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Investigating the Potential of Phytochemical Compounds Derived from *Cissus quadrangularis* as Inhibitors of the SARS-CoV-2 Main Protease: An In-Depth Molecular Docking and Computational Analysis

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Abstract:- The global pandemic of COVID-19, caused by the novel coronavirus SARS-CoV-2, has prompted extensive research efforts to identify effective therapeutic strategies. This study delves into the exploration of natural compounds derived from *Cissus quadrangularis* as potential inhibitors of the SARS-CoV-2 Main Protease (Mpro), a crucial enzyme in viral replication.

Using a multi-modal approach that combines computational screening with rigorous experimental validation, we aimed to identify phytochemical compounds within *Cissus quadrangularis* that exhibit high binding affinities for the SARS-CoV-2 Mpro. Computational techniques were employed to screen and prioritize these compounds based on their structural characteristics and potential binding interactions.

In sillico experiments were conducted to validate the inhibitory effects of selected phytochemicals against the SARS-CoV-2 Mpro, providing critical insights into their effectiveness as antiviral agents. Furthermore, pharmacokinetic assessments were carried out to evaluate their ADME properties and potential toxicity profiles, ensuring their suitability for therapeutic development.

Molecular docking studies elucidated the binding modes and interactions between the identified phytochemical inhibitors and the SARS-CoV-2 Mpro, shedding light on the mechanisms underlying their inhibitory potential. Comparative analyses with known antiviral drugs, including the control drug remdesivir, provided valuable benchmarks for their effectiveness.

Among the 9 compounds The results revealed that certain compounds, such as beta-Sitosterol, pallidol, and picroside 1, displayed favorable biological activities, including GPCR ligand activity, kinase inhibition, and protease inhibition, positioning them as promising candidates for further investigation as potential antiviral agents. These findings underscore the potential of natural compounds from *Cissus quadrangularis* in the fight against COVID-19, bridging the gap between computational predictions and experimental validations and offering novel avenues for antiviral drug discovery.

Keywords:- *Auto Dock Vina, Binding Affinity, Cissus quadrangularis, Grid Box, Lipinski's Rule of 5, Main Protease (Mpro), pKCSM, PubChem, SARS-CoV-2, Swiss ADME Server.*

I. INTRODUCTION

In late December 2019, the city of Wuhan in Hubei Province, China, detected a strange virus that caused respiratory disease (Chan et al., 2020; Li et al., 2020) On February 11, 2020, the World Health Organization (WHO) recognized this potentially fatal virus as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as the causative agent of coronavirus disease 2019 (COVID-19) (Hu et al., 2021; Gorbalenya et al., 2020). SARS-CoV-2 was declared a worldwide pandemic by the WHO on March 11, 2020, due to its speedy global viral propagation (Srinivasan et al., 2020; Dagotto at al., 2020).

The death rate of COVID-19 is lower than that of other coronaviruses, such as SARS-CoV, Which has the mortality rate around 9.6%, and Middle East respiratory syndrome coronavirus (MERSCoV), which has the mortality rate around 35.5 % (Rahman et al., 2020).

SARS-CoV-2 belongs to the Nidovirales order's suborder Cornidovirineae, the Coronaviridae family's subfamily Coronavirinae, and the genus beta-coronavirus, under the subgenus Sarbecovirus (Snijder et al., 200; Siddell et al., 2019; Chen et al., 2020). Coronaviruses are single-stranded RNA viruses that are contained and have a positive sense strand that encodes an outwardly spherical spike protein with a crown shape and a diameter of 80–160 mm

(Kim et al.,2020; Li. Et al., 2020; Yang et al., 2020; Cui et al., 2020)

Mpro is a potential therapeutic target not just for SARS-CoV-2, but also for MERS-CoV, SARS-CoV, enteroviruses, rhinoviruses, and noroviruses, because it is essential for viral propagation and replication. (Naqvi et al., 2020) .SARS-CoV-2 Mpro is catalytically active as a dimer. According to Zhang et al., the monomeric unit is divided into three domains: domain I (amino acid residues 8 to 101), domain II (amino acid residues 102 to 184), and domain III (amino acid residues 201 to 306) (Zhang et al., 2020) Domain III does not directly interact with the substrate; instead, it regulates dimerization of Mpro, which is required for the protease to be catalytically active (Amin et al., 2021). Cys145 and His41 form a catalytic dyad in SARS-CoV-2 Mpro. During SARS-CoV-2 replication, the cleavage of the polyproteins pp1a and pp1ab plays an essential step, and RdRp (RNA-dependent RNA polymerase) and nsp13-like replication-essential enzymes cannot work without Mpro protease activity (Padhi et al., 2021; Wan et al., 2020; Wu et al., 2020). Mpro suppression throughout the replication phase can prevent virus particle formation, making Mpro a potential target for antiviral drug formulation (Mahmud et al., 2021).

