, SARS-CoV and MERS-CoV). It was found that the furin-like cleavage site on protein S in SARS-CoV-2 was not found in SARS-CoV. This furin-like cleavage site could contribute to 2019-nCoV for efficient spread in human populations compared to other betacoronaviruses[13].

Sequence analysis was carried out to compare the SARS-CoV-2 and SARS-CoV spike genomes. In a study, western blot analysis was performed to observe proteases processed by the cleavage site[14]. A pseudovirus entry assay was performed to observe other proteases that could contribute to the SARS-CoV-2 spike and obtained some results. It was found that there were cleavage sites on the S SARS-CoV-2 protein that was not present in SARS-CoV. Furin is a protease that plays a role in the cleavage site of S SARS-CoV-2 protein. Even in the absence of furin, SARS-CoV-2 can still infect cells, which means that the virus is independent of a single protease. Because there are still other proteases that can activate SARS-CoV-2, namely TMPRSS2 and Cathepsin L. The presence of cleavage sites on the SARS-CoV-2 spike that is not present in the SARS-CoV spike may be able to expand virus tropism and increase transmission of SARS-CoV- 2 than SARS-CoV[14].

An experimental study comparing SARS-CoV-2 and other Betacoronaviruses, including SARS-CoV was done by using sequence analysis and phylogenetic analysis. Furin cleavage site was found in SARS-CoV-2 which was not present in other betacoronavirus subtypes B. This furin cleavage site can play an important role for infection and fusion of viral membranes against host cells[15].

Another experimental research also conducted on the SARS-CoV-2 and SARS-CoV spikes. Furin Cleavage site was found on the SARS-CoV-2 spike which was not found in SARS-CoV or other SARS-like coronaviruses. The furin Cleavage site causes SARS-CoV-2 to infect organs that produce furin, such as the brain, lungs, gastrointestinal tract, liver, pancreas and reproductive tissue which can cause systemic infections and even expand the distribution of the spread of COVID-19. This ability is not shared by other coronaviruses that do not have a furin cleavage site, so it is possible for SARS-

CoV-2 to have high transmission[16].

A variety of proteases that may be involved in the Spike protein process in SARS-CoV-2 was tested in an experimental study[17]. Biochemical peptide cleavage assay was used to confirm protease cleaving of the SARS-CoV-2 Spike protein. It was found that furin played a role in the activation of the SARS-CoV-2 spike and not in SARS-CoV. However, there are other proteases (trypsin and cathepsin) that can also activate the SARS-CoV-2 spike. Overall, the cleavage site on the SARS-CoV-2 spike can increase the work of the Spike protein over other coronaviruses[17].

Five studies regarding the difference between the spike of the two viruseshave been analyzed[13–17] and found The difference in the SARS-CoV-2 and SARS-CoV spikes is the presence of furin-like cleavage sites on protein S in SARS-CoV-2 which is not present in SARS-CoV. This site plays a role in COVID-19 for efficient spread in the human population. This furin cleavage site will activate the Spike protein on SARS-CoV-2 so that the virus can fuse with the host cells, then the infection process can occur. This furin cleavage site is not present in SARS-CoV[13–17].

The role of the Furin Cleavage Site in the SARS-CoV-2 Spike

The Furin Cleavage site found on the SARS-CoV-2 spike and not on SARS-CoV is thought to have an important role. By using in vitro and in vivo methods, it was found that the absent of furin cleavage site in SARS-CoV-2 affects the infection and pathogenesis processes. It also showed a decrease in viral replication in host cells and a weaker disease pathogenesis process when the virus was infected in hamsters. But the experiments carried out in hamsters showed only weak pathogenesis without decreased viral replication. With these two results, shows that the furin cleavage site in the SARS-CoV-2 spike is important for the process and the pathogenesis of viral infections[18].

Anothe study[20], also shows the role of cleavage sites in this virus. An experimental study was done and proved that the cleavage site that could be used for furin would be able to activate the spike protein in SARS-CoV-2 so that the virus can then be active and then can infect host cells. There is another protease that can activate the SARS-CoV-2 spike besides furin, namely TMPRSS2. Furin inhibitors shown to inhibit SARS-CoV-2 infection. This indicates that furin plays an important role in the activation and multiplication of viruses in host cells[19].

