DOI : https://doi.org/10.31351/vol29iss2pp231-238

Docking Study of Naringin Binding with COVID-19 Main Protease Enzyme

Narmin H. Amin Hussen*,1

*Department of Pharmaceutical Chemistry, College of Pharmacy, University of Sulaimani, Iraq

Abstract

Recently the pandemic coronavirus disease 2019(COVID-19) has spread quickly all over the world caused by SAR-CoV2. In the present study, it has been used molecular docking to the binding affinity between COVID-19 main protease enzyme and flavonoids with evaluations based on docking scores calculated by AutoDock Vina. Results showed that naringin interacted with COVID-19 main protease, and it has the highest binding affinity than other flavonoids include quercetin, hesperetin, and naringenin. An important finding in this study is that naringin with poly hydroxyl groups can serve as an inhibitor of COVID-19 main protease bind to the pocket of the protein. It is shown that residues His163, Glu166, Asn142, His41and Gln189 participate in the hydrogen bonding interactions, the same as happened with decahydroisoquinoline as a novel structure as a protease inhibitor for SARS 3CL.On the other hand, some of the known protease inhibitors and anti-influenza drugs docked with COVID-19main protease, it has a low binding affinity than naringin.

Keywords: COVID-19 main protease, Flavonoids, Naringin, Molecular docking, Protease inhibitor.

Introduction

The novel coronavirus disease (COVID-19) was first identified in Wuhan, China, in December 2019 caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV2) which can be transmitted effectively between human to human and animal to human through droplets or direct contact, causing fever, cough, shortness of breath, pneumonia and kidney failure [1,2]. Recently, it has been reported that the number of infected human outside of China are suddenly increased, as of May 28, 2020, there have more than 5,700,000 cases, 357,533 deaths and 2,500,000 recovered in 216 countries and territories, most of the cases, and deaths have occurred in United States of America, Brazil, Russia, Spain, United Kingdom, Italy and France[3].

SARS-CoV2 is a Beta coronavirus, which is an enveloped, positive-sense, single-stranded RNA virus in the family of *Coronaviridae*. In general, coronaviruses (CoVs) are a large group of viruses that can be divided into four genera, including alpha, beta, delta, and gamma. Alpha- and Beta coronaviruses mainly infect bats, but they also infect other species like humans, camels, and rabbits [4-6].COVID-19 is closely related to two high pathogenic responsible for Severe Acute Respiratory Syndrome (SARS-CoV) in 2002 and Middle East Respiratory Syndrome (MERS-CoV) in 2012[7-9].

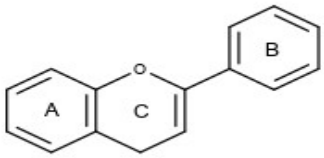
Drug development against coronavirus includes inhibition of viral replication through acting on its critical enzymes [10]. CoVs encode proteases such as papain-like protease (PLpro) and main protease (Mpro), which are involved in the proteolytic processing of the polyproteins into individual non-structural proteins (nsps) to control viral gene expression and replication [11,12]. The crystallized form of COVID-19 main protease (Mpro) was demonstrated by a Chinese researcher Liu et al[13], that it is a potential drug target protein for the inhibition of SARS-CoV-2 replication. The Mpro is a key protein required for the proteolytic maturation of the virus [14]. Thus, targeting Mpro has the potential to provide effective treatment against SARS-CoV-2 by inhibition of the viral polypeptide cleavage [15]. Further, studies have found that SARS-COV-2 requires angiotensin-converting enzyme 2 (ACE2) and Transmembrane Serine Protease 2 (TMPRSS2), to enter lung cells, the same cellular entry receptor as SARS-CoV to infect humans [16-120].

As of now, few antiviral strategies are being used to treat patients, lack of specific antiviral drugs or vaccines against SARS-CoV-2 is further aggravating the situation [21]. Thus, there is an urgent need to identify and develop effective antivirals against SARS-CoV-2 to fight this deadly virus. In this study, it has been used flavonoids with SARS-CoV-2 main protease against COVID-19 [22-24].

