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Original Research Article

In silico Evaluation of the Inhibitory Potential of Cymbopogonol from *Cymbopogon citratus* Towards Falcipain-2 (FP2) Cysteine Protease of *Plasmodium falciparum*

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ABSTRACT

Antimicrobial resistance is a major challenge militating against the health of people globally. *Plasmodium falciparum* has developed resistance to current drugs used in tackling malaria, and this has remarkably contributed to an increased mortality rate in Sub-Saharan Africa. The inhibitory potential of cymbopogonol against Falcipain-2 (FP2) of the *Plasmodium falciparum* parasite was evaluated and achieved using a computational approach in this study. SwissADME, ADMETLab, and PROTOX-II servers were used to evaluate cymbopogonol's absorption, distribution, metabolism, excretion, and toxicity (ADMET) properties in comparison to the other ligands. The test compound had a better docking score of – 8.40 kcal/mol compared to the standard and co-crystallized ligand. The compound also had a hydrophobic interaction with LEU I:78, MET I:29, VAL I:44, VAL I:47, LEU I:25, and ARG I:43 present in the FP2 receptor-binding motif of the malaria parasite. The compound also possesses a favorable ADMET characteristics and demonstrated no tendency towards hERG inhibition, hepatotoxicity, carcinogenicity, mutagenicity, or drug-liver injury. Therefore, cymbopogonol may be used for experimental research and future medication development for the successful treatment of malaria.

***Keywords*:** Malaria, Cymbopogonol, Falcipain-2 (FP2), Molecular docking, Drug discovery.

Introduction

Human survival is linked to the response of science to disease outbreaks as typified by the recent COVID-19 pandemic which engendered an increased mortality rate globally.[1] In Sub-Saharan Africa, there is an increased rate of mortality and morbidity resulting from the prevalence of malaria.[2, 47] Malaria which is an infectious disease has not only engendered deaths but has also resulted in economic hardship in Sub-Saharan Africa.[3,4,5] Malaria has been reported to affect a large population of children under 5 years of age, and this accounts for close to 70% of malaria deaths globally.[6] Discovered in 1880, to date various efforts towards combating the parasite seem to be rather ineffective due to the growing resistance to standard drugs.[7, 8] Plasmodium is the genus of the parasitic protozoans of the sporozoan subclass Coccidiasina that causes malaria. And its species are *P. falciparum P. vivax, P. ovale, P. knowlesi, and P. malariae*.[9] The *P. falciparum* species being the most life-threatening species for humans utilize its cysteine protease, falcipain-2 (FP2) and FP3, in hemoglobin hydrolysis and the phase completion of the parasite's development in man.[9, 10] It has been reported that the knock-out of FP2 blocks hemoglobin hydrolysis.[11] Thus, the inhibition of the hydrolysis of hemoglobin leads to the death of the *P. falciparum*, most especially at its trophozoite phase of development.[12]

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This, therefore, makes FP2 an important target for the development of new antimalarial drugs resulting from its essentiality to the survival of the parasite. [51] Previous works by different research groups have identified hits against FP2.[13,46,49, 50] A molecular docking study was conducted and a similar binding pose of 3-(1-benzoyl-5-(4-fluorophenyl)-4,5-dihydro-1H-pyrazol-3yl)-7-(diethylamino)-2H-chromen-2-one and epoxysuccinate in FP2 inhibition was observed.[13] Also, an *in vitro* validation study on the antiplasmodial activity of an acyl-hydrazone-based molecular hybrid against cysteine protease FP2 *in silico* was conducted.[14] The advent of bio-computational tools has replaced the employment of traditional methods in the development and discovery of drugs.[15] This is because using the bioinformatics technique is viable for designing and producing new medications of biomedical interest and takes less time and effort.[1] Also, compared to the traditional method, an advantage of the bio-computational approach is its ability to predict the binding affinity, ligand-protein interaction, and ADMET properties of the ligand of interest.[1] In traditional medicine, *Cymbopogon citratus* which belonging to the family Poaceae, has been reported to have bioactivity against inflammation, pain, protozoa infections, and malaria.[16,17,18] *In vivo* studies have shown antimalarial activity of *Cymbopogon citratus* in mice.[16,19] Also, Gas chromatography-mass spectrometry (GC-MS) analysis has revealed the presence of cymbopogonol in lemongrass.[20] In this study, the activity of the phytochemicals present in lemongrass was explored against FP2, and cymbopogonol was found to have the best inhibitory potential towards the target. Also, triterpenoids have been reported to play important pharmacological activities like anti-inflammatory, analgesic, antipyretic, and hepatoprotective.[21] They are also reported to have antioxidant, antimicrobial, antiviral, antiallergic, antiangiogenic, and spasmolytic activities.[22] These classes of compounds have also been reported to have antimalarial activity. For example, synthetic oleanane triterpenoids were observed to improve survival in experimental cerebral malaria.[23] Interestingly, Iridal, a triterpenoid extracted from *Iris germanica* L., was reported to have antiplasmodial activity against