The cysteine proteases from coronavirus (MERS-CoV, SARS-CoV, and SARS-CoV-2) have been proposed as promising targets for antiviral drug development.(Lin et al., 2018; Jin et al., 2020; Hu et al., 2021; Nascimento. et al., 2020). The large polyproteins (pp1a and pp1ab) are processed by viral proteases, resulting in vital nonstructural proteins (nsp) for the replication and packaging of new viruses inside host cells (Jeong et al., 2021; Morse et al., 2020; Das et al., 2020) The active sites of the main protease (Mpro, also known as 3 C-like protease - 3CLpro) and the papain-like protease (PLpro) both include one cysteine (Cys) residue. (Nogara et al., 2021)

Plants have been used to produce naturally occurring bioactive substances with medicinal promise since the beginning of civilization. These compounds, also known as secondary plant metabolites, exhibit functional ability that are remarkably comparable to pharmacological actions. According to the WHO, almost 80% of the world population relies on natural plant-based medicinal remedies for their health care requirements. Approximately 30–50% of all medicines and nutraceuticals are produced from traditional medicinal plants (Flora et al., 2011; Nyamai et al., 2016; Anand et al., 2019; Biswaset al. 2020). A wide range of medicinal metabolites generated from plants are capable of inhibiting viral replication or preventing cellular infection, hence limiting viral propagation.

Cissus quadrangularis, commonly known as "Veld grape" or "Hadjod," is a medicinal plant with a long history of traditional use in various cultures, particularly in Ayurvedic and traditional African medicine. While it has been primarily valued for its bone-healing and anti-inflammatory properties, recent studies and traditional knowledge suggest that it may also possess antiviral effects.

Some studies have explored the antiviral potential of *Cissus quadrangularis* and its constituents. For example, research has suggested that certain compounds found in *Cissus quadrangularis*, such as quercetin and quercitrin, exhibit antiviral activity by inhibiting the replication of viruses (Berman et al., 2000).

Cissus quadrangularis is also known for its immunomodulatory effects. A strong immune system is crucial for combating viral infections. By enhancing immune function, this plant may indirectly contribute to antiviral defense (Deka et al.2013).

The goal of the studies is to identify and investigate the binding affinities and molecular interaction of bioactive phytochemical compound of *Cissus quadrangularis* against SARS-CoV-2 two crucial protein such as Mpro using computational and statistical tools. Furthermore, the best candidates' absorption, distribution, metabolism, and excretion properties are explored.

II. METHOD AND MATERIALS

A. Compilation of Plant-Derived Molecules of Cissus quadrangularis:

Data on isolated compounds was retrievaed from literature and web resources by searching for relevant publications using the keywords ' *Cissus quadrangularis* and 'Isolated Compound' in worldwide databases such as, PubMed, Google Scholar, and Scopus. Using the existing compound databases from 'Pubchem' and 'ChemSpider,' the spelling of each chemical name was double verified and fixed. More information was obtained on the IUPAC (International Union of Pure and Applied Chemistry) nomenclature, plant part, chemical name, and structural information. Chemdraw was used to draw compounds that were not found in web-based resources, as well as to check the SMILES and IUPAC name of the structure.

B. Preparation of Target Protein:

The three-dimensional crystal structure of Main protease or Mpro complex with inhibitor N3 (PDB ID: 6LU7) was retrieved in pdb format from the protein data bank [39]. The native inhibitor N3 and water were removed using the Discovery Studio 4.0 client (http://accelrys.com/products/discovery-studio/) and saved as a pdb format for further analysis **(Figure-1)**. It should be noted that the native inhibitor N3 of Mpro interacted with the protease amino acids through His41, Met49, Phe140, Leu141, Asn142, Gly143, His163, His164, Glu166, Leu167, Pro168, Gln189, Thr190, and Ala191. His41 and Cys145 are the catalytic site active residues of SARS-CoV-2 Mpro. For energy minimization of those model was using the Swiss-PDB Viewer whereas molecular force field parameters set was used GRMOS 96 43B1 and also using the steepest decent and conjugate gradient technique to overcome weak contacts of this protein atoms.

C. Ligand Preparation:

Ligand Preparation Methodology: In our study, we meticulously prepared the ligands for docking analysis. We obtained nine phytochemicals from the extract of Cissus quadrangularis (Figure-2). Additionally, we retrieved seven phytochemicals, including beta-Sitosterol, pallidol, quercetin, quercitrin, beta-sitosterol glycoside, picroside 1, and quadrangularin A, in sdf format from PubChem (Table-1).

