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Covid-19 treatment: Investigation on the phytochemical constituents of *Vernonia amygdalina* as potential Coronavirus-2 inhibitors

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ABSTRACT

The upsurge in the current cases of COVID-19 poses a major threat on human health and population all over the globe. The emergence of new infectious diseases and increase in frequency of drug resistant viruses demand effective and novel therapeutic agents. In this study, we used bioinformatics approach to investigate the possible inhibitory potentials of phytochemical constituents of *Vernonia amygdalina* towards coronavirus-2 major protease. Pharmacodynamics, pharmacokinetics and toxicological profiles of the compounds were also examined using the pkCSM server. All the phytochemicals showed good binding affinity to the binding pocket of PDB ID 6LU7. It was observed that veronicoside A exhibited the highest binding affinity when compared to remdesivir, hydroxy-vernolide, vernodalin, vernodalol, and vernolide. The amino acids LEU272, LEU287, GLY275, TYR237, LYS236, THR198, THR199, ARG131, and LYS5 were showed as the key residues for veronicoside A binding to human SARS-COV2 major protease. The Pharmacodynamics and pharmacokinetics results suggested that all the tested phytochemicals have significant drug likeness properties and they could be absorbed through the human intestine. Furthermore, all the tested phytochemicals are not hepatoxic and also exhibited non or relatively low toxic effects in human. Taken together, the results of this study indicated that all the tested phytochemicals are potential putative inhibitors of SARS-COV2 major protease with non or low toxicity effects. However, further experimental and clinical studies are needed to further explore their activities and validate their efficacies against COVID-19.

1. Introduction

Novel Coronavirus disease 2019 (COVID-19), ranked among the ninth deadliest world pandemic, is a highly infectious and severe acute respiratory disorder caused by a morbific virus called SARS-CoV-2 which is transmitted to humans via contact with infected persons and/or feeding on infected animals. The COVID-19 clinical manifestations are very similar to viral pneumonia such as fever, fatigue, cough, shortness of breath, and other complications. According to reports obtained on WHO and NCDC websites as of 27th June 2020, the coronavirus breakout in Wuhan, a city in Hubei Province of China in November 2019 and has spread to many countries in the world. This global pandemic has forced many nations to lock down their social and economic activities which in turn have adverse effects on the economy. Globally, more than ten million people have been confirmed infected with over 500,000 deaths. Nigeria is one of the countries seriously affected by the virus having over 25,000 cases and more than 500 mortalities [1,2]. Thus, there is an exigent need for effective and non-invasive treatment.

Coronaviruses (SARS-CoV) are non-segmented positive-sense single-stranded RNA viruses with a large viral RNA genome of diameter 80–120 nm [26]. They belong to the family of Coronaviridae, in the subfamily Orthocoronaviridae which consists of four genera namely: Alpha, Beta, Gamma, and Delta coronavirus [3]. Some of the modes of actions of SARS-CoV-2 include hyper-inflammation characterized by a sudden and fatal hyper-cytokinaemia with multi-organ failure [4]; immunosuppression; reduction of Angiotensin-Converting Enzyme 2 (ACE2) to enhance pulmonary vascular permeability and damage the alveoli [5]; and activated by ORF3a, ORF3b, and ORF7a via JNK pathway which induces lung damage [6].

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At present, there is no known effective treatment that can mitigate/inhibit the pathogenesis of coronavirus in infected patients, however, there is vaccine to prevent the wide spread of the virus. Available clinical interventions for COVID-19 patients are only palliative and limited to support. There is still urgent need for therapeutic agents as cases of infections is increasing on the daily basis. Therefore, many research groups around the world are currently focusing on developing novel antivirals using various in silico methods [28–30]. This research investigated the inhibitory potentials of some naturally occurring phytochemicals present in *Vernonia amygdalina* against COVID-19 major protease (6LU7). *Vernonia amygdalina*, commonly known as bitter leaf, is an indigenous African plant with a number of scientific proven medicinal importance [10,31–33]. These phytochemicals can be repurposed to mitigate the pathogenesis of the SARS-CoV-2 and thus put an end to the frightening associated mortality rate.

