



RESEARCH ARTICLE

Hepatoprotective Efficacy of Phytochemicals in *Clerodendrum volubile* as Potential Natural Inhibitor on CYP2E1 Protein Implicated for Toxicity in Liver

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Abstract

This study delves into the effects of secondary metabolites in *Clerodendrum volubile* on CYP2E1, a protein target implicated in hepatotoxicity. Among implicated in cis,cis-linoleic acid, 3-hydroxydecanoic acid, n-pentadecanoic acid, palmitic acid, and 3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one demonstrate strong binding affinities and stability within the protein's active site compared to standard compounds with lower values. These compounds exhibited notable binding affinities against CYP2E1 with energies of -10.746, -10.203, -9.974, -9.177, and -8.447 kcal/mol, respectively. The inhibition of cytochromes P450 (CYPs) activities, responsible for procarcinogen activation, is implicated in the mechanism. This investigation focuses on the inhibitory potential of *C. volubile* compounds on CYP2E1 enzymatic activities to mitigate liver toxicity. Further analysis of the five hit compounds for drug-likeness and toxicity using SWISSADME reveals that all compounds possess desirable drug-like properties and are readily absorbable. The hit ligands effortlessly met the criteria molecular weight (MW) of ≤ 500 and violated none in Lipinski rule of 5 (Ro5).

Keywords

Clerodendrum volubile, Phytochemicals, CYP2E1, Hepatoprotection, Pharmacokinetics

Introduction

Hepatotoxicity is a significant health concern, and it is characterized by liver damage a life-threatening disease that causes hepatocyte death and inflammation

of the liver. There are several causes of this clinical complication some of which are due to exposure to viral infection, improper drug use, excessive alcohol intake, and sepsis. Liver failure, is a severe consequence of abrupt massive hepatocyte death, evolving over days or weeks to a lethal outcome, with a clinical mortality of 25-75%. The liver is one of the cogent organs in the body, It is a major public health concern because the liver plays a crucial role in metabolism, detoxification, and immune function coupled with its functions in protein, lipid, and glucose metabolism, and synthesizes most plasma protein. Different injuries to liver cells result in a consistent sequence of events characterized by a sudden increase in aminotransferases, changes in mental function, and disruptions in blood clotting. The causes of acute liver failure encompass acetaminophen toxicity, hepatic ischemia, viral and autoimmune hepatitis, and drug-induced liver injury resulting from prescription medications as well as herbal and dietary supplements. Hepatotoxicity is a liver injury caused by various factors, including drugs, toxins, and viral infections. CYP2E1 protein is a transcription factor that plays a crucial role in the regulation of inflammatory responses, cell proliferation, and apoptosis. Cytochrome P450 2E1 (CYP2E1, EC 1.14. 13. n7) is a member of the cytochrome P450 mixed-function oxidase system, which is involved in the metabolism of xenobiotics in the body



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[1-3]. CYP2E1 has major contribution in liver diseases such as nonalcoholic fatty liver disease, alcoholic liver disease, and nonalcoholic steatohepatitis, probably through hepatic lipid peroxidation. The deleterious effects of CYP2E1 are also due to the fact that significant levels are located within mitochondria [4-6]. Chronic ethanol use can induce CYP2E1 activity, leading to a greater percentage of acetaminophen metabolized to NAPQI, increasing the risk of hepatotoxicity from acetaminophen use. Many scientists question the existence of an important ethanol-acetaminophen interaction [7,8]. Several studies have linked CYP2E1 activation to the pathogenesis of liver diseases, including hepatotoxicity.

CYP2E1 is considered one of the common pathogenic factors for both Alcoholic fatty liver diseases (AFLD) and non-alcoholic fatty liver diseases (NAFLD) due to its inducibility under these conditions with the increased ROS, causing deleterious effects on mitochondrial dysfunction and hepatotoxicities [4]. Therefore, the inhibition of CYP2E1 activation has been suggested as a potential therapeutic target for liver diseases [9]. CYP2E1 is associated with liver toxicity through two mechanisms: Firstly, by activating substrates into reactive metabolites, and secondly, by potentially generating reactive oxygen species (ROS). The lack of close coordination between substrate binding, oxygen binding, and electron delivery during catalysis can lead to the conversion of diatomic oxygen into superoxide or hydrogen peroxide, which is released instead of being utilized to monooxygenate the substrate [10]. These ROS have been implicated in various damaging events, including lipid peroxidation, protein oxidation, and DNA oxidation. Notably, CYP2E1 exhibits a significantly higher propensity for ROS production compared to other cytochrome P-450 enzymes, and some drugs have been explored for their potential in treating hepatotoxicity targeting the CYP2E1 protein. Several drugs have been used in the treatment of liver diseases, including Prednisolone. However, the adverse effects associated with these drugs have limited their use in clinical practice. Natural products have gained increasing attention in the treatment of liver diseases due to their low toxicity and potential therapeutic effects [11,12]. Flavonoids are a class of natural compounds that have been reported to protect against hepatotoxicity by inhibiting the CYP2E1 pathway [13]. *C. volubile* is a leafy vegetable found in certain regions of Nigeria, is not fully utilized despite its growing popularity due to its various benefits, such as its traditional uses, nutritional content, and potential therapeutic properties. Nonetheless, there is still much to uncover regarding the plant's pharmacological potential [14].

Molecular docking, however, offers a faster and more efficient way to screen for potential compounds with the desired pharmacological activity. By predicting the binding affinity of small molecules to target

proteins, molecular docking enables researchers to identify compounds that are likely to bind to the target protein and exhibit the desired pharmacological activity [15]. This study aimed at investigating the molecular docking of *C. volubile* compounds on hepatotoxicity targeting CYP2E1 protein with resveratrol as a natural inhibitor, Sulforaphane, and chlormethiazole drugs are also used as standards, selecting the hit compounds for pharmacokinetics and drug-likeness properties and also evaluating the free energies and binding interactions.

