Cellular and Molecular Biology
E-ISSN : 1165-158X / P-ISSN : 0145-5680
www.cellmolbiol.org

The discovery of the natural compound Boeravinone-C as a potential antiobesity drug candidate targeting pancreatic lipase using chemo-informatics-based approaches

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ARTICLE INFO

Original paper

Article history:
Received: April 05, 2023
Accepted: May 30, 2023
Published: July 31, 2023

Keywords:

Boerhaavia Diffusa, Anti-Obesity; Pancreatic Lipase; Molecular Modeling; Docking; Molecular Dynamic Simulations

ABSTRACT

Obesity is a metabolic disorder distinguished by excess fat deposition in fatty tissues. Pancreatic lipase is one of the promising drug targets for treating obesity due to its critical role in the hydrolysis of triglycerides into mono-glycerides and free fatty acids. Due to unsatisfactory results and severe side effects of the current drugs available for treating obesity, there is an urgent need to identify novel therapeutic options. *Boerhaavia diffusa* is one of the widely known species of flowering plant commonly known as Punamava. Extracts from Punamava plants have been widely used in treating countless ailments in traditional medicine. Recently, multiple reports demonstrated the potential antiobesity activity of *B. diffusa* plant extracts. In this scenario, we have evaluated numerous reported *B. diffusa* against pancreatic lipase drug targets to identify which reported phytochemicals to have the most promising potential to act as an inhibitor for pancreatic lipase using computational approaches. All the twenty-four phytochemicals from Boerhaavia diffusa were identified as significantly strong binders with a range of binding energies between -6.0 to -8.0 Kcal/mol inside the pancreatic lipase active binding site. On the other hand, we calculated 2D Quantitative Structure-Activity Relationship (QSAR) molecular descriptor properties adhered to Lipinski's rule of five. Between twenty-four phytochemicals evaluated, Boeravinone-C, with a range binding energy of -8.0 Kcal/mol, was discovered as the best lead-like molecule, compared to marketed Orlistat, which has shown -5.6 Kcal/mol of binding energy. Conclusively, Boeravinone-C from *B. diffusa* extract showed promising inhibitory potential against pancreatic lipase worth further evaluation.

Doi: http://dx.doi.org/10.14715/cmb/2023.69.7.10

Introduction

Obesity is a metabolic disorder distinguished by excess fat deposition in the fatty tissues (1). Pancreatic lipase is one of the promising drug targets for treating obesity due to its critical role in the hydrolysis of triglycerides into mono-glycerides and free fatty acids (2–4). Due to unsatisfactory results and severe side effects (5) of the current drugs available for treating obesity, there is an urgent need to identify novel therapeutic options. Pancreatic lipase is a protein consisting of a single polypeptide chain of approximately 45 kDa, and it contains a catalytic triad of serine, aspartate, and histidine residues essential for its enzymatic activity. The catalytic domain of pancreatic lipase has a characteristic alpha-beta hydrolase fold and is structurally similar to other lipolytic enzymes (6). Boerhaavia *diffusa* is a widely available flowering plant species commonly known as Punamava. Extracts from Punamava plants have been widely used in treating countless ailments in traditional medicine (7). Extracts from *B. diffusa* were demonstrated to be having diuretic (8), hepatoprotective (9), anti-inflammatory (10), anti-fibrinolytic (11), anti-cancer (12–14), anti-diabetic (15), immuno-modulatory (16), immunosuppressive (17), pulmonary tuberculosis (18), anti-lymphoproliferative (19), analgesic (20), antibacterial (21), anti-fungal (22), adaptogenic (23), antiamoebic (24), lipotropic (25) and anticonvulsant activity (26) etc., numerous phytochemicals e.g. flavonoids (5,7-dihydroxy-3',4'-dimethoxy- 6,8- dimethyl flavone, C-methyl flavone, 6', 5'-dimethoxy-5, 7, 3-trihydroxyflavone, 3,5,4′-dihydroxy-6,7-dimethoxyflavone, borhavone, (4',7-dihydroxy-3'-methylflavone), 3,3',5-trihydroxy-

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7-methoxyflavone, alkaloids (punarnavine), 3,4-àglycosides (punarnavoside, 3-O-β-D-galactopyranosyl-(1'''3')-O-β-D-glucopyranoside, eupalitin), 2")-O-β-D-galactopyranoside, rotenoids (boeravinone A-H), àdimethoxyphenyl-1-O-β-D apiofuranosyl-(1''steroids, triterpenoids, lignans, proteins, lipids, carbohydrates and the herb has been linked to glycoproteins and other compounds (11,27). Multiple reports (28–32) recently demonstrated the potential antiobesity activity of *B. diffusa* plant extracts. In this scenario, this study aims to evaluate the phytochemicals mentioned above of *B. diffusa* against pancreatic lipase drug target towards identifying which reported phytochemicals have the most promising potential to act as an inhibitor for pancreatic lipase using computational approaches.