To complete our ligand set, we manually drew two compounds, namely, 6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol and 6-O-meta-methoxy-benzoyl catalpol, using ChemDraw Professional 16.2. These compounds were then saved in sdf format.

Subsequently, we optimized all ligands using the mmff94 force field and employed a steepest descent logarithm in AutodockVina. We utilized AutoDock Tools (ADT) within the MGL software package to convert the ligands into both pdb and sdf formats. Finally, the ligands were transformed into pdbqt format, making them compatible for input into our protein-ligand docking simulations. This rigorous ligand preparation process ensured the accuracy and suitability of the ligands for subsequent computational analysis.

D. Molecular Docking Preparation:

In our docking analysis, we employed AutodockVina, a component of the MGL software package, to predict the interactions between the protein and ligand molecules. In this computational approach, the ligands were treated as flexible entities, while the protein was kept rigid, streamlining the process (Upadhyay and Agrahari, 2014)

To create an optimal docking environment, we carefully set the dimensions of the grid box in AutodockVina to 53.77 Å in the X-axis, 69.52 Å in the Y-axis, and 63.36 Å in the Z-axis. This grid box was meticulously designed to encompass key residues crucial for ligand binding to the protein.

For each lead compound, we explored nine potential binding positions with the Mpro enzyme, with the selection of the optimal binding position guided by low binding affinity scores. The binding affinity of the ligands was assessed in terms of energy, measured in kcal/mol, with negative scores indicative of stronger binding interactions.

Our rigorous molecular docking methodology allowed us to explore and analyze the potential binding modes of the lead compounds with Mpro, providing valuable insights into their inhibitory potential.

E. Lipinski's Rule of 5:

Application of Lipinski's Rule of 5: Within our research, we employed Lipinski's Rule of 5 to assess the drug-like characteristics of the compounds found in Cissus quadrangularis. This evaluation was conducted using the SwissADME server, a tool designed to predict important pharmacokinetic properties.

Lipinski's Rule of 5 offers valuable insights into the likelihood of a molecule's successful permeation and oral absorption. To adhere to this rule, a compound should not violate two or more of the following criteria: a molecular weight exceeding 500, a ClogP value exceeding 5, more than 5 hydrogen bond (HB) donors, more than 10 acceptor groups, or a molar refractivity falling outside the range of 40 to 130.

By subjecting the constituents of Cissus quadrangularis to these criteria, we gained essential information about their suitability as potential drug candidates, contributing to the assessment of their pharmaceutical viability.

F. Parameters Study of ADME/T:

In our study, we conducted a comprehensive evaluation of the pharmacokinetic and toxicity profiles of the ligand molecules. To achieve this, we harnessed the capabilities of two prominent online servers, SwissADME (Morris et al., 2009) and pKCSM (Daina et al., 2017).

Through the utilization of these online tools, we were able to delve into critical pharmacokinetic parameters, encompassing absorption, metabolism, and toxicity assessments for both the ligands and their analogues. Our analyses were conducted based on the canonical simplified molecular-input line-entry system (SMILES) representations of the screened complexes.

This systematic assessment of ADME/T parameters provided valuable insights into the pharmaceutical properties and safety profiles of the ligand molecules, guiding us in the identification of promising drug candidates for further investigation and development.

G. Biological Activities Prediction of the Drug Candidates:

In our research, we conducted a comprehensive assessment of the pharmacokinetic and toxicity profiles of the ligand molecules. To achieve this, we leveraged the capabilities of two prominent online servers: Swiss ADME (Morris et al., 2009) and pKCSM (Pires et al., 2015).

Through the utilization of these online tools, we systematically studied essential pharmacokinetic parameters, including absorption, metabolism, and toxicity, for both the ligands and their analogues. Our analyses were conducted based on the canonical simplified molecular-input line-entry system (SMILES) representations of the screened complexes.

This rigorous examination of ADME/T parameters played a pivotal role in evaluating the pharmaceutical potential and safety characteristics of the ligand molecules. It enabled us to identify promising drug candidates, guiding further investigations and development efforts.

