Jurnal Farmasi dan Ilmu Kefarmasian Indonesia
Pharmacy and Pharmaceutical Sciences Journal
Published by Faculty of Pharmacy Universitas Airlangga
OPEN ACCESS
E-ISSN 2580-8303
P-ISSN 2406-9388

Jurnal Farmasi dan Ilmu Kefarmasian Indonesia Vol. 10 No. 1 April 2023, 11-21
DOI: 10.20473/jfiki.v10i12023.11-21
Available online at https://e-journal.unair.ac.id/JFIKI/

Molecular Docking of Bicycloproline Derivative Synthetic Compounds on Envelope Protein: Anti-SARS-CoV-2 Drug Discovery

Syaiful Prayogi[1,2], Binar Asrining Dhiani[1], and Asmiyenti Djaliasrin Djalil[1]*
[1]Department of Pharmacy, Faculty of Pharmacy, Universitas Muhammadiyah Purwokerto, Purwokerto, Indonesia
[2]Department of Pharmacy, Faculty of Science and Technology, University of Peradaban, Brebes, Indonesia

*Corresponding author: asmiyentidjaliasrindjalil@ump.ac.id

Submitted: 29 August 2022
Accepted: 26 December 2022
Published: 30 April 2023

Abstract

Background*: Although a SARS-CoV-2 vaccine is readily available, new cases of COVID-19 are still occurring. New drug discovery is needed to treat COVID-19. Protein E is one of the potential targets. Two synthetic compounds of bicycloproline derivatives have the potential to be developed.* ***Objective:*** *This study aimed to estimate the interaction of bicycloproline compounds to protein E in-silico.* ***Methods:*** *There were two bicycloproline-derived compounds, MI-09 and MI-30, used in docking. Remdesivir was used as a reference ligand. The crystal structure of the E protein was created using homology modeling, while the test compound was drawn using the Marvin Sketch. MOE 2022.02 and BDS 2021 were used for docking and visualization processes.* ***Results:*** *The pentamer of the SARS-CoV-2 E protein obtained a clash score (1.06); poor rotatomer (0.00%); favored rotamers (98.11%); Ramachandran favored (96.43%); Ramachandran outlier (1.78%); Rama Z-score (-1.08); and mol probity (1.04). Research shows promising inhibition potential of the MI-09 and MI-30. The MI-30 has the best binding energy of -10.3326 kcal/mol.* ***Conclusion:*** *The docking results show that MI-30 has potency as an inhibitor of protein E and can be developed in treating COVID-19. Further research is needed to confirm the result by in vitro and in vivo studies.*

Keywords*: COVID-19, homology modeling, envelope protein, bicycloproline, molecular docking*

How to cite this article:
Prayogi, S., Dhiani, B. A., & Djalil, A. D. (2023). Molecular Docking of Bicycloproline Derivative Synthetic Compounds on Envelope Protein: Anti-SARS-CoV-2 Drug Discovery. *Jurnal Farmasi dan Ilmu Kefarmasian Indonesia*, 10(1), 11-21. http://doi.org/10.20473/jfiki.v10i12023.11-21

INTRODUCTION

Although a vaccine for Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) is already available, new cases of COVID-19 are still occurring due to low immunization coverage and emerging new variants. Therefore, new drugs are needed to treat severe COVID-19. CoV is a sheathed virus consisting of a single-stranded RNA as its genome. The genome size ranges from 26 to 32 kilobases, considered one of the largest among retroviruses (Woo et al., 2010). Pore-forming proteins in the SARS-CoV-2 envelope protein use amphipathic α-helix for pore formation. Pore openings are essential for transporting ions, toxins, and viroporin activity (Khader & Mohideen, 2021). For this reason, knowledge about the SARS CoV-2 protein is essential.

SARS-CoV-2 is a class of betacoronaviruses that have the polyproteins ORF1a and ORF1ab as well as 4 structural proteins: spike glycoprotein (S), membrane (M), nucleocapsid (N), and envelope (E) (Abdelrahman et al., 2020; M et al., 2020; Prajapat et al., 2020). Among these components, protein E is an ideal protein target for molecular docking (Chernyshev, 2020; Das et al., 2021). Research on the drug development of COVID-19, which targeted the SARS-CoV-2 E protein, was still limited. Some research was focused on drug repurposing of existing antiviral drugs. Therefore, drug development initiated with molecular docking targeting protein E of SARS-CoV-2 has significant novelty value.

In previous studies, many researchers have done extensive work uncovering SARS-CoV structural proteins. Structural proteins of SARS-CoV, such as proteins E, S, and M, can be suitable candidates for studying target drug interactions (J. Torres et al., 2006). Comparable to SAR-CoV, the SARS CoV-2 E protein plays a role in the infection process Protein E plays a role in the infection process (Alsaadi & Jones, 2019), virus assembly, virion release, and pathogenesis (Schoeman & Fielding, 2019) and, together with the M protein, plays a role in the spike maturation of SARS-CoV-2 (Boson et al., 2021). Its involvement in various aspects of protein targeting potentially stops the spread of infection while reducing symptoms and managing complications such as *Acute Respiratory Distress Syndrome (ARDS)* in severe SARS-CoV-2 infection (Schoeman & Fielding, 2020). Among the other structural proteins of SARS-CoV-2, the E protein has not received much investigation. We, therefore, have exploited to target the viral *E* protein as a therapeutic intervention against COVID-19. Some drug candidates that inhibit the SARS-CoV-2 E protein have been discovered from the ab initio-designed drugs based on structure characterization (Chernyshev, 2020; Das et al., 2021).

Meanwhile, a research group from China reported the activity of bicycloproline-derived compounds in inhibiting Mpro SARS-CoV-2 with an IC50 value of 7.6-748 nM. A total of 32 compounds (MI-01 to MI-32) were successfully synthesized by modifying the antiviral structure of telaprevir-boceprevir (Qiao et al., 2021). Mpro plays a role in the process of replication and transcription of viruses (Prajapat et al., 2020; Zhang et al., 2020). Protease can divide and produce proteins needed for virus survival (Jo et al., 2019). Two compounds (MI-09 and MI-30) among them have the potential to be developed and explored for possible targets of other proteins, including protein E (Li & Huang, 2021).

