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Investigating Potential Antihypertensive Bioactive Agents In Hibiscus Sabdariffa Through Molecular Docking, Pharmacokinetic, And Admet Prediction Studies

Frank C. EGBA[1] and Ijeoma ESIABA[2]

[1-2] Babcock University, Department of Biochemistry, Ilishan-Remo, Ogun State, Nigeria

Emails: egbafrank72@gmail.com, esiabai@babcock.edu.ng

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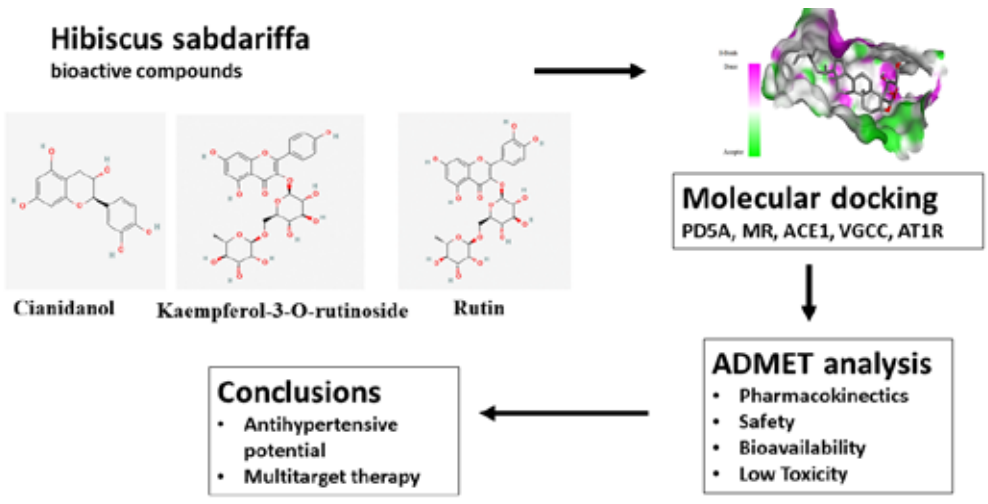
ABSTRACT:

Hypertension remains a critical global health concern, contributing significantly to cardiovascular morbidity and mortality. In different cultures, traditional remedies, such as plant-based therapeutics, have been widely employed to manage this condition. However, the limited scientific understanding of their mechanisms hinders their integration into standardized treatment frameworks. This study investigated the antihypertensive potential of Hibiscus sabdariffa (HS) by employing molecular docking and ADMET analyses to elucidate its bioactive compounds and their molecular interactions. Seventy-four compounds from HS were docked against five antihypertensive protein targets. ADMET and pharmacokinetic analyses were done on the top-ranking compounds. Molecular docking analysis revealed promising interactions between key bioactive compounds of HS, including kaempferol-3-O-rutinoside, beta-sitosterol 3-O-beta-D-galactopyranoside, cianidanol, and Rutin, and the following crucial antihypertensive targets: phosphodiesterase 5A (PDE5A), angiotensin II type 1 receptor (AT1R), angiotensin-converting enzyme I (ACE 1), mineralocorticoid receptor (MR), and voltage-gated L-type calcium channel (VGCC). The best docking scores for the receptors ranged from -9.4 to -10.6. Complementary ADMET analysis provided valuable insights into the pharmacokinetic properties and safety profiles of these compounds, underscoring their therapeutic potential. Notably, cianidanol exhibited favorable docking scores and pharmacokinetic attributes, including high bioavailability and low toxicity. These findings establish a molecular basis for the traditional use of Hibiscus sabdariffa as a multitarget therapy for the management of hypertension and support its potential development as a natural therapeutic agent. Future experimental studies are essential to validate and optimize these bioactive compounds for antihypertensive drug development.

KEYWORDS:

ADMET analysis, Bioactive compounds, Hibiscus sabdariffa, Hypertension, Molecular docking.

GRAPHICAL ABSTRACT



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1. INTRODUCTION

Hypertension, a cardiovascular disorder characterized by persistently elevated blood pressure, is a leading preventable cause of cardiovascular diseases and premature death worldwide. Affecting over one billion adults globally, it imposes a significant health burden, particularly in low- to middle-income countries, where nearly 46% of cases are undiagnosed, owing to its often asymptomatic nature [1-4].

Current treatment strategies typically combine lifestyle modifications, such as dietary changes and increased physical activity, with pharmaceutical interventions. However, the high cost and side effects of these medications have spurred interest in alternative natural therapies, which may offer safer, cost-effective solutions [5]. Among these natural therapies, *Hibiscus sabdariffa* (HS), known for its rich array of bioactive compounds, has emerged as a promising candidate [6].

Commonly referred to as Roselle, Jamaica sorrel, or red sorrel, HS is an annual, flowering subshrub from the *Malvaceae* family. Its vibrant red calyces (sepals) and reddish stems are widely recognized and cultivated across subtropical and tropical regions, including Nigeria, Mexico, and Thailand. Its long history of traditional use across various cultures highlights its potential health benefits [7-8].

Preclinical and clinical studies have shown that HS contains bioactive components that are beneficial for managing various health conditions, including hypertension, inflammation, and diabetes, while maintaining a favorable safety profile and minimal side effects [7, 9-10]. Its widespread availability, cost-effectiveness, and proven efficacy in hypertension management have contributed to its popularity as a natural therapeutic agent in diverse cultural and socioeconomic settings [6, 11].

Despite its long history of use in hypertension management, the specific phytochemicals responsible for its antihypertensive effects and its mechanisms of action are not fully understood. Bridging these knowledge gaps is crucial for integrating HS into standardized treatment protocols, as understanding its molecular interactions is key to confirming its therapeutic efficacy in hypertension management.

This study utilizes in silico methods — molecular docking and ADMET profiling — to examine the antihypertensive potential of bioactive compounds found in HS. By assessing the binding affinities, physicochemical properties, and toxicological properties of these compounds in the context of hypertension, this research contributes to the development of natural treatments for hypertension. Building on existing research that has highlighted the general therapeutic potential of HS [7-9], this study focused on identifying specific bioactive compounds to elucidate their molecular mechanisms in hypertension management. These findings will also help bridge the gap between traditional herbal medicine and modern drug discovery, potentially expanding treatment options for this prevalent health concern.