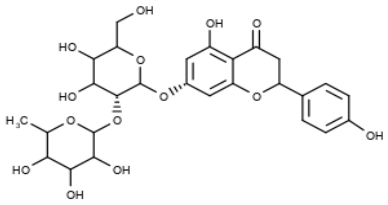
[1]Corresponding author E-mail**:** narmin.hussen@univsul.edu.iq
Received: 25/3 /2020
Accepted: 6/ 8/2020

The flavonoids, a large group of naturally occurring low molecular weight compounds widely distributed in the plant kingdom; particularly, they belong to a class of plant secondary metabolites having a polyphenolic structure, widely found in fruits, vegetables, and certain beverages. These compounds share a common structural core with two benzene rings (A and B) joined by a third heterocyclic ring (C) (Figure 1) [25-27]. Studies have suggested that flavonoids exhibit biological activities, including anti-allergenic, antiviral, anti-inflammatory, and vasodilating actions [28,29]. Therefore, the inhibition of proteases was proposed as a new function mechanism for flavonoids by several independent laboratories. Flavonoids such as naringin, quercetin, hesperetin and naringenin possess a variable degree of antiviral activity (Figure 1). [30,31].

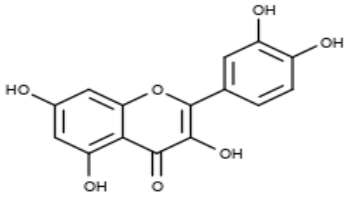
In this study, we performed molecular docking to understand the interaction between 9 flavonoids and 14 FDA approved antiviral drugs such as lopinavir, indinavir, ribavirin, ritonavir, favipiravir, and remdesivir (Figure1) with COVID-19 main protease were performed to identify these drugs inhibiting COVID-19 main protease enzyme.



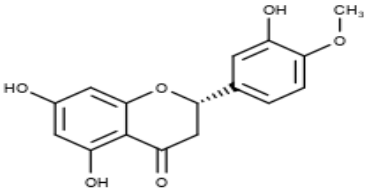
Basic structure of flavonoids (I)



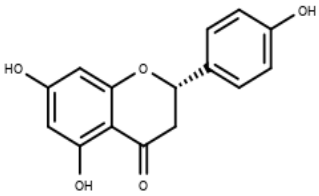
Naringin (II) .



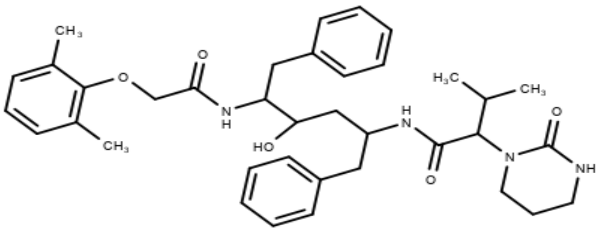
Quercetin (III)



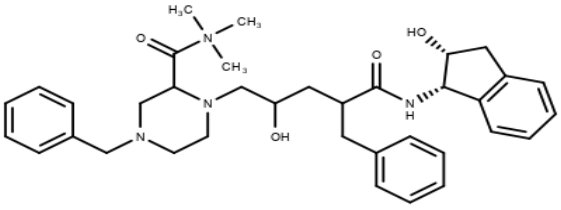
Hesperetin (IV)



Naringenin(V)



Lopinavir (VI)



Indinavir (VII)

Figure 1. (I) Basic structure of flavonoids, (II) structure of naringin, (III) structure of quercetin, (IV) structure of hesperetin, (V) structure of naringenin, (VI) structure of lopinavir and (VII) structure of indinavir.

Methods

Ligand preparation

The two and three-dimensional models of the drug was obtained from the Pub Chem data base (https://pubchem.ncbi.nlm.nih.gov/) in the structure-data file (SDF). Then Open Babel was used to converting SDF to pdb format (https :// sourceforge .net/projects/openbabel/). Ligands used in this docking study are 9 flavonoids and 14 FDA approved antiviral drugs. Among these drugs, naringin is most promising, since it demonstrates the highest docking score to the COVID-19 protein (Table 1).

Protein preparation

Protein Data Bank (PDB) is a structural repository for biological macromolecules such as proteins and their complexes (www.rcsb.org/pdb)[32]**.** The crystal structure of COVID-19 main protease with N3 as inhibitors(6LU7.pdb)(http://www.rcsb.org/structure /6LU7)[15](Figure2),available in Protein Data Bank was used as a receptor. The three-dimensional structure of the target protein was retrieved from PDB by giving the PDB ID in the database. Protein Data Bank (PDB) files may have a variety of problems that need to be corrected before they can be used for docking. Before docking, the entire N3as inhibitors were removed from the protein molecule.

Lipinski's rule of five.

The rule of five is beneficial to assess in vivo absorption abilities of the designed compounds. A ligand has a molar mass less than 500, hydrogen bond donors (-OH, NH) less than five, hydrogen bond acceptors (N, O) less than ten and calculated CLogP is less than five satisfy the rule of five. ClogP, the number of hydrogen donors, and number of hydrogen acceptors of the drugs were obtained from the PubChem database (https: // pubchem .ncbi.nlm.nih.gov/).