2. Materials and methods

2.1. Protein preparation

The crystal structure of SARS-COV2 major protease (PDB ID 6LU7) with resolution 2.16 Å was retrieved from the protein databank (www.rcsb.org). Prior to docking and analysis, the crystal structure was prepared by removing existing ligands and water molecules. Also, missing hydrogen atoms were added using Autodock v4.2 program, Scripps Research Institute [27]. Thereafter, non-polar hydrogens were merged while polar hydrogen was added and subsequently saved into pdbqt format in preparation for molecular docking.

2.2. Ligand preparation

The SDF structures of selected phytochemical constituents of the leaf extract of *Vernonia amygdalina*: hydroxyvernolide, vernodalin, vernodalol, vernolide, and veronicoside were retrieved from the PubChem database (www.pubchem.ncbi.nlm.nih.gov). The phytochemicals were converted to mol2 chemical format. Polar hydrogens were added while non-polar hydrogens were merged with the carbons and the internal degrees of freedom and torsions were set. The protein and ligand molecules were further converted to the dockable PDBQT format using Autodock tools.

2.3. Molecular docking

Docking of the phytochemicals to the targeted protein and determination of binding affinities was carried out using AutodockVina [7]. The PDBQT formats of the receptor and that of the phytochemicals were positioned at their respective columns and the software was run. The binding affinities of phytochemicals for the protein target were recorded. The phytochemicals were then ranked by their affinity scores. The molecular interactions between the receptor and phytochemicals with most remarkable binding affinities were viewed with Discovery Studio Visualizer, BIOVIA, 2016. The respective binding free energy was calculated by the Molecular Mechanics/Generalized Born Surface Area (MM/GBSA) using HawkDock Server (http://cadd.zju.edu.cn/hawkdock/).

2.4. Molecular dynamics simulation

The conformational stability of the protein–ligand interactions was evaluated using molecular dynamics simulations analysis performed through iMODS server (http://imods.chaconlab.org) by normal mode analysis (NMA) predicting properties such as deformability, mobility profiles, eigenvalues, variance, co-variance map and elastic network of the protein–ligand interactions [8].

2.5. ADMET analysis

The solubility, pharmacodynamics, pharmacokinetics and toxicological profiles of hydroxyvernolide, vernodalin, vernodalol, vernolide, veronicoside and remdesivir were computed based on their ADMET (absorption, distribution, metabolism, elimination, and toxicity) studies using pkCSM tool (http://biosig.unimelb.edu.au/pkcsm/prediction) as described by Pires et al. (2015). The canonical SMILE molecular structures of the compounds used in the studies were obtained from PubChem (https://pubchem.ncbi.nlm.nih.gov).

3. Results

3.1. Molecular docking analysis

The molecular docking analysis and visualization of 6LU7 binding with remdesivir, hydroxy-vernolide, vernodalin, vernodalol, vernolide, and veronicoside A is shown in Fig. 1, Fig. 2 and Table 1. Out of the all the tested compounds, veronicoside A exhibited the best docked score (−7.4 kcal/mol) with the SARS-COV2 major protease (6LU7). LEU272, LEU287, GLY275, TYR237, LYS236, THR198, THR199, ARG131, LYS5 amino acid residues participating in the interaction at the binding pocket of the SARS-COV2 major protease (6LU7) (Fig. 1F). Vernolide exhibited (−7.2 kcal/mol) binding affinity with 6LU7. HIS41, MET49, GLN189, GLU166, MET165, LEU167, THR190 amino acid residues participating in the interaction at the binding pocket of 6LU7 (Fig. 1E). Vernodalin displayed (−6.7 kcal/mol) binding affinity with 6LU7. ASP153, GLN110, and PHE294 amino acid residues participating in the interaction at the binding pocket of 6LU7 (Fig. 1C). Remdesivir exhibited (−6.6 kcal/mol) binding affinity with 6LU7. ARG131, THR199, LYS137, ASP289, LEU272, LEU287, and MET276 amino acid residues participating in the interaction at the binding pocket of 6LU7 (Fig. 1A). Hydroxy-vernolide showed (−6.2 kcal/mol) binding affinity with 6LU7. HIS41, MET49, GLN189, GLU166, MET165, LEU167, and THR190 amino acid residues participating in the interaction at the binding pocket of 6LU7 (Fig. 1B). Vernodalol exhibited (−6.1 kcal/mol) binding affinity with 6LU7. ASP197, THR199, TYR237, TYR239, LEU272, LEU286, and LEU287 amino acid residues participating in the interaction at the binding pocket of 6LU7 (Fig. 1D).

