Original article

# MOLECULAR DOCKING ANALYSIS OF SOME PHYTOCHEMICALS ON TWO SARS-COV-2 TARGETS

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Received: date; Accepted: date; Published: date

**Abstract:**

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) (previously called 2019 novel coronavirus (2019-nCoV) is the causative agent of coronavirus disease 2019 (COVID-19), a disease recently declared a global public health emergency by the World Health Organization. At the moment there is no available drug(s) and vaccine(s) for the treatment or prevention of COVID-19. SARS-CoV-2 spike envelope glycoprotein (S) and main protease ($M^{pro}$) are crucial determinants in the virus infectious process and have been recognized as key targets for therapeutics designs. In the present *in silico* study, a library of 22 phytochemicals with antiviral activity obtained from PubChem Database was screened for activity against 6lu7 and 6vsb with the PyRX software. Six lead compounds with binding energies within the range of -9 to -9.6 Kcal/mol were selected for molecular docking analyses against 6lu7. SwissADMET and Molinspiration Cheminformatics for CLogP (mean range of 0.77-8.72) of the lead compounds showed no correlation observed between lipophilicity and interaction with receptors and all the compounds except for baicalin exhibited drug-like properties based on Lipinski and Veber filter. The ADMET profile showed that lead compounds lack hepatotoxicity and mutagenicity effects while

they show variable immunotoxicity, carcinogenicity and cytotoxicity. The compounds Scopodulic acid and Dammarenolic acid showed the best-fit value of activity against SARS-CoV-2 spike glycoprotein 6vsb and main protease $M^{pro}$ 6lu7 targets, respectively. Our data suggest silibinin a repurposing candidate drug may have multitarget activity against SARS-CoV-2. So further in vitro and in vivo evaluations are recommended.

**Keywords**: *Covid-19, s-glycorotein, $M^{pro}$, Virtual screening*

## 1. Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), family *Coronaviridae*, genus *Betacoronavirus*, is spreading widely in China, causing coronavirus disease 2019 (COVID-19)(1). Since 2003, three Coronaviruses have been associated with pneumonia, the first was severe acute respiratory syndrome coronavirus (SARS-CoV)(2) which affected 8,098 people causing 774 deaths between 2002 and 2003(3), the second was Middle-East respiratory syndrome coronavirus (MERS-CoV)(4) which affected 27 countries and infecting a total of 2,494 individuals and claiming 858 lives(4). SARS-CoV-2 is a human pathogen which has been declared a global pandemic by the World Health Organisation (5). SARS-CoV and SARS-CoV-2 are closely related and originated in bats, who most likely serve as a reservoir host for these two viruses (4). To date, no therapeutics or vaccines are approved against any human-infecting coronaviruses(4).

The entry into the host cell by the Coronaviruses is usually mediated by spike (S) glycoprotein (4). This glycoprotein interacts with the angiotensin-converting enzyme 2 (ACE2) enabling the virus penetration into the host. The main protease ($M^{pro}$ also known as $3CL^{pro}$) is one of the best-characterized drug targets among coronaviruses (6). The protease enzyme is essential for processing the polyproteins that are translated from the viral RNA(7). For this study, these two drug targets were selected for SARS-CoV-2 using plant-based compounds screened against them.

Therefore, potent inhibitors of these two targets will be able to interfere with the SARS COV-2 replication process and thus serves as potential drugs for the management of the COVID-19. Hence, this work is aimed at identifying other potential lead compounds of plant origin that can serve as candidates for testing against the SARS COV2 virus.

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## 2. Results

A library of 22 compounds of plant origin known to have antiviral activity was obtained from Pubchem database. Though the compounds are chemically diverse they consist of largely flavonoids and terpenes. Some compounds from the citrus family made were found among the library and demonstrated some good binding affinities.

Most of the compounds have shown similar binding affinities to the selected protein targets (6lu7 and 6vsb) compared to the training sets of known ligands to the selected targets. (See table 1)

Table 1. Comparison of Binding affinities to some known ligands and the co-crystalised ligand

| S/N | Group | PubChem ID | PyRx Binding Affinity (Kcal/mol) on 6lu7 |
|---|---|---|---|
| **1.** | Co-crystalised Ligand | **7885280** | **4.9** |
| **2.** | **Known Ligands** | **11313622** | **-7.5** |
| | | **121304016** | **-5.6** |
| | | **235905** | **-6.6** |
| | | **5284592** | **-6.2** |
| | | **5475158** | **-5.1** |
| **3** | **Phytochemicals (Query Set)** | **52803443** | **-7.1** |
| | | **57347487** | **-7.2** |
| | | **65727** | **-7.0** |
| | | **1548994** | **-6.9** |
| | | **11729855** | **-6.7** |
| | | **479503** | **-6.6** |
| | | **72303** | **-5.6** |
| | | **68077** | **-5.5** |
| | | **72344** | **-5.1** |

However, the top six compounds with most favourable binding affinity were selected for each of the targets.