Materials and Methods

Software and program

Discovery Studio visualizer software (33) was utilized to visualize ligand and receptor structures H - bonding networks, calculate bond lengths, and render images. PubChem (34) database was chosen to retrieve phytochemical structures. For the docking studies, the initial docking program in this project is Autodock 4.0 (35). Auto-Dock Tools version 1.5.6 was used to prepare the receptors and ligands in pdbqt files and fix a grid box's size. Use the Molinspiration, Orisis, property explorer program (36,37) to investigate the ADMET properties of phytochemicals. Schrodinger's Maestro ver.9.5 was utilized to visualize the screened compounds' pharmacophore features and simulate molecular dynamics results.

Ligand and protein receptor Preparation

The pancreatic lipase crystal structure [PDB: 1N8S] was taken from Protein Data Bank (PDB) (38). Autodock's repair commands module added numerous missing atoms to the crystal structure. Clean a protein's crystal structure before docking by removing any water molecules. H-atoms were introduced into these protein targets to ensure that amino acid residues maintained their correct ionization and tautomeric states. Used the resulting modified structure to perform the semi-flexible dockings. The ligand and receptors' energies were minimalized using the Conjugate Gradient and Steepest Descent methods in Discovery Studio. The methods for minimization were implemented using the CHARMM force field (39).

Semi-flexible docking

The binding pose and associated energy of phytochemicals via the drug target pancreatic lipase are predicted using Autodock Version 4.0. The docking studies followed the protocol described in Autodock version 4.0 using the default protocol for predicting IC_{50} values and binding posture and related binding energies. Briefly, set the energy scoring grid box to 120, 120, and 120 Ao (x, y, and z) centered at X = 27.857; Y = 22.737, and Z = 97.37 with a grid point spacing of 0.375 A^{o} assigned using the default atomic solvation parameters created the grid box such a way that a centered three-dimensional grid box on it encircled the pancreatic lipase. Choose the docking engine as the Lamarckian Genetic Algorithm (LGA) (40), with all docking parameters set to default. The optimal docking solution for each docked complex is reported by Autodock, following each LGA run based on cluster analysis. The Gibbs free energy of binding (G) in chemical bonding comprises six energy variables: repulsion and dispersion, electrostatic interactions, H- bonding, desolvation effects, torsional limitations, and internal ligand divergence from covalent geometry. A total of ten docking modes offered by LGA cluster analysis were used in this study, with the lowest energy docking option selected from each docking simulation. The presence of active rotatable bonds in each molecule was permitted as this gave the chemical flexibility.

Phytochemical pharmacological properties

The online server Osiris Property Explorer (www. organic-chemistry. org/prog/peo/) (41) was used to assess the pharmaceutical reliability of the drug candidates, to examine molecular descriptors such as Log P, the number of H- bond acceptors, the number of H- bond donors, the molecular weight of phytoconstituents, and compounds' toxicology profile.

Molecular dynamic simulations

Simulations of molecular dynamics were performed to comprehend the binding interactions at the molecular level and analyze those interactions at the atomic level using the default protocol. In brief, OPLS 2005 force field (42) TIP3P water models have been simulated using parameters at neutral pH conditions (43). The particular size and shape of the water box buffered at ten distances. The volume of the water box was determined using periodic boundary conditions and calculated to be 545000 cubic s of the simulation box volume, eliminating short-range electrostatic and Van der Waals interactions during the equilibration process and long-range electrostatic interactions figured out using the Particle Mesh Ewald method (44) and used a RESPA integrator (45) with a time step of 2 fs to compute long-range electrostatics every six fs. Each simulated condition contained approximately 54415, 54590, and 54570 atoms for apo in composite with Orlistat and Boeravinone-C, respectively. The simulated conditions were equilibrated using Desmond in the NPT ensemble (46) at 300 K temperature and 1 bar using the Nose-Hoover chain relaxation thermostat method (47), as well as the Martyna-Tobias-Klein relaxation barostat method (48). We ensured all simulations were conducted at the same temperature, volume, and pressure conditions with an isotropic coupling style throughout the simulated timescale. For simulation quality analysis, it was established that the average total energy of the simulated systems remained approximately -137500 Kcal/mol for apo and in complex with Orlistat but was -138000 Kcal/mol for complex with Boeravinone-C.