3.2. Molecular properties of the phytochemicals

Data in Table 2 revealed the results of the molecular properties of the phytochemicals used in this study. Veronicoside A was found to have the highest molecular weight of 814.699, followed by vernodalol with 392.404, hydroxy-vernolide with 378.377, vernolide with 362.378 and vernodalin with 360.362. Similarly, the surface area of the phytochemicals: veronicoside A, vernodalol, hydroxy-vernolide, vernolide, and vernodalin are 319.905, 162.317, 155.582, 150.788 and 150.152 respectively. Vernodalin has the highest lipophilicity of 0.8498, vernodalol has 0.4583, hydroxy-vernolide has 0.141 while veronicoside A has the least lipophilicity of 2.2824 (see Fig. 3.).

3.3. Predicted absorption properties of the phytochemicals

The predicted absorption properties of each of the phytochemicals were reported in table 3. The result showed that veronicoside A has the highest water solubility value of −2.886 while vernolide has the lowest valve of −3.936. Vernolide has the highest permeability value of 0.804, but veronicoside A having the least permeability valve of −1.469. Likewise, vernolide can be readily absorbed by the intestinal cells (100%) whereas veronicoside A may not be absorbed intestine. All the phytochemicals are substrate of P-glycoprotein and none of the phytochemicals is neither inhibitor of P-glycoprotein-I nor P-glycoprotein-II.

