Chapter 7

Omicron: Emergence, Detection, and Response

Serap Çetinkaya[1]
Nil Özbilüm Şahin[1]
Ali Fazıl Yenidünya[1]
and Burak Tüzün[2,*]

[1] Department of Molecular Biology and Genetics,
Science Faculty,
Sivas Cumhuriyet University, Sivas, Turkey
[2] Plant and Animal Production Department,
Technical Sciences Vocational School of Sivas,
Sivas Cumhuriyet University, Sivas, Turkey

Abstract

COVID-19 pandemic emerged in December 2019, and it is still a global threat with quite a few variants. The B.1.1.529, Omicron, identified in South Africa in 2021, was one of the most notorious variants due to its high infection and mutability capacity. The Omicron variant had mutations in the S region of the key RBD which boosted the transmission ability of the virus.

Resistance to antibodies and vaccines has been the key features of this variant. The rise of antibody-evading variants has reached alarming proportions and discovery of small molecule inhibitors has been thought to be a solution to this problem. Presently scientific attention has been substantially directed towards computational drug design performing molecular simulations to generate effective chemical agents to tackle the Omicron variant.

* Corresponding Author's Email: theburaktuzun@yahoo.com.

In: Advances in Health and Disease. Volume 67
Editor: Lowell T. Duncan
ISBN: 979-8-88697-790-5

Keywords: COVID-19, omicron, SARS-CoV-2, spike protein, vaccine, molecular docking

Introduction

The world at large has been living with the severe acute respiratory syndrome Coronavirus-2 (SARS-CoV-2), renown as COVID-19 pandemic, for more than two years, and yet a definite solution as to the eradication of the virus does not seem visible in the foreseeable future. Infection generally propagates by aerial means, like through droplets formed during coughing and sneezing. The pandemic involved more than 300 million people and 5.5 million casualties (Figure 1) [1]. The variants Alpha, Beta, Gamma, Delta, and Omicron, differ in their genetic contents, molecular structures, and in susceptibility to vaccines available, implicating insurmountable encounters in the clinical sense.

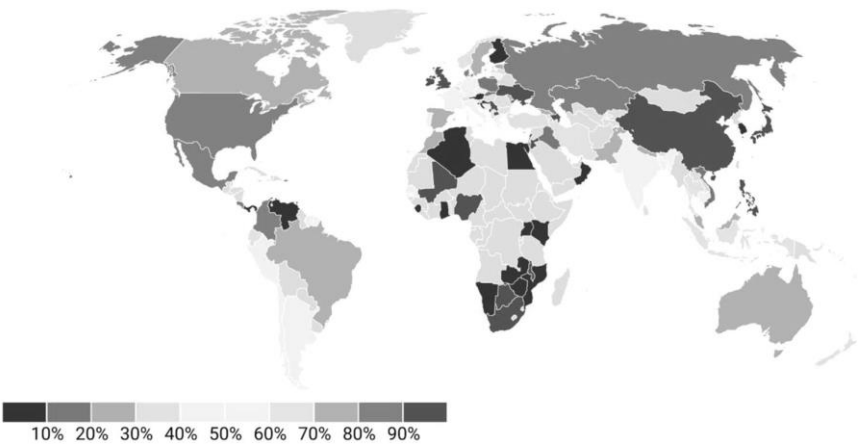

Figure 1. Omicron pandemic worldwide.

The spike protein (S) found on the virus outer surface demonstrate a very high binding affinity to the angiotensin converting enzyme (ACE) receptors on host cells (Figure 2). Mutations within the spike protein vary greatly among the variants. During the first step involves the binding of the S protein to ACE2 receptor on the host cell surface. SARS-CoV-2 then penetrates into the cell membrane. The virus particle enters the cell through a membrane fusion event,

involving furin and a type II transmembrane serine protease (TMPRSS2) or cathepsin L [2, 3].

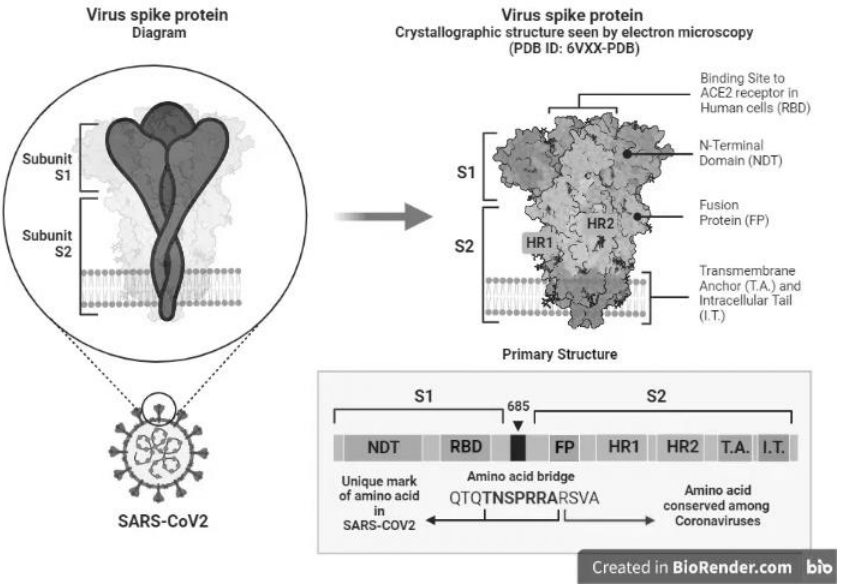

Figure 2. Structure of the SARS-CoV2 spike glycoprotein.

SARS-CoV-2 in the Past and Present

Coronaviruses (CoVs) have been known for many years as enveloped viruses with single-stranded RNA genomes that cause diseases in domestic and wild animals and humans (Figure 3). Although its seven types are known to date, four of them (HCoV 229E, HCoV NL63, HCoV HKU1 and HCoV OC43) cause only mild colds in humans, while SARS-CoV (severe acute respiratory syndrome Coronavirus), MERS-CoV (Middle East respiratory syndrome coranavirus) cause severe respiratory tract infections and death [4]. On 31 December 2019, Chinese authorities have first reported the Coronavirus SARS-CoV2, later named as COVID-19 (Coronaviridae Study Group of the International Committee on Taxonomy of Viruses, 2020). COVID-19 is an enveloped virus with 100-130 nm in diameter. Its genome is a positive-sense RNA with 29,727 nucleotides [5]. The World Health Organization (WHO) declared the worldwide disease a Public Health Emergency on 30 January

2020 and a pandemic on 11 March 2020 [6]. Common symptoms of COVID-19 infection are fever, fatigue, dry cough, diarrhoea, sore throat, myalgia, stomachache, loss of smell and sense of smell. Besides, respiratory failure, chest pain, haemoptysis, and confusion have also been reported [6]. Fortunately, most of the patients, having mild to moderate respiratory complaints, overcome the infection at home. Serious cases expectedly have involved elderly patients with susceptible backgrounds such as cardiovascular disease, diabetes, cancer, and chronic respiratory disease [7].

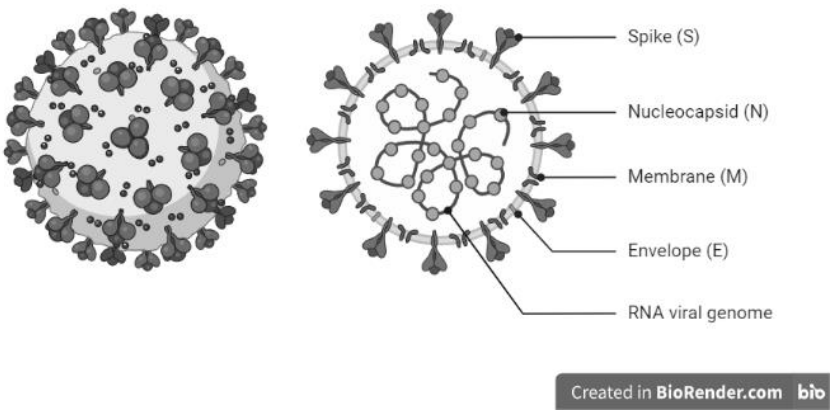

Figure 3. Human Coronavirus structure.