Materials and Method

Ligand identification and optimization

C. volubile, phytochemicals were extracted using GC-MS, and their 2D structures were obtained from the NCBI PubChem database. The 3D structures were created with Schrödinger Maestro 11.5's LigPrep and an OPLS3 force field. Ligand ionization states were created at pH ranges ranging from 7.02.0 (general pH of biological systems). The computation of 25 stereoisomers was left at the specified chiralities (various other chiral centers), and the best potential stereochemical structures were generated per ligand.

Protein preparation

The three-dimensional crystal structure of 3E6I was obtained from the RCSB protein data bank. The protein was equipped for calculations using the protein preparation wizard of Schrodinger Maestro 11.5 by filling missing loops and side chains using Prime, assigning bond orders, adding hydrogens, and deleting water beyond 5.00 Å. Using Epik, Tautomeric states of heteroatom groups were generated at a pH of 7.0 ± 2.0, then optimized using PROPKA at pH 7.0. Restrained minimization was carried out by setting heavy atom root-mean-square-deviation to 0.30 Å using an OPLS3 force field.

GRID generation

The maestro receptor grid generation tool was used to generate the receptor grid of CYP2E1, based on the space occupied by its cognate ligands. It is worth noting that the binding pocket of this target protein is located at the X, Y, and Z coordinates.

Compound preparation for molecular docking against the CYP2E1 protein target

The compounds generated were subsequently docked in the Maestro's glide tool, using standard precision SP and extra precision XP algorithm. To obtain probable binding orientations and binding energy of ligands at the specified binding domain of CYP2E1, the grid box with dimensions was used to cover a region on the distal portion of the protein preceded by running of the docking procedure. Following the docking run, compounds that have docking scores (binding energies) lower than the control drug, were exposed to molecular

visualization to examine their binding poses (3D) and molecular interaction fingerprints (2D) using PyMOL, subjected to MMGBSA.

Drug-Likeness and ADMET studies

ADMET (Absorption, Distribution, Metabolism, Excretion, and Toxicity) is critical at the final stage of the drug discovery and design route to analyze the pharmacokinetics and pharmacodynamics of proposed compounds that could serve as potential drugs [16]. The SWISSADME was used to predict the ADMET characteristics of compounds with the greatest hits following the molecular docking study.

MMGBSA-binding free energy (ΔG_{bind})

This technique forecasts the free energy of small drug molecules binding to biological macromolecules. The free energy of binding (ΔG_{bind}) was estimated using Prime Schrodinger Maestro 11.5, the OPLS3 force field, and VSGB solvation model with other parameters set to default [17].

Results and Discussion

The present study employs computational methods such as molecular docking, 2D-Interaction, Prime MMGBSA, and ADMET to discover potential compounds that may exhibit inhibitory effects on the CYP2E1 protein.

Molecular docking simulation and validation

Docking simulations were conducted against the target of interest, employing the 49 compounds identified from *C. volubile* in comparison to Ciprofloxacin, to assess their potential binding affinity to CYP2E1 protein- a therapeutically significant target in hepatotoxicity. The docking outcomes indicated that the compounds exhibit inhibitory potential against the examined protein better than Resveratrol, Sulforaphane, and chlormethiazole standards. The docking analysis depicts the interactions that significantly affect the inhibition (Table 1).

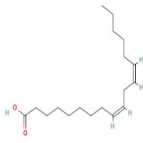
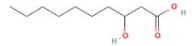
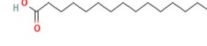
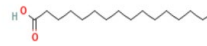
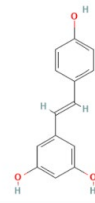
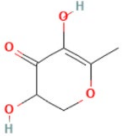
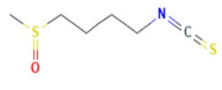
Analyzing the binding energies reveals that *cis,cis*-linoleic acid, 3-hydroxy decanoic acid, n-pentadecanoic

Table 1: Docking scores (kcal/mol) of Hit ligands from *Clerodendrum volubile* docked against CYP2E1 protein of Cytochrome P450.

S/N	PUBCHEM ID	COMPOUNDS NAME	DOCKING SCORE	MMGBSA Δd BIND
1.	5280450	<i>cis, cis</i> -linoleic acid	-10.746	-63.43
	26612	3-hydroxydecanoic acid	-10.203	-24.26
	13849	n-pentadecanoic acid	-9.974	-44.63
	985	palmitic acid	-9.177	-56.88
	119838	3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one	-8.447	-38.52
	7478	<i>p</i> -anisic acid	-7.56	-33.41
	445154	Resveratrol	-6.472	-41.24
	481107206	α -monostearin	-6.466	-51.05
	31292	stearamide	-6.334	-25.27
	5367459	2-hydroxy-1-(hydroxymethyl) ethyl (9 <i>E</i> ,12 <i>E</i> ,15 <i>E</i>)-9,12,15-octadecatrienoate	-5.865	-50.18
	798	2,3-benzopyrrole	-5.493	-13.33
	5280590	methyl elaidate	-5.423	-41.18
	188323	cirsimaritin	-5.354	-8.1
	753	glycerol	-5.283	-16.83
	5280442	acacetin	-5.024	-22.92
	5363758	3-[(2 <i>E</i>)-2,4-pentadienyl]-2,4-pentanedione	-4.939	-33.94
	69421	palmitic amide	-4.896	-44.54
	164174842	α -glyceryl linolenate	-4.895	-40.75
	164174842	α -glyceryl linolenate	-4.895	-40.75
	12366	palmitic acid, ethyl ester	-4.773	-35.5
	49862999	glycerol α -monoacetate	-4.596	-23.48
	5319706	linolenic acid, methyl ester	-4.019	-40.51
	8181	palmitic acid, methyl ester	-3.994	-34.05
	5283387	oleamide	-3.951	-39.6
	5284421	linoleic acid, methyl ester	-3.894	-32.61
	17785621	n-propyl decyl ether	-3.858	-20.85