All of the flavonoids in the present study satisfy the rule of five except naringin, quercetin, laurifolin and elatin.

Molecular dockin

Docking between the protein and ligand was performed using AutoDock 4.2.6 (http://vina. Scripps.edu.). AutoDock Tools were used for preparing the input files and analyzing the result. A program for molecular docking and virtual screening is Auto Dock Vina [33]. The virtual screening program has been used is AutoDockVina, implemented in an application called PyRx 0.8 (https://pyrx .source forge.io/), which is open-source software to perform virtual screening. To determine the scoring function in this method, specification of search space inside the coordination system of the protein is necessary, in which different positions of the ligand should be examined. The magnitude of the search space was determined with the grid center of X: -25, Y: -52, Z: -4.4, and the number of points in each magnitude was X:45, Y:45, Z:45 in angstrom. Each output file has several models ranked in the ascending order in terms of binding energy. The predicted binding energy of the ligand with the target protein is represented in kcal/mole. In each case, only the best mode is usually selected and used for subsequent analysis.

Visualization

In order to sketch, visualize, and analyze ligand molecules, a suite of applications called Marvin has been used. All the Marvin tools were accessible from the Marvin Sketch 19.9 application (https: // chemaxon .com / products / mar-vin). H-bonds interactions between ligands and amino acids of targeted protein were visualized on UCSF Chimera [34].

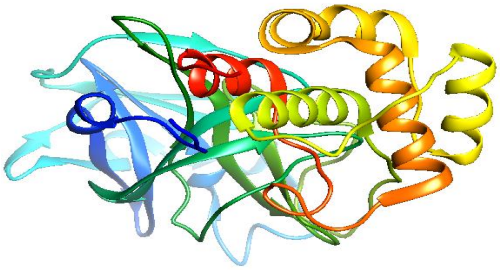


Figure 2. The 3D structure of COVID- 19 main protease

Results and Discussion

Table 1 shows the binding energy of several flavonoids with COVID-19 main protease sorted according to the docking scores (binding affinities) calculated from the Autodock Vina. Naringin showed lower binding energy, and higher binding affinity than the other docked flavonoids. Hydrogen-bonding contributes most to the binding affinities of all flavonoids with the receptor protein. Flavonoids through hydrogen-bonding interact with COVID-19 main protease, with binding energies between -10.2.to-6.0kcal/mol

Table 1. The docking score (kcal/mol), MW, CLogP, No. of H bond donor, No. of H bond acceptor, and Lipinski's rule of five for flavonoids.

Flavonoids	Docking score (kcal/mol)	MW (g/mol) <500	ClogP <5	No. of H bond donor <5	No. of H bond acceptor ≤ 10	Lipinski's rule of five
Naringin	-10.2	580.5	-0.5	8	14	NO
Quercetin	-8.0	302.23	1.5	5	7	NO
Hesperetin	-7.9	302.28	2.4	3	6	Yes
Naringenin	-7.7	272.25	2.4	3	5	Yes
Ternatin	-7.0	374.3	3.1	2	8	Yes
Hydroxyflavone	-6.1	238.2	3.4	1	3	Yes
Alvocidib	-6.1	401.8	3.3	3	6	Yes
Laurifolin	-6.0	356.4	11.8	2	5	NO
Elatin	-6.0	594.5	-2.1	11	15	NO

Upon study, Naringin has a high binding affinity with binding energy (-10.2 kcal/mol); however, it could not pass the rule of five criteria due to its molecular mass greater than 500 g/mol and the number of hydrogen bond acceptor exceed the allowed range. As shown in Figure 3, naringin could fit well to the binding pocket of COVID-19 protease through five hydrogen-bonding interactions. Naringin with poly hydroxyl groups may serve as inhibitors of COVID-19 protease, it is shown that residues His163, Glu166, Asn 142, His41, and Gln 189 participate in the hydrogen bonding interaction, the same as happened with decahydroisoquinoline as a novel structure protease inhibitor [35]. The one hydrogen bond is occurred between one N-H group of His163 of COVID-19 interact with one hydroxyl group of naringin, distance is 1.97 Å (Figure 3). Next hydrogen bonds between carboxyl oxygen of Glu166 and Asn142, chains of COVID-19 protein, and the hydroxyl group of ligands, with bond length 2.43 and 3.09Å, respectively (Figure 3). More, one hydrogen bonds between carboxyl oxygen of Gln189 of backbone and hydroxyl group of the ligand has occurred with bond length 2.63Å (Figure 3). Finally, other hydrogen bond can be seen between N-H His41 side chain and the hydroxyl group of phenol in the ligand with bond length of 3.61 Å (Figure 3). An important finding in this work is that the poly hydroxyl group of naringin can function as a protease inhibitor bind to the COVID-19 main protease.