Results

Virtual screening of the phytochemicals from *B. diffusa* with pancreatic lipase

We have performed the virtual screening for the twenty-four phytochemicals (Figure 1) with the pancreatic lipase to know the molecular interactions and binding energies responsible for this target-specific inhibition. Virtual screening has been performed targeting its critical site core residues. Virtual screening results are presented in Table 1. As per the virtual screening experiment, all the twenty-

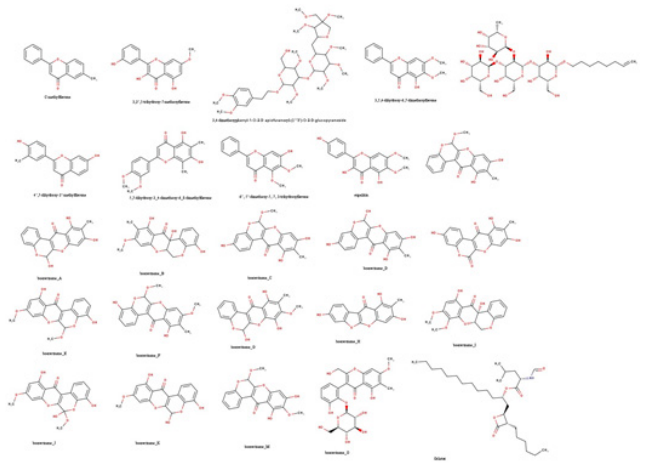

Figure 1. 2D structural representation of the phytochemicals from *B. diffusa* used in this study provided individual PubChem compound ID as the title.

four phytochemicals investigated in this work show binding energies of -6.0 to -8.0 Kcal/mol. In comparison, the marketed drug Orlistat taken as control has shown binding energy of -5.6 Kcal/mol.

The molecular interactions between Orlistat and Boeravinone-C compounds with pancreatic lipase were observed from the docking analysis. (Figure 2).

Prediction of pharmacological properties

Pharmacological properties of the phytochemicals were predicted using Osiris Property Explorer following Lipinski's Rule of Five and Bioavailability via Oral Administration. Expected pharmacological attributes are presented in Table 2.

The oral bioavailability of compounds can be considered by employing Veber's rule (49). Table 3 represents the toxicology profile predicted for the compounds from

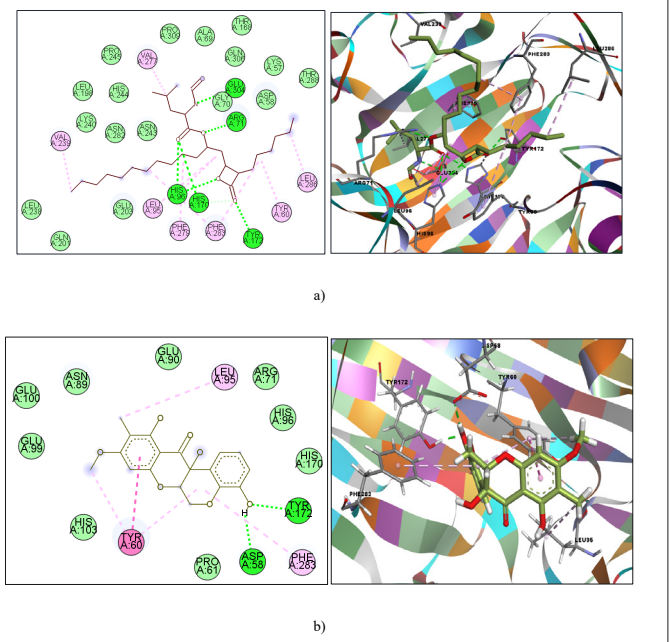

Figure 2. Molecular interactions were observed between a) Orlistat and b) Boeravinone-C compounds with pancreatic lipase from the docking analysis. The left panels represent the 2D molecular interactions network, whereas the right panels depict the molecular interaction in 3D representation.

Table 1. Virtual screening results of Phytochemicals targeting pancreatic lipase (PDB ID: 1NS8).