The primary purpose of this study was to trace other targets of synthesized bicycloproline derivative compounds (MI-09 and MI-30) against the E protein of SARS-CoV-2. Although the crystal structure of protein E of different organisms is predetermined, structural information is still needed. Therefore, this study is also focused on homological modeling of the SARS-CoV-2 E protein. Molecular docking was carried out between the E protein of SARS-CoV-2 and 2 compounds derived from bicycloproline to study protein-ligand interactions by adopting an in-silico strategy to explain its antiviral properties.

MATERIALS AND METHODS

Hardware applied for calculation, molecular modeling, and docking molecule were a personal computer with a specification of OS Windows 10 Pro-64-bit; Processor: Intel ® Core™ i5-10400 CPU @2.9GHz (12 CPUs); RAM 8192 MB DDR4; Direct 12; 1 TB HDD; 802.11b/g WLAN; Display: NVIDIA GeForce 210 Display Mode: 1366 x 768 (32bit) (60Hz). The program package Molecular Operating Environment MOE 2022.02 was the software applied. Program package MOE dock applied for and draws up parameter docking and simulation of process docking. Program Bio Discovery Visualization 2021 was applied to view the interaction of the ligand with macromolecules.

Protein preparation

Because there is no crystal structure of SARS-CoV-2 E protein eligible for docking, homology modeling was carried out. The monomer and pentamer structure of the SARS-CoV-2 E protein was built using the SWISS-MODEL web server

P-ISSN: 2406-9388
E-ISSN: 2580-8303

(https://swissmodel.expasy.org). *Wuhan-1 isolate* (GenBank ID: QHD43418.1) (National Library of Medicine) was used to predict *the sequence of the E protein* of SARS-CoV-2 (Dey et al., 2020). The E protein structure of the SARS-CoV (PDB ID: 5X29) was used as a template. The model with the best QMEAN was selected for follow-up. GalaxyRefine server was used to fine-tune the model (Heo, Park, and Seok 2013). The selected SARS-CoV-2 E protein monomer was constructed into a pentamer model (Chernyshev, 2020) using the GalaxyHomomer server (https://galaxy.seoklab.org) (Baek et al., 2017). MolProbity (http://molprobity.biochem.duke.edu) was used to validate the SARS-CoV-2 protein E pentamer (Chen et al., 2010). The generated protein structure was energy minimized using MOE with CHARMM27 Forcefield parameters; Gradient 0.1 RMS kcal/mol/A^2; and refinement to RMS Gradient: 0.001 kcal/mol/A to improve the protonated state of amino acid residues and add polar hydrogen using MOE.

Ligand preparation

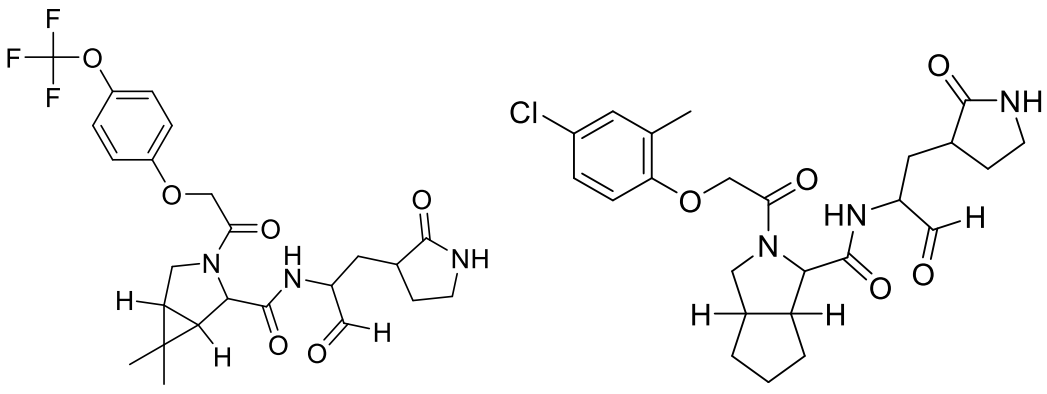
This study used 2 bicycloproline derivative compounds (Figure 1) that actively inhibit SAS-CoV-2 in vitro from previous studies by Qiao and team (Qiao et al., 2021). Remdesivir molecule is used as a ligand for protocol validation. The 2D model of the compound structure was drawn using Marvin-Sketch software and stored in the *.mol extension. The 2D model converted to 3D. Furthermore, geometry optimization was carried out using the Semi-Empirical AM1 method (Asmara and Dwi 2015) via command compute-geometry optimization with parameters convergance limit: 0.01; iteration limit: 32767; algorithm: Polak-Ribiere (conjugate gradient); RMS gradient of: 0.01 kcal/(Å mol) or 32767 maximum cycle. Then the ligands were stored in mol format.

Docking

Molecular docking was performed using MOE. Docking was focused at the docking site obtained using MOE software with the command 'Compute-Site Finder,' selected dummy atoms. The location of the ligand's pocket was focused on the area of amino acids responsible for forming interactions with ligands. The previous studies identified *Asn15, Leu19, Ala22, and Phe26 as the amino acids responsible for forming the interaction with the ligand (Dey et al., 2020; Park et al., 2021).*

Before docking the bicycloproline derivative compounds, the protocol and algorithm were validated by docking the remdesivir to the target protein. Process docking was done to apply two scoring methods: the GBVI/WSA dG scoring function and the london dG scoring function in dock MOE. There were several settings during docking: the placement method was triangle matcher, and the refinement was rigid receptor. The result of the process docking was then kept in mdb format. The *root-mean-square deviation* (RMSD) (<2 Å) suggested that the method could consistently predict the natural conformation of the complex ligand-protein.