Molecular Features of Omicron Variant (B.1.1.529)

Viral pandemics often operate at different mutation levels and accordingly create viral strains specific to each mutation level [8]. RNA viruses are very much prone to high mutation rates within a very short period of time because they do not possess proofreading machinery [9]. Omicron, B.1.1.529, has first been identified in South Africa and Botswana [10]. Further research has intriguingly maintained that Omicron could not be evolutionarily related to the previously identified variants.

Subsequently, three proposals have been suggested as to the development of Omicron: silent evolution, evolution taken place during, chronic, long-term, infections, or in other animals, rodents in particular [11]. These suggestions are quite plausible because Omicron is not a single type but has three strains, BA.1, BA.2, and BA.3. The first one, once being the commonest global strain, gradually left its space to BA.2 sporadically. BA.3 is on the other hand

spatially a very restricted strain [12]. Omicron has globally become formidable, evading the available treatment methods. Its genome, ca 30 000 bs, harbours more than 18 000 mutations in exon sequences. These mutations are made up of insertions, deletions, nonsynonymous, and synonymous single nucleotide polymorphisms (SNPs) [13]. Around 32 mutations have now been reported in its Spike (S) protein.

Some of these mutations, K417N, E484K, N501Y, D614G, and T478K, residing within RBD, show similarities to those of the previously identified variants [13]. K417N involves an amino acid substitution, from lysine to asparagine and is shared with Beta variants. E484K, causing reinfection, converts glutamic acid to lysine and it is shared with both Beta and Gamma variants [12, 14]. E484K mutation in the gamma variant was capable of causing reinfection). N501Y involving the substitution from asparagine to tyrosine, is shared with the Alpha, Beta, and Gamma variants. D614G causing the change of aspartic acid to glycine is also mapped in the Alpha, Beta and Gamma [15, 16].

Transmissibility of Omicron

Mutation Type

Concern about the rapid transmissibility of the Omicron variant has increased in recent months as the number of cases folds dramatically, involving days and weeks, especially in the US, in Britain, and in South Africa [17]. The reports have maintained that Omicron was four-fold more infectious than the wild type while Gamma variant had similar infection rates to that of the wild type; Delta was almost twice efficient; the least infectious strain being the Beta variant (Table 1).

Table 1. Characteristic comparisons of Omicron and Delta variants [18]

Characteristic	Omicron	Delta
Country first found	South Africa	India
Time frst reported	November 24, 2021	December 5, 2020
Mutation residues	43	18
Δ69-70S deletion	+	-
Transmissibilty	3-6 fold	1 fold
R in Gauteng SA	>2	<1

Table 1. (Continued)

Characteristic	Omicron	Delta
Morbidity	Mild	Severe
Death	Rare	Common
Age of patients	Middle aged people	Children or elderly
Vaccine effect	33%	80%

Pathogenesis of Omicron

SARS-CoV-2 is able to infect a diverse range of host cells via their ACE2 receptor (Figure 4). The host cells can be from respiratory- or endothelial system: alveolar epithelial cells, and macrophages in particular [19]. Macrophage and neutrophil cells are the members of the innate immune system, and they define molecular patterns, activating the host immune system via Toll-like receptors. The cytokines interleukin-6 (IL-6), IFN-γ, and monocyte chemoattractant protein-1, recruit monocytes, macrophages, and neutrophils to the site of infection [20, 21]. These recruited cells release cytotoxic chemicals to kill the infected host cells [22]. It is also worth to mention that T cell responses to Omicron are maintained or even boosted in the majority of infected and vaccinated individuals [23]. It appears that T-cells can reduce the binding of the viral spike protein to ACE by more than 50% [24].

The growth of monocyte macrophages and neutrophils occurs at the site of infection due to the excessive presence of proinflammatory cytokines. Overproduction of proinflammatory cytokines in SARS-CoV-2 causes respiratory distress in the lung, resulting from the immune response to the virus. The cytotoxic substances released by these cells cause tissue damage and even cell death [25]. Although this immune response has been clearly understood in COVID-19, the immune reaction in the case of Omicron infection is still unclear [26]. The Omicron variant utilizes a new pattern to enter the host cell. Due to this pattern, the Omicron variant is more effective than the Delta variant. Omicron can cause upper respiratory tract infections but not more serious lung diseases. This makes the activity of Omicron clinically similar to previous Coronaviruses [27]. Replication of Omicron is reduced in cells overexpressing TMPRSS2. Omicron enters the cell by inhibiting TMPRSS2-mediated cell surface fusion. However, it follows endosomal fusion [28, 29].

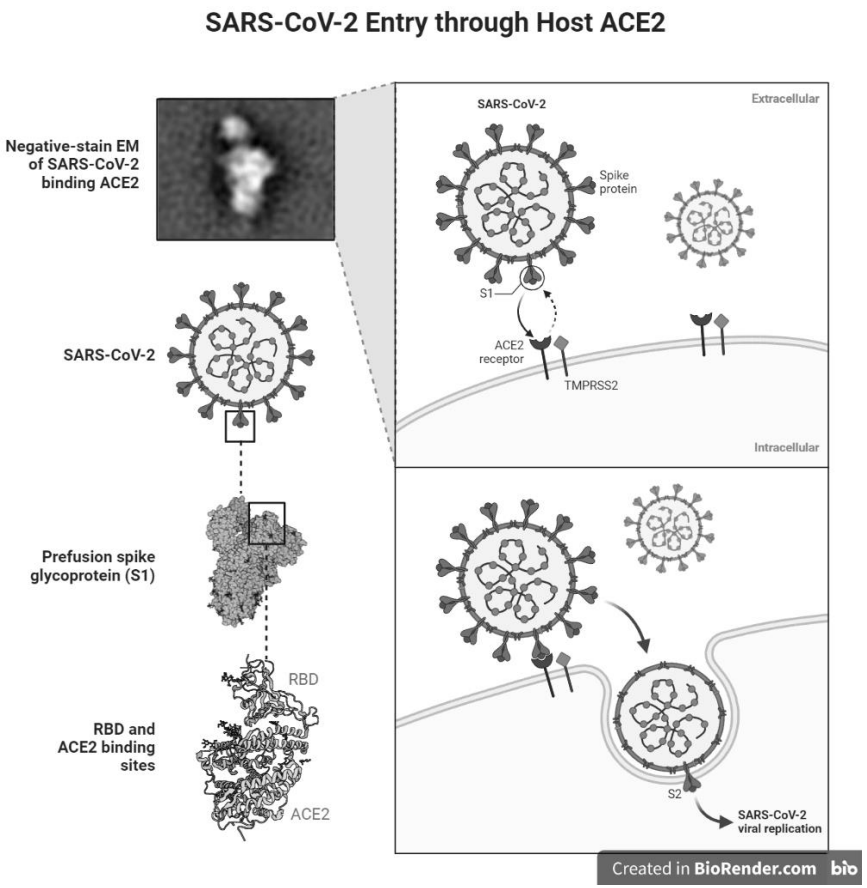

Figure 4. SARS-CoV-2 targeting of ACE2 receptor and entry in infected cell.

Clinical Symptoms of Omicron

Omicron infections involve common, less common, and severe symptoms. The former usually manifests as headache, runny nose, tiredness (mild or severe), sore throat, and sneezing. The second includes pharyngitis, headaches, discoloration of fingers and toes, skin rashes, red or itchy eyes, diarrhea, and stomach cramps. The last one is characterized by chest distress, difficulty in breathing and/or moving, and disorientation [30]. It might be useful to note that these symptoms are shared with those of the Delta variant.