5350	Sulforaphane	-3.767	-26.74
5365074	(11E)-10,13,13-trimethyl-11-tetradecyl acetate	-3.655	-34.08
7476	<i>p</i> -acetylanisole	-3.565	-20.31
10783	chlormethiazole	-3.47	-12.37
5370052	3-oxo-.alpha.-ionol	-3.266	-10.06
119838	3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one	-3.265	-16.62
537123	1-(6-oxabicyclo[3.1.0]hex-1-yl) ethanone	-3.165	-19.38
181561	5-methoxypyrrolidin-2-one	-3.087	-24.37
27209	(2,6,6-trimethyl-2-hydroxycyclohexylidene) acetic acid lactone	-3.046	4.95
556188	tricyclo[5.2.1.0(1,5)]decane-8,9-diol	-2.902	8.66
557353	2-oxadamantan-6-ol	-2.328	16.76
91693822	fumaric acid, 2-dimethylamino ethyl heptadecyl ester	-1.872	-3.59
5363508	(2Z,6E)-10,11-dihydroxy-3,7,11-trimethyl-2,6-dodecadienyl acetate	-1.84	-38.24
29075	nonadec-1-ene	-1.049	-33.73
23217	1-heptadecene	-0.012	-32.71
5541	fungacetin	0.141	-10.75
5205941	2,3-bis(1-methylallyl)pyrrolidine	1.653	-20.27
91693822	fumaric acid, 2-dimethylamino ethyl heptadecyl ester	4.051	-37

Table 2: Prime MMGBSA of hit Compounds from *Clerodendrum volubile* against the target protein.

S/N	PUBCHEM ID	COMPOUNDS NAME	MMGBSA Δ d BIND (kcal/mol)	COMPOUND STRUCTURES
1.	5280450	<i>cis, cis</i> -linoleic acid	-63.43	
2.	26612	3-hydroxydecanoic acid	-24.26	
3.	13849	n-pentadecanoic acid	-44.63	
4.	985	palmitic acid	-56.88	
5.	445154	Resveratrol	-41.24	
6.	119838	3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one	-38.52	
7.	5350	Sulforaphane	-26.74	
8..	10783	chlormethiazole	-12.37	

acid, palmitic acid, and 3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one stand out as the compounds with the most robust inhibitory potentials. They exhibit notable binding affinities against CYP2E1 with energies of -10.746, -10.203, -9.974, -9.177, and -8.447 kcal/mol, respectively. In comparison, several compounds have lower binding energies than the standard Resveratrol with energies of -6.472 kcal/mol respectively. These show that the compounds of *C. volubile* possess a potential inhibitory effect on the target protein (CYP 2E1).

Binding free energy calculation

To further validate the results obtained from the docking procedure, the complexes were subjected to the MM-GBSA post-docking analysis. This analysis is often employed to rigorously assess the stability of complexes formed after docking simulation, by computing the binding free energy of the ligand with the receptor [17]. Intriguingly, results obtained from MM-GBSA analysis have been reported to be correlated with the biological activities derived from experimental procedures [18]. Table 2 presents the results of the MM-GBSA analysis conducted in this study. Interestingly, the compound *cis,cis*-linoleic acid, which was ranked among the five hit compounds based on the docking results, was found to possess the highest binding free energy measured in kcal/mol -63.43. The other hit compounds which include palmitic acid, *n*-pentadecanoic acid, 3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one, and 3-hydroxy decanoic acid, have binding free energies of -56.88, -44.63, -38.52, and -24.26 kcal/mol respectively. The standards Resveratrol, Sulforaphane, and chlormethiazole have binding free energies of -41.24, -26.74, and, -12.37 kcal/mol respectively which is lower than the hit compounds this indicates that the hit compounds have better binding strength compared to the standards (the lower the binding energy the better its affinity to the target).

These calculations suggest that the enduring and robust complex stability consistently observed with *cis,cis*-linoleic acid, and palmitic acid across all lead targets implies that the hit compounds might competitively induce prolonged and heightened biological responses compared to standard drugs and other hits.

Interaction profiling of hit compounds with target protein

The diversity in binding scores and affinities among the identified hit compounds implies that these molecules have different degrees of effectiveness in interacting with the target protein. In drug discovery and molecular biology, understanding the interaction between small molecules (such as potential drugs) and specific amino acid residues in target proteins is crucial. The binding affinity refers to the strength of the interaction between a molecule and its target protein, and a higher affinity generally indicates a stronger binding. The figures 1 below show the 2D and 3D interactions of each hit compound and the standards [19] (Table 3).

Figure 1 (1A, 2A, 3A, 4A, 5A, and 6A), indicate the 2D interaction of *cis,cis*-linoleic acid, 3-hydroxydecanoic acid, *n*-pentadecanoic acid, palmitic acid, and 3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one and standard respectively to the active site of CYP2E1 receptor while Figure 1 (1B, 2B, 3B, 4B, 5B, and 6B) display their 3D interactions. Notably, the hydrogen bond is represented by a purple arrow, the green region represents the hydrophobic interaction. The interacting residues of the hit compounds and the two standards with the CYP2E1 target protein are displayed in Figure 1 (1A-6A) above.

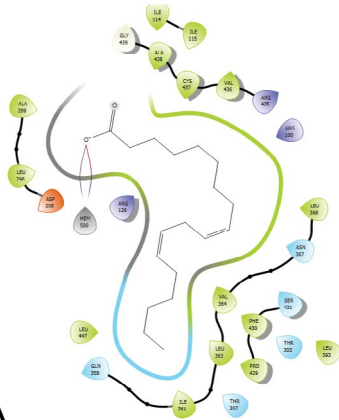
ADMET profiling

The pharmacokinetic properties of the hit ligands were assessed after evaluating their potency and

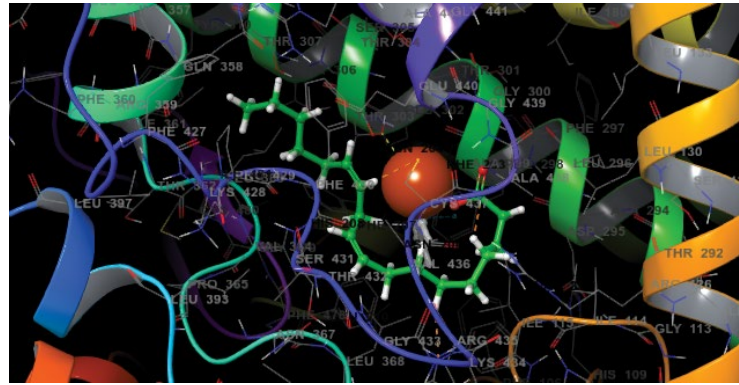
Table 3: Shows interactions of all ligands' atoms with amino acid residues in the binding site of CYP2E1 after molecular docking.