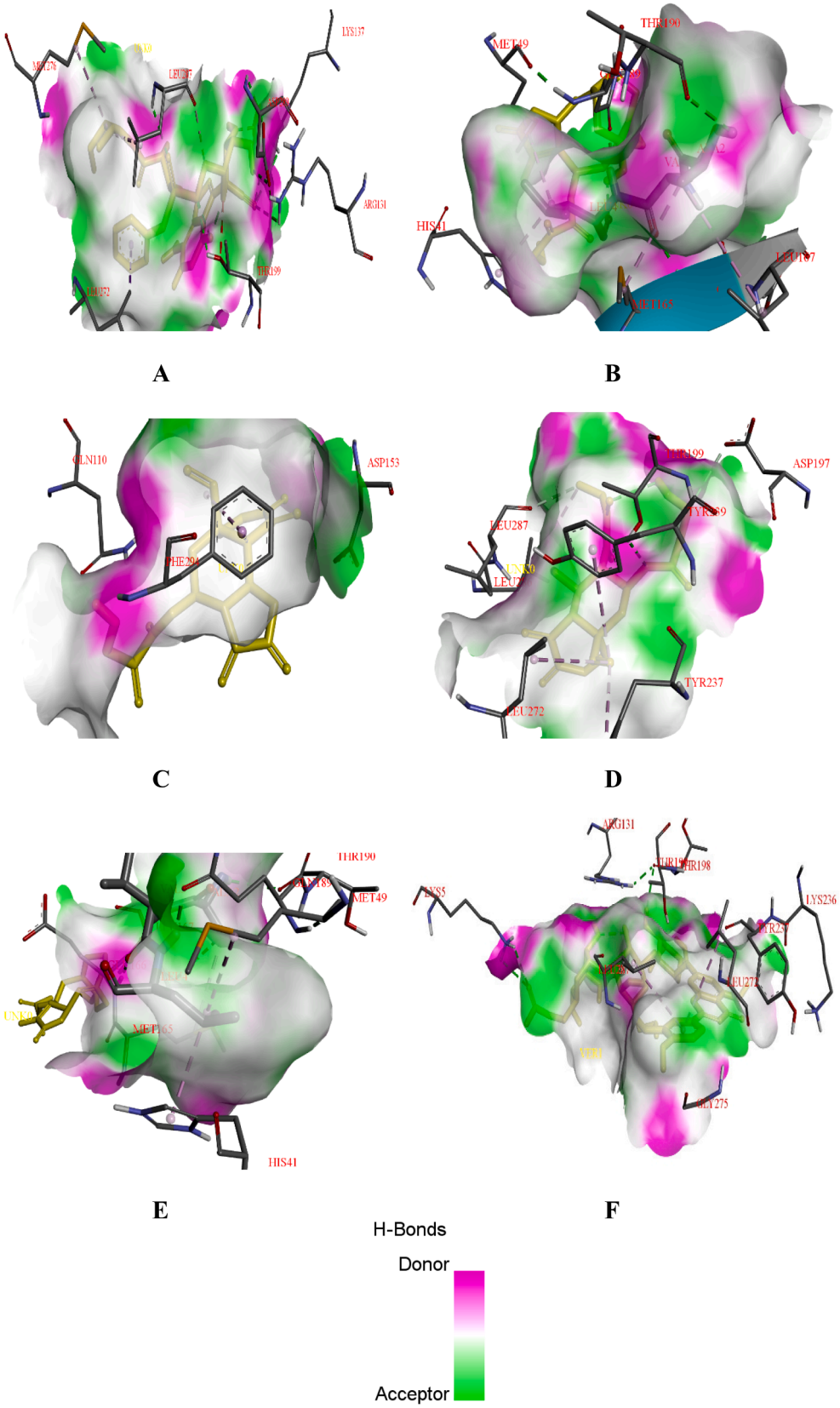

Fig. 1. Docking analysis and visualization of 6LU7 binding with (A) Remdesivir, (B) Hydroxy-vernolide, (C) Vernodalin, (D) Vernodalol, (E) Vernolide, (F) Vernonicoside A.

Fig. 2. Binding-interaction analysis of 6LU7 binding with (A) Remdesivir, (B) Hydroxy-vernolide, (C) Vernodalin, (D) Vernodalol, (E) Vernolide, (F) Veronicoside A.

Table 1
Molecular docking analysis of the tested compounds against COVID-19 major protease (6LU7).

Compound	PubChem CID	Binding energies (kcal/mol)	ligand-amino acid interactions
Remdesivir	121,304,016	−6.6	ARG131, THR199, LYS137, ASP289, LEU272, LEU287, MET276
Hydroxy-vernolide	5,281,472	−6.2	HIS41, MET49, GLN189, GLU166, MET165, LEU167, THR190
Vernodalin	179,375	−6.7	ASP153, GLN110, PHE294
Vernodalol	442,318	−6.1	ASP197, THR199, TYR237, TYR239, LEU272, LEU286, LEU287
Vernolide	5,281,508	−7.2	HIS41, MET49, GLN189, GLU166, MET165, LEU167, THR190
Veronicoside A	44,258,142	−7.4	LEU272, LEU287, GLY275, TYR237, LYS236, THR198, THR199, ARG131, LYS5

3.4. Predicted in vivo distribution and cytochrome P450 promiscuity of the phytochemicals

Table 4 showed the predicted *in vivo* distribution of the phytochemicals. All the phytochemicals tested have relatively low steady-state volume of distribution. Also, the predicted result revealed that hydroxy-vernolide has the highest unbound fraction in the human blood. All the phytochemicals have relatively low blood–brain barrier and CNS permeability values.

Table 5 displayed the predicted human cytochrome P450 promiscuity of the screened phytochemicals. All the compounds were neither substrate of CYP2D6 and CYP3A4 nor inhibitor of CYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A4. Expect vernodalin and vernolide that are substrate of CYP3A4.

3.5. Predicted in vivo clearance of the phytochemicals

The predicted clearance of each of the phytochemicals were reported in Table 6. Hydroxy-vernolide has the highest total clearance rate of 1.267 while Veronicoside A has the least clearance rate of −0.356. None of the phytochemicals is substrate of renal organic cation transporter.

3.6. Predicted toxicological profile of the phytochemicals

Table 7 reported the predicted toxicological profile of all the tested phytochemicals. Hydroxy-vernolide, vernolide, and veronicoside A has no mutagenic potentials against bacteria (AMES toxicity) but vernodalol and vernodalin could be toxic to bacteria. None of the phytochemicals has adverse effects on the hepatic or dermal cells. The phytochemicals are not inhibitors of human ether-a-go-go-related gene (hERG) hERG I and hERG II except veronicoside A which may inhibit hERG II. All the phytochemicals have relatively low maximum recommended tolerated dose values.

4. Discussion

Plants belonging to the *Vernonia* genus is principally known to house huge amounts of sesquiterpene lactones with several documented pharmacological and biological activities [9]. *Vernonia amygdalina* is a member of this group of plants and normally called bitter leaf. It is a medicinal plant used traditionally in the management and treatment of different diseases such as respiratory diseases, cough, reproductive diseases [10]. This present study examines the possible inhibitory activity of selected phytochemical constituents (hydroxyvernolide, vernodalin, vernodalol, vernolide, and veronicoside) of the leaf extract of *Vernonia amygdalina* against SARS-COV2 major protease (6LU7).