Plasmodium falciparum chloroquine-resistant and -sensitive strains.[24] Likewise, karavote B amongst other triterpenoids from *Momordica balsamina* was identified to have the highest activity against liver stages of *Plasmodium berghei.*[25] Cymbopogonol is a triterpenoid whose biological activity is yet to be determined. This study will be the first in the open literature to report the *in silico* bioactivity of cymbopogonol. Owing to the urgency for further therapeutic interventions against malaria, this study employed a computational approach to evaluate the therapeutic potential of the compound against malaria.

Materials and Methods

Ligand preparation

The test ligand, cymbopogonol (Fig. 1), and standard ligands—sulfadoxine, pyrimethamine, lumefantrine, artemether, artemether-lumefantrine were obtained from the PubChem (https://pubchem.ncbi.nlm.nih.gov) with the respective IDs: 1317[52]084, 3000518, 4993, 6437380, 68911 and 6450800.[26] Two co-crystallized ligands, PDB:GOL and PDB:E64, were extracted from the active site of the FP2 receptor. The compound and standard ligand were uploaded to PyRx software in MOL SDF format, and the OpenBabel plugin was used to convert it to PDBQT format. To get the lowest energy for the ligand docking, the output files were minimized at a force field called uff.

Preparation of the protein target

The plasmodium cysteine falcipain-2 (FP2) was the protein target for this study. The RCSB Protein Data Bank (PDB) repository (www.rcsb.org) was used to find the crystal structure of FP2. The X-ray diffraction method was used to derive the FP2 in complex with the inhibitor E64 (PDB: 3BPF) and PDB:GOL, with a resolution of 2.9, R-Value free 0.325, R-Value work 0.2775, and R-Value observed 0.278.[27] The non-standard residues, such as ions, water, and bound ligands (PDB:GOL and PDB:E64), which were retrieved from the active binding pocket by specified methods, were deleted from the structure's PDB format before it was uploaded to the PyMol display tool workspace. For a molecular docking investigation, the produced protein (Figure 2) was subsequently uploaded to the PyRx software.

Molecular docking

Using the PyRx workspace tool and AutoDock Vina (Scripps Research, La Jolla, CA, USA), molecular docking analysis was carried out after the target protein and ligand were prepared.

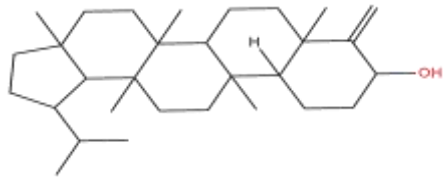

Figure 1: Structure of cymbopogonol

Figure 2: Structure of falcipain-2

The ligands' energy was reduced to the absolute minimum before being transformed to PDBQT. To precisely identify the binding site of the target receptor, the ligands and the receptor were chosen for docking analysis at the resolution of the grid box, which was taken along the x, y, and z axes, respectively, at a maximum dimension of 55.7097 63.9421 42.8786. The standards were initially docked against the FP2 receptor, and the interactions that resulted were compared with those of cymbopogonol. Additionally, the complex created by the ligands and receptor docking position was visualized using the BIOVIA Discovery Studio21 (Dassault Systèmes, San Diego, CA, USA) to examine the interactions and bonding between the receptor and the ligands.