S.No	Phytochemical Name	Pubchem compound ID	Binding Energy (Kcal/mol)
1.	C-methylflavone	689013	-6.9
2.	3,3',5-trihydroxy-7-methoxy flavone	90963381	-7.0
3.	3,4-dimethoxy phenyl-1-O-β-D apiofuranosyl-(1''3')-O-β-D-glucopyranoside	132993749	-6.0
4.	3,5,4-dihydroxy-6,7-dimethoxyflavone	471722	-6.9
5.	3-O-β-D-galactopyranosyl-(1'''2'')-O-β-D-galactopyranoside	66679288	-6.4
6.	4',7-dihydroxy-3'-methyl flavone	90761690	-7.9
7.	5,7-dihydroxy-3_4-dimethoxy-6_8-dimethyl flavone	11002377	-7.3
8.	6', 5'-dimethoxy-5, 7, 3-trihydroxy flavone	5481647	-6.9
9.	eupalitin	5748611	-7.5
10.	boeravinone_A	14018346	-7.5
11.	boeravinone_B	14018348	-7.6
12.	boeravinone_C	13940641	-8.0
13.	boeravinone_D	15081178	-7.5
14.	boeravinone_E	11537197	-7.6
15.	boeravinone_F	12004175	-7.9
16.	boeravinone_G	11537442	-7.3
17.	boeravinone_H	16745324	-7.4
18.	boeravinone_I	16203334	-7.5
19.	boeravinone_J	16203335	-7.8
20.	boeravinone_K	71662492	-7.5
21.	boeravinone_M	102500671	-7.8
22.	boeravinone_O	71662493	-7.8
23.	boeravinone_P	102103085	-7.1
24.	boeravinone_Q	102103083	-7.3
25.	Orlistat	3034010	-5.6

Table 2. ADME properties of the phytochemicals based on Lipinski's rule of five.

S.No	Phytochemical Name	Molecular Formula	Molecular Weight	H-Donor	H-acceptor	No. of rotatable bonds	TPSA	LogP
1.	C-methylflavone	$C_{16}H_{12}O_2$	236.269	0	2	1	26.3	3.717
2.	3,3',5-trihydroxy-7-methoxy flavone	$C_{16}H_{12}O_6$	300.265	3	6	2	96.22	2.112
3.	3,4-dimethoxy phenyl-1-O-β-D apiofuranosyl-(1''3')-O-β-D-glucopyranoside	$C_{35}H_{58}O_{16}$	734.83	1	16	20	158.68	0.043
4.	3,5,4-dihydroxy-6,7-dimethoxyflavone	$C_{17}H_{14}O_5$	298.293	1	5	3	64.99	2.887
5.	3-O-β-D-galactopyranosyl-(1'''2'')-O-β-D-galactopyranoside	$C_{32}H_{56}O_{20}$	760.777	12	20	17	316.6	-3.830
6.	4',7-dihydroxy-3'-methyl flavone	$C_{16}H_{12}O_4$	268.267	2	4	1	66.76	3.025
7.	5,7-dihydroxy-3_4-dimethoxy-6_8-dimethyl flavone	$C_{19}H_{18}O_6$	342.346	2	6	3	85.22	3.229
8.	6', 5'-dimethoxy-5, 7, 3-trihydroxy flavone	$C_{17}H_{14}O_5$	298.293	1	5	3	64.99	2.887
9.	eupalitin	$C_{18}H_{14}O_6$	326.303	2	6	1	85.22	2.564
10.	boeravinone_A	$C_{17}H_{12}O_6$	312.276	3	6	0	96.22	2.136
11.	boeravinone_B	$C_{18}H_{16}O_7$	344.318	3	7	1	105.45	1.817
12.	boeravinone_C	$C_{18}H_{14}O_7$	342.302	3	7	1	105.45	2.218
13.	boeravinone_D	$C_{17}H_{12}O_7$	328.275	4	7	0	116.45	1.790
14.	boeravinone_E	$C_{17}H_{10}O_7$	326.259	3	7	0	113.29	1.575
15.	boeravinone_F	$C_{18}H_{14}O_7$	342.302	2	7	2	94.45	2.150
16.	boeravinone_G	$C_{19}H_{16}O_7$	356.329	2	7	2	94.45	2.494
17.	boeravinone_H	$C_{18}H_{14}O_7$	342.302	3	7	1	105.45	2.066
18.	boeravinone_I	$C_{16}H_{10}O_6$	298.249	3	6	0	100.13	3.476
19.	boeravinone_J	$C_{18}H_{16}O_7$	344.318	2	7	2	94.45	1.748
20.	boeravinone_K	$C_{18}H_{14}O_8$	358.301	3	8	2	114.68	2.455
21.	boeravinone_M	$C_{17}H_{12}O_7$	328.275	3	7	1	105.45	1.722
22.	boeravinone_O	$C_{18}H_{14}O_7$	342.302	2	7	2	94.45	2.150
23.	boeravinone_P	$C_{24}H_{26}O_{12}$	506.458	7	12	6	195.6	-0.345
24.	boeravinone_Q	$C_{16}H_{12}O_6$	300.265	3	6	2	96.22	2.112
25.	Orlistat	$C_{29}H_{53}NO_5$	495.742	1	6	23	81.7	7.618

B. diffusa.

Drug likeliness and docking studies for screening of best lead compound

Boeravinone-C was discovered to be a potential lead-like compound against pancreatic lipase because of its comparative highest negative ΔG binding energy and ADMET descriptor parameters. Established on the predicted pharmacological properties, all the investigated twenty-four phytochemicals showed promising pharmacological attributes. These phytochemicals were primarily consistent with Lipinski's rule, toxicology profile parameters, oral bioavailability, and Veber's rule. We have mapped their pharmacophore features to further understand the screened best compound, Boeravinone-C, compared to Orlistat (Figure 3).