The outcome of the binding affinities of the selected compounds on the 6lu7 and 6vbs targets are presented in **Table 2 and Table 3** respectively.

**TABLE 2** Binding affinities of the compounds on the 6vsb and their Interaction with the binding site

| S/N | Ligands | Binding Affinity (Kcal/Mol) | Hydrogen Bond Interaction with Residues | Hydrophobic Bond Interaction with Residues |
|---|---|---|---|---|
| **1.** | Scopadulcic Acid | -9.6 | GLN B: 913 | TYR C:904, GLY B:1093, VAL B:911, ARG B: 1107, ASN B:907, THR B:912, ASN B:1119, GLN B:1113, GLY B: 910, GLN B:1106, GLU B:1092, PHE A:1121, ARG A:1091 |
| **2.** | Baicalin | -9.4 | ARG B:1039, ARG A:1039, ARG C:1039, ALA B:1020, ASN C:1023 | ALA C:1026, LEU B:1024, GLN C:784, SER C:1030, ASP B:1041, LEU C:1024, THR C:1027, PHE B:1042, PHE C:1042, PHE A:1042, THR B:1027, SER B:1021, GLU C:780. |
| **3.** | Sylibinin | -9.2 | GLU A:954, ARG B:765 | GLU A:1017, ARG A:1014, ALA B:766, LYS B:776, LYS A:947, LEU A:948, PRO A:728, VAL A:951, ILE A:1018, ALA B:766, GLN A:957, GLN B:762, GLN A:1010, ILE A:1013, LEU B:1012, ARG B:1019, GLU B:773. |
| **4.** | Solanidine | -9.1 | | TYR A:369, TYR C:489, ARG C:454, PRO C:491, TYR C:421, ASN C:460, LEU C:461 |
| **5.** | Naringenin | -9.0 | GLU C:1092, ARG C:1107, ASN C:1108, GLY C:910, ILE C:909 | ASN C:907, THR C:912, GLN C:1113, ARG B:1091, GLU B:1092, GLY C:1093, GLN C:1106, TYR A:904 |
| **6.** | Oleanane | -9 | | LEU A:1141, ASP C:1118, LEU C:1141, PRO A:1140, ASP A:1118, THR A:1117, THR B:1116, ASP B:1118, PRO B:1140, GLU B:1144, ASP A:1139, GLU A:1144 |

**TABLE 3** Binding affinities of the compounds on the 6lu7

| S/N | Ligands | Binding Affinity (Kcal/Mol) |
|---|---|---|
| **1.** | Dammarenolic acid | -7.2 |
| **2.** | Quercetin | -7.1 |

| | | |
|---|---|---|
| **3.** | Solanidine | -7.0 |
| **4.** | Silybinin | -6.8 |
| **5.** | Loliolide | -6.7 |
| **6.** | Shikonin | -6.6 |

The binding affinities of the top six compounds on the 6vsb target are comparable to each other that is they all lie within a close range of 9 to 9.6 kcal/mol indicating that they might likely have equal or comparable potential as lead compounds for the 6vsb spike glycoprotein.

**Table 4.** Comparison of the calculated cLog P values for the selected compounds

| S/N | Compound | SwisADME cLog P | Molinspiration cLog p | Mean calculated cLog P |
|---|---|---|---|---|

| | | | | |
|---|---|---|---|---|
| 1. | Scopodulcic acid | 4.57 | 5.01 | 4.79 |
| 2. | Baicalin | 0.22 | 0.55 | 0.77 |
| 3. | Sylibinin | 1.59 | 1.47 | 3.06 |
| 4. | Solanidine | 5.01 | 5.93 | 5.47 |
| 5. | Naringenin | 1.84 | 2.12 | 1.98 |
| 6. | Oleanane | 8.57 | 8.86 | 8.72 |
| 7. | Dammarenolic acid | 6.74 | 8.08 | 7.41 |
| 8. | Quercetin | 1.23 | 1.68 | 1.46 |
| 9. | Loliolide | 1.53 | 1.84 | 1.69 |
| 10. | Shikonin | 2.08 | 2.02 | 2.05 |

clog P = octanol/water coffecient

One of the compounds sylibinin (8)is an FDA approved drug, which showed up as active on both $M^{Pro}$ and spike glycoprotein will make a good candidate of repurposing. Finding Quercetin as a potential inhibitor of the $Mp^{ro}$ Protein (6flu7) of the SARS-COV-2 corresponds with an earlier report(9)

Looking at the cLog P of the compounds, there was no correlation observed between the lipophilicity and the interaction with the receptors. However, for the compounds acting on 6lu7 (S/N 3,4,7,8,9 and 10 in Table 4), interaction with the receptor is correlated with low lipophilicity except for solanidine and Dammarenolic acid that have high cLogP values. Though both compounds also use their polar functional groups in interacting with the receptor. Bacailin and Naringenin showed good hydrogen bond interaction with the 6vsb receptor due to their polarity.