There are not many cases of convulsion in children, but the data were not sufficient to infer that these cases are the result of infection.

The Omicron variant has been shown to replicate more rapidly in nasal epithelial cells than in lung or intestinal cell lines [29]. It has also been shown, in accordance with the previous inference, that Omicron multiplies rapidly in human respiratory organs and bronchi but is less potent in human alveolar organs and lungs [31]. These results indicate that Omicron infections mostly involve the upper respiratory tract. Most studies attributed this to the inefficient TMPRSS2 usage of Omicron [28]. Its high transmissibility rate might be to do with this specificity of the infection site. It can also mean that Omicron prognosis is less severe.

Severity of Omicron

Omicron infections can lead to organ dysfunction and sometimes to death. Its severity is explained by the need for and length of hospitalization, and the necessity for aeration [32]. The commonest infection manifestations are asymptomatic. As the Omicron diversity spread rapidly, there were serious increases in hospitalizations (Figure 5).

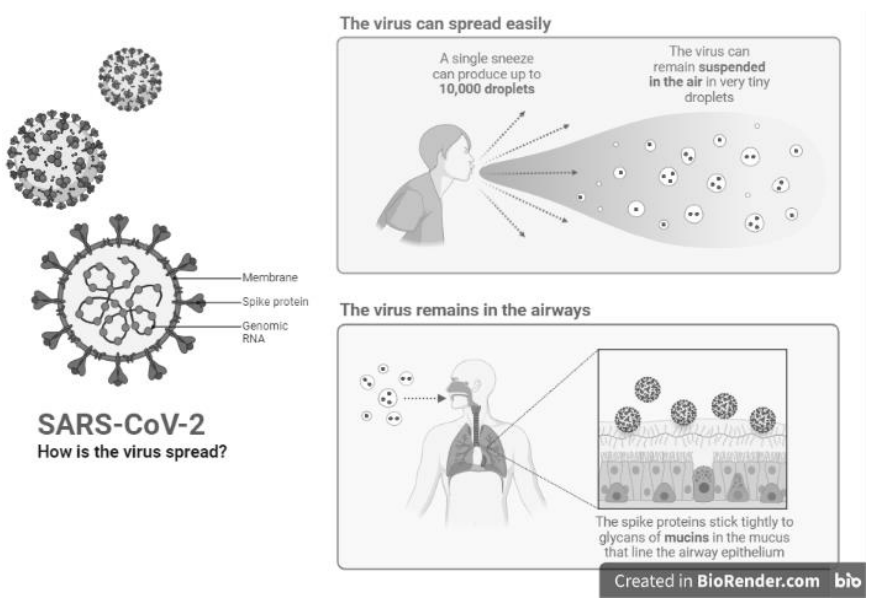

Figure 5. Spread of SARS-CoV-2 virus.

Unvaccinated people, the elderly, and people with a pre-existing chronic illness are at higher risk of developing a serious infection. These clinical features do not seem to differ according to the strains of Omicron [33], despite the possible growth advantage of BA.2.93. Vaccination before infection, economic status of the patient, and the quality of the healthcare service appear to greatly contribute to the outcome of Omicron infections [34].

Mutations in Omicron and Their Biological Consequences

The combination of K417N + E484K + N501Y mutations has been known to be lethal in the infections with Beta or Gamma variants. It also seems to increase transmissibility by 50%, and result in higher hospitalization rates, and death [35].

Omicron spike protein harbours at least 30 amino acid conversions, affecting its infection frequency [36]: Ala67Val, Thr95Ile, Gly142Asp, Δ69-70, Δ143-145, Leu212Ile, Gly339Asp, Lys417Asn, Δ211, Asn440Lys, Gly446Ser, Ser373Pro, Ser375Phe, Ser371Leu, Ser477Asn, Glu484Ala, Gln498Arg, Gln493Arg, Gly496Ser, Asn501Tyr, Tyr505His, Thr547Lys, Asn679Lys, Thr478Lys, Asn764Lys, Pro681His, Asp614Gly, Asp796Tyr, His655Tyr, Gln954His, Asn969Lys, Asn856Lys, and Leu981Phe.

Some exemplary effects of the above amino acid conversions were as follows: Asn 501Tyr positively influences Omicron's receptor binding capacity [37], and its rate of transmission. D614G, N501Y, and K417 also enable Omicron to be more transmissible [38]. N501Y also involves in transmissibility, and some other mutations enhance the binding affinity of S protein to ACE2.

Cellular kinases, such as PI3K/AKT, appear to be essential for the entry of Omicron. N501Y mutants can also imitate the epidermal growth factor in accomplishing the first step of the viral entry [39]. Moreover, His655Tyr adjacent to the furin cleavage point, can hasten the cleavage of S protein and the infectivity [40].

It seems that electrogenic mutations modulating the electrostatic forces between RBD of the spike protein and its receptor, ACE2, are implicated in the infection process, where higher Coulomb attractions increase the virus binding on the host cell surface [41, 42].

Cryo-EM structural analysis of the Omicron variant spike protein complexing with human ACE2 resolved hydrogen bonds and additional salt

bridges in mutant RBDs harboring the R493, R498, and S496 transformations [43].

RNA-dependent RNA polymerase (Nsp12) and protein 14 (Nsp14) are central in the replication of Omicron genome replication, and it has been suspected that mutant forms of these proteins might in part be responsible for the high mutation rate in the Omicron genome [34]. Omicron nucleocapsid protein also has mutant forms harbouring R203K and G204R conversions [44-46].

Directed amino acid conversions, Q493R, S373P, S375F, N501Y, S371L, T478K, and Q498R have indicated that amino acid at these positions in the wild type spike protein could be accounted for Omicron's high binding propensity to ACE2. Receptor binding domain, RBD, of the spike protein appears to be stabilized by the hydrophobic residues phenylalanine and leucine.

The pathogenicity of Omicron is mainly attributed to the unstructured protein regions. Omicron proteins have variant relatively few disordered regions. In its spike protein such a region resides between amino acids 468 and 473 [47].

Prediction of Escape Mutants through Bioinformatics Analysis

Deep mutational screening assays make it easy to estimate the influence of mutations on virus antigenicity. In this context, mutations in the RBD of the spike protein have been scrutinized using an "escape calculator" on the basis of experimental data. The mutational background of Omicron has been pictured using amino acid interaction (AAI) networks. Experimental data included the results obtained with tens of monoclonal antibodies. Calculations have been found to be in agreement with the experimental neutralization results [48].

Efficacy of COVID-19 Vaccines against the Omicron Variant

Recently, Immunotherapy has highlighted the state of treatment approaches and the effectiveness of COVID-19 vaccines that have received urgent approval against many SARS-CoV-2 variants [49]. Researchers have partially demonstrated the efficacy of various vaccine formulations, including

inactivated vaccines, nucleic acid vaccines, viral vector-based vaccines, and protein subunit vaccines, to reduce COVID-19 infection [49].

The Omicron variant of SARS-CoV-2 has been identified in COVID-19 vaccinated patients. While this reduces the immune invasion of the new variant, it increases the need for new vaccines. The Omicron virus spike protein 30 mutation is thought to be associated with an increase in circulating antibodies against the infection [50].

Compared to the Omicron Delta variant, it infects more of the upper respiratory tract than the lungs and promotes the spread of this variant [51, 52]. Studies have shown that CoronaVac and mRNA vaccines (RNA-1273 and BNT162b2) have a significantly lower neutralizing effect against the Omicron variant [51-53]. None of the 25 CoronaVac recipient serum samples collected in the vaccination tests conducted at the vaccination centers showed any neutralizing antibody titers against the Omicron isolates [54].