2. MATERIAL AND METHODS

2.1 Target Selection

All the targets used in this study were sourced from the *Open Targets platform* [12] (https://platform.opentargets.org/). Protein targets relevant to hypertension pathways and those with the highest potential for therapeutic intervention were selected. The X-ray crystallographic 3D structures of the selected targets were downloaded from the Research Collaboratory for Structural Bioinformatics (*RCSB*) online protein data bank repository [13] (https://www.rcsb.org/). The following protein targets were used: phosphodiesterase 5A (PD5A; PDB ID: 1xp0), angiotensin II type 1 receptor (AT1R; PDB ID: 4zud), angiotensin-converting enzyme I (ACE 1; PDB ID: 7z70), mineralocorticoid receptor (MR; PDB ID: 6gev), and voltage-gated L-type calcium channel (VGCC; PDB ID: 8we8).

2.2 Target preparation

Protein structures were prepared via the BIOVIA Discovery Studio visualizer. Water molecules were removed, and hydrogen atoms were added to stabilize the protein structures. The cocrystalized ligands were used to determine the binding sites of the targets before removal. Protein chain A, which contained the active site for each target, was used in the analysis.

2.3 Ligand Selection

The 74 ligands used in this study were sourced from the *PubChem* database [14] (https://pubchem.ncbi.nlm.nih.gov/). Silicon-containing compounds were excluded because of

incompatibility with the PyRx software used for molecular docking.

2.4 Ligand Preparation

The 2D/3D structures of the bioactive compounds used were retrieved from *PubChem*. *PyRx* was used in the 3D transformation, optimization, and energy minimization of the ligands to determine their most stable pose.

2.5 Molecular analysis

Docking was performed using the *AutoDock Vina* module within *PyRx*, followed by visualization of ligand–target interactions via *BIOVIA Discovery Studio*. The results obtained were ranked on the basis of their root mean square deviation values and binding energies in kcal/mol. Ligands with the lowest binding energies, indicative of strong target interactions and favorable poses, were selected for further analysis.

2.6 Setting the Grid Dimension for AutoDock Calculations

The grid was generated to define the position and size of the protein's active site for ligand docking. The search space coordinates were provided by *AutoDock Vina* via *PyRx*. The target's active site was determined via the position of the cocrystallized ligand at the binding site and validated via the web-based tool *Castp* [15] (http://sts.bioe.uic.edu/castp/index.html?1ycs). The grid settings used in the PyRx interface for the site-defined docking analysis are as follows:

Phosphodiesterase 5A: Center (Angstrom): X: -20.2738, Y: 31.8800, Z: 65.3799; Dimensions (Angstrom): X: 18.2444, Y: 20.8713, Z: 25.4134

Mineralocorticoid receptor: Center (Angstrom): X: 7.5056, Y: 16.5037, Z: 15.9444; Dimensions (Angstrom): X: 17.9585, Y: 20.3215, Z: 27.7966

Angiotensin converting enzyme 1: Center (Angstrom): X: 8.8189, Y: 4.4763, Z: 23.5860; Dimensions (Angstrom): X: 29.4904; Y: 34.9122; Z: 32.0535

Angiotensin II type 1 receptor: Center (Angstrom): X: -40.7332, Y: 67.5631, Z: 28.7825; Dimensions (Angstrom): X: 23.7163, Y: 18.0038, Z: 26.2902

L-type voltage-gated calcium channel: Center (Angstrom): X: 159.4740, Y: 166.1762, Z: 147.6636; Dimensions (Angstrom): X: 25.0000, Y: 25.0000, Z: 27.2547

Default settings were applied for all other parameters to ensure consistency across the analyses.

2.7 Docking Validation Protocol

To validate the docking protocol, the cocrystallized ligands for each target protein were extracted and re-docked into their respective binding sites using Discovery Studio 2021 and PyRx's *AutoDock Vina* module. The same docking parameters applied to the test compounds were used. The accuracy of redocking was assessed by calculating the root-mean-square deviation (RMSD) between the docked pose and the experimental crystallographic pose of the ligand. An RMSD ≤ 3.0 Å was considered indicative of a valid docking protocol.

2.8 Pharmacokinetic and Toxicity Analysis

The ADMET properties of the selected lead compounds were evaluated using in silico predictive models to assess their potential efficacy, safety, and bioavailability. The *SwissADME* web application (16) (http://www.swissadme.ch/) was used to assess the ADME properties of the compounds - lipophilicity (mean Log Po/w), aqueous solubility (via the estimated SOLubility (ESOL) model), human gastrointestinal absorption, and interactions with metabolizing enzymes. The drug likeness of the compounds was evaluated on the basis of bioavailability scores and adherence to Lipinski's Rule of Five. Compounds meeting these criteria are more likely to exhibit favorable oral bioavailability and therapeutic potential [16]. The *ProTox-III* online server [17] (https://comptox.charite.de/protox3/) was used to predict toxicological endpoints, including acute toxicity class, lethal dose (LD50), hepatotoxicity, carcinogenicity, mutagenicity, cytotoxicity, and immunotoxicity of the lead compounds.

3. RESULTS AND DISCUSSION

3.1 Molecular Docking Analysis

Hypertension continues to pose a significant global health challenge, with its prevalence and associated burden projected to escalate in the coming years. Several enzymes and receptors have been identified as key in the development and progression of hypertension and are often the targets of common antihypertensive medications. Effective modulation of these targets

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is essential in managing this prevalent condition [18]. The use of combination pharmacotherapy, which has demonstrated superior efficacy compared with single-pill therapy, may be hindered by its higher costs, increased risk of side effects, and complexity of multidrug regimens. Consequently, the exploration of alternative therapeutic strategies, particularly those derived from natural sources, is imperative. HS offers a promising avenue for the development of safe, cost-effective antihypertensive agents [5, 10].

This study elucidates the molecular interactions between key bioactive compounds from HS and hypertensive targets, revealing several compounds with binding affinities surpassing those of established drugs. These findings highlight the therapeutic potential of HS-derived compounds, particularly their capacity to modulate critical enzymes and receptors involved in hypertension pathophysiology.