Compounds were studied for their interaction with receptors by carrying out molecular docking. The observed parameter was the affinity energy between ligands and receptors which was assessed through the scoring bond energy and compound conformation at the binding site in the form of a mode of interaction between two molecules, such as hydrophobic interaction, electrostatic interaction, and hydrogen bond formed. The molecules' docking results were visualized using Biovia Discovery Studio 2021 (Dassault Systèmes, 2021).



A B

Figure 1. 2D structure of compound. MI-09 (A) and MI-32 (B)

RESULTS AND DISCUSSION

Modeling envelope protein SARS-CoV-2

Homology modelling of SARS CoV-2 E protein was done which showed the sequence identity of 91.38% with the template PDB ID:5X29 (SARS CoV)) (Figure 2). The SARS-CoV-2 E protein was already available in the PDB repository (PDB ID: 7K3G), but the structure was obtained using the solid-state NMR method (Mandala et al., 2020). The validation results show that the E structure has a high clash score and side chain outlier value. The results indicate that the NMR model wass less precise. Therefore, we tried to build a homology model in this study.

The monomer structure of the SARS-CoV-2 E protein with the lowest QMEAN (0.46) and Seq Identity (91.38%) was selected for refining. QMEAN is a combined assessment function that can determine global (i.e., for the entire structure) and local (i.e., per amino acid residue) total quality estimates based on a single model. The QMEAN terms range from 0 to 1, where a value of 1 indicates good agreement. The values were converted to Z-Scores to correlate with what we expect from high-resolution X-ray structures. The identity of seq will affect the proximity of the model to the actual state. The greater the percentage of identity values, the more similar the model will be to the actual state (Komari et al., 2020).

The final model was selected with a clash score (1.0), Rama favored (96.4%), and MolProbity (1.039). A favored value of 96.4% indicates that the protein has a structure with a quality that most amino acids are in the favored region than the outlier (Sharma et al., 2013). MolProbity combines log-weighted clash scores, Ramachandran outlier, and poor side-chain rotamer. The MolProbity value of 1.039 indicates that the model is quite good. If the model structure has a lower MolProbity than the actual crystallography resolution, then the model is said to be better in quality. This score shows one value expected to describe crystallographic resolution (Chen et al., 2010). As the SARS-CoV-2 E protein forms pentamer in physiological conditions (Pervushin et al., 2009), we built the SARS-CoV E protein pentamer structure using the GalaxyHomomer server (Baek et al., 2017; Pervushin et al., 2009). The final validation of the SARS-CoV-2 E protein pentamer obtained a clash score (1.06), poor rotatomer (0.00%), favored rotamers (98.11%), Ramachandran favored (96.43%), Ramachandran outlier (1.78%), rama Z-score (-1.08), and MolProbity (1.04). Outlier residues with a maximum value of 2% can be improved to obtain a better model through energy minimization preparations (Agnihotry et al., 2022). The resulting homology modeling of SARS-CoV-2 E protein is shown in Figure 3. The value of the parameters obtained shows that the modeling proteins can be used for the docking process (Bordoli et al. 2009; Fiser 2010; Komari et al. 2020; Sharma et al. 2013). The SARS-CoV-2 E pentamer model has completed docking preparations (energy minimization) (Ramasami, 2020).

Molecular docking

A research group from China revealed that oral or intraperitoneal treatment with two compounds of 32 new SARS-CoV-2 Mpro inhibitors (MI-09 and MI-30) showed effective antiviral activity in a transgenic mouse model of SARS-CoV-2 infection. Therefore, we were interested to investigate the interaction of these two compounds against the SARS-CoV-2 E protein (Li & Huang, 2021; Qiao et al., 2021).

The MI-09 and MI-30 compounds can bind to the SARS-CoV-2 protein envelope. The binding energy was close to remdesivir (Table 1-2). Remdesivir was the first small-molecule antiviral drug approved by the FDA to treat SARS-CoV-2 infection. The mode of action of remdesivir was to inhibit RdRp (RNA-dependent RNA polymerase) during viral pathogenesis in patients (Gö Tte, 2021). As of today, we could not find any other FDA-approved drug for SARS-CoV-2 SAR with a mechanism of action on the E protein of SARS-CoV-2, remdesivir used as a reference ligand to validate the docking protocol. The interactions of potential drug candidates against SARS-CoV-2 protein E were ranked based on posing and scoring parameters. Binding affinity is used to determine the final rank of the ligand docking pose.