The two BNT162b2 vaccines, which can provide more than 90% protection against the Delta variant, are less effective against SARS-CoV-2 of the Omicron type [55]. In South Africa, two doses of BNT162b2 were associated with 33% efficacy against SARS-CoV-2 infection during the Omicron surge, while 70% efficacy against COVID-19 hospitalization [56]. In one study, polyclonal serum from individuals vaccinated with two doses of BNT162b2 COVID-19 vaccine and from recovered individuals reported complete resistance to several clinically used monoclonal antibodies in addition to neutralizing activity against Omicron [57].

Moreover, although the additional booster vaccine dose is four to six times lower than the wild-type SARS-CoV-2 strain, it produces a highly effective antibody response and reduces severe morbidity and mortality in Omicron-infected patients [58, 59]. Neutralization tests comparing human serum samples from individuals who received two and three doses of mRNA vaccine (BNT162b and mRNA-1273) found significantly lower efficacy against Beta and Omicron variants in subjects who received two doses. A similar reduction in neutralizing activity was observed in Coronavac and AstraZeneca, and neutralizing antibodies against the Omicron variant were not detected in any of them. Recent data highlight the importance of a third dose of mRNA vaccine, showing that two doses of vaccine are not sufficient to protect against Omicron infection [53, 60]. Laboratory studies with Pfizer-BioNTech vaccines show that a repeat dose alone can increase neutralizing antibody titers 25-fold compared to two other doses [61].

Vaccination programs against new variants should continue at full speed and SARS-CoV-2 variants should be closely monitored as part of global surveillance initiatives to ensure necessary countermeasures are taken [62].

A conserved T-cell activity was demonstrated in both previously infected and vaccinated sera conferring broad immunity against the Omicron variant. Confirmation of a pre-existing non-spiking memory T cell would justify the development of second-generation vaccines based on non-spiking antigens [63]. Some institutions have already started developing Omicron variant-specific COVID-19 vaccines and are confident enough to launch the vaccine by the end of July 2022 [64, 65].

Theoretical Calculation

Theoretical calculations provide significant time and financial savings for experimental processes by comparing the activities of molecules before experimental processes. molecular docking calculations are made in Schrödinger's Maestro Molecular modeling platform (version 12.8) [66]. As a result of the theoretical calculations made, large are achieved by determining the molecules with high activity. In this study, calculations were made for proteins that are omicron variants of SARS-CoV-2 (PDB ID: 7MRV, 7QO7, and 7U0N) [67-69]. These proteins are compared with their activities using molecules used for 30 selected diseases. The results obtained are given in Table 2.

In the calculations made, the numerical values of the molecules docking score parameter are compared in Table 2. In this comparison, it is thought that the molecules with the most negative docking score parameter value have higher activity. Therefore, the inhibitory potential of these molecules to the omicron variant of SARS-CoV-2 virus was compared [70-73].

Table 2. Activity values of molecules against omicron proteins

		Proteins		
		7MRV	7QO7	7U0N
1	Carvacrol	-5.08	-5.16	-6.29
2	Cynaropicrin	-5.69	-5.64	-6.53
3	Remdesivir	**-6.99**	-6.09	-7.40
4	Santalic acid	-3.26	-2.85	-3.98
5	Alpha-Bisabolol	-3.85	-3.43	-4.73
6	Chlorogenic Acid	-5.51	-6.03	-5.61
7	Bharangin	-4.54	-4.06	-4.78
8	Beta-pinene	-4.25	-3.79	-4.29

		Proteins		
		7MRV	7QO7	7U0N
9	BetaSitosterol	-3.47	-3.88	-4.27
10	Cissamine	-5.27	-6.04	-5.57
11	Eugenol	-3.71	-4.01	-4.86
12	Gallic acid	-4.83	-4.84	-5.58
13	Ribavirin	-6.79	-6.82	**-8.04**
14	6-Gingerol	-2.54	-2.89	-3.34
15	Favipiravir	-4.76	-5.56	-5.92
16	Quercetin	-5.72	-5.34	-5.68
17	Vitexin	-5.45	-6.41	-6.75
18	Apigenin	-5.79	-5.29	-6.24
19	Linoleic acid	0.24	0.35	-0.31
20	Rutin	-6.81	**-6.90**	-6.88
21	Cucurbitacin B	-5.82	-5.09	-5.60
22	Costunolide	-4.16	-5.13	-5.30
23	6-Shogaol	-2.30	-2.12	-2.77
24	Pellitorine	-0.86	-0.29	-0.61
25	Andrographolide	-6.20	-5.03	-5.28
26	Piperine	-4.88	-4.32	-5.52
27	Vasicine	-6.31	-6.05	-6.73
28	Thymol	-5.32	-4.58	-6.08
29	Piperidine	-4.77	-4.13	-5.29
30	Spathulenol	-4.25	-4.74	-6.54

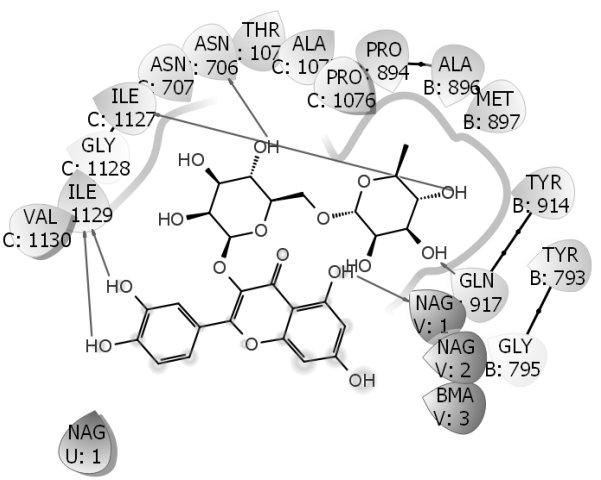

Figure 6. (Continued).

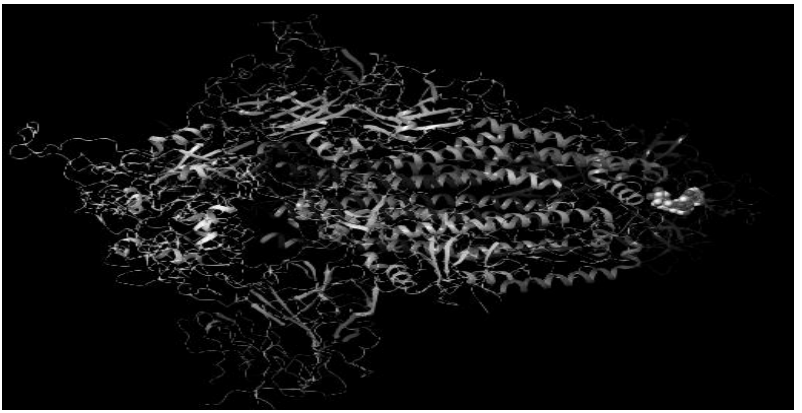

Figure 6. Illustration of molecule rutin interaction with 7QO7- protein.