Docking protocol validation

By redocking the standard ligands into the catalytic domain or binding site of the proteins utilized for the study using the PyRx tool, the docking pose generated from the PyRx docking tool was validated.[1]

ADMET predictions

Model predictions on the SwissADME, ADMETLab server, and PROTOX II, respectively, were used to estimate the lead compounds' absorption, distribution, metabolism, excretion, and toxicity (ADMET) properties.[28, 29]

Results and Discussion

Test compound's binding affinity with FP2 drug targets

The compounds' binding affinities (ΔG kcal/mol) ranged from -8.4 to -4.3 for FP2. The standard ligand with the highest binding affinity (-7.5 kcal/mol) to the FP2 pharmacological target is artemether-lumefantrine. Also, of the co-crystallized ligands, E64, exhibits the highest binding energy for the FP2 drug target (-6.5 kcal/mol). The test compound, cymbopogonol gave a binding affinity higher than all the standard inhibitors and the co-crystallized ligands (Table 1).

Cymbopogonol exhibits the highest affinity for binding. Cymbopogonol's interaction towards the *P. falciparum* target protein, FP2, in this study is an indicator of the compound's inhibitory potential against the FP2 biomolecule and its prospective use as a malaria treatment agent (Table 1). Due to structural similarities with other triterpenoids of pharmacological significance (such as iridal, ursolic acid, balsaminol A, balsaminol B, balsaminoside A, cucurbalsaminol C, karavilagenin C, karavoate A, karavoate B, karavoate C, karavoate D, karavoate E, karavo), cymbopogonol can alter how the malaria-causing protein drug target performs.[24, 30, 31] According to this work, cymbopogonol has a higher affinity for binding to the FP2 target proteins than the standard and co-crystallized ligands. This could be linked to the hydrophobic interactions of the potential inhibitor's skeleton as visualized from the two-dimensional molecular interaction, supporting its binding of triterpene to FP2 (Fig. 3). Because 55% of amino acids occur as non-polar, interactions involving non-polar groups are critical for biological recognition, including protein-ligand binding.[1] Hydrophobic interaction is known to be a structural parameter that determined the binding affinity during drug design.[32, 48] When compared to the standard ligand, the triterpenoid molecule-cymbopogonol, possesses the best molecular docking score. This possible inhibitor showed a binding behavior resembling that of the co-crystallized ligand, E-64. Recently, Scientific Reports published that E-64 is a potential drug and inhibitor in the treatment of malaria as it binds tightly to FP2 and blocks access to the catalytic residue of FP2.[12] In this molecular docking study, the molecular interaction of the triterpenoid is compared to that of E64. The binding affinity results demonstrate that cymbopogonol presented favorable binding with FP2 receptors compared to that of E64 (Table 1).

Analysis of selected hit compounds' molecular docking

Figures 3 and 4 show the two-dimensional (2D) and three-dimensional (3D) interactions of FP2 drug target in complex with cymbopogonol and E-64 respectively. Table 2 highlights the hydrophobic interaction of the FP2- cymbopogonol complex. Amino acids LEU I:78, MET I:29, VAL I:44, VAL I:47, LEU I:25, and ARG I:43 interacted with the test compound (Figures 3-4, Table 2). The amino acids that interact with FP2 and E-64 are VAL I:44 and LEU I:78.