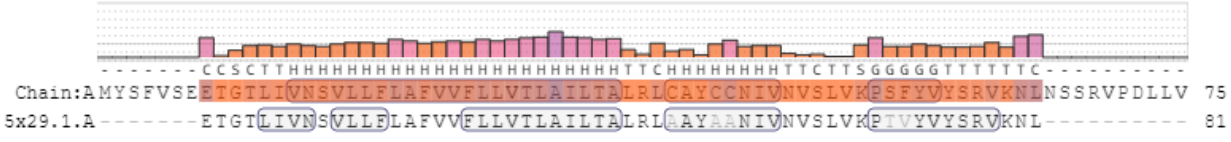


Figure 2. Sequence envelope modeling

P-ISSN: 2406-9388
E-ISSN: 2580-8303

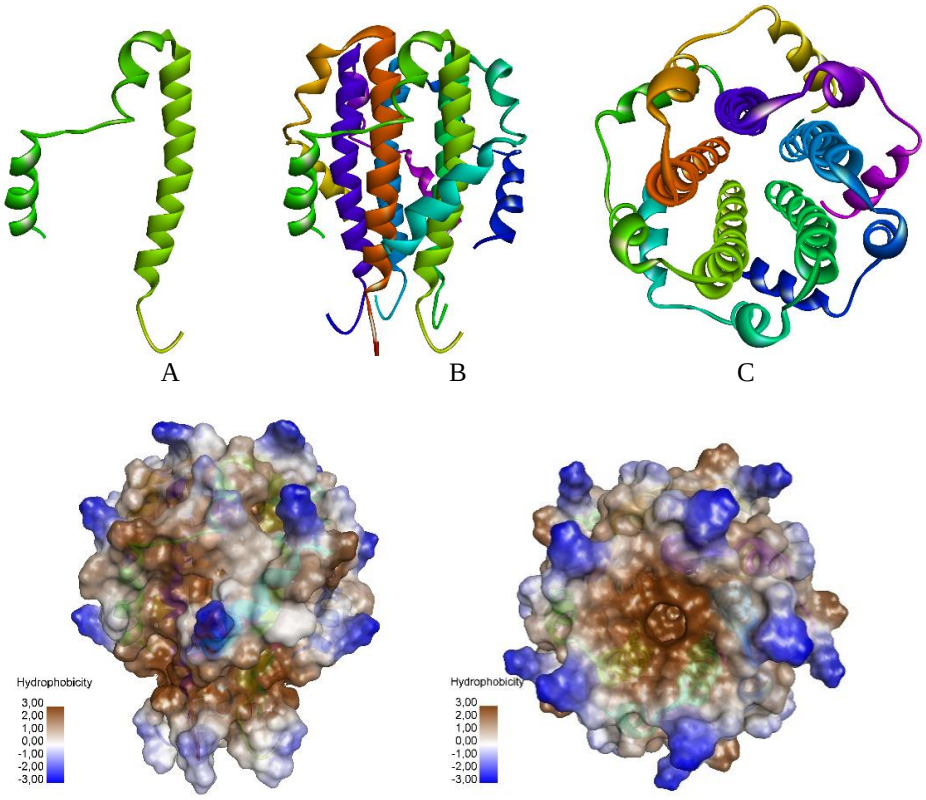


Figure 3. Protein E is the result of modeling. (A) monomer structure of E protein, (B) pentamer structure of E protein, (C) upper-view pentamer structure of E protein, (D) surface pentamer structure of E protein, (E) upper-view surface pentamer structure of E protein

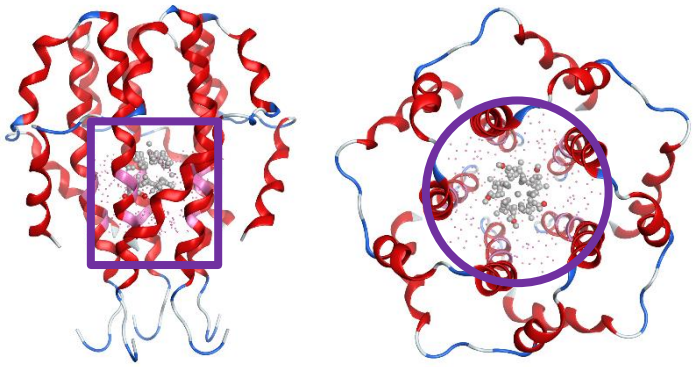


Figure 4. Site docking position. (A) view site docking, (B) upper-view site docking.

Table 1. Reference ligand docking results against SARS-CoV-2 protein envelope

Compound	S (kcal/mol)	RMSD (Å)	Hydrogen Bond	Active Site
Remdesivir	-10.8653	1.6114	Leu A: 19; Leu D: 19; Phe C:23	Hidrophobic: Leu B:19; Leu E:19; Ala E:22; Phe A:23; Phe C:23; Phe A:26; Phe B:26 Electrostatic: Phe C:26

P-ISSN: 2406-9388
E-ISSN: 2580-8303

The docking site was predetermined based on previous publications (Dey et al., 2020; Park et al., 2021). *The study* identified *Asn15, Leu19, Ala22, and Phe26 as the amino acids responsible for forming the interaction with the ligand.* The docking of remdesivir give RMSD value of 1.6114 Å (< 2 Å). The interaction visualization was depicted in Figure 4.

The bicycloproline derivative compounds were docked against the SARS-CoV-2 E protein through the same docking method and conditions. Protein-ligand binding occurs only when the free energy change is negative. The free energy of the bond was proportional to the stability of the protein-ligand interaction. Therefore, protein ligands occur with low binding affinity energies in the system (Afriza, Suriyah, and Ichwan 2018; Du et al. 2016; Sergeev, Dolinska, and Wingfield 2014). The binding affinity energy of the bond indicates the stability of the ligand-protein complex, which is an essential characteristic of drug efficacy (Mohamad Rosdi et al., 2017). The results showed that all compounds could bind to the envelope protein of SARS-CoV-2. The binding energy was close to remdesivir (-10.8653 kcal/mol) (Table1 and Figure 5). MI-30 has the lowest bond energy (-10.3326 kcal/mol) (Table 2 and Figure 6). The result indicates that MI-30 has the most spontaneous tendency to bind to protein E compared to MI-09.

In addition to bond energy, types of molecular interactions such as hydrogen bonds, hydrophobic, and electrostatic interactions, with essential amino acid residues exhibiting docking ligands in conformation into parameters to consider (Hariono et al., 2016). Remdesivir forms 3 hydrogen bonds and 7 hydrophobic bonds SARS-CoV-2 E protein. Both MI-30 and MI-09 form hydrogen bonds, which are formed between H and O atoms. The proton carrier pair (the so-called hydrogen bond donor) in the biological system (protein/receptor) is usually the NH_3 or OH group. The bond reaches excellent strength because the hydrogen atoms of the donor group are bound to highly electronegative atoms, where the electron density of hydrogen atoms shifts to neighboring atoms (Klebe, 2013). The bond between O... H is strong enough (Itoh et al., 2019; N Baker 2006; Panigrahi and Desiraju, 2007). It shows that both compounds have a reasonably good bond strength, with the greatest strength being MI-30, with a more significant number of hydrogen bonds.