3.1.1 Target 1: Phosphodiesterase 5A (PD5A)

Table 1 summarizes the docking scores for the top five bioactive compounds and the standard drug sildenafil against PD5A. Kaempferol-3-O-rutinoside, HS03, and Rutin showed similarly high predicted binding affinities (-10.2 to -10.1 kcal·mol^{-1}), while epigallocatechin gallate and quercitrin scored slightly lower (-10 to -9.6 kcal·mol^{-1}).

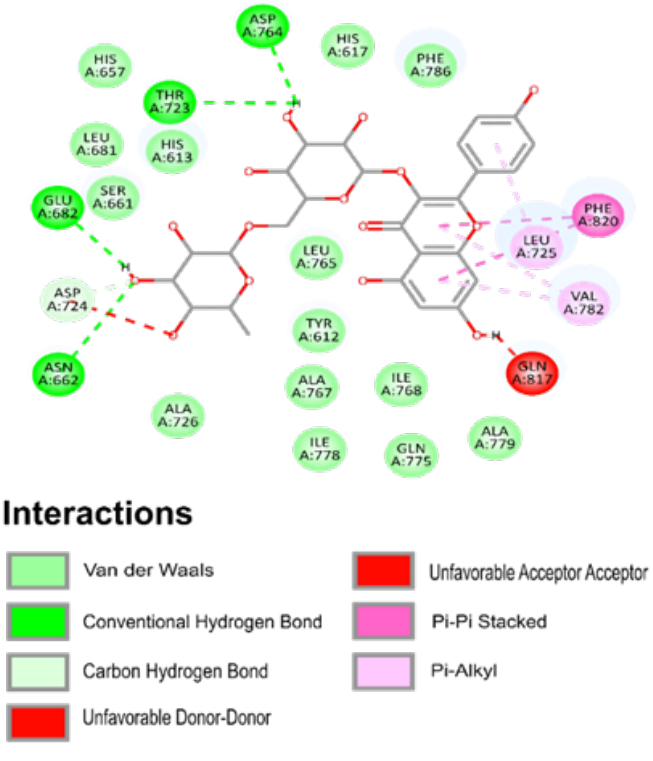
Kaempferol-3-O-rutinoside formed several hydrogen bonds with residues ASN662, GLU682, THR723, and ASP764 within the active site of the target. Pi–alkyl and pi–pi stacking interactions were also observed with the hydrophobic amino acid residues LEU725 and VAL782 and the aromatic amino acid residue PHE820 (Fig. 1). Taken together, the docking poses and interaction patterns suggest these compounds warrant further investigation as potential PD5A binders. The interaction between the reference drug, sildenafil, and PD5A is shown in Fig. 2.

The docking protocol reproduced the crystallographic binding pose of the co-crystalized ligand, Vardenafil, with an RMSD of 0 Å and binding affinity of −9.0 kcal/mol, which is within the acceptable cutoff for validation.

Table 1: Docking scores of the five top-scoring compounds of HS and standard drug against the antihypertensive target phosphodiesterase 5A (PD5A)

Compound (ligand)	Structural class	Docking score (Kcal/mol)
Kaempferol-3-O-rutinoside	Trihydroxyflavone	-10.2
HS03	Terpenoid	-10.1
Rutin	Tetrahydroxyflavone	-10.1
Epigallocate-chin Gallate	Flavans	-10
Quercitrin	Tetrahydroxyflavone	-9.6
Sildenafil (Standard)	PD5A Inhibitor	-9.6

HS03 = (3R,5S,9S,10R,13S,14R,17S)-17-[(E,2S,5S)-5-ethyl-6-methylhept-3-en-2-yl]-10,13-dimethyl-2,3,4,5,6,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol



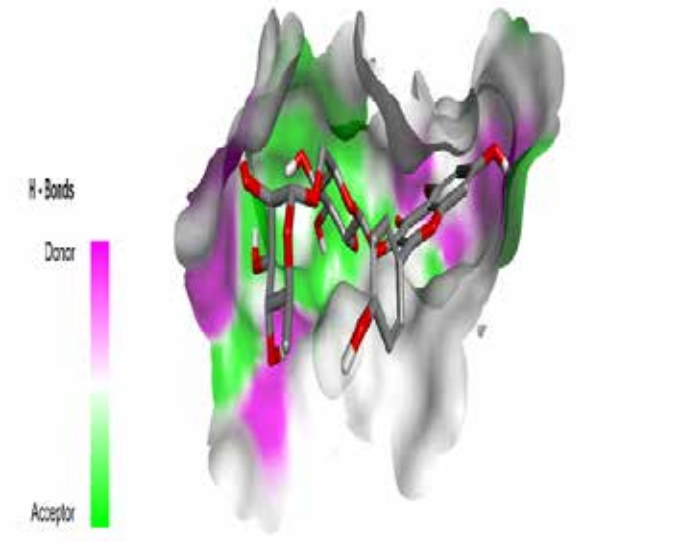


Figure 1: 2D (top) and 3D (bottom) interactions of kaempferol3-O-rutinoside with the phosphodiesterase A5 active site

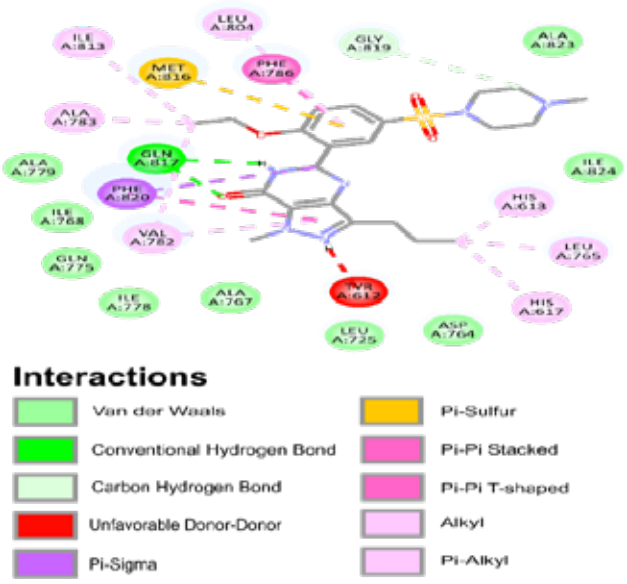


Figure 2: 2D ligand interaction of sildenafil (standard) with the phosphodiesterase A5 active site

PD5A is an enzyme involved in the degradation of cyclic guanosine monophosphate (cGMP) in vascular smooth muscle cells. The inhibition of this enzyme by HS-derived flavonoids, including kaempferol-3-O-rutinoside and structurally related compounds, could increase cGMP levels, promote vasodilation, and reduce blood pressure. These results are consistent with studies that highlighted the antihypertensive effects of kaempferol-3-O-rutinoside (19-21). This study demonstrates that kaempferol-3-O-rutinoside and similar HS compounds may serve as promising antihypertensive agents through PD5A inhibition.