Table 1: Test compounds' binding affinities (ΔG in kcal/mol) against FP2 drug targets

Ligand	Binding Affinity(ΔG Energy, Kcal/mol)	RMSD/ ub	RMSD/ lb
Test Ligand			
Cymbopogobol	-8.4	0	0
Standard ligands			
Sulfadoxine	-6.9	0	0
Pyrimethamine	-7.1	0	0
Lumefantrine	-6.4	0	0
Artemether	-7.1	0	0
Artemether-Lumefantrine	-7.5	0	0
Co-crystallized ligands			
E64	-6.5	0	0
Glycerol	-4.3	0	0

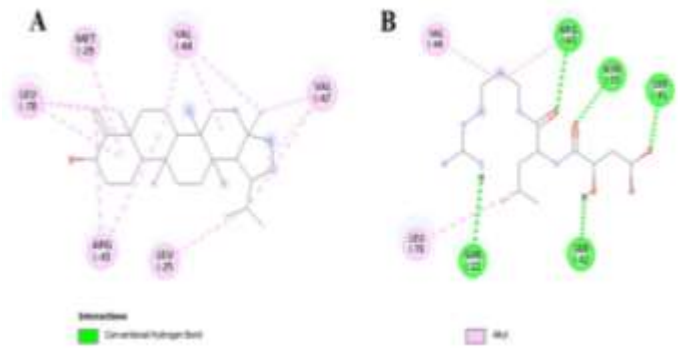

Figure 3: The 2D representation of the molecular interaction between amino-acid residues of FP2 with (a) cymbopogonol (b) E-64.

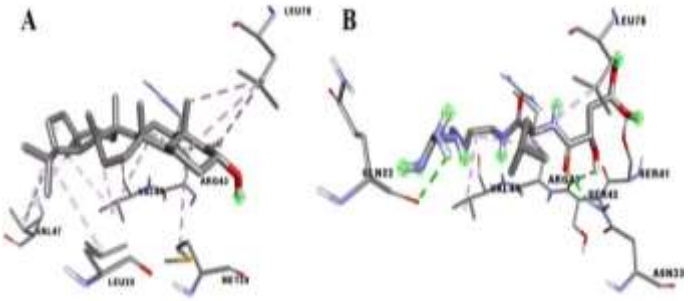

Figure 4: The 3D representation of the molecular interaction between amino-acid residues of FP2 with (a) cymbopogonol (b) E-64.

Due to the electrostatic attraction of the molecules in these two compounds, the interactions between FP2 and E-64 involved the amino acids ARG I:43, ASN I:33, SER I:41, SER I:42, and GLN I:22 through hydrogen bonding (Table 3). FP2 target and the test compound, cymbopogonol, was docked and analysis of the target's 3D and 2D structures revealed interactions between the compounds. Similar interactions with the protein were observed for cymbopogonol and E64 via hydrophobic (alkyl-alkyl) interaction. The potential inhibitor had other hydrophobic interactions with the amino acids LEU I:78, MET I:29, VAL I:44, VAL I:47, LEU I:25, and ARG I:43 on the FP2 receptor. The structural characteristic of the hydrophobic interaction, which is measured by the amount of hydrophobic surface buried when ligands bind, best corresponds with binding free energy.[32] The co-crystallized ligand, E64, interacted with amino acids ARG I:43, ASN I:33, SER I:41, SER I:42, and GLN I:22 on the receptor of FP2. This is due to hydrogen bonding between the polar moieties of the compound and the amino acids. It is crucial to note that the interaction of the putative inhibitor with the amino acids in the binding pocket may prevent FP2 from acting as a catalyzer, thereby preventing the spread of malaria. The hydrophobicity of this compound compared to E64 makes it a better potential inhibitor of FP2 as evident by their relative binding affinities. This lemon grass-derived triterpenoid has not been worked on scientifically before now, and there is no recorded bioactivity of cymbopogonol in the open literature. Therefore, these results suggest that cymbopogonol will potentially interfere with the associated Falcipain-2 (FP2). Further investigations to validate these findings and understand the mechanism of the compound against malaria disease *in vivo* are recommended. We recommend that the test compound is considered for optimization and future studies *in vivo* studies and possible clinical trials.

Molecular docking protocol validation

In Fig. 5, the validation of the docking methodology is demonstrated by the re-docking of cymbopogonol and E-64 into the catalytic domain. The outcome demonstrates that there was a considerable overlap when E-64 and cymbopogonol were re-docked against FP2 (Fig. 5a and 5b). The perfect overlap of the re-docked pose with the experimental orientation shows that Autodock vina on PyRx accurately re-docked the co-crystallized ligand, then the test ligand back into the binding pocket of FP2. This demonstrates the validity of the docking methodology employed in this study and the accuracy of the docking ratings. Additionally, a study reporting a similar docking approach showed that by re-docking the co-crystallized ligand (PDB Ligand ID: 2WR) with the researched mutant EGFR (PDB: 3W2S), there was a considerable overlap.[33] It was clear from the analysis that the re-docked co-crystallized ligand had almost perfect overlap.