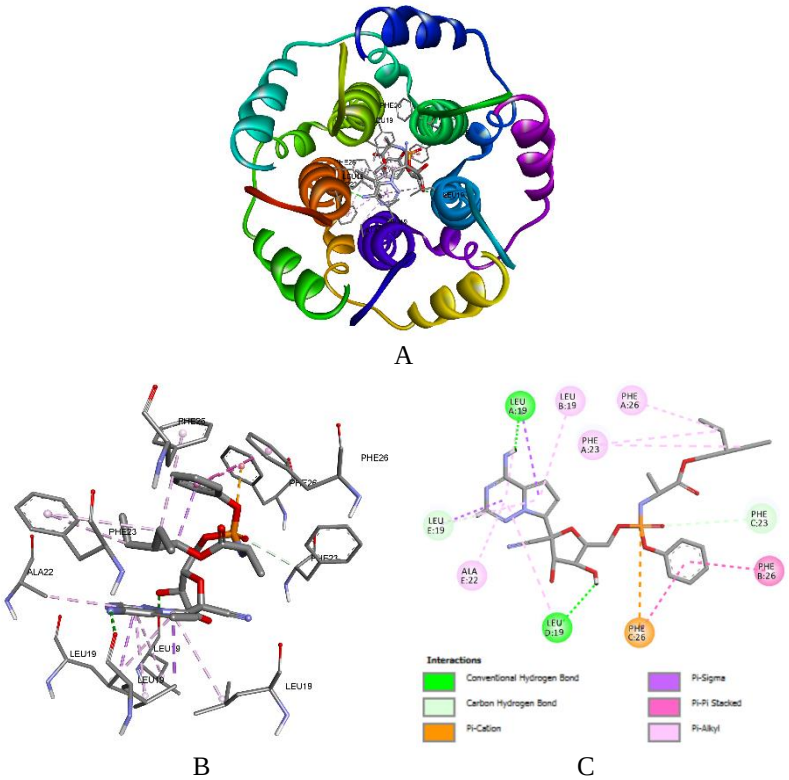


Figure 5. Docking interaction of remdesivir with SARS-CoV-2 E protein. A. Bottom view of ligand-protein, B. 3D display of ligand interaction with amino acid residue, and C. 2D display of ligand interaction with amino acid residue.

P-ISSN: 2406-9388
E-ISSN: 2580-8303

Table 2. Docking parameters of MI-09 and MI-30 compounds against protein E

Compound	S (kcal/mol)	Hydrogen Bond [Distance] {Atom Ligand-Atom Protein}	Hydrophobic Interactions	Electrostatic Interactions
MI-30	-10.3326	Leu A:19 [2.55] {H --- O} Leu A:19 [2.44] {H --- O} Leu E:19 [2.61] {H --- O} Phe A:23 [3,06] {O --- H}	Ala E:22(Alkyl); Ala A:22(Pi-Alkyl); Phe A:23(Pi-Alkyl); Phe E:23(Pi-Alkyl); Phe A:26(Pi-Alkyl); Phe B:23(Pi-Pi T-shaped)	-
MI-09	-10.0403	Leu A:19 [3.02] {O --- H} Leu E:19 [2.57] {H --- O} Phe B:23 [2.39] {O --- H}	Leu D:19(Alkyl; Halogen F, Alkyl); Leu E:19(Alkyl); Ala D:22(Alkyl); Ala E:22(Alkyl); Phe E:26(Pi-Alkyl) Phe D:26(Pi-Alkyl) Phe C:26(Pi-Alkyl, Pi-Alkyl) Phe B:26(Pi-Alkyl) Phe A:26(Pi-Alkyl) Ala C:22(Alkyl) Leu C:19(Alkyl, Pi-Alkyl) Ala B:22(Pi-Alkyl) Leu B:19(Pi-Alkyl)	-

In addition to hydrogen bonds, both MI-30 and MI-09 form hydrophobic bonds. The most hydrophobic bond type is alkyl. Hydrophobic interactions are the main contributors to protein stability compared to hydrogen bonds. Hydrogen bonds also support protein stability, but to a lesser extent than hydrophobic bonds (Pace et al., 2011). Hydrophobic bonding is also vital in combining drug molecules' non-polar regions with biological receptors' non-polar regions. The non-polar regions of drug molecules that are insoluble in water and the surrounding water molecules will combine through hydrogen bonds to form quasi-crystalline structures (icebergs) (Patrick, 2013; Siswandono, 2016). Therefore, hydrophobic bonds are the main determinants of complex equilibrium(Pace et al., 2011). Previous studies suggest that tretinoin compounds form hydrophobic interactions with protein E involving the Leu18 (Chain C, D), Leu21 (Chain C), Ala22 (Chain C), Val25 (Chain C), and Phe26 (Chains A-E), which may be the primary ligand binding site in the SARS-CoV-2 E protein. Blocking of SARS-CoV-2 E ion channels by small molecules can inhibit the activity of viroporin's E activity and consequently eliminates its contribution to viral assembly (Dey et al., 2020). In other previous NMR studies on the SARS-CoV-2 E, protein has shown inhibitor-mediated binding of residue hydrophilic interactions (Glu8, Thr9, Thr11, and Asn15) primarily by Asn15 hexamethylene ameloride (HMA) (Park et al., 2021; Pervushin et al., 2009). The E protein plays a multifunctional role in infection, viral assembly, virion release, and pathogenesis. E protein plays a role in the spike in SARS-CoV-2 maturation (Alsaadi & Jones, 2019; Boson et al., 2021; Schoeman & Fielding, 2020). Being involved in various aspects of the SARS-CoV-2 cycle, targeting this protein can potentially stop the spread of infection, reduce symptoms, and manage complications such as Acute Respiratory Distress Syndrome in SARS-CoV-2 infection (Schoeman & Fielding, 2020).