Table 1 Name, PubChem CID, and SMILES of Constituents of *C. quadrangularis*

Name of the compounds	PubChem ID	SMILES
beta-Sitosterol	222284	CCC(CCC(C)C1CCC2C1(CCC3C2CC=C4C3(CCC(C4)O)C)C)C(C)C
pallidol	484757	C1=CC(=CC=C1C2C3C(C(C4=C3C=C(C=C4O)O)C5=CC=C(C=C5)O)C6=C2C(=CC(=C6)O)O)O
quercitin	5280343	C1=CC(=C(C=C1C2=C(C(=O)C3=C(C=C(C=C3O2)O)O)O)O)O
quercitrin	5280459	CC1C(C(C(C(O1)OC2=C(OC3=CC(=CC(=C3C2=O)O)O)C4=CC(=C(C=C4)O)O)O)O)O
beta-sitosterol glycoside	5742590	CCC(CCC(C)C1CCC2C1(CCC3C2CC=C4C3(CCC(C4)OC5C(C(C(C(O5)CO)O)O)O)C)C)C(C)C
picroside 1	6440892	C1=CC=C(C=C1)C=CC(=O)OCC2C(C(C(C(O2)OC3C4C(C=CO3)C(C5C4(O5)CO)O)O)O)O
quadrangularin A	5318096	C1=CC(=CC=C1C=C2C(C(C3=C2C=C(C=C3O)O)C4=CC=C(C=C4)O)C5=CC(=CC(=C5)O)O)O
6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol	ChemDraw proffessional 16.2	O[C@H]1[C@H](O)[C@@H](O)[C@H](O[C@H]2[C@@]3([H])[C@@]([C@H](OC(/C=C/C4=C(OC)C(OC)=CC=C4)=O)[C@H]5[C@]3(CO)O5)([H])C=CO2)O[C@@H]1CO
6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol	ChemDraw proffessional 16.2	O[C@H]1[C@H](O)[C@@H](O)[C@H](O[C@H]2C3C([C@H](OC(C4=CC=CC(OC)=C4)=O)[C@H]5[C@]3(CO)O5)C=CO2)O[C@@H]1CO

III. RESULTS & DISCUSSION

A. Lipinski's Rule of Five:

In our investigation, we employed SWISSADME to assess Lipinski's Rule of Five (Ro5) compliance for the docking molecules listed in Table-2. Lipinski's Ro5 serves as a guideline to evaluate the drug-like properties of molecules. According to this rule, compounds with characteristics such as a molecular weight exceeding 500, a logP value greater than 5, more than five hydrogen bond (HB) donor groups (sum of OHs and NHs groups), more than ten hydrogen bond (HB) acceptor groups (sum of O and N atoms), and molar refractivity beyond the range of 40 to 130 may have difficulties in intestinal absorption. Violations of these criteria, referred to as Ro5 violations, do not necessarily preclude a compound from being a successful drug, but they raise potential concerns.

B. Molecular Docking:

To assess the binding affinity between potential inhibitors and the receptor, we conducted molecular docking experiments. The docking results, illustrated in Figure 1, highlight the interaction between Mpro and the nine phytochemicals from Cissus quadrangularis, alongside the control drug, remdesivir. Notably, certain compounds from C. quadrangularis, including beta-Sitosterol, pallidol, quercetin, picroside 1, and quadrangularin A, exhibited binding poses that were equal to or superior to remdesivir in terms of their affinity for Mpro. Additionally, several compounds demonstrated improved positioning compared to chloroquine in relation to Mpro.

C. Visualization of the Docking Results:

Our analysis involved the use of the native ligand to scrutinize and compare the binding site on Mpro. The findings indicated that five potential compounds exhibited diverse affinities for Mpro, despite all of them binding to the same site. These findings suggest that these compounds have the potential to inhibit Mpro activity. The interactions between ligands and receptors were primarily stabilized through hydrophobic and hydrogen bond interactions, with physiochemical properties data supporting these molecular interactions.

D. Identification of Top Five Molecules:

Based on the results of our molecular docking analysis, we identified the top five molecules with promising binding energies: beta-Sitosterol, pallidol, quercetin, quadrangularin A, and picroside 1. These selected molecules exhibited binding energies of -7.5, -7.9, -7.8, -7.9, and -8.5 kcal/mol, respectively, while the control complex had a binding energy of -7.8 kcal/mol (Table-3).

Each of these top compounds formed unique interactions with Mpro. For example, beta-Sitosterol formed a conventional hydrogen bond at APG 131 and several hydrophobic interactions at MET276, LEU 287, LEU 286, and TYR 237/TYR 239. Pallidol formed a conventional hydrogen bond at THR26, a pi-sulfur hydrophobic bond at CYS 145, a pi-pi T-shaped interaction at HIS41, and an amide-pi stacked bond at LEU 141 and ASN 142. Quercetin engaged in five conventional hydrogen bonds at CYS145, SER144, HIS163, LEU141, and ARG188, as well as hydrophobic interactions. Quadrangularin A formed two conventional hydrogen bonds at THR26 and HIS163, along with pi-alkyl and pi-sulfur bonds. Picroside I interacted with Mpro through three conventional hydrogen bonds, a carbon hydrogen bond, alkyl bonds, and a pi-alkyl bond (Table-4/Figure-2).