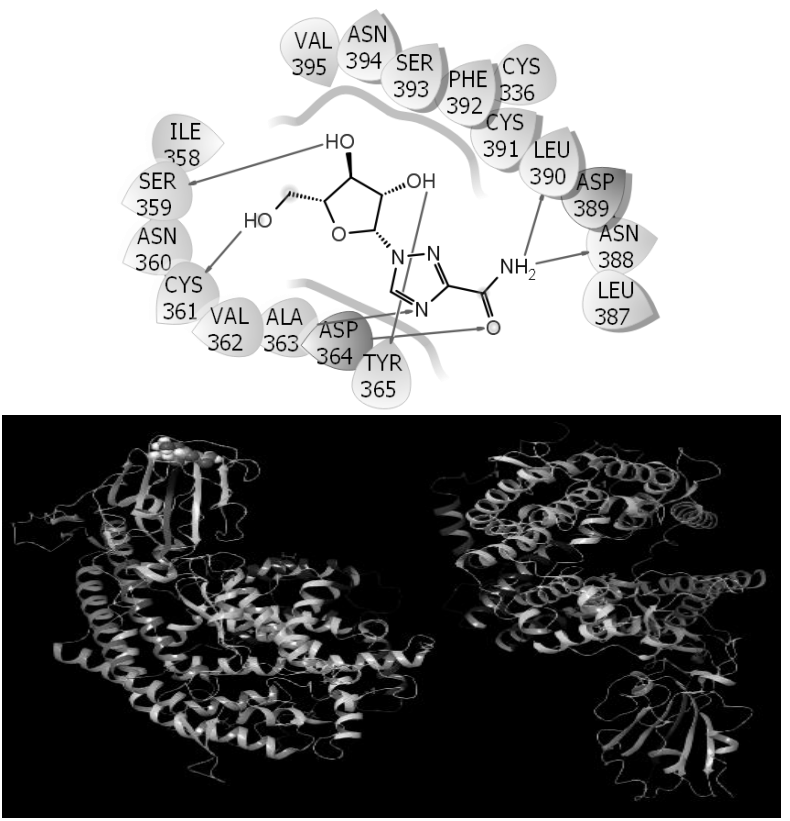

Figure 7a. Illustration of molecule ribavirin interaction with 7U0N protein.

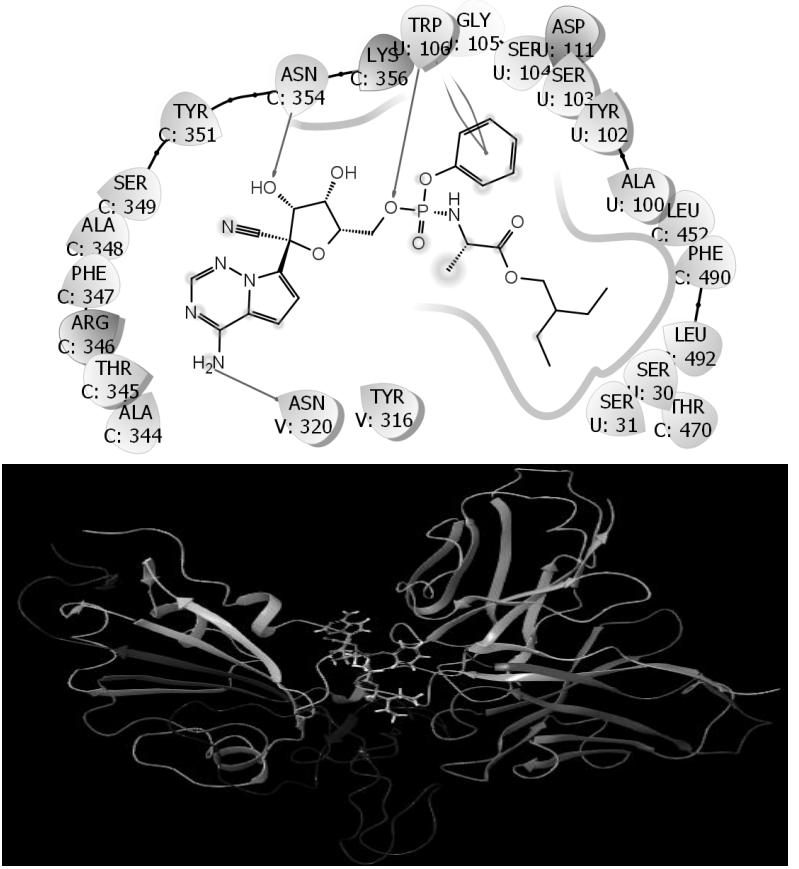

Figure 7b. Illustration of molecule remdesivir interaction with 7WRV protein.

Conclusion

The Omicron variant, which has been reported in many countries, has been declared an alarming variant by WHO. However, it is known that the severity of the disease is lower than other variants. Due to S and N protein mutations, capsid assembly and cell permeability increased 3 times in Omicron variant compared to Delta variant. The more stable structure of the omicron variant is supported by the fact that its RBD and the entire spike protein have more alpha-helical structures compared to the delta variant [74].

Although the Omicron variant is known to increase the contagiousness of COVID-19 infection, it is not yet clear whether it can trigger the disease or interfere with the immunity provided by the vaccine [75, 76]. People are currently receiving a third vaccine or booster that increases the level of neutralizing antibodies. Our immune cells exert an action against the Omicron invaders. This attenuated form is expected to cause herd immunity due to its high infectivity and lower mortality.

There is considerable uncertainty surrounding the various features of Omicron. Due to the rapid increase in the global availability of the Omicron variant, the urgency of administering repeated doses has been emphasized. There is much uncertainty about the benefits and advantages of vaccines against the Omicron variant. Although several studies have shown that the effectiveness of the vaccines against COVID 19 compared to the Omicron variant is low, people should still be vaccinated because the vaccines help to significantly reduce the severity of the disease. The Omicron variant is still a very new concept and therefore requires a lot of research to be clearly understood. Therefore, the administration of a booster dose of the COVID-19 vaccine can be tailored according to vaccination strategies. Effectively, the focus must be on accelerating vaccination booster programs and reintroducing stricter and preventive measures that reduce the spread and infectivity of the virus.

The big question will be whether the next variant will reverse this trend and cause more severe disease, or will lower pathogenicity, Omicron level, or better SARS-CoV-2 variants be introduced? While we have so far completely failed to predict the course of the pandemic, perhaps it is time to reconsider how new variants evolve, bearing in mind that various variants may evolve in different ways, we can predict what will happen next [77, 78].

In the calculations, the activities of 30 molecules whose molecules were studied were compared against the SARS-CoV-2 virus omicron variant. It was observed that the routine molecule against the omicron variant 7QO7 protein with -6.90 docking score parameter values, the ribavirin molecule against 7U0N protein with -8.04 docking score parameter value, and the molecule remdesivir with -6.99 docking score parameter value against 7WRV protein than other molecules. It is hoped that the calculations made will be an important guide before many *in virtro* and in vivo procedures in future.

Acknowledgments

The numerical calculations reported in this paper were fully/partially performed at TUBITAK ULAKBIM, High Performance and Grid Computing Center (TRUBA resources). This work was supported by the Scientific Research Project Fund of Sivas Cumhuriyet University (CUBAP) under the project number RGD-020.

References

[1] Sharma, V., Rai, H., Gautam, D. N., Prajapati, P. K., & Sharma, R. (2022). Emerging evidence on Omicron (B. 1.1. 529) SARS-CoV-2 variant. *Journal of Medical Virology*, 94(5), 1876-1885.

[2] Dong, Y., Dai, T., Liu, J., Zhang, L., & Zhou, F. (2020). Coronavirus in Continuous Flux: From SARS-CoV to SARS-CoV-2. *Advanced Science* (Weinheim, Baden-Wurttemberg, Germany), 7(20), 2001474.

[3] Garcia-Beltran, W. F., St Denis, K. J., Hoelzemer, A., Lam, E. C., Nitido, A. D., Sheehan, M. L., Berrios, C., Ofoman, O., Chang, C. C., Hauser, B. M., Feldman, J., Roederer, A. L., Gregory, D. J., Poznansky, M. C., Schmidt, A. G., Iafrate, A. J., Naranbhai, V., & Balazs, A. B. (2022). MRNA-based COVID-19 vaccine boosters induce neutralizing immunity against SARS-CoV-2 Omicron variant. *Cell*, 185(3), 457-466.e4.

[4] Yin, Y., & Wunderink, R. G. (2018). MERS, SARS and other Coronaviruses as causes of pneumonia: MERS, SARS and Coronaviruses. *Respirology*, 23(2), 130–137. https://doi.org/10.1111/resp.13196.