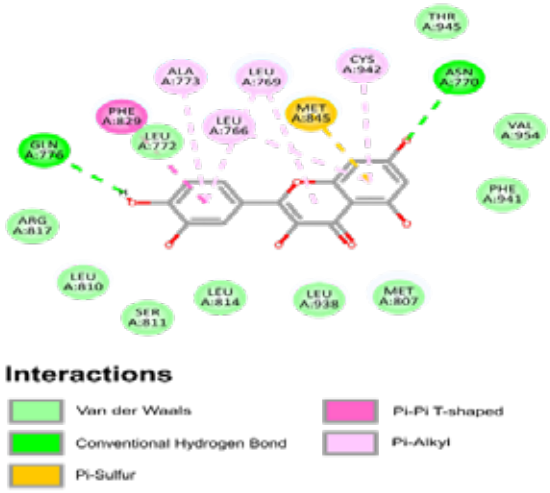
3.1.2 Target 2: Mineralocorticoid Receptor (MR)

Table 2 summarizes the docking scores for the top five bioactive compounds and the standard drug spironolactone against the MR. Quercetin, gossypetin, and cianidanol formed multiple hydrogen bonds with key residues GLN776 and MET807, alongside pi–sulfur interactions at MET845, pi–alkyl interactions with hydrophobic residues, and pi–pi stacking interaction with PHE829. These interactions, depicted in Figs. 3–5, contributed to their strong predicted binding affinity (–9.3 kcal/mol), which is comparable to that of spironolactone (–10.4 kcal/mol, Fig. 6). Therefore, these flavonoids may be considered to have a similar binding potential to the standard drug.

Redocking yielded a perfect overlap with the crystallographic pose for the co-crystalized ligand, RSCB ligand ID: EWN, with an RMSD of 0 Å, and a binding affinity of –12.2 kcal/mol, indicating excellent docking reliability.

Table 2: Docking scores of the five top-scoring compounds of HS and standard drug against the antihypertensive target mineralocorticoid receptor (MR)

Compound (ligand)	Structural class	Docking score (Kcal/mol)
Spironolactone (Standard)	MR antagonist	-10.4
Quercetin	Pentahydroxyflavone	-9.3
Gossypetin	Hexahydroxyflavone	-9.3
Cianidanol	Flavan-3-ols	-9.3
Hibiscetin	Flavonoids	-9.2
Apigenin	Trihydroxyflavone	-9.2



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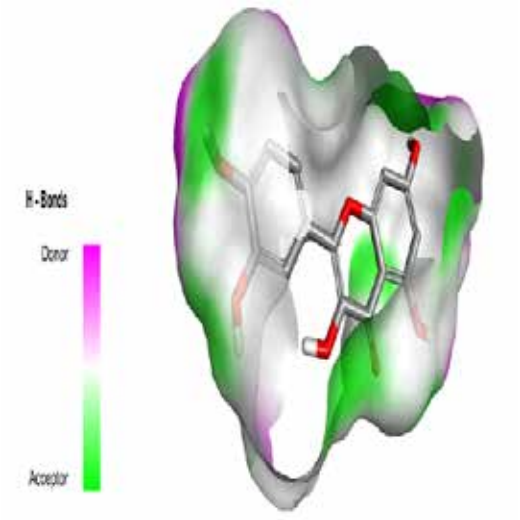


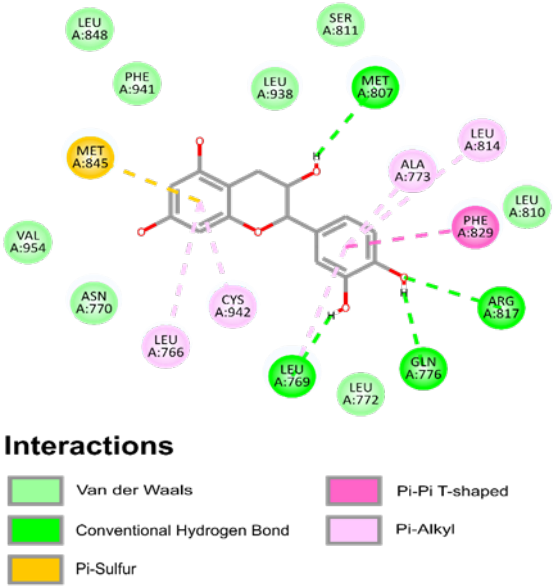
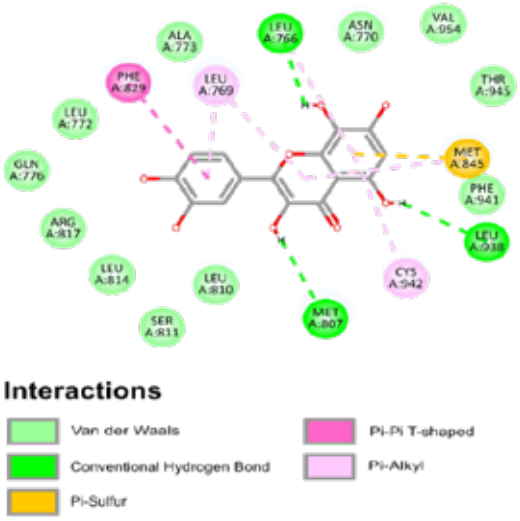


Figure 3: 2D (top) and 3D (bottom) interactions of quercetin with the MR active site