Compound Name	H-Bond	Hydrophobic Interactions
<i>cis,cis</i> -linoleic acid		Leu 296, Asn 367, Val 364, Arg 126, Asp 295, Leu 447, Gln 358, Ile 361, Leu 363, Val 364, Leu 368, The 307, Pro 429, Phe 430, Ser 431, Thr 303, Leu 393, Gly 439, Ala 438, Cys 437, Val 436, Arg 435, Arg 100, Ile 114, Ile 115.
3-hydroxydecanoic acid		Leu 447, Ala 443, Thr 307, Thr 304, Thr 303, Gln 358, Leu 363, Val 364, Pro 429, Phe 430, Leu 368, Cys 437.
<i>n</i> -pentadecanoic acid	Arg 126, Arg 100, Trp 122, Arg 435.	Leu 447, Ala 443, Thr 307, Thr 304, Thr 303, Gln 358, Pro 429, Phe 430, Ser 431, Ile 115, Ile 114, Val 436, Cys 437, Ala 438, Leu 368, Leu 393, Asn 367, Val 364, Leu 363.
palmitic acid	Thr 307, Thr 304, Thr 303, Asn367	Leu 447, Ala 443, Thr 307, Thr 304, Thr 303, Gln 358, Leu 363, Val 364, Asn 367, Leu 368, Pro 429, Phe 430, Ser 431, Leu 393, Arg 100, Arg 435, Val 436, Cys437, Ala 438, Arg 126, Ile115, Ile 114, Ala 299.
3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one	Arg 435	Pro 429, Phe 430, Ser 431, Leu 393, Val 364, Asn 367, Leu 368, Hip 370, Arg 100, Arg 435, Val 436, Cys 437, Ala 438.
Resveratrol	His 1345	Leu 393, Hip370, Leu 368, Asn 367, Ser 431, Phe 430, Pro 429, Val 364, Leu 363, Ile 361, Gln 358, Leu 447, Thr 307, Ala 443, Thr 304, Thr 303, Cys437.