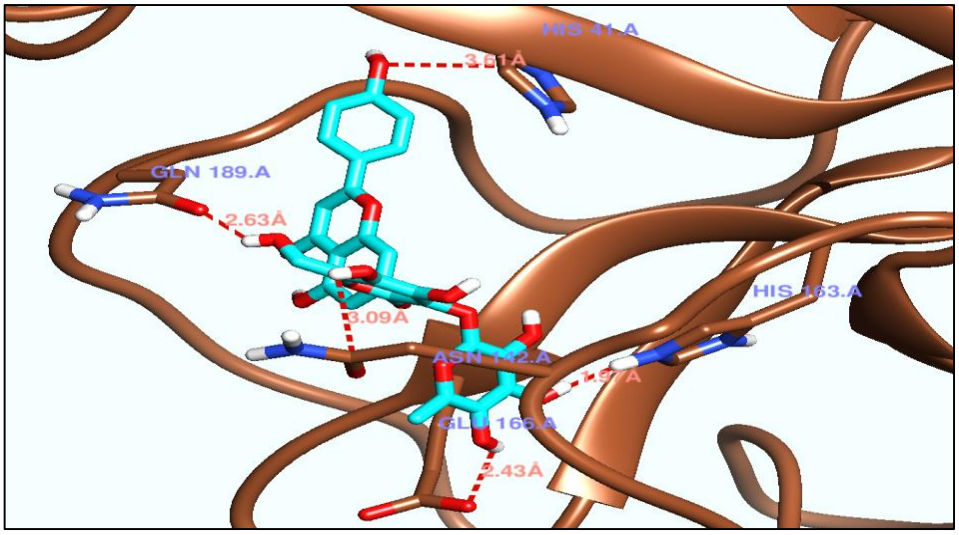


Figure 3. Molecular docking of naringin has interacted with COVID-19 main protease enzyme.

Therefore, the binding energies calculated for other flavonoids lower than naringin including quercetin, hesperetin, naringenin, ternatin, 3-hydroxy flavone, and elatin are -8.0, -7.9, -7.8,-7.0 -6.1 ,and -6.0 kcal/mol respectively. They bind with the same COVID-19 main protease pocket, but it could not fit well to binding pocket (Figure 4) because it has low poly hydroxyl group for hydrogen bonding interaction (Figure 1) and also the number of hydrogen bonding interaction with the amino acid of COVID-19 main protease lower than naringin.

It was found the Figure 4A, quercetin formed one hydrogen bonding between hydroxyl group of ligand with amino acids His 163, distance is 2.26Å .Next hydrogen bond between the hydroxyl group of quercetin with side-chain amino acids Gln 189, His41and Glu 166 with bond length,2.02 Å,3.06 Å and 3.76 Å respectively (Figure 4 A). The hydroxyl group of hesperetin formed two hydrogen bonding between the N-H group of His164 and carboxyl oxygen of Gln 189 chains of COVID-19 protein with bond length 4.48 Å and 4.67Å respectively.

As shown in Figure 4 C, naringenin could bind with the COVID-19 main protease pocket through hydrogen bonding with the amino acid Glu 166 and Asn 142, distances are 4.52 Å and 4.79 Å respectively.Ternatin with the hydroxyl group can be formed hydrogen bonding with amino acids His 163 and Gln 189, bond length 4.13 Å and 5.43 Å respectively (Figure 4 D).