MD simulations events analysis of Pancreatic lipase in its apostate; in complex with Orlistat and complex with Boeravinone-C compound

To further understand and validate the molecular level interactions, binding ability, and influence of Boeravinone-C in complex pancreatic lipase, we have performed 100 nanoseconds of molecular dynamic simulations. We have also completed two other simulations: one with pancreatic lipase in its apo form and the second with pancreatic lipase in complex with Orlistat. Root mean square deviation (RMSD) of the protein backbone and root mean initially analyzed square fluctuations (RMSF) of individual residues to understand the impact of compound binding on the protein conformation changes (Figures 4 & 5).

To further validate Boeravinone-C compounds' ability to inhibit pancreatic lipase, we have analyzed its energy throughout the simulated timescale. The analysis revealed that the energy of the Orlistat was maintained at an average of approximately -30 kcal/mol. At the same time, it found that the energy of the Boeravinone-C compound was to be keeping an average of approximately -60 kcal/mol (Figure 6).

Molecular interactions of Orlistat in complex with Pancreatic lipase observed during Molecular dynamic simulations

Schrodinger's Desmond software was used to detail

Table 3. Predicted toxicology profile of the compounds from *B. diffusa.*

S.No	Phytochemical Name	Pubchem compound ID	Mutagenic	Tumorigenic	Reproductive effect	Eye irritant
1.	3,4-dimethoxy phenyl-1-O-β-D apiofuranosyl-(1''3')-O-β-D-glucopyranoside	132993749	None	None	None	None
2.	3,5,4-dihydroxy-6,7-dimethoxyflavone	471722	None	None	None	None
3.	3-O-β-D-galactopyranosyl-(1'''2'')-O-β-D-galactopyranoside	66679288	None	None	None	High
4.	4',7-dihydroxy-3'-methyl flavone	90761690	None	High	None	None
5.	5,7-dihydroxy-3_4-dimethoxy-6_8-dimethyl flavone	11002377	None	High	None	None
6.	6',5'-dimethoxy-5, 7, 3-trihydroxy flavone	5481647	None	None	None	None
7.	eupalitin	5748611	None	None	None	None
8.	boeravinone_A	14018346	None	None	Low	Low
9.	boeravinone_B	14018348	None	None	Low	None
10.	boeravinone_C	13940641	None	None	None	None
11.	boeravinone_D	15081178	None	None	Low	Low
12.	boeravinone_E	11537197	None	None	Low	None
13.	boeravinone_F	12004175	None	None	High	None
14.	boeravinone_G	11537442	High	None	Low	Low
15.	boeravinone_H	16745324	None	None	Low	Low
16.	boeravinone_I	16203334	High	None	Low	None
17.	boeravinone_J	16203335	None	None	None	None
18.	boeravinone_K	71662492	None	None	High	None
19.	boeravinone_M	102500671	High	None	Low	Low
20.	boeravinone_O	71662493	High	None	Low	None
21.	boeravinone_P	102103085	None	None	Low	Low
22.	boeravinone_Q	102103083	None	None	Low	None
23.	Orlistat	3034010	None	None	None	None
24.	C-methylflavone	689013	None	None	None	None
25.	3,3',5-trihydroxy-7-methoxy flavone	90963381	High	None	None	None

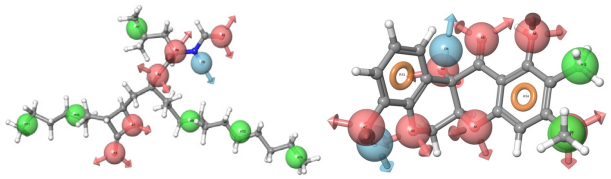

Figure 3. Pharmacophore features of a) Orlistat and b) Boeravinone-C compounds. Red spheres represent hydrogen acceptors; Cyan spheres represent hydrogen donors; green spheres represent hydrophobic regions; and circular donut shapes represent aromatic rings.