5280450 - 3E6I - minimized

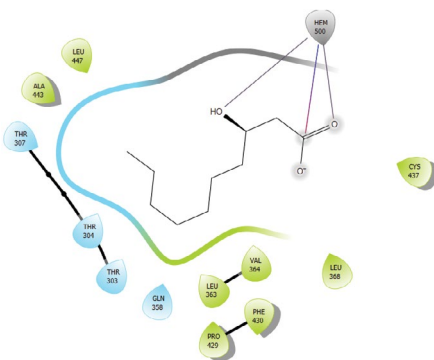


1A

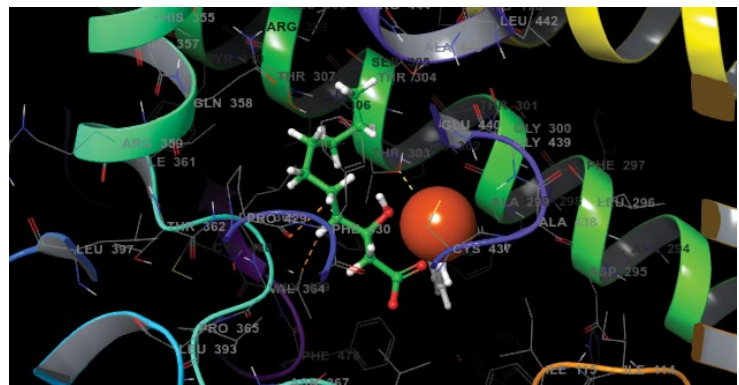


1B

26612 - 3E6I - minimized

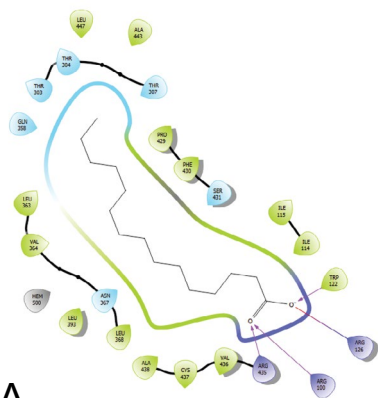


2A

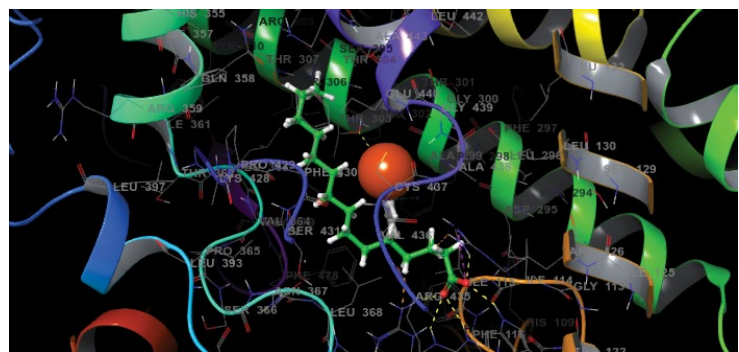


2B

13849 - 3E6I - minimized

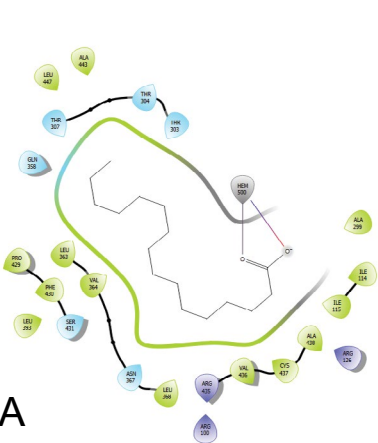


3A

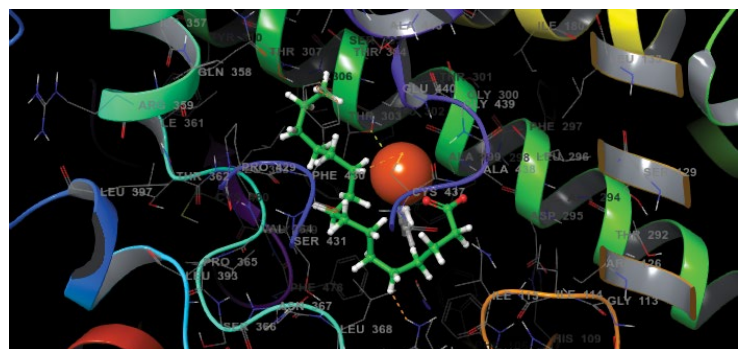


3B

985 - 3E6I - minimized

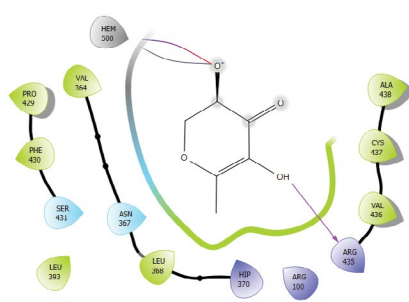


4A

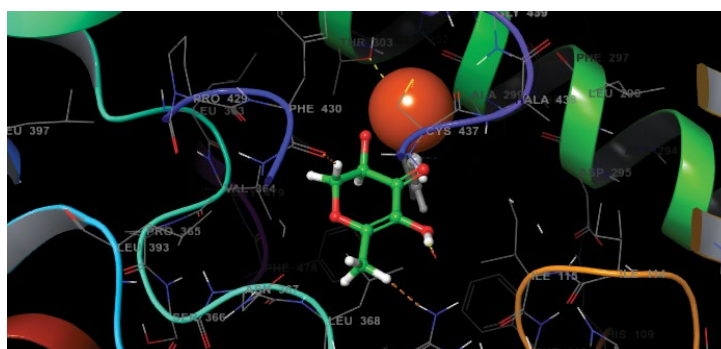


4B

119838 - 3E6I - minimized

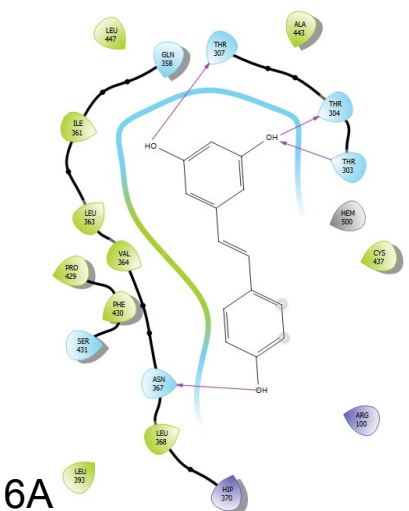


5A

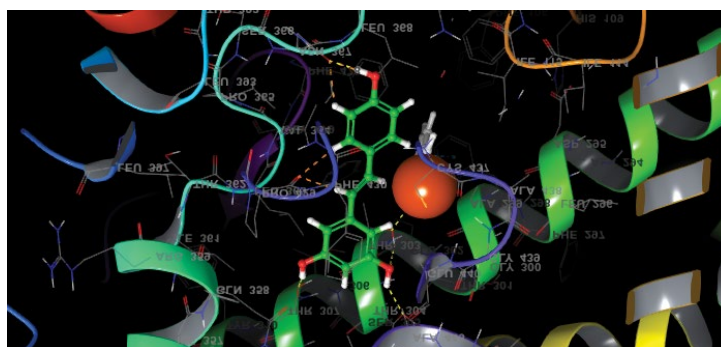


5B

445154 - 3E6I - minimized



6A



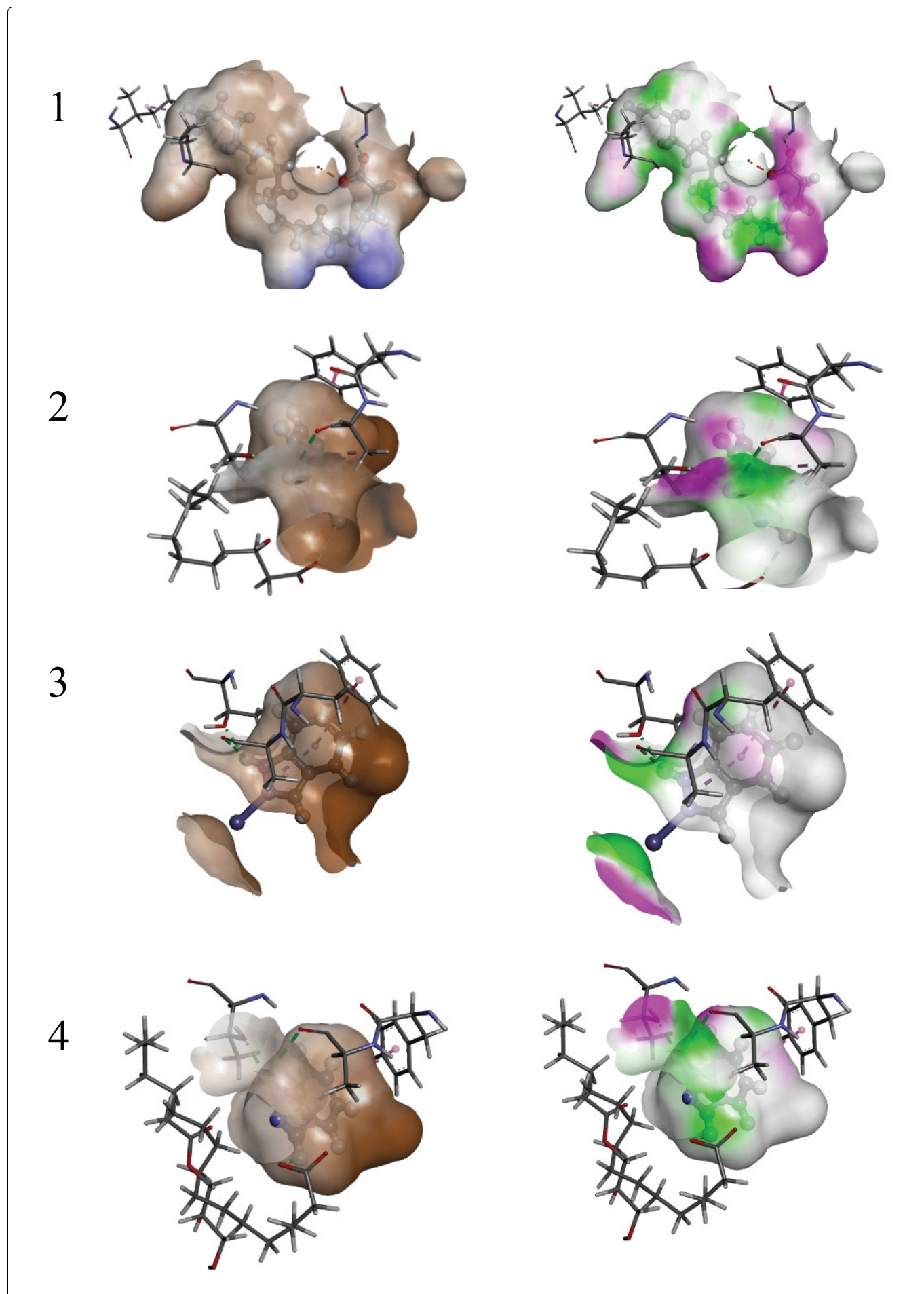
6B



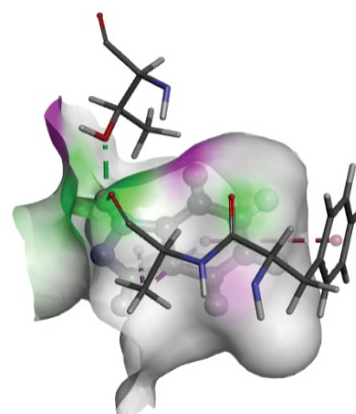
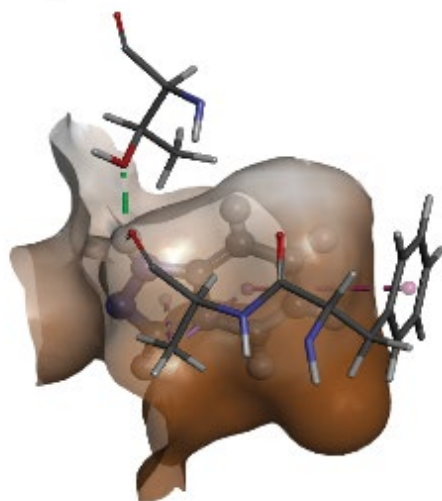
Figure 1: The 2D and 3D interaction of the 5 hit compounds using Maestro.

varied affinities. This investigation is crucial to address issues such as potential side effects, toxicity, limited absorption, and inadequate clearance, commonly encountered during drug development. Understanding these factors are essential to avoid hurdles that often lead to eliminating promising compounds with therapeutic potential. The focus on pharmacokinetics serves as a vital transition from preclinical investigation to clinical trials in the drug development process. The properties of the candidate inhibitors were assessed using SWISSAdme [20], and all components were evaluated for evaluation of critical parameters in the context of drug development. These parameters include the count of hydrogen bond acceptors and donors, molecular weight, and the octanol/water partition coefficient (QPlogPo/w). The evaluation process was thorough, and the results were promising. The key observation is that all the hit ligands, which are potential drug candidates, consistently fall within