P-ISSN: 2406-9388
E-ISSN: 2580-8303

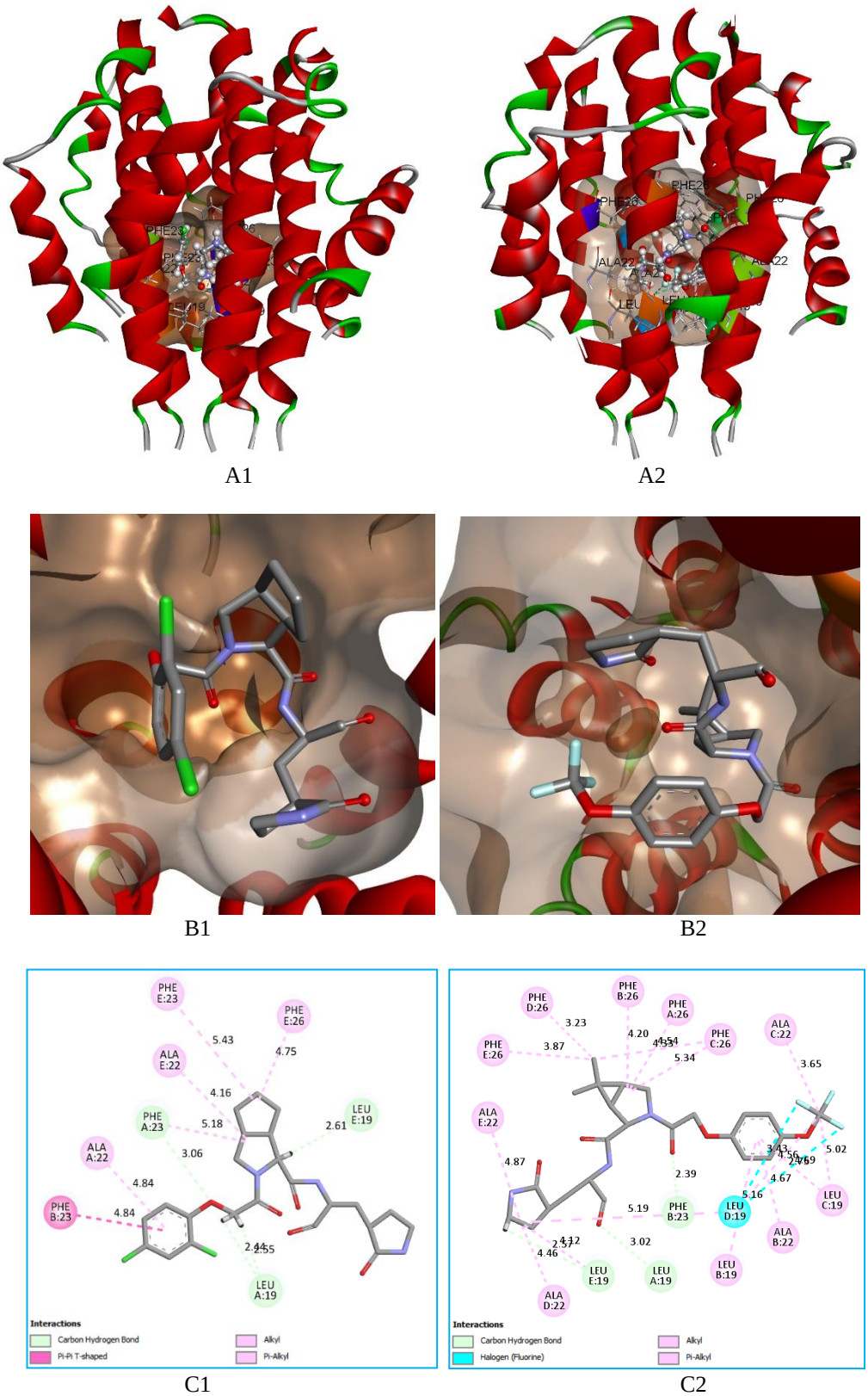


Figure 6. Docking interaction of MI-30 and MI-09 with SARS-CoV-2 E protein. A1-B1. The position of the ligand on the docking site of MI-30, A2-B2. The position of the ligand on the docking site of MI-09, C1. Ligand interaction with amino acid residues of MI-30, C2. Ligand interaction with amino acid residues of MI-09.

P-ISSN: 2406-9388
E-ISSN: 2580-8303

CONCLUSIONS

The bicycloproline compound MI-30 showed the best docking results with the lowest binding energy (-10.3326 kcal/mol) compared to MI-09 (-10.0403 kcal/mol). MI-30 forms a complex with the E protein of SARS-CoV-2 firmly and stably with the most hydrogen and hydrophobic bonds with successive amounts of 4 and 6 bonds. Meanwhile, the protein-ligand interaction of MI-09 with SARS-CoV-2 E protein formed 3 hydrogen bonds and 13 hydrophobic interactions. Overall, this discovery provides new knowledge about the possible mechanisms of inhibition of two bicycloproline-derived compounds to block the E protein cycle of SARS-CoV-2. Nonetheless, modifications of the compound, in-vitro as well as in-vivo investigations are needed to confirm this discovery.

ACKNOWLEDGMENT

This study was supported by The Ministry of Education, Culture, Research and Technology (MoECRT) of the Republic of Indonesia through the Master Thesis Research Grant 2022 (Award No 158/E5/PG.02.00.PT/2022).

REFERENCES

Abdelrahman, Z., Li, M., & Wang, X. (2020). Comparative Review of SARS-CoV-2, SARS-CoV, MERS-CoV, and Influenza A Respiratory Viruses. *Frontiers in Immunology*, *11*(September). https://doi.org/10.3389/fimmu.2020.552909

Afriza, D., Suriyah, W. H., & Ichwan, S. J. A. (2018). In silico analysis of molecular interactions between the anti-apoptotic protein survivin and dentatin, nordentatin, and quercetin. *Journal of Physics: Conference Series*, *1073*(3). https://doi.org/10.1088/1742-6596/1073/3/032001

Agnihotry, S., Pathak, R. K., Singh, D. B., Tiwari, A., & Hussain, I. (2022). Protein structure prediction. *Bioinformatics*, 177–188. https://doi.org/10.1016/B978-0-323-89775-4.00023-7

Alsaadi, E. A. J., & Jones, I. M. (2019). Membrane binding proteins of coronaviruses. *Future Virology*, *14*(4), 275. https://doi.org/10.2217/FVL-2018-0144

Asmara, A. P., & Dwi. (2015). Analisis Hubungan Kuantitatif Struktur Dan Aktivitas Senyawa Turunan Triazolopiperazin Amida Menggunakan Metode Semiempirik AM1. *Elkawnie*, *1*(2), 125–138.