interpret molecular interactions of the pancreatic lipase and Orlistat complex. There found 22 contacts between Orlistat and Pancreatic lipase, which involved six contacts in hydrogen bonds. Hydrophobic interactions occurred at 13 contacts, and ten contacts in water bridging interactions were observed, respectively. Details are shown in Figure 7 of the molecular interaction profile of Orlistat with the Pancreatic lipase. Hydrogen bonds with Glu83; Ser110; Arg111; Thr112; Ile251 and Trp252. Hydrophobic interactions with Phe77; Tyr114; Leu153; Ala178; Pro180; Ile209; Phe215; Ile248; Ile251; Trp252, Phe258, Ala260 and Leu264. Water bridging interactions with Ile78; Asp79; Lys80; Glu83; Lys107; Ser110; Arg111; Thr112;

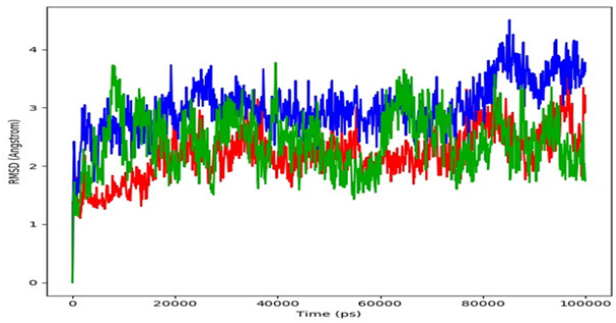

Figure 4. RMSD of the Pancreatic lipase backbone in its apostate (red), complex with Boeravinone-C (green), and complex with Orlistat (blue).

Trp252 and Glu253 were observed (Figure 7). Molecular interactions observed between Pancreatic lipase in complex with Orlistat during the 100 ns MD simulation timescale were studied.

Molecular interactions of Boeravinone-C in complex with Pancreatic lipase domain observed during Molecular dynamic simulations

When the Pancreatic lipase in complex with Boeravi-

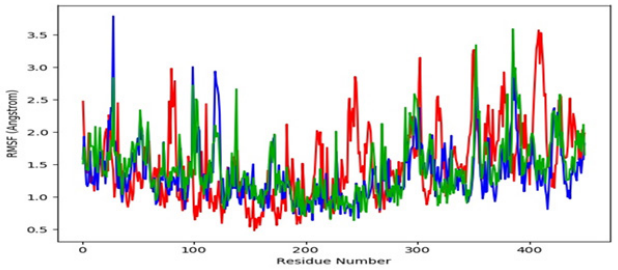

Figure 5. RMSF of the Pancreatic lipase backbone is complex with Boeravinone-C (green) and complex with Orlistat (blue).

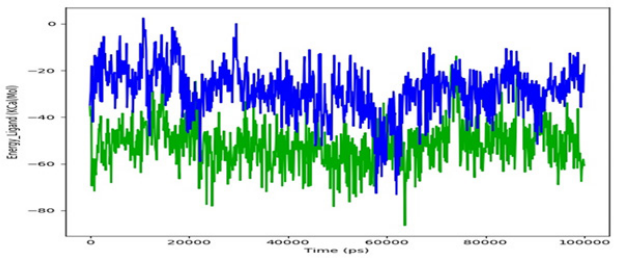

Figure 6. Calculated energy of the Orlistat (blue) and Boeravinone-C compounds inside the active binding site of Pancreatic lipase.

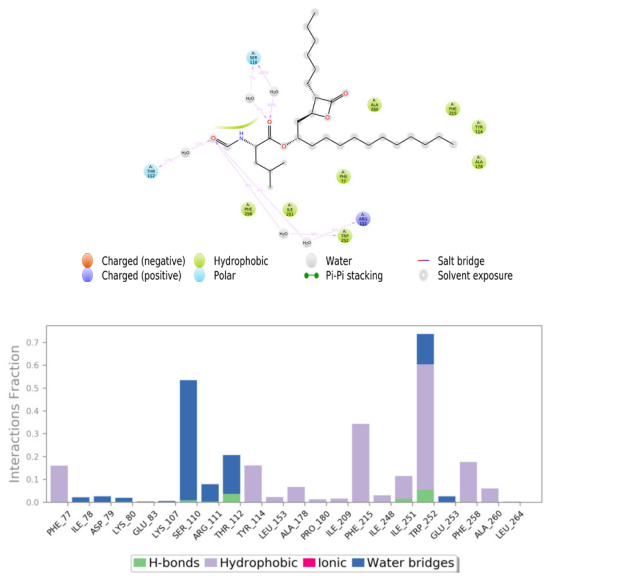

Figure 7. Molecular interactions were observed between the Pancreatic lipase domain in complex with Boeravinone-C during the 100 ns MD simulation timescale.