[5] Wu, A., Peng, Y., Huang, B., Ding, X., Wang, X., Niu, P., Meng, J., Zhu, Z., Zhang, Z., Wang, J., Sheng, J., Quan, L., Xia, Z., Tan, W., Cheng, G., & Jiang, T. (2020). Genome Composition and Divergence of the Novel Coronavirus (2019-nCoV) Originating in China. *Cell Host & Microbe*, 27(3), 325–328. https://doi.org/10.1016/j.chom.2020.02.001.

[6] Zheng, C., Huang, W. Y., Sheridan, S., Sit, C. H.-P., Chen, X.-K., & Wong, S. H.-S. (2020). COVID-19 Pandemic Brings a Sedentary Lifestyle in Young Adults: A Cross-Sectional and Longitudinal Study. *International Journal of Environmental Research and Public Health*, 17(17), 6035. https://doi.org/10.3390/ijerph17176035.

[7] Rothe, C., Schunk, M., Sothmann, P., Bretzel, G., Froeschl, G., Wallrauch, C., Zimmer, T., Thiel, V., Janke, C., Guggemos, W., Seilmaier, M., Drosten, C., Vollmar, P., Zwirglmaier, K., Zange, S., Wölfel, R., & Hoelscher, M. (2020). Transmission of 2019-nCoV Infection from an Asymptomatic Contact in Germany. *The New England Journal of Medicine*, 382(10), 970–971. https://doi.org/10.1056/NEJMc2001468.

[8] Burki, T. (2021). Understanding variants of SARS-CoV-2. *Lancet* (London, England), 397(10273), 462.

[9] Wang, Q., Wu, J., Wang, H., Gao, Y., Liu, Q., Mu, A., Ji, W., Yan, L., Zhu, Y., Zhu, C., Fang, X., Yang, X., Huang, Y., Gao, H., Liu, F., Ge, J., Sun, Q., Yang, X., Xu, W., … Rao, Z. (2020). Structural Basis for RNA Replication by the SARS-CoV-2 Polymerase. *Cell*, 182(2), 417-428.e13.

[10] Burki, T. K. (2022). Omicron variant and booster COVID-19 vaccines. The Lancet. *Respiratory Medicine*, 10(2), e17. https://doi.org/10.1016/S2213-2600(21)00559-2.

[11] Li, Q., Nie, J., Wu, J., Zhang, L., Ding, R., Wang, H., Zhang, Y., Li, T., Liu, S., Zhang, M., Zhao, C., Liu, H., Nie, L., Qin, H., Wang, M., Lu, Q., Li, X., Liu, J., Liang, H., … Wang, Y. (2021). SARS-CoV-2 501Y.V2 variants lack higher infectivity but do have immune escape. *Cell*, 184(9), 2362-2371.e9. https://doi.org/10.1016/j.cell.2021.02.042.

[12] Kannan, S., Shaik Syed Ali, P., & Sheeza, A. (2021). Evolving biothreat of variant SARS-CoV-2—Molecular properties, virulence and epidemiology. *European Review for Medical and Pharmacological Sciences*, 25(12), 4405–4412. https://doi.org/10.26355/eurrev_202106_26151.

[13] Daria, S., Bhuiyan, M. A., & Islam, M. R. (2022). Detection of highly muted Coronavirus variant Omicron (B.1.1.529) is triggering the alarm for South Asian countries: Associated risk factors and preventive actions. *Journal of Medical Virology*, 94(4), 1267–1268. https://doi.org/10.1002/jmv.27503.

[14] Resende, P. C., Bezerra, J. F., Teixeira Vasconcelos, R. H., Arantes, I., Appolinario, L., Mendonça, A. C., Paixao, A. C., Duarte, A. C., Silva, T., Rocha, A. S., Lima, A. B. M., Pauvolid-Corrêa, A., Motta, F. C., Teixeira, D. L. F., de Oliveira Carneiro, T. F., Neto, F. P. F., Herbster, I. D., Leite, A. B., Riediger, I. N., … Siqueira, M. M. (2021). Severe Acute Respiratory Syndrome Coronavirus 2 P.2 Lineage Associated with Reinfection Case, Brazil, June-October 2020. *Emerging Infectious Diseases*, 27(7), 1789–1794. https://doi.org/10.3201/eid2707.210401.

[15] Weissman, D., Alameh, M.-G., de Silva, T., Collini, P., Hornsby, H., Brown, R., LaBranche, C. C., Edwards, R. J., Sutherland, L., Santra, S., Mansouri, K., Gobeil, S., McDanal, C., Pardi, N., Hengartner, N., Lin, P. J. C., Tam, Y., Shaw, P. A., Lewis, M. G., … Montefiori, D. C. (2021). *D614G Spike Mutation Increases SARS CoV-2 Susceptibility to Neutralization. Cell Host & Microbe*, 29(1), 23-31.e4. https://doi.org/10.1016/j.chom.2020.11.012.

[16] Yi, C., Sun, X., Ye, J., Ding, L., Liu, M., Yang, Z., Lu, X., Zhang, Y., Ma, L., Gu, W., Qu, A., Xu, J., Shi, Z., Ling, Z., & Sun, B. (2020). Key residues of the receptor binding motif in the spike protein of SARS-CoV-2 that interact with ACE2 and neutralizing antibodies. *Cellular & Molecular Immunology*, 17(6), 621–630.

[17] Dyer, O. (2021). COVID-19: South Africa's surge in cases deepens alarm over omicron variant. *BMJ* (Clinical Research Ed.), 375, n3013. https://doi.org/10.1136/bmj.n3013.

[18] Ren, S.-Y., Wang, W.-B., Gao, R.-D., & Zhou, A.-M. (2022). Omicron variant (B.1.1.529) of SARS-CoV-2: Mutation, infectivity, transmission, and vaccine resistance. *World Journal of Clinical Cases*, 10(1), 1–11. https://doi.org/10.12998/wjcc.v10.i1.1.

[19] Shi, Y., Wang, Y., Shao, C., Huang, J., Gan, J., Huang, X., Bucci, E., Piacentini, M., Ippolito, G., & Melino, G. (2020). COVID-19 infection: The perspectives on

immune responses. *Cell Death and Differentiation*, 27(5), 1451–1454. https://doi.org/10.1038/s41418-020-0530-3.

[20] Mortaz, E., Tabarsi, P., Varahram, M., Folkerts, G., & Adcock, I. M. (2020). The Immune Response and Immunopathology of COVID-19. *Frontiers in Immunology*, 11, 2037. https://doi.org/10.3389/fimmu.2020.02037.

[21] Valizadeh, H., Abdolmohammadi-Vahid, S., Danshina, S., Ziya Gencer, M., Ammari, A., Sadeghi, A., Roshangar, L., Aslani, S., Esmaeilzadeh, A., Ghaebi, M., Valizadeh, S., & Ahmadi, M. (2020). Nano-curcumin therapy, a promising method in modulating inflammatory cytokines in COVID-19 patients. *International Immunopharmacology*, 89(Pt B), 107088. https://doi.org/10.1016/j.intimp.2020.107088.

[22] Schultze, J. L., & Aschenbrenner, A. C. (2021). COVID-19 and the human innate immune system. *Cell*, 184(7), 1671–1692. https://doi.org/10.1016/j.cell.2021.02.029.

[23] Shen, X. (2022). Boosting immunity to Omicron. *Nature Medicine*, 28(3), 445–446. https://doi.org/10.1038/s41591-022-01727-0.

[24] Kodera, S., Rashed, E. A., & Hirata, A. (2022). Estimation of Real-World Vaccination Effectiveness of mRNA COVID-19 Vaccines against Delta and Omicron Variants in Japan. *Vaccines*, 10(3), 430. https://doi.org/10.3390/vaccines10030430.