accepted ranges for these physicochemical attributes. This conformity reaffirms the suitability of these ligands for further consideration and development in the field of drug development. As shown in Table 4, all 5 hit ligands effortlessly met the criteria molecular weight (MW) of ≤ 500 and violated none in Lipinski rule of 5 (Ro5) -one of the rules that determines drug efficacy cis,cis-linoleic acid and palmitic acid violated just one of the 5 rules which is still acceptable. Cytochrome P450 (CYP) is a family of enzymes that catalyze the phase 1 metabolism of xenobiotics at large. Any compound that inhibits selected isoforms would induce a drug-drug interaction. The entire compounds show different inhibitions to the other isoforms of Cytochrome P450 (CYP). They are all moderately soluble and have high GI absorption. The BOILED-egg allows for intuitive evaluation of passive gastrointestinal absorption (HIA) and brain penetration (BBB) in the function of the position of the molecules in the WLOGP-versus-TPSA referential. P-glycoprotein



5



6

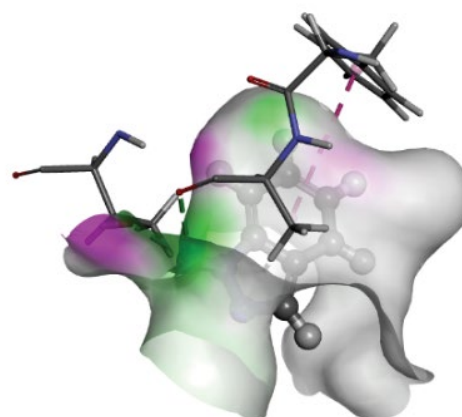
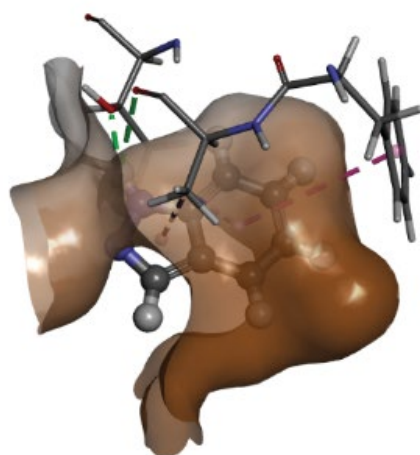


Figure 2: Hydrophobic bonds (X) and Hydrogen bonds (Y) maps between CYP2E1 and five chosen inhibitors including Resveratrol the standard. 1) *cis,cis*-linoleic acid. 2) 3-hydroxydecanoic acid 3) n-pentadecanoic acid 4) palmitic acid. 5) 3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one 6) Resveratrol. Ligands are presented in ball and stick while amino acid residues of CYP2E1 are in lines executed in BIOVIA's Discovery Studio 2016.