none-C compound was analyzed concerning the simulated timescale of 100 ns; there found 29 contacts between Boeravinone-C and Pancreatic lipase, out of which 14 associates were involved in hydrogen bonds, involved 12 contacts during hydrophobic interactivity, and 24 contacts in water bridging interactions were observed respectively. The molecular interaction profile of Orlistat with the Pancreatic lipase. Hydrogen bonds with Ile78; As79; Lys80; Glu82; Glu83; Lys107; Ser110; Arg111; Lys238; Ile251; Trp252; Glu253; Phe258 and Ala260. Hydrophobic interactions with Arg37; Phe77; Ile78; Lys80; Lys107; Arg111; Phe225; Lys238; Ile251; Trp252; Phe258 and Ala250. Water bridging in-teractions with His75; Gly76; Ile78; Asp79; Lys80; Glu82; Glu83; Asn84; Asp105; Trp106; Lys107; Ser110; Arg111; Thr112; Lys238; Ile251; Trp252; Glu253; Gly254; Arg256; Asp257; Phe258; Ala260 and Cys261 were observed. The molecular interactions observed between pancreatic lipase domains in complex with Boeravinone-C during the 100ns MD simulation timescale were examined.

Discussion

The computer-based software for studying the phytochemical ligand database has significantly reduced the time necessary for bioactivity assessments. The phytochemical ligand database from *B. diffusa* was docked against the pancreatic lipase. Out of the twenty-four phytochemicals, the virtual screening results of the evaluated phytochemicals observed that all of the compounds had shown better binding energies when compared to the marketed control drug Orlistat. The binding energies revealed that Boeravinone-C, with a binding energy of -8.0 Kcal/mol, was discovered as lead like molecule, compared to marketed Orlistat, which has shown -5.6 Kcal/mol of binding energy targeting pancreatic lipase. When the best conformations from the docking analysis of Orlistat and Boeravinone-C compounds were analyzed, Orlistat was noted to form a carbon-hydrogen bond with Arg111, pi-sigma bond with Trp252, and alkyl bonds with His75, Phe77 & Ile78. Whereas, Boeravinone-C was observed to develop a couple of pi-alkyl bonds with Arg111 and a solid covalent bond (Figure 2).

The pharmacokinetics (ADME) properties were investigated using Osiris Property Explorer following Lipinski's Rule of Five to predict the pharmacological properties of the twenty-four chosen ligands. We observed that phytochemicals, with the exceptions of 3,4-dimethoxy phenyl-1-O-β-D apiofuranosyl-(1’’3')-O-β-D-glucopyranoside and 3-O-β-D-galactopyranosyl-(1’’’2")-O-β-D-galactopyranoside. Lipinski's rule parameters fol-

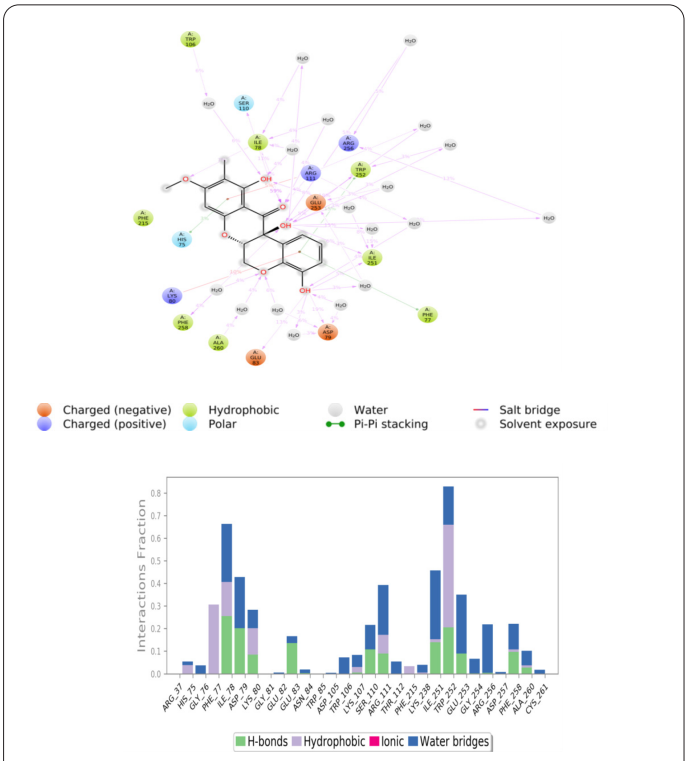

Figure 8. Molecular interactions were observed between pancreatic lipase domains in complex with Boeravinone-C during the 100 ns MD simulation timescale.