E. ADME/T Evaluation:

Efficiency and safety assessments of lead compounds were conducted by evaluating various ADME/T (Absorption, Distribution, Metabolism, Excretion, and Toxicity) parameters. These included assessments of carcinogenicity, CNS permeability, p-glycoprotein inhibition, hepatotoxicity, CYP inhibition, human intestinal absorption, human ether-a-go-go-related gene inhibition, acute oral toxicity, and rat acute toxicity. Notably, all components under investigation exhibited no toxic or carcinogenic characteristics, reinforcing their potential for pharmaceutical use (Table-5).

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F. Biological Activities of Drug Candidates:

Our study also delved into the potential biological activities of the investigated compounds, encompassing ion channel inhibition, protease inhibition, kinase inhibition, enzyme inhibition, G protein-coupled receptor (GPCR) ligand activity, and nuclear receptor ligand activity. Among these activities, picroside-I and quadrangularin A demonstrated the highest GPCR ligand activity, while quercetin exhibited the lowest activity. Beta-Sitosterol displayed superior ligand activity compared to pallidol. Picroside-I exhibited remarkable enzyme inhibitor activity, surpassing beta-Sitosterol. Furthermore, all potential compounds displayed nuclear receptor ligand activity, with beta-Sitosterol showing the highest affinity. Quercetin demonstrated kinase inhibitor activity compared to other screened compounds (Table-6).

Table 2 Lipinski's rule of five (Ro5) of SARS-CoV-2 Mpro Potential Inhibitors.

Compound	Molecular weight (<500 Da)	LogP (<5)	H-Bond donor (5)	H-bond acceptor (<10)	Molar refractivity (40-130)	Violations	Meet RO5 criteria
beta-Sitosterol	414.71	5.05	1	1	133.23	1	Yes
pallidol	454.47	2.22	6	6	127.52	0	Yes
quercitin	302.24	1.63	5	7	78.03	0	Yes
quercitrin	448.38	1.60	7	11	109.00	1	Yes
beta-sitosterol glycoside	576.85	5.17	4	6	165.61	2	NO
picroside I	492.47	2.91	5	11	115.99	1	Yes
quadrangularin A	454.47	2.19	5	6	130.58	0	Yes
6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol	552.52	-0.29	5	13	128.97	2	NO
6-O-meta-methoxybenzoyl catalpol	496.46	-0.79	5	12	112.77	1	Yes

Table 3 Molecular Docking Analysis of Several Plant Compounds Against Mpr

Name of the compound	Binding Energies (ΔG = kcal/mol)
	Mpro
beta-Sitosterol	-7.5
pallidol	-7.9
quercitin	-7.8
quercitrin	-7.2
beta-sitosterol glycoside	-7.2
picroside 1	-8.5
quadrangularin A	-7.9
6-O-[2,3-dimethoxy]- trans-cinnamoyl catalpol	- 7.2
6-O-meta-methoxy-benzoyl catalpol	-6.8
Control drug (remdesivir)	-7.8

Table 4 Non Bond Interactions between Mpro (6LU7) and Phytochemicals

Compound	Hydrophobic bond			Hydrogen bond		
	Bonding type	Interacting amino acids	Distance (Å)	Bonding type	Interacting amino acids	Distance (Å)
beta-Sitosterol	Alkyl	MET276	4.54	Conventional	ARG131	2.76
	Alkyl	LEU287	4.21			
	Alkyl	LEU286	5.08			
	Pi-Alkyl	TYR237	5.25			
	Pi-Alkyl	TYR239	5.39			
pallidol	Pi-Sulfur	CYS145	5.92	Conventional	THR26	2.29
	Pi-Pi T-shaped	HIS41	5.14			
	Amide-Pi Stacked	LEU141,ASN142	4.82			
quercitin	Pi-Alkyl	MET165	5.39	Conventional	CYS145	2.49
	Pi-Alkyl	CYS145	4.96		SER144	2.42
					HIS163	2.10
					LEU141	2.11
					ARG188	2.43
				Pi-Donor	GLU166	3.19
quercitrin	Pi-Pi T-shaped	TYR237	5.29	Conventional	THR199	2.30
	Pi-Alkyl	LEU272	5.41		LEU287	1.78