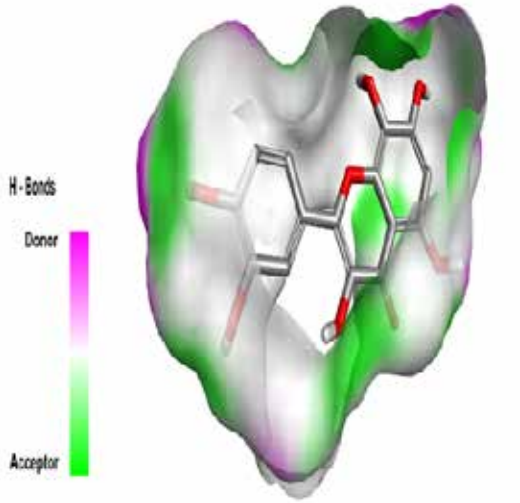


Figure 4: 2D (top) and 3D (bottom) interactions of gossypetin with the MR active site

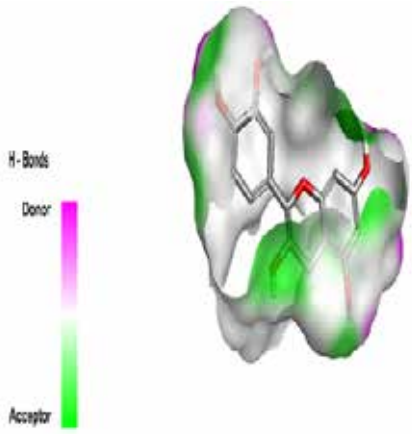


Figure 5: 2D (top) and 3D (bottom) interactions of cianidanol with the MR active site

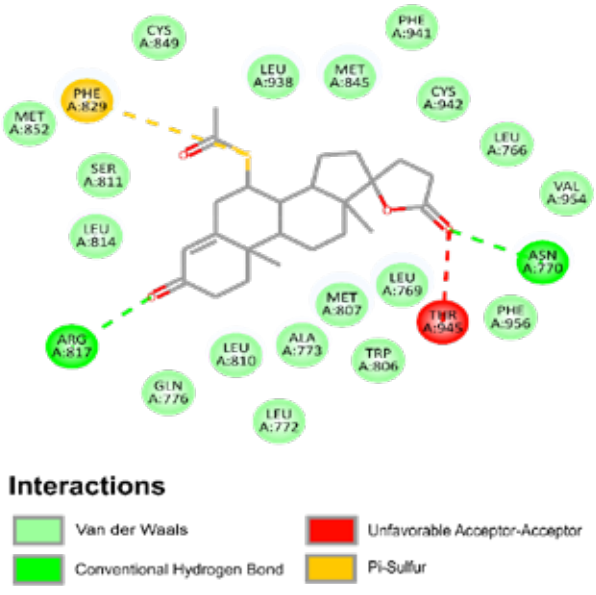


Figure 6: 2D ligand interaction of spironolactone (Standard) with the MR active site

The MR, a nuclear receptor central to sodium and water homeostasis, plays a key role in blood pressure regulation. This study revealed that quercetin, gossypetin, and cianidanol scored slightly lower than the standard drug spironolactone. However, these compounds exhibited structural similarities and favorable binding profiles, highlighting their potential as scaffolds for developing novel MR antagonists. The diverse biological activities of quercetin, including its antidiabetic, neuroprotective, and antihypertensive effects, are well documented [22-23]. The antihypertensive actions of quercetin have been attributed to multiple mechanisms, including attenuation of oxidative stress, modulation of the renin–angiotensin system, and improvement of endothelial function [24]. Similarly, cianidanol (catechin) has shown therapeutic potential in regulating lipid metabolism and stimulating nitric oxide production — mechanisms that collectively contribute to its antihypertensive effects [25-27].

Consistent with prior studies reporting the diuretic effects of quercetin and its analogs [28-29], our findings suggest that these compounds may modulate MR activity. By potentially influencing aldosterone-mediated sodium and water retention, these HS-derived flavonoids may contribute to the observed diuretic and antihypertensive effects of HS.

3.1.3 Target 3: Angiotensin II Type I Receptor (AT1R)

Table 3 summarizes the docking scores for the top five bioactive compounds and the standard drug Valsartan against AT1R. Several HS-derived compounds, including beta-sitosterol 3-O-beta-D-galactopyranoside, gossypol, and kaempferol-3-O-rutinoside, demonstrated high predicted binding affinities (−10.1 to −9.6 kcal/mol), which were comparable or slightly more favorable than valsartan (−9.0 kcal/mol). These binding affinities suggest that multiple structurally diverse HS compounds may serve as potential AT1R modulators. Beta-sitosterol 3-O-beta-D-galactopyranoside formed hydrogen bonds with key residues such as PRO19, ALA21, and ARG23, alongside pi–pi interactions with ILE288 and TRP84 (Fig. 7). These interactions suggest that multiple structurally diverse HS compounds may serve as potential AT1R modulators.

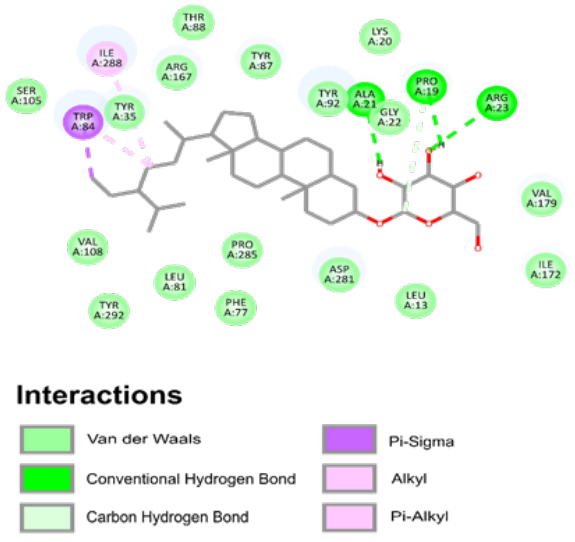
Redocking of the cocrystallized ligand, Olmesartan, yielded RMSD values of 2.62 Å (lower bound) and 3.87 Å (upper bound) for the best pose. The lower-bound RMSD indicates a near-native alignment, while the upper-bound value is slightly above the commonly used 3.0 Å threshold. Thus, the redocking result is borderline: a near-native pose was recovered, but conservative alignment metrics suggest some deviation from the crystallographic orientation. The resulting pose showed a molecular overlay RMSD of 0.94 Å compared to the crystallographic conformation via Discovery Studio.