ADMET Profile

Table 4 lists the compounds' expected scores for drug-likeness, water-solubility, bioavailability, and lipophilicity according to SwissADME. The molecular weight of cymbopogonol from *Cymbopogon citratus* is 426.72 g/mol which is higher than the molecular weights of all other ligands except Lumefantrine and Artemether-Lumefantrine. Log S value of cymbopogonol is -6.96 (poorly soluble) as compared to other ligands which range from 1.08 (glycerol - highly soluble) to -11.79 (lumefantrine - poorly soluble). Also, the Log P value of the compound is 7.20 (poorly soluble) as compared to the standard ligands and E64 which range from -1.09 to 7.91. The bioavailability score of 0.55 and one Lipinski violation for cymbopogonol point to its potential for use as an oral medication. Also, cymbopogonol has MWt ≤500 (426.72 g/mol) although it has a consensus Log P value of 7.20 (Table 4). Furthermore, the MR of cymbopogonol is 135.14. Also, the compound obeyed the Veber rule and had a similar bioavailability score of 0.55. Additionally, it generated ratings for synthetic accessibility (SA) of 5.52. The compound showed a TPSA score of 20.23. The GI absorption potential is low for the test ligand compared to other ligands. cymbopogonol showed no BBB permeability with a skin permeation value (Log Kp) of – 2.28 cm/s (Table 5). Since the putative inhibitor is not a Pgp substrate, it can be prevented from accessing its site of action even though it is predicted to have a low GI absorption value (Table 5). The pharmacokinetics prediction and cytochrome P450 inhibitory potential of cymbopogobol are further displayed in Table 5. In comparison to the other compounds, cymbopogonol is not a potent inhibitor of CYP1A2, CYP2C19, CYP2C9, and CYP3A4 (Table 5). In addition, the inhibitor did not exhibit any potential to inhibit hERG or to be carcinogenic, or hepatotoxic (Table 6).

Table 2: Table of chemical interactions of the FP2 binding pocket with cymbopogonol

Name	Parent	XYZ:X	XYZ:Y	XYZ:Z	Category	Types	From Chemistry	To Chemistry
I:MET29 - I:ARG43	Ligand Non-bond Monitor	103.69	-21.8794	-113.015	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1		103.15	-19.0089	-116.01	Hydrophobic	Alkyl	Alkyl	Alkyl
I:VAL44 - N:UNK1	Ligand Non-bond Monitor	100.564	-20.6483	-115.313	Hydrophobic	Alkyl	Alkyl	Alkyl
I:VAL44 - N:UNK1	Ligand Non-bond Monitor	98.7027	-20.7281	-115.814	Hydrophobic	Alkyl	Alkyl	Alkyl
I:VAL47 - N:UNK1	Ligand Non-bond Monitor	94.508	-20.6368	-114.707	Hydrophobic	Alkyl	Alkyl	Alkyl
I:LEU78 - N:UNK1	Ligand Non-bond Monitor	105.682	-17.9094	-111.36	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:ARG43	Ligand Non-bond Monitor	105.259	-19.2497	-115.82	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:LEU78	Ligand Non-bond Monitor	106.867	-17.7452	-112.215	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:LEU78	Ligand Non-bond Monitor	105.778	-16.7933	-111.757	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:VAL44	Ligand Non-bond Monitor	97.5705	-21.2037	-116.507	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:VAL47	Ligand Non-bond Monitor	94.6408	-21.32	-115.83	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:LEU25	Ligand Non-bond Monitor	97.2733	-21.6535	-111.76	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:VAL47	Ligand Non-bond Monitor	94.5613	-21.3735	-113.647	Hydrophobic	Alkyl	Alkyl	Alkyl