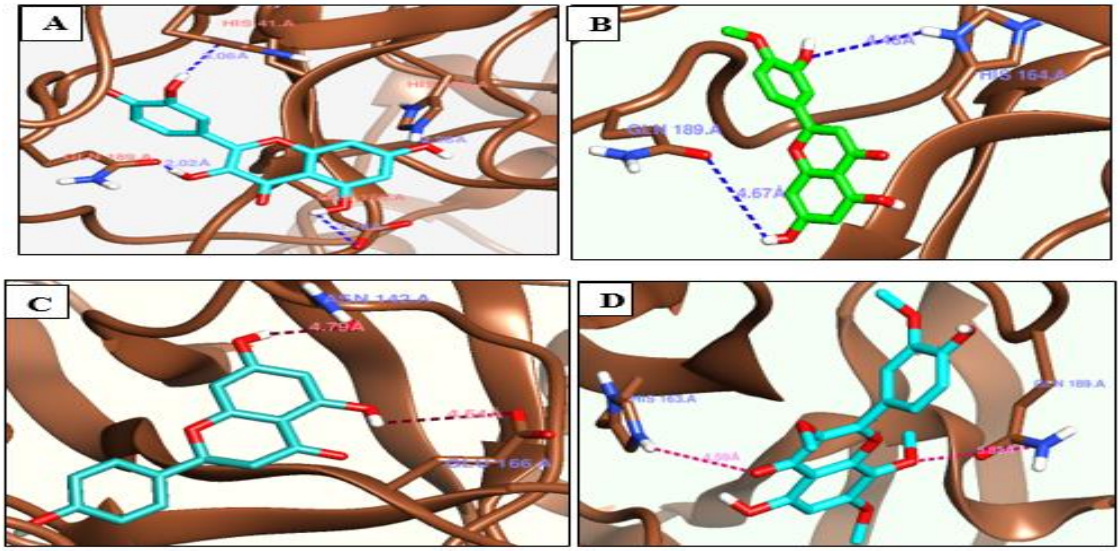


Figure 4. (A) Molecular docking of quercetin interacted with COVID-19 main protease enzyme. **(B)** Molecular docking of hesperetin. **(C)** Molecular docking of naringenin. **(D)** Molecular docking of ternatin.

On the other hand, numerous recent studies have been suggested some of the drugs against COVID-19 disease especially protease inhibitors drugs [36,37]. To understand and compare that naringin is a reasonably better binding affinity with COVID-19 main protease enzyme than other drugs; also it has been used molecular docking to the binding affinity between COVID-19 main protease enzyme and 14 FDA approved drugs. Results showed that the low binding affinities calculated for ligands such as lopinavir, indinavir, ritonavir, and ribavirin than naringin, and they do not interact effectively with the COVID-19 main protease. As shown in Figure 5, all of the protease inhibitors having a different binding pockets with flavonoids.

For comparison, the docking energy between the COVID-19 main protease and lopinavir calculated and the score was−7.9 Kcal/mol. As shown in Figure 6, lopinavir could bind with the COVID-19 main protease pocket through hydrogen bonding with the amino acid Gln 110 and Asn 151, distances are 2.77 Å and 3.34 Å respectively. The docking energy between the COVID-19 main protease and indinavir, ritonavir, and ribavirin were calculated to be −7.7, -7.5, -7.0 kcal/mol respectively. All of these scores appear lower than naringin (-10.2 kcal/mol). Table 2 shows the binding energy of several ligands with COVID-19 main protease sorted according to the docking scores (binding energies) calculated from the Autodock Vina.

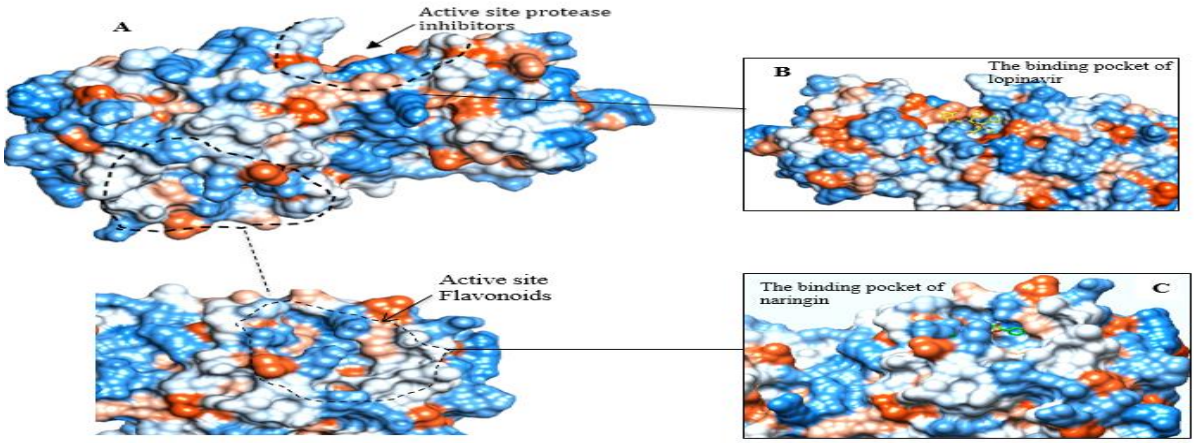


Figure 5. (A) The structure of COVID-19 main protease enzyme generated by using USCF Chimera with two active sites for flavonoids and protease inhibitors. (B) Ligand-protein interaction for lopinavir. (C) Ligand-protein interaction for naringin.