As shown in the result, veronicoside A exhibited the highest affinity with 6LU7. This indicates that veronicoside can be a putative inhibitor of coronavirus-2. Veronicoside has been reported to have radical scavenging and antioxidant activities. It has also been documented to have cytotoxicity activities against Hep-2 (human larynx epidermoid carcinoma), RD (human rhabdomyosarcoma), and L-20B (transgenic murine cells) cell lines [11]. Several species of plants containing veronicoside are being used in traditional medicine to treat influenza, respiratory diseases, hernia, cough, laryngopharyngitis, cancer, hemoptysis, and are also used as an antiscorbutic and expectorant [12].

Vernodalin and vernolide also showed a good binding affinity with the coronavirus-2 major protease, suggesting them as potential suitable inhibitors of the virus. Vernodalin and vernolide have been reported to exhibit antiproliferative activities [13] against lung A549 (adenocarcinomic human alveolar basal epithelial cells), HeLa, and MDA-MB-23 (human breast cancer) cell lines and induced apoptosis on HepG2 cells with G2/M phase cell cycle arrest [14]. They have potential to be used as lead compounds in the development of a therapeutic natural product for treatment of cancers in the lungs, breast or liver. These phytochemicals may also offer help in inhibiting the proliferative activities of SARS-COV2 in the host thereby mitigate the pathogenesis of COVID-19.

Sinisi *et al.* [15] has reported vernodalol has a good activator of Nrf2. NF-E2-related factor-2 (NRF2) is a transcriptional factor that binds to and facilitates the activation of the ARE-dependent gene. Under basal conditions, NRF2 is sequestered in the cytoplasm and its expression is maintained to be low due to constant polyubiquitination. In response to different kinds of stress, NRF2 is significantly induced and translocates into the nucleus, where it activates the antioxidant response element (ARE)-dependent gene expression in association with small Maf proteins and other coactivators. Thus, causing the release of phase II cytoprotective enzymes such as γ-glutamylcysteine ligase (γ-GCS), NAD[P]H: quinone oxidoreductase-1 (NQO1), heme oxygenase-1 (HO-1), and glutathione S-transferase (GST) which protect the cells against the attack of the stress. Since oxidative stress has been reported as one of the features of COVID-19, vernodalol can help to extenuate it by activation of NRF2.

Nuclear Factor kappa-light-chain-enhancer of activated B-cells (NF-kB) pathway has been implicated in the mode of actions of SARS-COV2 [16,17] leading to a cytokine profile resembling secondary haemophagocytic lymphohistiocytosis (sHLH) with a hyperinflammatory syndrome characterized by a fulminant and severe hypercytokinaemia with multiorgan failure. This is characterized by increased tumor necrosis factor-α, interleukin (IL)-2, IL-7, interferon-γ inducible protein 10,

Table 2
Molecular properties of the phytochemicals.

Descriptor	Remdesivir	Hydroxy vernolide	Vernodalin	vernodalol	Vernolide	Veronicoside A
Molecular weight	602.585	378.377	360.362	392.404	362.378	814.699
Lipophilicity (Log P)	2.31218	0.141	0.8498	0.4583	1.1686	−4.0253
Number of rotatable bonds	13	3	4	6	2	11
Number of acceptors	13	8	7	8	7	22
Number of donors	4	2	1	2	1	12
Surface area	242.488	155.582	150.152	162.317	150.788	319.905