Table 4: ADMET profile and drug likeness of the five hit compounds including the natural substrate using SWISS-ADME server.

Molecule	Compounds	Mol MW	dHB	aHB	RO5	GI absorption	logKp (cm/s)	BBB permeant	PSA	Pgp substrate
Molecule 1	<i>cis,cis</i> -linoleic acid	280.4	1	2	1	High	-3.05	Yes	37.3	No
Molecule 2	3-hydroxydecanoic acid	188.26	2	3	0	High	-5.66	Yes	57.53	No
Molecule 3	n-pentadecanoic acid	242.4	1	2	0	High	-3.07	Yes	37.3	No
Molecule 4	palmitic acid	256.42	1	2	1	High	-2.77	Yes	37.3	No
Molecule 5	Resveratrol	228.24	3	3	0	High	-5.47	Yes	60.69	No
Molecule 6	3,5-dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one	144.13	2	4	0	High	-7.44	No	66.76	No
Molecule 7	Sulforaphane	177.29	0	2	0	High	-6.38	No	80.73	No
Molecule 8	Chlormethiazole	161.65	0	1	0	High	-5.78	Yes	41.13	No

(P-gp), the most extensively studied ATP-binding cassette transporter, functions as a biological barrier by extruding toxic substances and xenobiotics out of cells serving to protect the integrity of tissues and cellular environments (Figure 2 and Figure 3).

Conclusion

The study investigated selected natural compounds that have inhibitory and pharmacological properties against the CYP2E1 to reduce the risk of liver diseases, druggable compounds demonstrated a superior binding

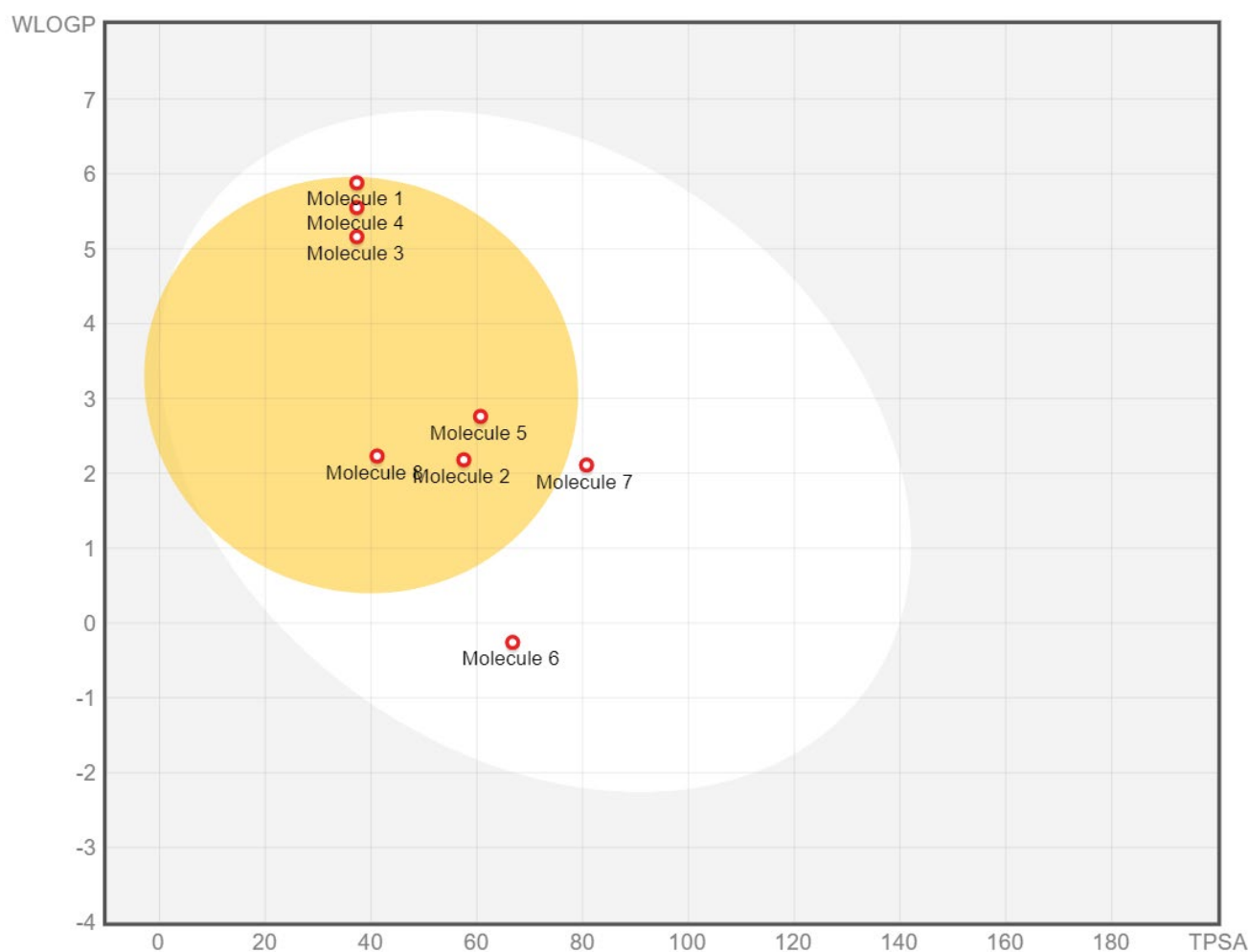


Figure 3: Showing the boiled-egg diagram of the 5 hit compounds and the three standard drugs. The white region is for a high probability of passive absorption by the gastrointestinal tract, and the yellow region (yolk) is for a high probability of brain penetration. Yolk and white areas are not mutually exclusive. In addition, the points are coloured in blue if predicted as actively effluxed by P-gp (PGP+) and in red if predicted as non-substrate of P-gp (PGP-) [21].