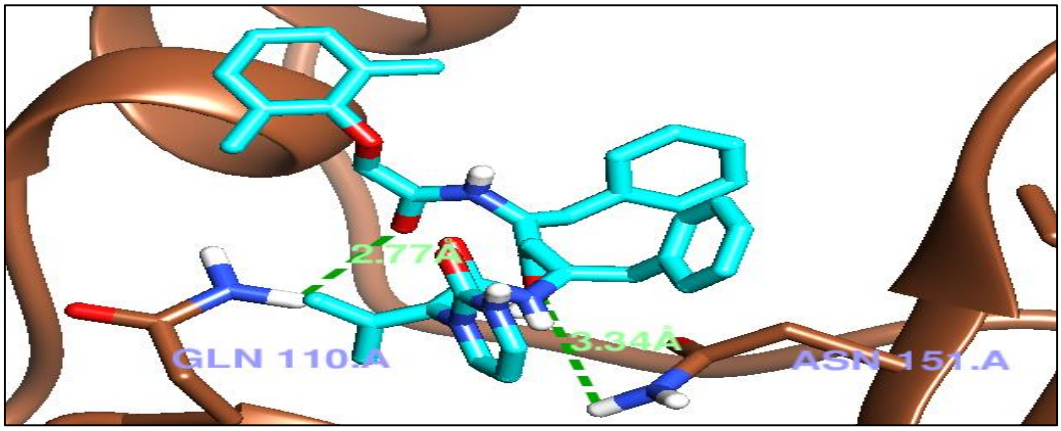


Figure 6. Molecular docking of lopinavir has interacted with COVID-19 main protease enzyme

Table 2. The docking score (kcal/mol), MW, CLogP, No. of H bond donor, No. of H bond acceptor, and Lipinski's rule of five for FDA approved drugs.

Drug name	Docking score (kcal/mol)	MW (g/mol) <500	ClogP <5	No. of H bond donor <5	No. of H bond acceptor ≤10	Lipinski's rule of five
Lopinavir	-7.9	628.8	5.9	4	5	NO
Indinavir	-7.7	613.8	2.8	4	7	NO
Ritonavir	-7.5	720.9	6	4	9	NO
Ribavirin	-7.0	212.2	-1.8	4	7	NO
Camostat mesilate	-6.4	494.5	3	9	9	Yes
Zanamivir	-6.3	332.3	-3.2	7	8	NO
Favipiravir	-5.8	157.1	-0.6	2	4	NO
Rimantadine	-5.7	179.3	2.6	1	1	Yes
Oseltamivir	-5.5	312.4	1.1	2	5	Yes
Simeprevir	-5.3	749.9	4.8	2	10	NO
Remdesivir	-5.3	602.6	1.9	4	13	NO
Baloxavir marboxil	-5.2	571.6	3.8	0	12	NO
Amantadine	-5.2	151.2	2.4	1	1	Yes
Boceprevir	-4.7	519.7	3.1	4	5	Yes

Conclusion

As noted before, COVID-19 has become a global concern, due to widespread outbreaks and lack of treatment. Therefore, to contribute to this fight against COVID-19, molecular docking was performed to identify novel compounds having the potential to bind main protease of COVID-19. Relying on this topic and repurposing concept, a procedure employing docking of flavonoids, protease inhibitors, and anti-influenza drugs was used to identify new potential molecule to bind the main protease of COVID-19 and the result indicates that naringin has a high binding affinity with low energy -10.2 kcal/mol to the main protease of COVID-19 than other natural molecules. However, further studies should be conducted for the validation of these compounds using in *vitro* and in *vivo* models to pave a way for these compounds in drug discovery.

References

1. Guangdi Li and Erik De Clercq. Therapeutic options for the 2019 novel coronavirus (2019-nCoV). Nature reviews | Drug Discovery. 2020.19: 149.
2. Yushun Wan 1, Jian Shang 1, Rachel Graham, Ralph S. Baric 2, Fang Li. Receptor recognition by novel coronavirus from Wuhan: An analysis based on decade-long structural studies of SARS. Journal of Virology. 2020.DOI:10.1128/JVI.00127-20.
3. Chan JF, Kok KH, Zhu Z, et al. Genomic characterization of the 2019 novel human-pathogenic coronavirus isolated from a patient with atypical neumonia after visiting Wuhan. Emerg Microbes Infect. 2020;9 (1):221–236.
4. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A novel coronavirus from patients with pneumonia in China, 2019. The New England Journal of Medicine. 2020; 382(8): 727–33.

5. Zhou P, Yang XL, Wang XG, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. Nature. 2020. doi:10.1038/s41586-0202012-7.

6. Daniel Wrapp, Nianshuang Wang, Kizzmekia S. Corbett, Jory A. Goldsmith, Ching-Lin Hsieh, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science. Marh 13, 2020; 367(6483):1260-1263. DOI: 10.1126/science. abb2507.