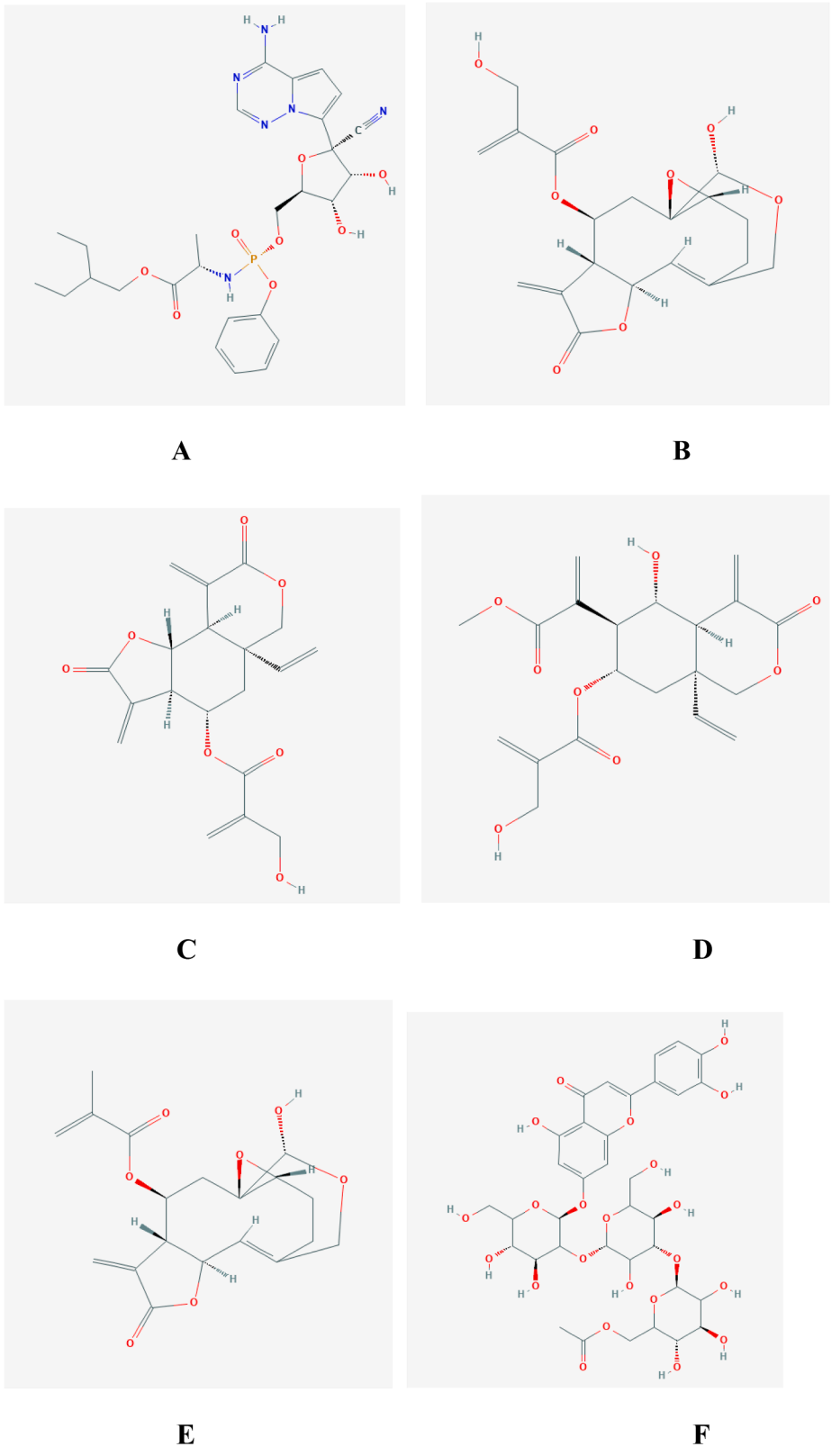

Fig. 3. 2D structures of (A) Remdesivir, (B) Hydroxy-vernolide, (C) Vernodalin, (D) Vernodalol, (E) Vernolide, (F) Veronicoside A.

granulocyte-colony stimulating factor, macrophage inflammatory protein 1-α, and monocyte chemoattractant protein 1 [18]. However, vernolide and vernodalol have been documented to show marked inhibitory activity on STAT3/NF-κB [15]. Therefore, vernolide and vernodalol could protect against COVID-19-induced multiorgan failure by suppressing the hyperinflammatory syndrome via inhibition of NF-kB.

The results of the solubility, pharmacodynamics, pharmacokinetics and toxicological profiles of remdesivir, hydroxy-vernolide, vernodalin, vernodalol, vernolide, and veronicoside A are presented in Tables 2–7.

Table 3
Predicted absorption properties of the phytochemicals.