Table 3: Table of interactions displaying how the co-crystallized ligand, E-64, interacts chemically with the FP2 binding pocket

Name	Parent	XYZ:X	XYZ:Y	XYZ:Z	Category	Types	From Chemistry	To Chemistry
I:ASN33:HD21 - N:UNK1:O	Ligand Non-bond Monitor	106.864	-21.124	-113.816	Hydrogen Bond	Conventional Hydrogen Bond	H-Donor	H-Acceptor
I:SER41:HG - N:UNK1:O	Ligand Non-bond Monitor	110.419	-17.98	-115.603	Hydrogen Bond	Conventional Hydrogen Bond	H-Donor	H-Acceptor
I:ARG43:HE - N:UNK1:O	Ligand Non-bond Monitor	102.898	-16.645	-116.335	Hydrogen Bond	Conventional Hydrogen Bond	H-Donor	H-Acceptor
N:UNK1:H - I:SER42:O	Ligand Non-bond Monitor	107.466	-20.9235	-115.572	Hydrogen Bond	Conventional Hydrogen Bond	H-Donor	H-Acceptor
N:UNK1:H - I:GLN22:O	Ligand Non-bond Monitor	99.069	-20.036	-108.255	Hydrogen Bond	Conventional Hydrogen Bond	H-Donor	H-Acceptor
I:ARG43 - N:UNK1	Ligand Non-bond Monitor	102.292	-19.1092	-116.492	Hydrophobic	Alkyl	Alkyl	Alkyl
I:VAL44 - N:UNK1	Ligand Non-bond Monitor	99.7063	-20.7487	-115.795	Hydrophobic	Alkyl	Alkyl	Alkyl
N:UNK1:C - I:LEU78	Ligand Non-bond Monitor	105.494	-17.7497	-110.805	Hydrophobic	Alkyl	Alkyl	Alkyl

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Table 4: Predicted Lipophilicity (Log P), Water Solubility (Log Sw), Druglikeness, and Bioactivity of test compounds.

Parameters	Cymbopogonol	Sulfadoxine	Pyrimethamine	Lumefantrine	Artemether	Artemether-Lumefantrine	E-64	Glycerol
Molecular weight (g/mol)	426.72	310.33	248.71	528.94	298.37	827.31	357.41	92.09
Consensus Log P	7.20	0.69	2.29	7.91	2.70	9.81	-0.58	-1.09
Log Sw (Silicos-IT)	-6.96	-4.03	-4.87	-11.79	-4.18	-11.79	-1.31	1.08
Solubility class	Poorly soluble	Soluble	Soluble	Poorly soluble	Moderately soluble	Insoluble	Soluble	Highly soluble
#Heavy atoms	31	21	17	35	21	56	25	6
#Aromatic heavy atoms	0	12	12	18	0	18	0	0
Fraction Csp3	0.93	0.17	0.17	0.33	1.00	0.57	0.73	1.00
#Rotatable bonds	1	5	2	10	1	11	13	2
#H-bond acceptors	1	6	2	2	5	7	6	3
#H-bond donors	1	2	2	1	0	1	5	3
MR	135.14	76.53	71.06	152.61	76.07	228.68	90.06	20.02
TPSA (Å2)	20.23	124.81	77.82	23.47	46.15	69.62	172.43	60.69
Lipinski violations	1	0	0	2	0	2	0	0
Ghose violations	3	0	0	3	0	4	1	4
Veber violations	0	0	0	0	0	1	2	0
Egan violations	1	0	0	1	0	1	1	0
Muegge violations	2	0	0	1	0	3	1	2
Bioavailability Score	0.55	0.55	0.55	0.17	0.55	0.17	0.55	0.55
Synthetic availability	5.52	2.99	2.43	4.52	6.65	8.97	4.18	1.31

Table 5: Pharmacokinetics prediction output of test compounds.