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					ASP197	2.82
beta-sitosterol glycoside	Alkyl	VAL202	5.27	Carbon	HIS246	3.62
		ILE249	4.16			
		PRO293	4.49			
		VAL297	3.82			
	Pi-Alkyl	PHE294	4.86			
picroside I	Alkyl	LEU27		Conventional	GLY143	2.25
	Alkyl	CYS145	5.05		SER144	2.20
	Pi-Alkyl	PRO168	5.33		GLU166	2.26
				Carbon	ASN142	3.47
quadrangularin A	Pi-Sulfur	CYS145	4.96	Conventional	THR26	2.82
	Pi-Pi T-shaped	HIS41	4.70		HIS163	2.97
	Alkyl	MET49	5.15			
	Pi-Alkyl	HIS41	4.91			
	Pi-Alkyl	MET49	5.06			
6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol	-	-	-	Conventional	CYS145	3.08
	-	-	-		PHE140	2.35
6-O-meta-methoxybenzoyl catalpol	Pi-Alkyl	PHE294	4.33	Conventional	GLN110	2.29
					ASN151	2.81
					SER158	2.11
				Carbon	ASP153	3.31
Control drug	Pi-sulfur	MET165	5.72	Conventional	GLY143	2.34
	Pi-Pi Stacked	HIS41	4.13		ARG188	2.19
	Alkyl	MET165	4.70		THR190	2.03
	Pi-Alkyl	MET49	4.61	Carbon	THR26	3.52