Filtering the compounds for drug-likeness based on Linpinski's and/or Veber's rule showed that all the compounds have a drug like properties except baicalin which

failed the two filtering scales applied (Table 5). This implies that baicalin is not worth considering further without any structural modification.

**Table 5. Drug likeness**

| S/N | Compound | Mol. Wt[a](g/mol) | TPSA[b] | HBA[c] | HBD[d] | RB[e] | cLogP[f] | Lipinski filter | Veber filter |
|---|---|---|---|---|---|---|---|---|---|
| 1. | Scopodulcic acid | 438.56 | 80.67 | 5 | 1 | 4 | 4.79 | + | + |
| 2. | Baicalin | 446.36 | 187.12 | 11 | 6 | 4 | 0.77 | - | - |
| 3. | Sylibinin | 482.44 | 155.14 | 10 | 5 | 4 | 3.06 | + | - |
| 4. | Solanidine | 397.64 | 23.47 | 2 | 1 | 0 | 5.47 | + | - |
| 5. | Naringenin | 272.25 | 86.99 | 5 | 3 | 1 | 1.98 | + | + |
| 6. | Oleanane | 412.75 | 0.00 | 0 | 0 | 0 | 8.72 | + | + |
| 7. | Dammarenolic acid | 458.72 | 57.53 | 3 | 2 | 1 | 7.41 | + | + |
| 8. | Quercetin | 302.24 | 131.36 | 7 | 5 | 1 | 1.46 | + | + |
| 9. | Loliolide | 196.24 | 46.53 | 3 | 1 | 0 | 1.69 | + | + |
| 10. | Shikonin | 288.3 | 94.83 | 5 | 3 | 3 | 2.05 | + | + |

[a]Mol. Wt.: Molecular weight
[b]TPSA: Total Polar Surface Area
[c]No. HBA: Number of hydrogen bond acceptors
[d]No. HBD: Number of hydrogen bond donors
[e]No. RB: Number of rotatable bonds
[f]Mean clog P: Mean of calculated log P values

Figure 1. (a) Dammarenolic acid in the binding pocket of 6lu7 (b) Binding interactions between dammarenolic acid and the 6lu7 protein

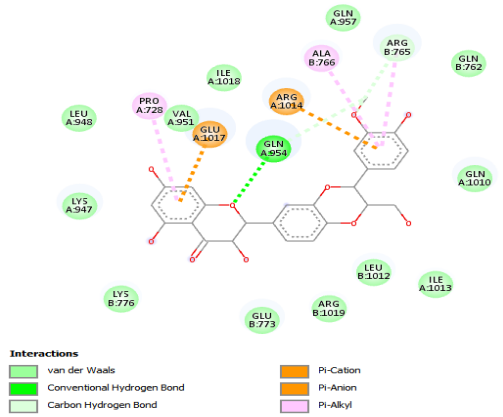


Figure 2: 2D interaction of 6VSB with Silibinin

Table 6. Predicted Toxicity Profile of the compounds using PROTOX II

| S/N | Compound | Hepatotoxicity | ImmunoToxicity | Carcinogenicity | Mutagenicity | Cytoxicity | Possible Toxicity Targets |
|---|---|---|---|---|---|---|---|
| 1. | Scopodulcic acid | - | ++ | - | - | - | AR, AO, PGS |
| 2. | Baicalin | -- | -- | + | - | -- | AO, PGS |
| 3. | Sylibinin | -- | ++ | - | -- | -- | PGS |
| 4. | Solanidine | -- | ++ | - | -- | + | AR, PGS |
| 5. | Naringenin | - | -- | - | -- | + | AR, PGS |
| 6. | Oleanane | -- | -- | ++ | -- | -- | |
| 7. | Dammarenolic acid | - | - | -- | -- | -- | AR, AO PGS |
| 8. | Quercetin | - | + | -- | - | -- | AO, AR, PGS |
| 9. | Loliolide | - | + | -- | - | -- | AO, PGS |
| 10. | Shikonin | - | - | ++ | - | + | PG |

Key: -- inactive, - less inactive, + Active and ++ More active

AR = Androgen Receptor, AO = Amine Oxidase A & PGS=Prostaglandin G/H Synthase 1

The predicted toxicity profile of the selected compounds shows that all the compounds are likely to be relatively safe. Which makes them good potential candidates for anti-infectives because the chances of achieving selective toxicity are high. Baicalin is most likely the safest.