Parameters/ID	Cymbopogonol	Sulfadoxine	Pyrimethamine	Lumefantrine	Artemether	Artemether-Lumefantrine	E-64	Glycerol
GI Absorption	Low	High	High	Low	High	Low	Low	High
Blood-brain permeant	No	No	Yes	No	Yes	No	No	No
Pgp substrate	No	No	No	Yes	No	Yes	Yes	No
CYP1A2 inhibitor	No	No	Yes	No	Yes	No	No	No
CYP2C19 inhibitor	No	No	Yes	No	No	No	No	No
CYP2C9 inhibitor	No	No	No	No	No	No	No	No
CYP2D6 inhibitor	No	No	No	Yes	No	No	No	No
CYP3A4 inhibitor	No	No	Yes	No	No	Yes	No	No
Skin permeant Log Kp (cm/s)	-2.28	-7.70	-5.91	-3.34	-5.61	-2.65	-9.12	-8.11

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Table 6: Toxicity profile prediction of test compounds

Target classification	Cymbopogonol	Sulfadoxine	Pyrimethamine	Lumefantrine	Artemether	Artemether-Lumefantrine	E-64	Glycerol
hERG (hERG Blockers)	-	---	---	+++	-		-	---
H-HT (Human Hepatotoxicity)	---	-	+++	---	+		+	---
AMES (Ames Mutagenicity)	---	---	---	---	-		---	---
LD50 (LD50 of acute toxicity)	3.393 -log mol/kg (172.644 mg/kg)	1.979 -log mol/kg (3257.097 mg/kg)	2.594 -log mol/kg (633.44 mg/kg)	3.117 -log mol/kg (404.032 mg/kg)	2.246 -log mol/kg (1693.434 mg/kg)	1000mg/kg	2.213 -log mol/kg (2188.608 mg/kg)	1.11 -log mol/kg (7148.77 mg/kg)
DILI (Drug Induced Liver Injury)	---	+++	++	---	-		---	---
FDAMDD (Maximum Recommended Daily Dose)	-	+++	+	-	+		++	+++
Toxicity Class	5	6	3	4	4	4	4	5

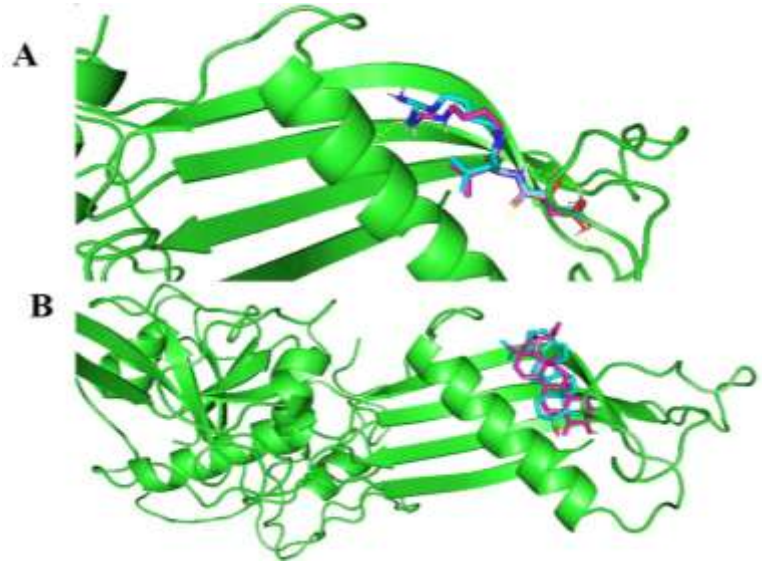

Figure 5: Validation of docking: comparability of the re-docked binding mode of (a) cymbopogonol in FP2 binding pocket (b) the co-crystallized pose of E-64 in FP2 binding pocket.