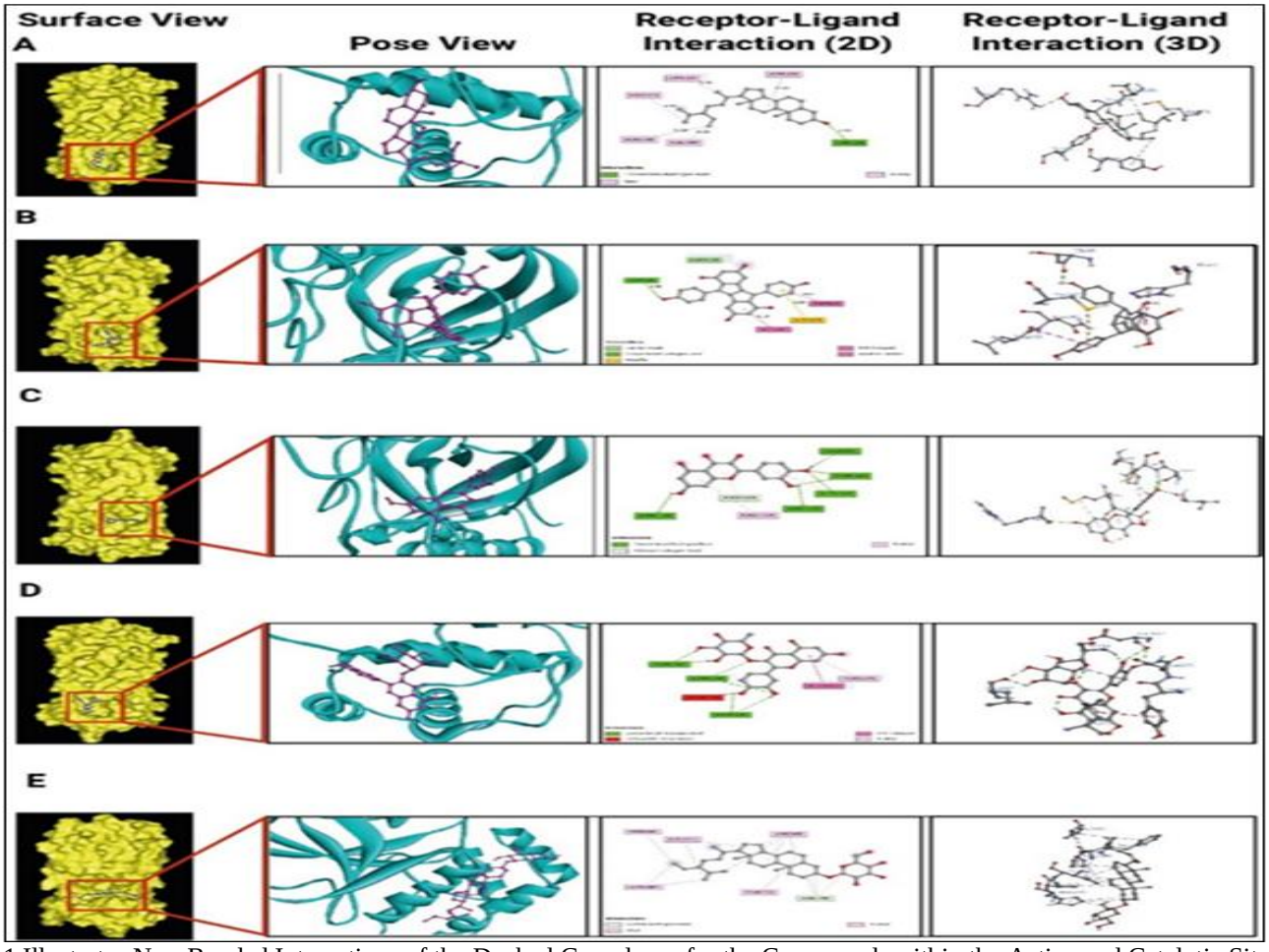

Fig 1 Illustrates Non-Bonded Interactions of the Docked Complexes for the Compounds within the Active and Catalytic Sites of the Main Protease. (A) beta-Sitosterol. (B) pallidol (C) quercitin (D) quercitrin (E) beta-sitosterol glycoside

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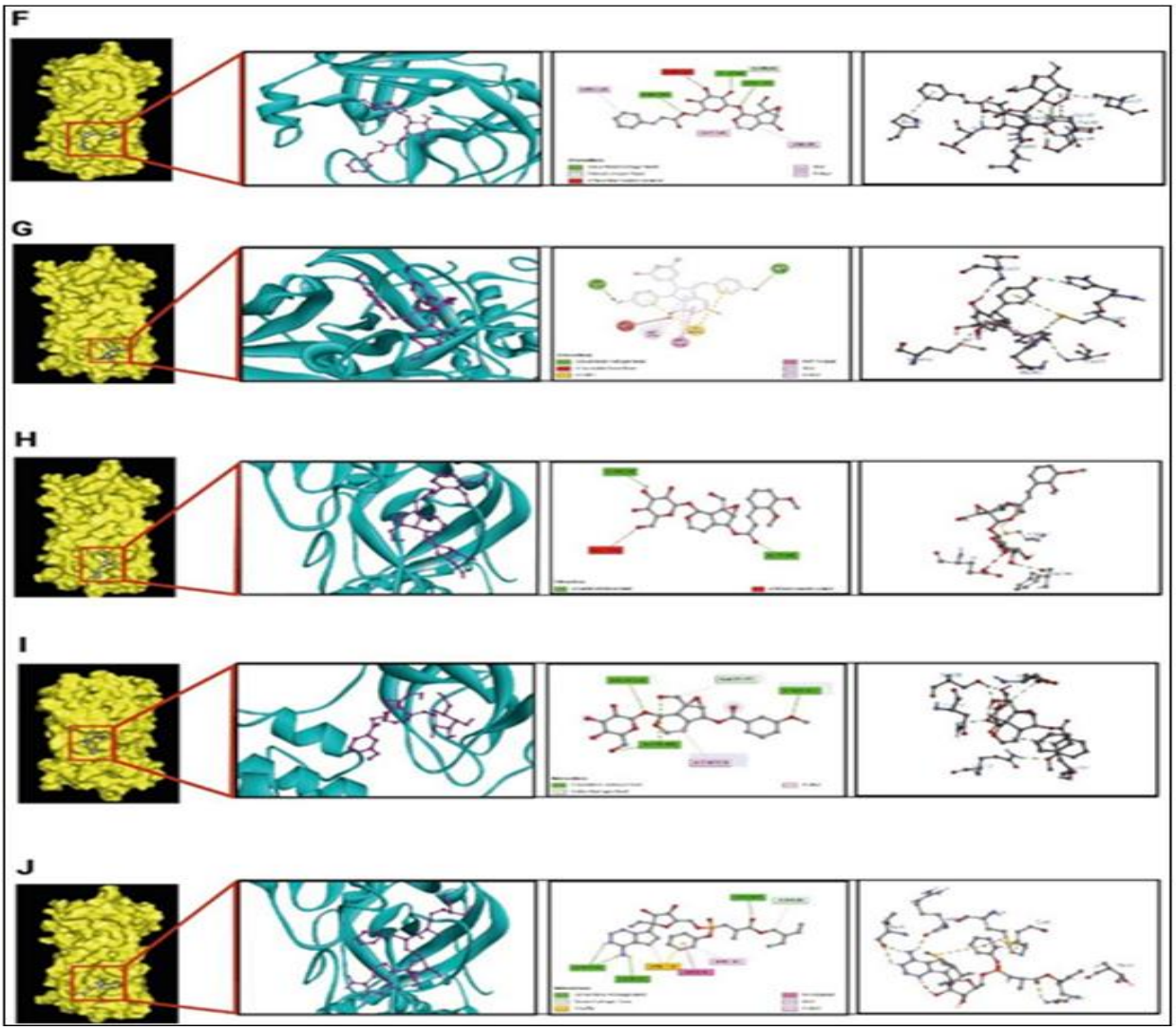

Fig 2 Illustrates Non-Bonded Interactions of the Docked Complexes for the Compounds within the Active and Catalytic Sites of the Main Protease. (F) picroside 1 (G) quadrangularin A (H) 6-O-[2,3-dimethoxy]- trans-cinnamoyl catalpol (I) 6-O-meta-methoxy-benzoyl catalpol (J) Control drug (remdisevir).