Baek, M., Park, T., Heo, L., Park, C., & Seok, C. (2017). GalaxyHomomer: a web server for protein homo-oligomer structure prediction from a monomer sequence or structure. *Nucleic Acids Research*, *45*(W1), W320–W324. https://doi.org/10.1093/NAR/GKX246

Bordoli, L., Kiefer, F., Arnold, K., Benkert, P., Battey, J., & Schwede, T. (2009). Protein structure homology modeling using SWISS-MODEL workspace. *Nature Protocols*, *4*(1), 1–13. https://doi.org/10.1038/NPROT.2008.197

Boson, B., Legros, V., Zhou, B., Siret, E., Mathieu, C., Cosset, F.-L., Lavillette, D., & Denolly, S. (2021). The SARS-CoV-2 envelope and membrane proteins modulate maturation and retention of the spike protein, allowing assembly of virus-like particles. *The Journal of Biological Chemistry*, *296*, 100111. https://doi.org/10.1074/JBC.RA120.016175

Chen, V. B., Arendall, W. B., Headd, J. J., Keedy, D. A., Immormino, R. M., Kapral, G. J., Murray, L. W., Richardson, J. S., & Richardson, D. C. (2010). MolProbity: all-atom structure validation for macromolecular crystallography. *Acta Crystallographica Section D: Biological Crystallography*, *66*(Pt 1), 12. https://doi.org/10.1107/S0907444909042073

Chernyshev, A. (2020). Pharmaceutical targeting the envelope protein of SARS-CoV-2: the screening for inhibitors in approved drugs. *ChemRxiv*, 1–13. https://chemrxiv.org

Das, G., Das, T., Chowdhury, N., Chatterjee, D., Bagchi, A., & Ghosh, Z. (2021). Repurposed drugs and nutraceuticals targeting envelope protein: A possible therapeutic strategy against COVID-19. *Genomics*, *113*(1), 1129–1140. https://doi.org/10.1016/J.YGENO.2020.11.009

Dassault Systèmes. (2021). *BIOVIA DISCOVERY STUDIO VISUALIZAION* (No. 2021). Dassault Systèmes. https://3ds.com/products-services/biovia/products

Dey, D., Borkotoky, S., & Banerjee, M. (2020). In silico identification of Tretinoin as a SARS-CoV-2 envelope (E) protein ion channel inhibitor. *Computers in Biology and Medicine*, *127*, 104063. https://doi.org/10.1016/J.COMPBIOMED.2020.104063

Du, X., Li, Y., Xia, Y. L., Ai, S. M., Liang, J., Sang, P., Ji, X. L., & Liu, S. Q. (2016). Insights into Protein–Ligand Interactions: Mechanisms, Models, and Methods. *International Journal of Molecular Sciences*, *17*(2). https://doi.org/10.3390/IJMS17020144

National Library of Medicine. (n.d.). *Envelope Protein [Severe acute respiratory syndrome coronavirus 2] - Protein - NCBI*. Retrieved April 1, 2022, from https://www.ncbi.nlm.nih.gov/protein/QHD43418.1?report=fasta

Fiser, A. (2010). Template-Based Protein Structure Modeling. *Methods in Molecular Biology (Clifton, N.J.)*, *673*, 73. https://doi.org/10.1007/978-1-60761-842-3_6

Gö Tte, M. (2021). *Remdesivir for the treatment of Covid-19: the value of biochemical studies*. https://doi.org/10.1016/j.coviro.2021.04.014

Hariono, M., Abdullah, N., Damodaran, K. v., Kamarulzaman, E. E., Mohamed, N., Hassan, S.

S., Shamsuddin, S., & Wahab, H. A. (2016). Potential New H1N1 Neuraminidase Inhibitors from Ferulic Acid and Vanillin: Molecular Modelling, Synthesis and in Vitro Assay. *Scientific Reports 2016 6:1*, *6*(1), 1–10. https://doi.org/10.1038/srep38692

Heo, L., Park, H., & Seok, C. (2013). GalaxyRefine: protein structure refinement driven by side-chain repacking. *Nucleic Acids Research*, *41*(W1), W384–W388. https://doi.org/10.1093/NAR/GKT458

Homology Modeling. (n.d.). Retrieved August 3, 2022, from https://www.umass.edu/molvis/workshop/homolmod.htm#readymade

Itoh, Y., Nakashima, Y., Tsukamoto, S., Kurohara, T., Suzuki, M., Sakae, Y., Oda, M., Okamoto, Y., & Suzuki, T. (2019). N+-C-H···O Hydrogen bonds in protein-ligand complexes. *Scientific Reports*, *9*(1). https://doi.org/10.1038/S41598-018-36987-9

Jo, S., Kim, S., Shin, D. H., & Kim, M.-S. (2019). Inhibition of SARS-CoV 3CL protease by flavonoids. *Https://Doi.Org/10.1080/14756366.2019.1690480*, *35*(1), 145–151. https://doi.org/10.1080/14756366.2019.1690480

Khader, A., & Mohideen, S. (2021). *Molecular Docking Analysis of Phytochemical Thymoquinone as a Therapeutic Agent on SARS-Cov-2 Envelope Protein Molecular Docking Analysis of Phytochemical Thymoquinone as a Therapeutic Agent on SARS-Cov-2 Envelope Protein View project Molecular Docking An*. *11*(1), 8389–8401. https://doi.org/10.33263/BRIAC111.83898401

Klebe, G. (2013). Protein–Ligand Interactions as the Basis for Drug Action. *Drug Design*, 61–88. https://doi.org/10.1007/978-3-642-17907-5_4

Komari, N., Hadi, S., & Suhartono, E. (2020). Pemodelan Protein dengan Homology Modeling menggunakan SWISS-MODEL. *Jurnal Jejaring Matematika Dan Sains*, *2*(2), 65–70. https://doi.org/10.36873/JJMS.2020.V2.I2.408

Li, L., & Huang, S. (2021). Newly synthesized Mpro inhibitors as potential oral anti-SARS-CoV-2 agents. *Signal Transduction and Targeted Therapy*, *6*(1), 3–4. https://doi.org/10.1038/s41392-021-00560-0

M, T. U. Q., SM, A., MA, A., & LL, C. (2020). Structural basis of SARS-CoV-2 3CL pro and anti-COVID-19 drug discovery from medicinal plants. *Journal of Pharmaceutical Analysis*, *10*(4), 313–319. https://doi.org/10.1016/J.JPHA.2020.03.009

Mohamad Rosdi, M. N., Mohd Arif, S., Abu Bakar, M. H., Razali, S. A., Mohamed Zulkifli, R., & Ya'akob, H. (2017). Molecular docking studies of bioactive compounds from Annona muricata Linn as potential inhibitors for Bcl-2, Bcl-w and Mcl-1 antiapoptotic proteins. *Apoptosis 2017 23:1*, *23*(1), 27–40. https://doi.org/10.1007/S10495-017-1434-7

N Baker, B. E. (2006). *22.2. Hydrogen bonding in biological macromolecules*.