Table 3: Docking scores of the five top-scoring compounds of HS against the antihypertensive target angiotensin II Type 1 receptor (AT1R)

Compound (ligand)	Structural class	Docking score (Kcal/mol)
Beta-Sitosterol 3-O-beta-D-galactopyranoside	Plant steroid	-10.1
Gossypol	Sesquiterpenes	-9.6
Kaempferol-3-O-rutinoside	Trihydroxyflavone	-9.6
HS03	Flavonoids	-9.6
HS05	Flavonoids	-9.6
Valsartan (Standard)	AT1R antagonist	-9.0

HS03 = (3R,5S,9S,10R,13S,14R,17S)-17-[(E,2S,5S)-5-ethyl-6-methylhept-3-en-2-yl]-10,13-dimethyl-2,3,4,5,6,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol

HS05 = 5-hydroxy-2-(4-methoxyphenyl)-8-(3-methylbut-2-enyl)-7-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy-3-[(2R,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxychromen-4-one



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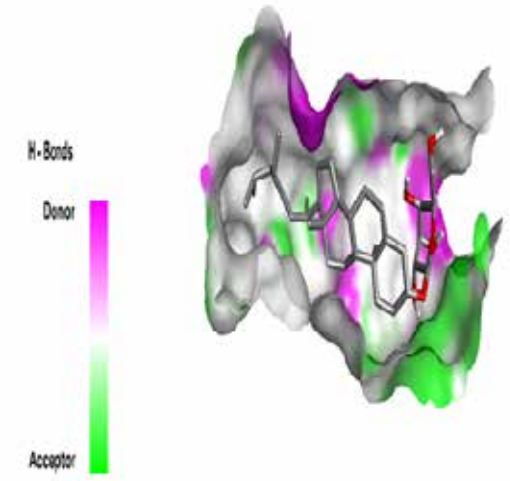

Figure7:2D(top)and3D(bottom)interactionsofbeta-sitosterol 3-O-beta-D-galactopyranoside with the AT1R active site

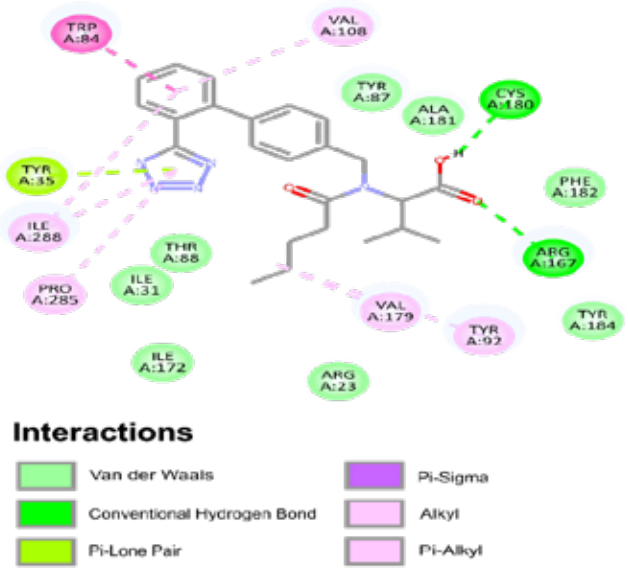

Figure 8: 2D ligand interaction of Valsartan (Standard) with the AT1R active site

AT1R is integral to hypertension pathophysiology, mediating vasoconstriction and sodium retention via the renin–angiotensin system. Several HS-derived compounds, including beta-sitosterol 3-O-beta-D-galactopyranoside, gossypol, and kaempferol-3-O-rutinoside, exhibited high predicted affinities for AT1R, suggesting potential for antagonistic activity. Such interactions could counteract angiotensin II-mediated effects and thereby contribute to blood pressure reduction. Notably, beta-sitosterol 3-O-beta-D-galactopyranoside has been reported to possess diverse biological activities, including antidiabetic and gastroprotective effects [30-31]. While its antihypertensive potential has been less explored, our findings suggest that this compound, alongside other HS phytochemicals with comparable docking profiles, may serve as promising candidates for AT1R-targeted therapies.

3.1.4 Target 4: Angiotensin-Converting Enzyme I

Table 4 summarizes the docking scores for the top five bioactive compounds and the standard drug lisinopril against ACE 1. Several flavonoids, including Rutin (−10.6 kcal/mol), kaempferol-3-O-rutinoside (−10.4 kcal/mol), and gossypitrin (−10.2 kcal/mol), demonstrated high predicted binding affinities, all more favorable than the standard drug lisinopril (−7.6 kcal/mol) (Fig. 10). For instance, Rutin formed hydrogen bonds with GLU162, THR282, and LYS511, along with a carbon–hydrogen bond with ASP377 (Fig. 9), contributing to its strong binding profile. These results suggest that multiple HS compounds may act as potential ACE1 inhibitors, offering scaffolds for the development of novel antihypertensive agents.

Redocking of the cocrystallized ligand, fosinoprilat, yielded RMSD values of 2.48 Å (lower bound) and 4.15 Å (upper bound) for the best pose. The lower-bound RMSD indicates a near-native alignment, while the upper-bound value is slightly above the commonly used 3.0 Å threshold. Thus, the redocking result is borderline: a near-native pose was recovered, but conservative alignment metrics suggest some deviation from the crystallographic orientation.