Other research on the furin cleavage site on SARS-CoV-2 has proven that the furin cleavage site is important in activating spike proteins so that the virus can infect host cells. Some furin inhibitors also could suppress furin activity in the virus. Furin inhibitors can also inhibit the entry and replication of the SARS-CoV-2 virus in host cell organs[20].

Another study of SARS-CoV-2 and SARS-CoV spikes was done by using Sequence analysis, and furin inhibitors. A site (cleavage site) was found on the SARS-CoV-2 Spike protein which is used for proteolytic processing that is not present in SARS-CoV, so that the spike from SARS-CoV-2 is activated and the virus can infect cells. Furin was found to play an important role in cleavage site protein S, especially for entry and fusion in host cells. This suggests that a viral variant with an optimal furin cleavage site may show increased viral spread and possible changes in pathogenicity. When this cleavage site is blocked (blockade), the virus is shown to still be able to enter the host cell. This indicates that SARS-CoV-2 can use proteases other than furin (the same as SARS-CoV, namely TMPRSS2 and Cathepsin L) to infect cells and is not dependent on furin alone, or vice versa. Viruses with a monobasic cleavage site (such as SARS-CoV) can only replicate in limited organs that have only certain proteases. While viruses with furin cleavage site or polybasic cleavage site (such as SARS-CoV-2) can spread in many organs, or even spread systemically and can cause broader disease symptoms[21].

A differences in the spikes of the two viruses also found in another experimental study[22]. Sequence analysis was carried out to compare the SARS-CoV-2 and SARS-CoV spike genomes. Proprotein convertase inhibitor (PPCI) was used to observe the role of the cleavage site for viruses. The small interfering rna (sirna) assay was used to determine the protease activating the SARS-CoV-2 spike. The pseudovirus entry assay was used to analyze the role of other proteases for SARS-CoV-2. It was found that there is a cleavage site on the SARS-CoV-2 Spike protein that was not found in the SARS-

CoV spike, this cleavage site is important for the process of viral infection. The efficiency of virus entry in host cells can be increased by the presence of cleavage sites on the S SARS-CoV-2 protein rather than SARS-CoV viruses that do not have a cleavage site. It also proven that furin is a PPC that activates the SARS-CoV-2 spike. As with SARS-CoV, entry of SARS-CoV-2 in host cells can be assisted by other proteases such as TMPRSS2 and Cathepsin L. This suggests that there are various proteases that can activate SARS-CoV-2 entry into host cells. The furin cleavage site contained in the SARS-CoV-2 spike allowed this virus to continue to infect cells and was not dependent on other proteases alone. This is important if the virus infects organs that have low expression of TMPRSS2 or Cathepsin L. The virus can then have high infectivity and can play a role in the wider spread of disease[22].

The five studies regarding the Role of Furin Cleavage Site on the SARS-CoV-2 Spike have been analyzed[18–22]. It can be concluded that the furin cleavage site in SARS-CoV-2 spike has a very important role for the virus. The Furin Cleavage site on the SARS-CoV-2 Spike plays a role in the activation of the virus spike, so that the virus can enter and fuse with host cells[19,20]. In addition, the SARS-CoV-2 virus spike can still be activated by proteases other than furin. So that the SARS-CoV-2 virus can still infect host cells even though there is a decrease in furin in the organ to be infected. This can also distinguish SARS-CoV-2 from SARS-CoV because SARS-CoV-2 does not only depend on a few proteases[19,21,22]. Some study conducted research by removing the furin cleavage site and with furin inhibitors proved it can inhibit the infection and virus replication. This suggests that a virus variant with an optimal furin cleavage site such as SARS-CoV-2 can show an increase in viral infection and possibly to cause the spread of disease in organs even in a wider population than viruses without a furin cleavage site such as SARS-CoV[18–22].

Conclusion

There is a difference between the SARS-CoV-2 spike and the SARS-CoV spike so that it can affect the spread of disease infections. SARS-CoV-2 spike has a furin cleavage site that does not found in SARS-CoV spike. The existence of this furin cleavage site can help the activation and efficiency of infection with the SARS-CoV-2 virus in various organs and increase its ability to spread to the population when compared to viruses without furin cleavage sites such as SARS-CoV.

Ethical Clearance – Not required since it is a literature review

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Conflict of Interest – Nil

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