Pace, C. N., Fu, H., Fryar, K. L., Landua, J., Trevino, S. R., Shirley, B. A., Hendricks, M. M. N., Iimura, S., Gajiwala, K., Scholtz, J. M., & Grimsley, G. R. (2011). Contribution of Hydrophobic Interactions to Protein Stability. *Journal of Molecular Biology*, *408*(3), 514. https://doi.org/10.1016/J.JMB.2011.02.053

Panigrahi, S. K., & Desiraju, G. R. (2007). Strong and weak hydrogen bonds in the protein–ligand interface. *Proteins: Structure, Function, and Bioinformatics*, *67*(1), 128–141. https://doi.org/10.1002/PROT.21253

Park, S. H., Siddiqi, H., Castro, D. v., de Angelis, A. A., Oom, A. L., Stoneham, C. A., Lewinski, M. K., Clark, A. E., Croker, B. A., Carlin, A. F., Guatelli, J., & Opella, S. J. (2021). Interactions of SARS-CoV-2 envelope protein with amilorides correlate with antiviral activity. *PLOS Pathogens*, *17*(5), e1009519. https://doi.org/10.1371/JOURNAL.PPAT.1009519

Patrick, G. L. (2013). An Introduction to Medicinal Chemistry Fifth Edition. In *Oxford University Press* (5th Ed). OXFORD. https://doi.org/10.1016/0307-4412(76)90096-0

Pervushin, K., Tan, E., Parthasarathy, K., Lin, X., & Jiang, F. L. (2009). Structure and Inhibition of the SARS Coronavirus Envelope Protein Ion Channel. *PLoS Pathog*, *5*(7), 1000511. https://doi.org/10.1371/journal.ppat.1000511

Prajapat, M., Sarma, P., Shekhar, N., Avti, P., Sinha, S., Kaur, H., Kumar, S., Bhattacharyya, A., Kumar, H., Bansal, S., & Medhi, B. (2020). Drug targets for corona virus: A systematic review. *Indian Journal of Pharmacology*, *52*(1), 56. https://doi.org/10.4103/IJP.IJP_115_20

Qiao, J., Li, Y.-S., Zeng, R., Liu, F.-L., Luo, R.-H., Huang, C., Wang, Y.-F., Zhang, J., Quan, B., Shen, C., Mao, X., Liu, X., Sun, W., Yang, W., Ni, X., Wang, K., Xu, L., Duan, Z.-L., Zou, Q.-C., … Yang, S. (2021). SARS-CoV-2 Mpro inhibitors with antiviral activity in a transgenic mouse model. *Science*, *371*(6536), 1374–1378. https://doi.org/10.1126/SCIENCE.ABF1611

Ramasami, P. (2020). *Computational Chemistry Methods: Applications* (P. Ramasami, Ed.). De Gruyter. https://doi.org/https://doi.org/10.1515/9783110631623

Schoeman, D., & Fielding, B. C. (2019). Coronavirus envelope protein: current knowledge. *Virology Journal 2019 16:1*, *16*(1), 1–22. https://doi.org/10.1186/S12985-019-1182-0

Schoeman, D., & Fielding, B. C. (2020). Is There a Link Between the Pathogenic Human Coronavirus Envelope Protein and Immunopathology? A Review of the Literature. *Frontiers in Microbiology*, *0*, 2086. https://doi.org/10.3389/FMICB.2020.02086

Sergeev, Y. v., Dolinska, M. B., & Wingfield, P. T. (2014). The thermodynamic analysis of weak protein interactions using sedimentation equilibrium. *Current Protocols in Protein Science / Editorial Board, John E. Coligan ... [et Al.]*, *77*, 20.13.1. https://doi.org/10.1002/0471140864.PS2013S77

Sharma, S., Sarkar, S., Paul, S. S., Roy, S., & Chattopadhyay, K. (2013). A small molecule chemical chaperone optimizes its unfolded state contraction and denaturant like properties. *Scientific Reports*, *3*. https://doi.org/10.1038/SREP03525

Siswandono. (2016). *Medicinal Chemistry* (Siswandono, Ed.; 2nd ed.). Airlangga University Press.

Torres, J., Parthasarathy, K., Lin, X., Saravanan, R., Kukol, A., & Ding, X. L. (2006). Model of a Putative Pore: The Pentameric α-Helical Bundle of SARS Coronavirus E Protein in Lipid Bilayers. *Biophysical Journal*, *91*(3), 938. https://doi.org/10.1529/BIOPHYSJ.105.080119

Woo, P. C. Y., Huang, Y., Lau, S. K. P., & Yuen, K. Y. (2010). Coronavirus Genomics and Bioinformatics Analysis. *Viruses*, *2*(8), 1804. https://doi.org/10.3390/V2081803

Zhang, L., Lin, D., Sun, X., Rox, K., & Hilgenfeld, R. (2020). X-ray Structure of Main Protease of the Novel Coronavirus SARS-CoV-2 Enables Design of α-Ketoamide Inhibitors. *BioRxiv*, 2020.02.17.952879. https://doi.org/10.1101/2020.02.17.952879

P-ISSN: 2406-9388
E-ISSN: 2580-8303