Table 4: Docking scores of the five top-scoring compounds of HS against the antihypertensive target angiotensin converting enzyme (ACE 1)

Compound (ligand)	Structural class	Docking score (Kcal/mol)
Rutin	Tetrahydroxyflavone	-10.6
Kaempferol-3-O-rutinoside	Trihydroxyflavone	-10.4
Gossypitrin	Flavonols	-10.2
Epigallocatechin Gallate	Flavans	-10.1
Quercitrin	Tetrahydroxyflavone	-9.8
Lisinopril (Standard)	ACE 1 inhibitor	-7.6

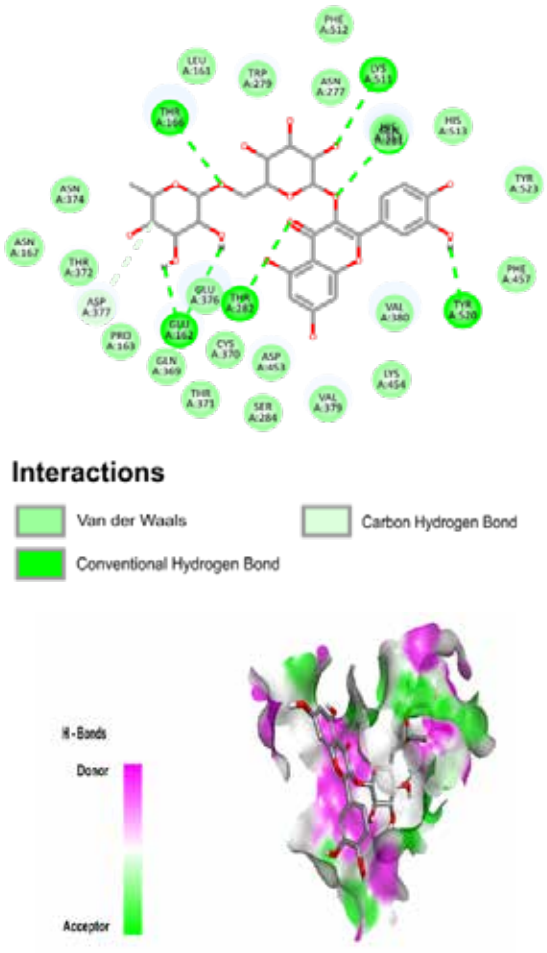

Figure 9: 2D (left) and 3D (right) interactions of Rutin with the ACE 1 active site

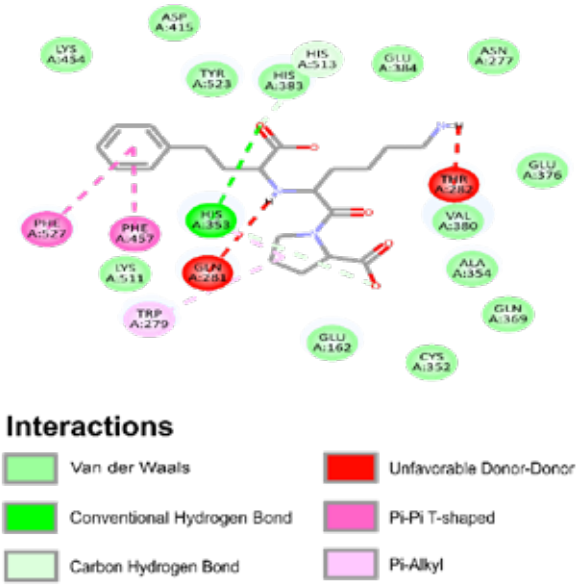

Figure 10: 2D ligand interaction of lisinopril (standard) with the ACE 1 active site

ACE 1 is a pivotal enzyme within the renin-angiotensin-aldosterone system that catalyzes the conversion of angiotensin I to the vasoconstrictive angiotensin II. Rutin exhibited a high predicted binding affinity for ACE 1, characterized by multiple hydrogen bonds and strong carbon–hydrogen bond interactions. These findings suggest that Rutin and structurally related compounds may act as potential ACE 1 inhibitors, thereby attenuating angiotensin II production and promoting blood pressure reduction. Rutin has been reported to exhibit various biological effects, including antioxidant, antidiabetic, anti-inflammatory, neuroprotective, and antihypertensive effects [30, 32]. Studies have linked its antihypertensive properties to mechanisms such as stimulation of the nitric oxide/guanylate cyclase pathway, ACE 1 inhibition, and antagonism of angiotensin II type 1 and mineralocorticoid receptors [32-34]. These actions are comparable to those of the known inhibitor lisinopril. Our findings further support Rutin's potential role as a multifaceted antihypertensive agent, with ACE1 inhibition forming a key mechanism underlying its efficacy.

3.1.5 Target 5: L-type Voltage-gated Calcium Channel (VGCC)

Table 5 summarizes the docking scores for the top five bioactive compounds and the standard drug amlodipine against VGCC. HS03 and beta-sitosterol 3-O-beta-D-galactopyranoside were the top-scoring compounds, each with a predicted binding affinity of −9.4 kcal/mol. Both formed multiple pi-interactions with hydrophobic residues, including VAL1053, ILE1046, MET1509, and PHE1513, within the active site. Beta-sitosterol 3-O-beta-D-galactopyranoside additionally engaged in two pi-interactions with PHE1181 and LEU1510 and a hydrogen bond with GLN1060, suggesting subtle differences in binding mode. These interactions, shown in Figs. 11 and 12, highlight their potential to modulate VGCC activity. The binding affinities of these compounds surpassed that of amlodipine, the known VGCC blocker (Fig. 13), which had a score of -6.9 kcal/mol.

Redocking of the VGCC cocrystallized ligand, amlodipine, did not reproduce the crystallographic pose within the docking protocol employed (closest RMSD = 3.071 Å). This likely reflects limitations of standard small-molecule docking for this ligand/target (e.g., large peptide ligand or induced-fit pocket).

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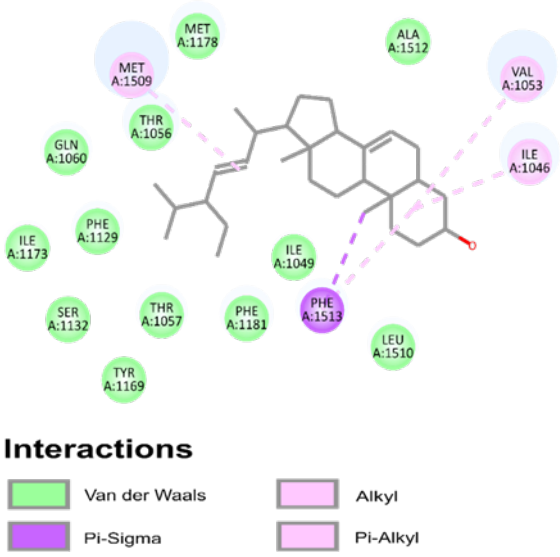
Table 5: Docking scores of the five top-scoring compounds of HS against the antihypertensive target VGCC