[25] Mehta, P., McAuley, D. F., Brown, M., Sanchez, E., Tattersall, R. S., Manson, J. J., & HLH Across Speciality Collaboration, UK. (2020). COVID-19: Consider cytokine storm syndromes and immunosuppression. *Lancet* (London, England), 395(10229), 1033–1034. https://doi.org/10.1016/S0140-6736(20)30628-0.

[26] Boechat, J. L., Chora, I., Morais, A., & Delgado, L. (2021). The immune response to SARS-CoV-2 and COVID-19 immunopathology—Current perspectives. *Pulmonology*, 27(5), 423–437. https://doi.org/10.1016/j.pulmoe.2021.03.008.

[27] Fall, A., Eldesouki, R. E., Sachithanandham, J., Morris, C. P., Norton, J. M., Gaston, D. C., Forman, M., Abdullah, O., Gallagher, N., Li, M., Swanson, N. J., Pekosz, A., Klein, E. Y., & Mostafa, H. H. (2022). The displacement of the SARS-CoV-2 variant Delta with Omicron: An investigation of hospital admissions and upper respiratory viral loads. *EBioMedicine*, 79, 104008. https://doi.org/10.1016/j.ebiom.2022.104008.

[28] Meng, B., Abdullahi, A., Ferreira, I. A. T. M., Goonawardane, N., Saito, A., Kimura, I., Yamasoba, D., Gerber, P. P., Fatihi, S., Rathore, S., Zepeda, S. K., Papa, G., Kemp, S. A., Ikeda, T., Toyoda, M., Tan, T. S., Kuramochi, J., Mitsunaga, S., Ueno, T., … Gupta, R. K. (2022). Altered TMPRSS2 usage by SARS-CoV-2 Omicron impacts infectivity and fusogenicity. *Nature*, 603(7902), 706–714. https://doi.org/10.1038/s41586-022-04474-x.

[29] Peacock, T. P., Goldhill, D. H., Zhou, J., Baillon, L., Frise, R., Swann, O. C., Kugathasan, R., Penn, R., Brown, J. C., Sanchez-David, R. Y., Braga, L., Williamson, M. K., Hassard, J. A., Staller, E., Hanley, B., Osborn, M., Giacca, M., Davidson, A. D., Matthews, D. A., & Barclay, W. S. (2021). The furin cleavage site in the SARS-CoV-2 spike protein is required for transmission in ferrets. *Nature Microbiology*, 6(7), 899–909. https://doi.org/10.1038/s41564-021-00908-w.

[30] Maisa, A., Spaccaferri, G., Fournier, L., Schaeffer, J., Deniau, J., Rolland, P., Coignard, B., regional COVID-19 investigation team, & EMERGEN consortium. (2022). First cases of Omicron in France are exhibiting mild symptoms, November 2021-January 2022. *Infectious Diseases Now*, 52(3), 160–164.

[31] Lamers, M. M., Mykytyn, A. Z., Breugem, T. I., Groen, N., Knoops, K., Schipper, D., van Acker, R., van den Doel, P. B., Bestebroer, T., & Koopman, C. D. (2022). SARS-CoV-2 Omicron efficiently infects human airway, but not alveolar epithelium. *BioRxiv*, 2022–01.

[32] Lewnard, J. A., Hong, V. X., Patel, M. M., Kahn, R., Lipsitch, M., & Tartof, S. Y. (2022). Clinical outcomes among patients infected with Omicron (B. 1.1. 529) SARS-CoV-2 variant in southern California. *MedRxiv*, 2022–01.

[33] Wolter, N., Jassat, W., group, D.-G. author, von Gottberg, A., & Cohen, C. (2022). Clinical severity of Omicron sub-lineage BA. 2 compared to BA. 1 in South Africa. *MedRxiv*, 2022–02.

[34] Gao, S.-J., Guo, H., & Luo, G. (2022). Omicron variant (B.1.1.529) of SARS-CoV-2, a global urgent public health alert! *Journal of Medical Virology*, 94(4), 1255–1256. https://doi.org/10.1002/jmv.27491.

[35] Wahid, M., Jawed, A., Mandal, R. K., Dailah, H. G., Janahi, E. M., Dhama, K., ... & Haque, S. (2021). Variants of SARS-CoV-2, their effects on infection, transmission and neutralization by vaccine-induced antibodies. *Eur. Rev. Med. Pharmacol. Sci*, 25, 5857-5864.

[36] Kupferschmidt, K., & Vogel, G. (2021). How bad is Omicron? Some clues are emerging. *Science*, 374(6573), 1304-1305.

[37] Ali, F., Kasry, A., & Amin, M. (2021). The new SARS-CoV-2 strain shows a stronger binding affinity to ACE2 due to N501Y mutant. *Medicine in Drug Discovery*, 10, 100086.

[38] Poudel, S., Ishak, A., Perez-Fernandez, J., Garcia, E., León-Figueroa, D. A., Romaní, L., ... & Rodriguez-Morales, A. J. (2022). Highly mutated SARS-CoV-2 Omicron variant sparks significant concern among global experts–What is known so far?. *Travel Medicine and Infectious Disease*, 45, 102234.

[39] Kazybay, B., Ahmad, A., Mu, C., Mengdesh, D., & Xie, Y. (2022). Omicron N501Y mutation among SARS-CoV-2 lineages: Insilico analysis of potent binding to tyrosine kinase and hypothetical repurposed medicine. *Travel Medicine and Infectious Disease*, 45, 102242.

[40] Chen, R. E., Zhang, X., Case, J. B., Winkler, E. S., Liu, Y., VanBlargan, L. A., ... & Diamond, M. S. (2021). Resistance of SARS-CoV-2 variants to neutralization by monoclonal and serum-derived polyclonal antibodies. *Nature Medicine*, 27(4), 717-726.

[41] Pawłowski, P. (2022). SARS-CoV-2 variant Omicron (B. 1.1. 529) is in a rising trend of mutations increasing the positive electric charge in crucial regions of the spike protein S. *Acta Biochimica Polonica*, 69(1), 263-264.

[42] Khandia, R., Singhal, S., Alqahtani, T., Kamal, M. A., Nahed, A., Nainu, F., ... & Dhama, K. (2022). Emergence of SARS-CoV-2 Omicron (B. 1.1. 529) variant, salient features, high global health concerns and strategies to counter it amid ongoing COVID-19 pandemic. *Environmental Research*, 209, 112816.

[43] Mannar, D., Saville, J. W., Zhu, X., Srivastava, S. S., Berezuk, A. M., Tuttle, K. S., ... & Subramaniam, S. (2022). SARS-CoV-2 Omicron variant: Antibody evasion and cryo-EM structure of spike protein–ACE2 complex. *Science*, 375(6582), 760-764.

[44] Leary, S., Gaudieri, S., Parker, M. D., Chopra, A., James, I., Pakala, S., ... & Mallal, S. (2021). Generation of a novel SARS-CoV-2 sub-genomic RNA due to the R203K/G204R variant in nucleocapsid: homologous recombination has potential to change SARS-CoV-2 at both protein and RNA level. *Pathogens and Immunity*, 6(2), 27.

[45] Mourier, T., Shuaib, M., Hala, S., Mfarrej, S., Alofi, F., Naeem, R., ... & Pain, A. (2021). Saudi Arabian SARS-CoV-2 genomes implicate a mutant Nucleocapsid protein in modulating host interactions and increased viral load in COVID-19 patients. *medRxiv*, 2021-05.

[46] Quarleri, J., Galvan, V., & Delpino, M. V. (2022). Omicron variant of the SARS-CoV-2: a quest to define the consequences of its high mutational load. *GeroScience*, 1-4.

[47] Kumar, S., Thambiraja, T. S., Karuppanan, K., & Subramaniam, G. Omicron and Delta variant of SARS-CoV-2: a comparative computational study of spike protein. *BioRxiv*. Published online December 3, 2021.