Model name	Remdesivir	Hydroxy vernolide	Vernodalin	vernodalol	Vernolide	Veronicoside A
Water solubility (log mol/L)	−3.07	−3.894	−3.382	−3.192	−3.936	−2.886
Caco2 permeability (log Papp in 10^{-6} cm/s)	0.635	0.8	0.469	0.279	0.804	−1.469
Intestinal absorption (% Absorbed)	71.109	96.455	96.144	75.395	100	0
Skin permeability (log Kp)	−2.735	−2.908	−3.222	−3.447	−3.086	−2.735
P-glycoprotein substrate	Yes	Yes	Yes	Yes	Yes	Yes
P-glycoprotein I inhibitor	Yes	No	No	No	No	No
P-glycoprotein II inhibitor	No	No	No	No	No	No

Caco2: Human colon adenocarcinoma-2.

Table 4
Predicted *in vivo* distribution of the phytochemicals.

Model name	Remdesivir	Hydroxy vernolide	Vernodalin	Vernodalol	Vernolide	Veronicoside A
VDss (human) (log L/kg)	0.307	0.198	−0.236	−0.197	0.156	0.246
Fraction unbound (human) (Fu)	0.005	0.551	0.419	0.509	0.452	0.225
BBB permeability (log BB)	−2.056	−0.423	−0.684	−0.48	−0.566	−2.686
CNS permeability (log PS)	−4.675	−3.179	−3.061	−3.049	−3.092	−6.228

VDss: Steady-state volume of distribution, BBB: Blood-brain barrier, CNS: Central nervous system.

Table 5
Predicted human cytochrome P450 promiscuity of the phytochemicals.

Model name	Remdesivir	Hydroxy vernolide	Vernodalin	vernodalol	Vernolide	Veronicoside A
CYP2D6 substrate	No	No	No	No	No	No
CYP3A4 substrate	Yes	No	Yes	No	Yes	No
CYP1A2 inhibitor	No	No	No	No	No	No
CYP2C19 inhibitor	No	No	No	No	No	No
CYP2C9 inhibitor	No	No	No	No	No	No
CYP2D6 inhibitor	No	No	No	No	No	No
CYP3A4 inhibitor	No	No	No	No	No	No

Table 6
Predicted *in vivo* clearance of the phytochemicals.

Model name	Remdesivir	Hydroxy vernolide	Vernodalin	vernodalol	Vernolide	Veronicoside A
Total clearance (log ml/min/kg)	0.198	1.267	0.725	0.747	1.184	−0.356
Renal OCT2 substrate	No	No	No	No	No	No

OCT2: Organic cation transporter 2.

Table 7
Predicted toxicological profile of the phytochemicals.

Model name	Remdesivir	Hydroxy vernolide	Vernodalin	vernodalol	Vernolide	Veronicoside A
AMES toxicity	No	No	Yes	Yes	No	No
Max. Tolerated dose (human) (log mg/kg/day)	0.15	0.11	0.236	0.501	−0.324	0.361
hERG I inhibitor	No	No	No	No	No	No
hERG II inhibitor	Yes	No	No	No	No	Yes
Oral Rat Acute Toxicity (LD50) (mol/kg)	2.043	3.949	2.285	2.388	3.467	2.475
Oral Rat Chronic Toxicity (LOAEL) (log mg/kg_bw/day)	1.639	2.087	1.768	1.971	1.107	5.441
Hepatoxicity	Yes	No	No	No	No	No
Skin sensitization	No	No	No	No	No	No
T. pyriformis toxicity (log ug/L)	0.285	0.287	0.314	0.29	0.295	0.285
Minnow toxicity (log mM)	0.291	3.852	2.36	3.646	3.007	10.719

AMES: Salmonella typhimurium reverse mutation assay, Max.: Maximum hERG: Human ether-a-go-go-related gene.

The profiles were investigated as a systemic virtual screening of drugs and potential drugs. This is done as alternative to *in vivo* examinations which are essential complements in drug discovery. The Lipinski's rule is a major criterion to evaluate drug likeliness and to determine if a compound with a particular pharmacological and biological actions has physical and chemical properties that could favour its activities in human. The molecular properties of the compounds based on the computed partition coefficient (log P) showed that the phytochemicals have relatively good lipophilicity as the logP values were less than 5 [19,20]. All the tested phytochemicals could be maintained in the system at appropriate concentrations.