The compounds have moderate ADMET properties in addition to the inhibitory potentials against FP2 in cymbopogonol. To retain its binding affinity, the chemical might need lead optimization of its characteristics. Absorption, distribution, metabolism, elimination, and toxicity are all parts of the ADMET analysis. It is a test that determines if a molecule can be quickly and effectively absorbed, transported to its intended location of the action, digested without impairing activity, and excreted from the body without having hazardous effects. A high-quality drug candidate should have the necessary ADMET qualities at a therapeutic dose in addition to being effective against the therapeutic target.[34,45]. As a result, numerous in silico models for predicting chemical ADMET properties have been developed, which is helpful since it identifies medication failure due to pharmacokinetics before moving on to the clinical phase.[35] Water solubility is another physicochemical feature that affects a drug's ADMET activities, while lipophilicity is typically regarded as a crucial factor in determining permeability across tissue membranes.[35, 36] Orally administered medications typically have a high lipophilic value, indicating simple passage and absorption through the intestinal lining, penetration of the membrane of target cells, and blood flow. The log P value of a chemical has a direct association with its lipophilicity but an inverse relationship with its water solubility.[37] Consequently, cymbopogonol had an interesting Log P values of 7.20 compared to the co-crystallized ligands (Table 4). Cymbopogonol's poor solubility may have been caused by the atoms, a higher MR value, and molecular weights. However, more studies on the aspect of lead optimization may be done for better *in silico* outcomes.[38] Prodrugs are inactive compounds that are created by chemically altering physiologically active molecules.[39] Therefore, inactive or less active derivatives of the potential inhibitor can be created towards increasing its lipophilicity and water solubility. These derivatives will then go through a biochemical or enzymatic transformation in vivo to release the active moiety responsible for eliciting pharmacological effects. A qualitative evaluation of oral bioavailability, drug-likeness is established based on chemical structures and physicochemical properties.[40] A ligand will be deemed orally inert if it breaches two or more of Lipinski's requirements, including those requiring five H-bond donors, ten H-bond acceptors, a molecular weight of 500 g/mol, and a log P of 5.43 for an orally active medication.[41] In light of these requirements, cymbopogonol satisfies the prerequisites for oral bioavailability as evidenced by its Consensus (Table 4).

Additionally, the inhibitor met Veber's requirement, which requires the presence of rotatable bonds and a polar surface area (TPSA) of 140, while E64 violated it.[42]. Further evidence that the prospective inhibitor will be a successful oral medication comes from the bioavailability score of 0.55 (Table 4). In comparison to the standard and co-crystallized ligands, this finding demonstrates the drug-likeness of cymbopogonol. The putative inhibitor will not participate in drug-drug interactions, according to pharmacokinetic projections (Table 5). Phase I drug metabolism is facilitated by the isoenzyme superfamily cytochrome P450 (CYP), which catalyzes a number of metabolic reactions.[43] One of the main causes of pharmacokinetics-related drug-drug interactions is the suppression of the five major isoforms CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4, which eventually become the substrates of pharmaceuticals.[37, 44]. From our study, cymbopogonol exhibits interactions with any of the CYP 450 isoforms, and thus may not be involved in drug-drug interactions.
Comparing cymbopogonol to the co-crystallized ligand E64, it is expected that it is not a Pgp substrate. An ATP-binding cassette transporter called Pgp is in charge of actively effluxing xenobiotics through biological membranes as this defends the body against foreign toxins and increases medication resistance.[35] The findings demonstrate that, unlike E64, which may be stopped from accessing its target site through active efflux of the ATP-binding cassette, the putative inhibitor is predicted to reach its target site. The toxicity prediction results, however, indicated that it does not tend to any of the toxicity criteria examined.

Conclusion

Cymbopogonol was tested for potential inhibitory effect against FP2, a crucial target in the treatment of malaria. The ligand's bioactivity has not been investigated in the open literature. The substance had the highest binding affinity of - 8.40 kcal/mol when compared to the co-crystallized ligand of FP2 and currently available conventional medications for malaria. The FP2 receptor-binding motifs consisting of amino acid residues- LEU I:78, MET I:29, VAL I:44, VAL I:47, LEU I:25, and ARG I:43- interacted with the compound. This triterpenoid has a good ADMET profile and exhibited no potential to block hERG, hepatotoxicity, cancer, mutagenicity, or cause drug-liver damage. For this reason, cymbopogonol may be considered for further research and development into a novel medication for the treatment of malaria.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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