[48] Greaney, A. J., Starr, T. N., & Bloom, J. D. (2021). An antibody-escape calculator for mutations to the SARS-CoV-2 receptor-binding domain. *BioRxiv*.

[49] Chavda, V. P., Kapadia, C., Soni, S., Prajapati, R., Chauhan, S. C., Yallapu, M. M., & Apostolopoulos, V. (2022). A global picture: therapeutic perspectives for COVID-19. *Immunotherapy*, 14(5), 351-371.

[50] Tiecco, G., Storti, S., Degli Antoni, M., Focà, E., Castelli, F., & Quiros-Roldan, E. (2022). Omicron genetic and clinical peculiarities that may overturn SARS-CoV-2 pandemic: a literature review. *International Journal of Molecular Sciences*, 23(4), 1987.

[51] Edara, V. V., Manning, K. E., Ellis, M., Lai, L., Moore, K. M., Foster, S. L., ... & Suthar, M. S. (2022). mRNA-1273 and BNT162b2 mRNA vaccines have reduced neutralizing activity against the SARS-CoV-2 omicron variant. *Cell Reports Medicine*, 3(2), 100529.

[52] England, P. H. (2021). SARS-CoV-2 variants of concern and variants under investigation in England. *Technical Briefing*, 23.

[53] Muik, A., Lui, B. G., Wallisch, A. K., Bacher, M., Mühl, J., Reinholz, J., ... & Şahin, U. (2022). Neutralization of SARS-CoV-2 Omicron by BNT162b2 mRNA vaccine–elicited human sera. *Science*, 375(6581), 678-680.

[54] Lu, L., Mok, B. W. Y., Chen, L. L., Chan, J. M. C., Tsang, O. T. Y., Lam, B. H. S., ... & To, K. K. W. (2022). Neutralization of severe acute respiratory syndrome Coronavirus 2 omicron variant by sera from BNT162b2 or CoronaVac vaccine recipients. *Clinical Infectious Diseases*, 75(1), e822-e826.

[55] Hoffmann, M., Krüger, N., Schulz, S., Cossmann, A., Rocha, C., Kempf, A., ... & Pöhlmann, S. (2022). The Omicron variant is highly resistant against antibody-mediated neutralization: Implications for control of the COVID-19 pandemic. *Cell*, 185(3), 447-456.

[56] Collie, S., Champion, J., Moultrie, H., Bekker, L. G., & Gray, G. (2022). Effectiveness of BNT162b2 vaccine against omicron variant in South Africa. *New England Journal of Medicine*, 386(5), 494-496.

[57] Gruell, H., Vanshylla, K., Tober-Lau, P., Hillus, D., Schommers, P., Lehmann, C., ... & Klein, F. (2022). mRNA booster immunization elicits potent neutralizing serum activity against the SARS-CoV-2 Omicron variant. *Nature Medicine*, 28(3), 477-480.

[58] World Health Organization. *Weekly epidemiological update on COVID-19* – 8 March 2022. Edition 82. (2022). www.who.int/publications/m/item/weekly-epidemiological-update-on-covid-19---8-march-2022.

[59] Garcia-Beltran, W. F., Denis, K. J. S., Hoelzemer, A., Lam, E. C., Nitido, A. D., Sheehan, M. L., ... & Balazs, A. B. (2022). mRNA-based COVID-19 vaccine boosters induce neutralizing immunity against SARS-CoV-2 Omicron variant. *Cell*, 185(3), 457-466.

[60] Planas, D., Saunders, N., Maes, P., Guivel-Benhassine, F., Planchais, C., Buchrieser, J., ... & Schwartz, O. (2022). Considerable escape of SARS-CoV-2 Omicron to antibody neutralization. *Nature*, 602(7898), 671-675.

[61] Hogan, A. B., Wu, S. L., Doohan, P., Watson, O. J., Winskill, P., Charles, G., ... & Ghani, A. C. (2022). The value of vaccine booster doses to mitigate the global impact of the Omicron SARS-CoV-2 variant. *medRxiv*, 2022-01.

[62] Chang, A., & Yu, J. (2022). Fighting fire with fire: immunogenicity of viral vectored vaccines against COVID-19. *Viruses*, 14(2), 380.

[63] Kundu, R., Narean, J. S., Wang, L., Fenn, J., Pillay, T., Fernandez, N. D., ... & Lalvani, A. (2022). Cross-reactive memory T cells associate with protection against SARS-CoV-2 infection in COVID-19 contacts. *Nature Communications*, 13(1), 80.

[64] Pfizer. Pfizer and BioNTech provide update on Omicron variant. www.pfizer.com/news/press-release/press-release-detail/pfizer-and-biontech-provide-update-omicron-varia.

[65] de Oliveira Campos, D. M., da Silva, M. K., Silva de Oliveira, C. B., Fulco, U. L., & Nobre Oliveira, J. I. (2022). Effectiveness of COVID-19 vaccines against Omicron variant. *Immunotherapy*, 14(12), 903-904.

[66] *Schrödinger Release 2021-3: Maestro*, Schrödinger, LLC, New York, NY, 2021.

[67] Geng, Q., Shi, K., Ye, G., Zhang, W., Aihara, H., & Li, F. (2022). Structural basis for human receptor recognition by SARS-CoV-2 Omicron variant BA. 1. *Journal of Virology*, 96(8), e00249-22.

[68] Ni, D., Lau, K., Turelli, P., Raclot, C., Beckert, B., Nazarov, S., ... & Trono, D. (2021). Structural analysis of the Spike of the Omicron SARS-COV-2 variant by cryo-EM and implications for immune evasion. *BioRxiv*, 2021-12.

[69] Chen, E., Reiss, K., Shah, D., Manjula, R., Allen, B., Murphy, E. L., ... & Lisi, G. P. (2021). A structurally preserved allosteric site in the MIF superfamily affects enzymatic activity and CD74 activation in D-dopachrome tautomerase. *Journal of Biological Chemistry*, 297(3).

[70] Poustforoosh, A., Hashemipour, H., Tüzün, B., Azadpour, M., Faramarz, S., Pardakhty, A., ... & Nematollahi, M. H. (2022). The impact of D614G mutation of

SARS-COV-2 on the efficacy of anti-viral drugs: A comparative molecular docking and molecular dynamics study. *Current Microbiology*, 79(8), 241.

[71] Tüzün, B., Sayin, K., & Ataseven, H. (2022). Could Momordica Charantia Be Effective In The Treatment of COVID19?. *Cumhuriyet Science Journal*, 43(2), 211-220.

[72] Karataş, H., Tuzun B., Kokbudak Z. (2022). Could pyrimidine derivative be effective against Omicron of SARS-CoV-2?. *Bratislava Medical Journal*, 123(7), 505-513.

[73] Ataseven, H., Sayin, K., Tüzün, B., & Gedikli, M. (2021). Could boron compounds be effective against SARS-CoV-2?. *Bratislava Medical Journal-Bratislavske Lekarske Listy*, 122(10).

[74] Jr., E. E. a. B. L. Omicron symptoms: What we know about illness caused by the new variant. *NCBNEWS*. 2021.

[75] Outlook Web Desk. January 3 2022. Is 'Florona' Another COVID-19 Variant? What We Know About The New Disease From Israel. *Outlook*.

[76] Sakay, Y. N. (2022). By the Numbers: COVID-19 Vaccines and Omicron. *Healthline*.

[77] Sigal, A. (2022). Milder disease with Omicron: is it the virus or the pre-existing immunity?. *Nature Reviews Immunology*, 22(2), 69-71.

[78] Mukherjee, A. G., Wanjari, U. R., Murali, R., Chaudhary, U., Renu, K., Madhyastha, H., ... & Gopalakrishnan, A. V. (2022). Omicron variant infection and the associated immunological scenario. *Immunobiology*, 152222.