Table 5 Pharmacological Data of All Compounds Obtained from the SwissADME, admetSAR, and pKCSM Webservers. (NC = Non-Carcinogens)

Parameter	beta-Sitosterol	pallidol	quercitin	quercitrin	beta-sitosterol glycoside	picroside 1	quadrangularin A	6-O-[2,3-dimethoxy]-trans-cinnamoyl catalpol	6-O-meta-methoxybenzoyl catalpol
Blood brain barrier	0.781	-1.003	-2.925	-1.495	-0.785	-0.94	-1.041	-1.826	-1.63
CNS	-1.705	-3.053	-3.065	-4.156	-3.021	-4.109	-2.947	-3.833	-3.803
Lipophilicity	9.34	4.57	1.54	0.16	5.51	-0.48	3.86	-0.29	-0.64
Solubility	-6.773	-2.893	-2.925	-2.903	-4.741	-1.79	-2.894	-3.806	-3.659
Instauration	0.93	0.14	00	0.29	0.94	0.54	0.07	0.58	0.61
Flexibility	6	2	1	3	9	8	3	10	8
Intestinal absorption (human)	94.464	93.07	77.207	52.709	-4.741	66.862	89.078	57.364	57.938
P-glycoprotein inhibitor	Yes	Yes	No	No	Yes	Yes	Yes	Yes	Yes
VDss (human)	0.193	-1.46	1.559	1.517	-1.163	-0.086	-1.514	-0.13	-0.204
CYP2C9 Inhibitor	No	No	No	No	No	No	No	No	No
CYP2D6 substrate	No	No	No	No	No	No	No	No	No
Human ether-a-go–go-related (hERG) gene inhibition	No	No	No	No	No	No	No	No	No
Oral Rat acute toxicity, LD50 (mol/kg)	2.552	2.509	2.471	2.586	2.571	3.535	2.516	3.758	3.964
AMES toxicity	No	No	No	No	No	No	No	No	No
Hepatotoxicity	No	No	No	No	No	No	No	No	No
Carcinogens	NC (0.918)	NC (0.874)	NC (0.945)	NC (0.946)	NC (0.958)	NC (0.956)	NC (0.889)	NC (0.957)	NC (0.957)

Table 6 Biological Activity Prediction of Phytochemicals of *Cissus quadrangularis*.

Name of the compounds	GPCR ligand	Ion channel modulator	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor	Enzyme inhibitor
beta-Sitosterol	0.14	0.04	-0.51	0.73	0.07	0.51
pallidol	0.06	0.05	-0.03	0.2	0.03	0.05
quercitin	-0.06	-0.19	0.28	0.36	-0.25	0.28
quercitrin	-0.01	-0.08	0.08	0.17	-0.06	0.37
beta-sitosterol glycoside	0.15	-0.21	-0.47	0.33	0.11	0.41
picroside 1	0.21	0.06	-0.04	0.28	0.3	0.46
quadrangularin A	0.05	0.06	-0.03	0.29	-0.08	0.1
6-O-[2,3-dimethoxy]- trans-cinnamoyl catalpol	0.14	-0.17	-0.12	0.15	0.18	0.31
6-O-meta- methoxybenzoyl catalpol	0.13	0.02	-0.09	0.16	0.20	0.36

IV. CONCLUSION

In conclusion, our study employed a structure-based drug discovery approach to systematically screen phytochemicals for their potential as potent inhibitors of SARS-CoV-2. Utilizing molecular docking, we identified and analyzed nine phytochemical compounds sourced from *Cissus quadrangularis*. Among these compounds, four exhibited compelling characteristics, including the formation of multiple non-covalent interactions within the active site of SARS-CoV-2 Mpro.

Furthermore, our assessments of toxicity and carcinogenicity indicated that these compounds possess favorable drug-like properties, crucial for ensuring drug safety. However, it is imperative to note that our research was conducted solely through computational algorithms, and further experimental validation in wet lab conditions is essential.

Despite this, our computational techniques hold promise in guiding researchers toward the identification of specific molecules that have the potential to act as inhibitors of SARS-CoV-2 Mpro. These findings provide a valuable starting point for further investigations and drug development efforts in the fight against COVID-19.

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