low the mol. Wt. It must be not more than 500 Da, Log P and the number of donor hydrogens must be below five, the required number of hydrogen acceptors be less than ten, and the refractivity molar range must be 40–130. The toxicity analysis result of compounds from *B. diffusa* is shown in Table 3. All the analyzed twenty-five compounds have mixed levels of toxicity for compounds; however, the compound Boeravinone-c has been predicted to be none toxicity for mutagenic, tumorigenic, and eye irritants but was noted to have high reproductive effect toxicity levels comparatively. The oral bioavailability of compounds can be considered by employing Veber's rule. Oral bioavailability was defined by low molecular weight (less than 500 Da); additionally, the number of rotatable bonds had to be less than ten, the total number of hydrogen bond acceptors and donors had to be less than twelve, and the complete polar surface area had to be less than 140. As shown in Table 2, all phytochemicals have favourable oral bioavailability.

The structure-based drug discovery is now an essential study for optimizing cost-efficient drugs. To screen the best compound for successful structure-based drug discovery, pharmacophore features of Orlistat (Figure 3a) and Boeravinone-C (Figure 3b) have been observed successfully. Our MD simulation study revealed that the RMSD of pancreatic lipase backbone kept an approximate average of 2 in its apostate, 3 in complex with Orlistat, and 2.2 Å̊s in complex with Boeranone-C correspondingly (Figure 4). Individual residues fluctuations were analyzed via RMSF; most residues active in the apostate were much minimized when complexes with compound, especially Boeravinone-C, compared to Orlistat. It was noticed that minimal fluctuations occurred when pancreatic lipase is complexed, indicating that the residues are pretty stable in the presence of the compound, with few peaks up to 3.5 A^{o} here and there, which are speculated to be caused by the loop movements (Figure 5). These restricted movements in the RMSD backbone and individual residual levels indicate that Boeravinone-C has much better inhibitory potential than Orlistat since the activity of the protein/enzyme mainly relies on its ability to undergo conformational changes. To further validate Boeravinone-C compounds' ability to inhibit pancreatic lipase, we have analyzed its energy throughout the simulated timescale. The analysis revealed that the energy of the Orlistat was maintained at an average of approximately -30 kcal/mol. At the same time, it found that the energy of the Boeravinone-C compound was to be keeping an average of approximately -60 kcal/mol (Figure 6). This much-minimized energy during the entire simulated timescale of about 100 ns is substantial proof that Boeravinone-C has better potential to stabilize its binding at the active site of the pancreatic lipase than the marketed drug Orlistat.

The molecular interactions of Orlistat in complex with Pancreatic lipase were observed during MD simulations. Throughout the simulated timescale, an average of 4 contacts from the interactions mentioned in the result section 3.5, Ser110, Phe215, and Trp252, were the most stable. Similarly, the molecular interactions of Boeravinone-C in complex with Pancreatic lipase domain observed during MD simulations showed that the average of 6 contacts from the interactions mentioned in the result section 3.6, Phe77; Ile78; Asp79; Ile251; Trp252, and Glu253 were found to be most stable among other interactions throughout the simulated timescale.

Conclusions

Our in silico studies demonstrated the ability of *B. diffusa* phytochemicals as valuable small ligand molecules. All the phytochemicals from *B. diffusa* have shown a significantly high binding affinity for pancreatic lipase for feasible antiobesity activity. All the phytochemicals under investigation in this study were noted with significant -ve binding energy via various molecular interactions. All or some of the molecular interactions were found with residues that fall under the active catalytic site, thereby increasing the thermodynamic stability of their complex. Molecular level interactions The knowledge gained from this current study is precious for future work related to computational screening of target-specific pancreatic lipase inhibitors. Instead, the promising ADMET drug-like profile of the currently investigated phytochemicals, particularly the Boeravinone-C compound, strongly supports further investigation into this phytochemical's ability to control obesity by targeting pancreatic lipase activity. The currently investigated phytochemical scaffold provides other changes that could result in more lead-like increased inhibitory activity and selectivity structures for pancreatic lipase.

Acknowledgments

The authors are thankful to the scientific research deanship at the University of Hail-Saudi Arabia for providing us the funding through project number RG 21146.

Interest conflict

The authors declare no conflict of interest.

Consent for publications

All the authors read and approved the final manuscript for publication.

Availability of data and material

All data generated during this study are included in this published article

Authors' Contribution

All authors had equal roles in study design, work, statistical analysis and manuscript writing.

Funding

This research has been funded by the scientific research deanship at the University of Hail-Saudi Arabia through project number RG 21146

Ethics approval and consent to participate

No humans or animals were used in the present research.

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