Intestinal absorption and Caco2 permeability are parameters that determine the ultimate bioavailability of drug candidates. The tested phytochemicals (hydroxy-vernolide, vernodalin, vernodalol, vernolide, and veronicoside A) have relatively low Caco2 permeability potential ($<8 \times 10^{-6}$ cm/s) and could be absorbed through the human intestine [21]. However, ADMETSAR1 as predicted that remdesivir is subcellular localization in the lysosome [22]. Furthermore, the observed

lipophilicities have an association with Caco2 permeability but correlated negatively with water solubility potentials of the tested phytochemicals. This result is in tandem with the findings of Yazdanian et al [23] who used the human colon adenocarcinoma (Caco-2) cell line assay to document no correlation between the drug permeability and measured lipophilicity. All the tested phytochemicals were predicted to be substrates of P-glycoprotein, a member of the ATP-binding cassette transporter and an efflux membrane transporter found chiefly in epithelial cells. On the other hand, none of the phytochemicals is predicted as P-glycoprotein inhibitors except remdesivir. This indicate that they don't alter the normal physiological activities of P-glycoprotein including restricting the active uptake and the distribution of drugs [24].

The volume of distribution calculated using a steady-state volume of distribution (VDss) as predicted showed that vernodalin has the lowest theoretical dose required for uniform distribution in the plasma when compared with other tested phytochemicals (hydroxy-vernolide, vernodalol, vernolide, and veronicoside A). VDss showed the distribution of drug in the tissue and plasma. The degree of diffusing across plasma membrane increases in this order hydroxyvernolide < vernodalol < vernolide < vernodalin < veronicoside A < remdesivir measured as the fraction that is in the unbound state. The predicted evaluation on the nervous system distribution of the compounds revealed that lipophilicity of the phytochemicals correlates to the degree of permeability across the central nervous system and the blood–brain barrier.

Cytochrome P450 is a group of enzymes that perform crucial functions in drug metabolism. They play a major role in the activation of drugs and also in the toxicity effects of the drugs. Only vernolide, vernodalin and remdesivir is substrate of CYP3A4, all other tested phytochemicals were neither substrate of CYP2D6 nor CYP3A4. None of the phytochemicals is inhibitors of CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4. The lipophilicity of the drug appears to correlate negatively to metabolism-related toxicity. Furthermore, none of the tested phytochemicals were a substrate of renal organic cation transporter, this implies that they are possibly cleared through other available routes such as sweat, bile, etc. Also, veronicoside A was observed to have the least total clearance while hydroxyvernolide has the highest. Drug clearance is related to bioavailability and is crucial for determining dosing rates to achieve steady-state concentrations.

The toxicological assessment of the tested phytochemicals revealed that all the tested compounds expect remdesivir are not skin sensitive (dermal toxic) or hepatoxic. Similarly, only vernodalin and vernodalol are bacterial mutagenic potential drugs using the AMES toxicity examination. However, all the compound showed high level of toxicity to *Tetrahymena pyriformis* toxicity test. None of the phytochemicals is an inhibitor to hERG I but veronicoside A and remdesivir may be inhibitors to hERG II. Inhibition of the hERG potassium channel could result in delayed ventricular repolarisation leading to a severe disturbance in the normal cardiac rhythm and disrupt hepatic functions [25]. Acute and chronic toxicity were also carried out on the tested phytochemicals to determine the safety of the compounds when administered. Exposure to low-moderate doses/concentrations of xenobiotics over long period of time is of significant concern in many treatment strategies or interventions. Chronic studies are designed to identify the lowest dose of a compound that can result in adverse effects (LOAEL), and the highest dose at which no adverse effects are observed (NOAEL).

5. Conclusion

Taken together, the results from this study showed that all the tested phytochemicals exhibited significant binding affinity to the binding pocket of SARS-COV2 major protease suggesting them as potential molecules that could mitigate/inhibit SARS-COV2. Binding of these phytochemicals to SARS-COV2 could inhibit or interfere the pathogenesis of COVID-19 thereby preventing its cellular entry and proliferation. Three of the tested phytochemicals (veronicoside A, vernodalin and vernolide) showed higher binding affinity with 6LU7 than remdesivir, an established antiviral drug, thus, suggested them as better antiviral agents. The pharmacodynamics and pharmacokinetics properties of the phytochemicals showed that they would be good drug candidates and the toxicological evaluations showed that the phytochemical has relatively low or no toxic effect in human. However, further experimental and clinical studies are needed to further explore their activities and validate their efficacies against COVID-19.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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