7. Han-Zhong Zhang, Hong Zangwillian Kemnitzer, Ben Tseng, Jindrich Cinatl, Jr., Martin Michaelis, Hans Wilhelm Doerr, and Sui Xiong Cai. Design and Synthesis of Dipeptidyl Glutaminyl Fluoromethyl Ketones as Potent Severe AcuteRespiratory Syndrome Coronavirus (SARS-CoV) Inhibitors.J. Med. Chem. 2006.49: 1198-1201

8. CatharineI.Paules,HilaryD.Marston,AnthonyS. Fauci.Coronavirus Infections More Than Just the Common Cold. JAMA. February 25,2020;8(323). DOI:10.1001/jama.2020.

9. Ghosh, Arun K., Jun Takayama, Yoann Aubin, Kiira Ratia, Rima Chaudhuri, Yahira Baez, Katrina Sleeman et al. Structure-Based Design, Synthesis, and Biological Evaluation of a Series of Novel and Reversible Inhibitors for the Severe Acute Respiratory Syndrome−Coronavirus Papain-Like Protease. J Med Chem. 2009; 52 :5228-5240.

10. Ratish Chandra Mishra, Rosy Kumari, Shivani Yadav, Jaya Parkash Yadav. Antiviral potential of phytoligands against chymotrypsin-like protease of COVID-19 virus using molecular docking studies: An optimistic. In Review/ Nature Research. 2020.DOI : 10.21203/rs.3.rs-23956/v1.

11. Sarma,et al.Therapeutic options for the treatment of 2019 -novel coronavirus: an evidence- based approach. Indian J. Pharmacol. 2020;52, 1-5.

12. Ghosh, Arun K., Jun Takayama, Kalapala Venkateswara Rao, Kiira Ratia, Rima Chaudhuri, Debbie C. Mulhearn, Hyun Lee et al. Severe acute respiratory syndrome coronavirus papain-like novel protease inhibitors: design, synthesis, protein-ligandd X-ray structure and biological evaluation. J Med Chem. 2010;53: 4968-4979.

13. Liu, X., Zhang, B., Jin, Z., Yang, H., Rao, Z. Structure of Mprofrom SARS-CoV-2 and discovery of its inhibitors. Nature. 2020;582: 289-293. DOI: 10.1038/s41586-020-2223-y.

14. Sevki Adem, Volkan Eyupoglu ,Iqra Sarfraz, Azhar Rasul, Muhammad Ali. Identification of potent COVID-19 main protease (Mpro) inhibitors from natural polyphenols:An insilico strategy unveils a hope against CORONA . Preprints. 2020. DOI : 10 .20944 /preprints 202003.0333.v

15. Zhenming Jin, Xiaoyu Du, Haitao Yang, et al. Structure of M^{pro} from SARS-CoV-2 and discovery of its inhibitors. Nature. April 09 2020.https://doi.org/10.1038/s41586-020-2223-y.

16. Markus Hoffmann, Hannah Kleine-Weber, Simon Schroeder, Marcel A. Müller, Christian Drosten,Stefan Pöhlmann. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. ACellPress journa1OpenAccess. (2020.02). DOI:https://doi.org/10.1016/j.cell.

17. Carolin Tarnow,a Géraldine Engels,b Annika Arendt,a Folker Schwalm,a Hanna Sediri,a Annette Preuss,b Peter S. Nelson,c Wolfgang Garten,a Hans-Dieter Klenk,a Gülsah Gabriel,b Eva Böttcher-Friebertshäusera. TMPRSS2 Is a Host Factor That Is Essential for Pneumotropism and Pathogenicity of H7N9 Influenza A Virus in Mice. Journal of Virology. 2014;.9:88.

18. Yan-Rong Guo, Qing-Dong Cao, Zhong-Si Hong, Yuan-Yang Tan, Shou-Deng Chen, Hong-Jun Jin, et al. The origin, transmission and clinical therapies on coronavirus disease 2019 (COVID-19) outbreak an update on the status. Guo et al. Military Medical Research. 2020; 7:11.

19. Michael Letko, Andrea Marzi and Vincent Munster. Functional assessment of cell entry and receptor usage for SARS-CoV-2 and other lineage B betacoronaviruses. Nature Microbiology. 2020; 5:562–569.

20. Yushun Wan, Jian Shang, Rachel Graham, Ralph S. Baric, Fang Li. Receptor Recognition by the Novel Coronavirus from Wuhan: An Analysis Based on Decade-Long Structural Studies of SARS Coronavirus. Journal of Virology. 2020; 94(7). https://doi .org/10.1128/JVI.00127-20.