## Discussion

Two compounds among the top six selected for each target, solanidine and sylibinin were observed to have a good binding affinity on both the 6vsb and the 6flu7. This makes them potential multitarget acting inhibitors on the SARS-COV2. Solanidine is a Steroidal glycoalkaloids found in potatoes(10), although toxic to humans and animals, Solanidine has been reported to be effective against herpes viruses (HSV), herpes genitalis and herpes zoster(11) Its activity against HSV is attributed to the presence of a sugar moiety(12). In silico method of drug screening using PROTOX II, showed that Solanidine is cytotoxic and immunotoxic. Prototox II is a cost and time conservative approach of testing and determining the toxicity of a compound to be considered a drug of choice(13). It incorporates molecular similarity, pharmacophores, fragment propensities and machine-learning models for the prediction of various toxicity endpoints; such as acute toxicity, hepatotoxicity, cytotoxicity, carcinogenicity, mutagenicity, immunotoxicity, adverse outcomes pathways (Tox21) and toxicity targets(13)

A safe drug must not be toxic to its host target. Based on the Protox II evaluation of Toxicity, Dammarenolic acid emerges as the compound of choice with the least toxicity. Dammarenolic acid has been reported as effective antiviral agents Dammarenolic acid potently inhibited the in vitro replication of other retroviruses, including simian immunodeficiency virus and Murine leukaemic virus in vector-based antiviral screening studies and has been proposed as a potential lead compound in the development of antiretrovirals. (14) The compound is cytotoxic

and demonstrates potential against the respiratory syncytial virus(15). We therefore propose that the evaluation of Dammarenolic acid will hold the key to COVID19 drug considering its drugability and low toxicity.

This study proposes a potential re-purposing of silybinin for the management of COVID19 diseases. Silybinin(Silymarin) possesses potent antiviral activities against numerous viruses, particularly hepatitis C virus (HCV)(16, 17) It has been reported to have activities against a wide range of viral groups including flaviviruses (hepatitis C virus and dengue virus), togaviruses (Chikungunya virus and Mayaro virus), influenza virus, human immunodeficiency virus, and hepatitis B virus(16). Silymarin inhibits HCV in both *in vitro* and *in vivo* by inhibiting HCV entry, RNA synthesis, viral protein expression and infectious virus production; in addition, it also acts by blocking off the virus cell-to-cell spread(18). As an FDA approved drug for the management of Hepatitis disease. In silico analysis of this drugs in this study has shown that it has activity against SAR COV 2 S-glycoprotein and proteas($M^{pro}$) targets making it a drug to be considered with multi-target ability in the management of this disease.

## 4. Materials and Methods

Plant Compounds with antiviral activities were mined from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/ ). Two proteins including the main protease (6lu7) and the crystal structure of COVID-19 main protease(19) in complex with an inhibitor N3 and the Spike glycoprotein, N-ACETYL-D-GLUCOSAMINE (6vsb10.1126/science.abb2507) were downloaded from the protein database (PDB). The proteins were prepared using Discovery studio (version)(20) and a rigid docking scoring function was carried out using PyRx software(21). The results of the dock poses were visualized using Discovery Studio.

The Physicochemical Properties and druggability of selected compounds were predicted using SwissADME(22) and Molinspiration(23) platforms and their predicted toxicity profile also compared using the PROTOX platform(24)

## 5. Conclusions

From the 22 phyto-compounds that were virtually screened, Scopodulcic acid and Dammarenolic acid showed the best binding energies with the Spike glycoprotein (6vsb) and the $M^{pro}$ (6flu7) respectively. This makes them potential lead compounds for development into candidates against the SARS-COV-2. Furthermore, the FDA approved drug silybinin (Legalon) with good binding affinity on the two targets can be evaluated further for possible repurposing against the SARS-COV-2 virus. We, therefore, propose that these lead compounds be tried for the COVID19 disease management

**Author Contributions:** For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used "Conceptualization, N.E. A.U and F.A; methodology, A.U,F.A ; software, A.U,F.A; validation N.E. A.U , N.S., S.O,J.C..A, A.U,U.U,L.P,and Z.Z formal analysis, X.X.; investigation, X.X.; resources, X.X.; data curation, F.A,A.U; writing—original draft preparation, N.E. A.U , N.S., S.O,J.C..A,

A.U,U.U,L.P,and; writing—review and editing, N.E. A.U , N.S., S.O,J.C..A, A.U,U.U,L.P,and; visualization, X.X.; supervision, J.CA,N.E;; funding acquisition, J.C.A All authors have read and agreed to the published version of the manuscript.", please turn to the CRediT taxonomy for the term explanation. Authorship must be limited to those who have contributed substantially to the work reported.

**Funding:** This research was funded by World bank-IDA, grant number ACE-126974" given to J.C.A.

**Conflicts of Interest:** Declare conflicts of interest or state "The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results".

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