affinity and favorable ADME profiles using Glide and SWISSADME. The study suggests that molecular docking and simulations are reliable methods for discovering new drugs to combat hepatotoxicity. This highlights the need for novel inhibitors due to adverse effect of existing drugs. These promising compounds should undergo further testing, including atomistic simulation, in-vitro, and in-vivo studies, to validate their potential as effective hepatoprotective drugs.

Conflict of Interest

We have no conflict of interest.

Data Availability Statement

The authors confirm that the data supporting the findings of this study are available within the article.

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References

1. Leung T, Rajendran R, Singh S, Garva R, Krstic-Demonacos M, et al. (2013) Cytochrome P450 2E1 (CYP2E1) regulates the response to oxidative stress and migration of breast cancer cells. *Breast Cancer* 15: R107.
2. Garcia-Suastegui WA, Ramos-Chavez LA, Rubio-Osornio M, Calvillo-Velasco M, Atzin-Méndez JA, et al. (2017) The role of CYP2E1 in the drug metabolism or bioactivation in the brain. *Oxid Med Cell Longev* 2017: 4680732.
3. Massart J, Begriche K, Hartman JH, Fromenty B (2022) Role of mitochondrial cytochrome P450 2E1 in healthy and diseased liver. *Cells* 11: 288.
4. Abdelmegeed MA, Ha SK, Choi Y, Akbar M, Song BJ (2017) Role of CYP2E1 in mitochondrial dysfunction and hepatic injury by alcohol and non-alcoholic substances. *Curr Mol Pharmacol* 10: 207-225.
5. Delli Bovi AP, Marciano F, Mandato C, Siano MA, Savoia M, et al. (2021) Oxidative stress in non-alcoholic fatty liver disease. An updated mini review. *Front Med* 8: 595371.
6. Harjumäki R, Pridgeon CS, Ingelman-Sundberg M (2021) CYP2E1 in alcoholic and non-alcoholic liver injury. Roles of ROS, reactive intermediates and lipid overload. *Int J Molecul Sci* 22: 8221.
7. Hinson JA, Roberts DW, James LP (2010) Mechanisms of acetaminophen-induced liver necrosis. *Handb Exp Pharmacol* 2010: 369-405.
8. Heit C, Dong H, Chen Y, Thompson DC, Deitrich RA, et

- al. (2013) The role of CYP2E1 in alcohol metabolism and sensitivity in the central nervous system. *Subcell Biochem* 67: 235-247.
9. Leung TM, Nieto N (2013) CYP2E1 and oxidant stress in alcoholic and non-alcoholic fatty liver disease. *J Hepatol* 58: 395-398.
10. Piver B, Berthou F, Dreano Y, Lucas D (2001) Inhibition of CYP3A, CYP1A and CYP2E1 activities by resveratrol and other non-volatile red wine components. *Toxicol Lett* 125: 83-91.
11. Gbenga-Fabusiwa FJ, Jeff-Agboola YA, Ololade ZS, Akinrinmade R, Agbaje DO (2022) Waste-to-wealth; nutritional potential of five selected fruit peels and their health benefits: A review. *Afr J Food Sci* 16: 172-183.
12. Ololade ZS, Olawore NO (2021) Health benefits of the fruit essential oil of eucalyptus citriodora: Secondary metabolites, radical scavenging, antioxidant, antiinflammatory, analgesic, antimicrobial potential. *Int J Drug Developm Res* 13: 1-7.
13. Rohilla R, Garg T, Goyal AK, Rath G (2016) Herbal and polymeric approaches for liver-targeting drug delivery: Novel strategies and their significance. *Drug Deliv* 23: 1645-1661.
14. Jaramillo-Morales OA, Díaz-Cervantes E, Via LD, Garcia-Argaez AN, Espinosa-Juárez JV, et al. (2023) Hepatoprotective activity, in silico analysis, and molecular docking study of verbascoside from leucophyllum frutescens in rats with post-necrotic liver damage. *Scientia Pharmaceutica* 91: 40.
15. Kumar V, Sharma N, Sourirajan A, Khosla PK, Kamal Dev (2020) Docking studies in targeting proteins involved in cardiovascular disorders using phytochemicals from Terminalia arjuna.
16. Benet LZ, Hosey CM, Ursu O, Oprea TI (2016) BDDCS, the rule of 5 and drug ability. *Adv Drug Deliv Rev* 101: 89-98.
17. Wang E, Sun H, Wang J, Wang Z, Liu H, et al. (2019) End-Point binding free energy calculation with MM/PBSA and MM/GBSA: Strategies and applications in drug design. *Chem Rev* 119: 9478-9508.
18. Genheden S, Ryde U (2015) The MM/PBSA and MM/GBSA methods to estimate ligand-binding affinities. *Expert Opin Drug Discov* 10: 449-461.
19. Patil R, Das S, Stanley A, Yadav L, Sudhakar A, et al. (2010) Optimized hydrophobic interactions and hydrogen bonding at the target-ligand interface leads the pathways of drug-designing. *PLoS One* 5: e12029.
20. Daina A, Michielin O, Zoete V (2017) Swiss ADME: A free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Sci Rep* 7: 42717.
21. Daina A, Zoete V (2016) A BOILED-Egg To Predict Gastrointestinal Absorption and Brain Penetration of Small Molecules. *ChemMedChem* 11: 1117-1121.