21. Liying Dong, Shasha Hu, Jianjun Gao. Discovering drugs to treat coronavirus disease2019(COVID19).DrugDiscoveris & Therapeutics. 2020; 14(1); 58-60.

22. Guangpu Xue, Lihu Gong, Cai Yuan, Mingming Xu, Xu Wang, Longguang Jiang, Mingdong Huang1. Structural mechanism of flavonoids in inhibiting serineproteases .FoodFunct. 2017;.DOI :10.1039/C6FO01825

23. Kumar S & Pandey AK. Chemistry and biological activities of flavonoids: an overview .Scientific WorldJournal .2013.DOI : https: //doi.org/10.1155/2013/162750.

24. N. C. Cook and S. Samman, Flavonoids Chemistry, metabolism, cardioprotective effects, and dietary sources. The Journal of Nutritional Biochemistry.(1996).7 :66-76.
25. A. N. Panche, A.D. Diwan and S. R. Chandra.Flavonoids:an overview. Journal of nutritional science. (2016).41 DOI:10.1017/jns.
26. Tej N. Kaul, Elliott Middleton, Jr., and Pearay L. Ogra. Antiviral Effect of Flavonoids on Human Viruses. Journal of Medical Virology. (1985).l5:71-79
27. Wenjiao Wu, Richan Li, Xianglian Li, Jian He 1, Shibo Jiang, Shuwen Liu, and Jie Yang. Quercetin as an Antiviral Agent Inhibits InfluenzaA Virus (IAV) Entry. Viruses (2016).8:6. DOI:10.3390/v801
28. Ye H, Xu HD, Yu CG, Dai YJ, Liu GY, Xu WP and Yuan S: Hydroxylation of naringin by Trichoderma harzianum to dramatically improve its antioxidative activity. Enzyme Microb Tech.(2009). 45: 282-287.
29. Camila a. Camargo1, Maria cristina c. Gomes-marcondes2, Nathalie c. Wutzki1 andhiroshi aoyama. Naringin inhibits tumor growth and reduces interleukin-6 and Tumor Necrosis Factor α Levels in Rats with Walker 256 Carcinosarcoma. Anticancer research. (2012) .32 : 129-134.
30. Hui-Kang Wang, Yi Xia, Zheng-Yu Yang, Susan L. Morris Natschke, and Kuo-Hsiung Leet. Recent advances in the discovery and development of flavonoids and their analogues as antitumor and anti-hiv agents J. Med. Chern.(1997). 40: 3049-3056.
31. Ribeiro IA and Ribeiro MHL: Naringin and naringenin determination and control in grapefruit juice by a validated HPLC method. Food Control. (2008).19: 432-438.
32. Helen M. Berman, John Westbrook, Zukang Feng, Gary Gilliland, T. N. Bhat, Helge Weissig, Ilya N. Shindyalov, Philip E. Bourne. The Protein Data Bank. Nucleic Acids Research.January,2000,28(1);Pages235242. DOI : https://doi.org/10.1093/nar/28.1
33. Oleg Trott, Arthur J. Olson. AutoDock Vina: Improving the speed and accuracy of docking with a new scoring function, efficien to ptimization , and multi-threading .Journal of computational chemistry. June 2009.DOI :https://doi.org/10.1002/jcc.21334
34. Eric F. Pettersen Thomas D. Goddard Conrad C. Huang Gregory S. Couch Daniel M. Greenblatt Elaine C. Meng Thomas E. Ferrin. UCSF Chimera—A visualization system for exploratory research and analysis. Journal of computational chemistry. 01 July 2004 https://doi.org/10.1002/jcc.20084
35. Shimamoto Y, Hattori Y, Kobayashi K, Teruya K, Sanjoh, Nakagawa A, Yamashita E, Akaji K.Fused-ring structure of decahydroisoquinolin as a novel scaffold for SARS 3CL protease inhibitors.BioorgMedChem.(2015).15:23(4).
36. James M. Sanders, Marguerite L. Monogue,Tomasz Z. Jodlowski, et al. Pharmacologic Treatments for Coronavirus Disease 2019(COVID-19). JAMA. 2020 ;323 (18):1824-1836. DOI : 10 .1001 / jama. 2020 .6019.
37. Sheahan TP, Sims AC, Leist SR, et al. Comparative therapeutic efficacy of remdesivir and combination lopinavir, ritonavir, and interferon beta against MERS CoV. NatCommun .2020 ;11(1):222. doi:10.1038/s41467-019-13940-6

Baghdad Iraqi Journal Pharmaceutical Sciences by bijps is licensed under a Creative Commons Attribution 4.0 International License. Copyrights© 2015 College of Pharmacy - University of Baghdad.