Compound (ligand)	Structural class	Docking score (Kcal/mol)
HS03	Terpenoids (Steroid)	-9.4
Beta-Sitosterol 3-O-beta-D-galactopyranoside	Plant steroid	-9.4
Cyanidin 3-(6''-acetyl-galactoside)	Anthocyanidin glycoside	-9.2
Epigallocatechin Gallate	Flavan	-9
HS07	Terpenoids (Steroid)	-8.9
Amlodipine (Standard)	VGCC Blocker	-6.9

HS03 = (3R,5S,9S,10R,13S,14R,17S)-17-[(E,2S,5S)-5-ethyl-6-methylhept-3-en-2-yl]-10,13-dimethyl-2,3,4,5,6,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol

HS07 = (3S,8S,9R,10R,13S,14S,17S)-10,13-dimethyl-17-[(2R)-6-methylheptan-2-yl]-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol



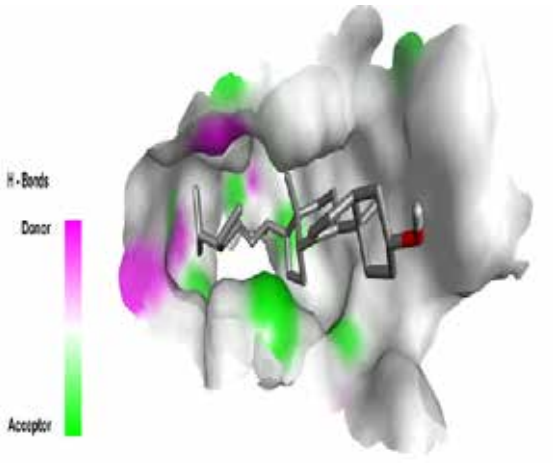


Figure 11: 2D (top) and 3D (bottom) interactions of HS03 with the VGCC active site

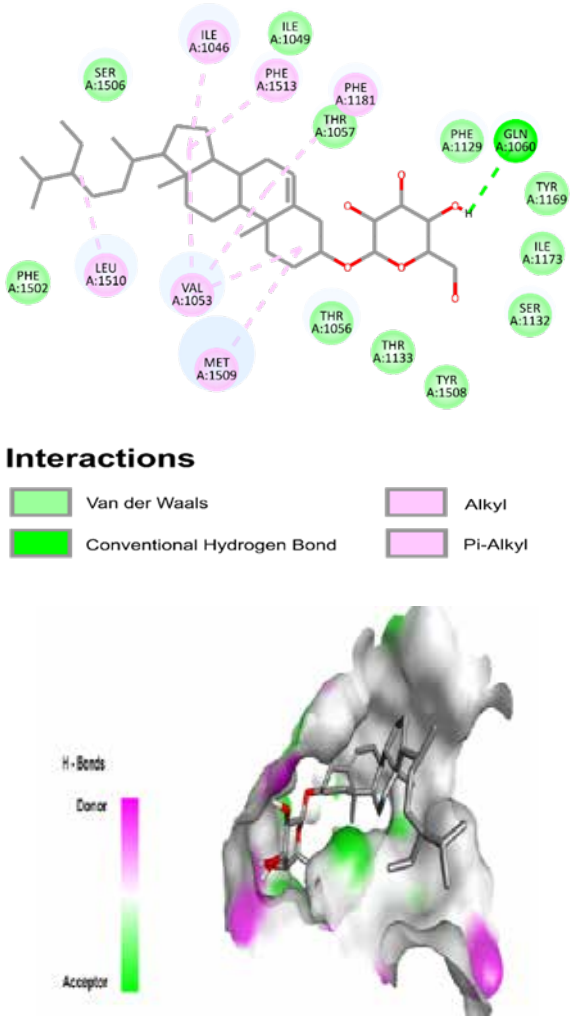


Figure 12: 2D (top) and 3D (bottom) interactions of beta-sitosterol 3-O-beta-D-galactopyranoside with VGCC active site

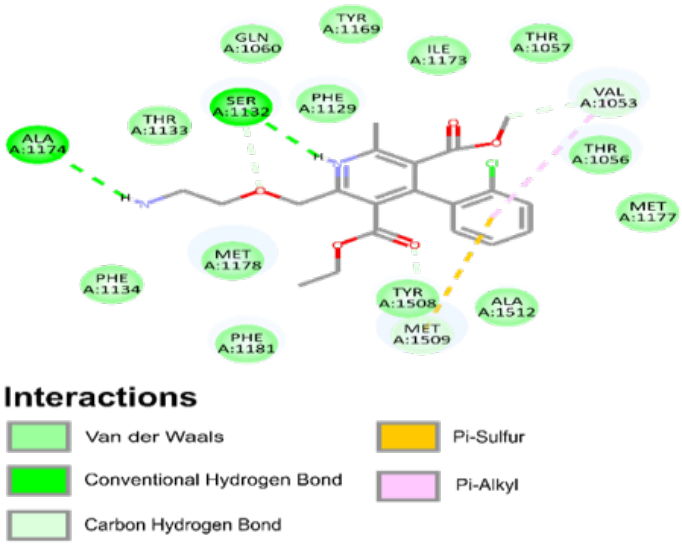


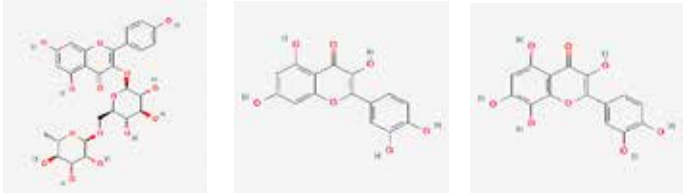
Figure 13: 2D ligand interaction of amlodipine (Standard) with VGCC active site

VGCC is a critical regulator of intracellular calcium levels and is essential for vascular smooth muscle contraction and cardiac function. HS03 and beta-sitosterol 3-O-beta-D-galactopyranoside exhibited significantly high predicted binding affinities for VGCC, with interactions primarily involving hydrophobic residues within the binding site. By modulating calcium influx, these compounds may induce vasorelaxation, contributing to the observed antihypertensive effects of HS. HS03, structurally similar to campesterol, has been previously reported to exhibit hypotensive effects, although the precise mechanisms remain unclear (35). Our findings indicate a potential role for HS03 and HS phytochemicals with comparable docking profiles in VGCC modulation, providing a plausible pathway for their antihypertensive effects. Similarly, beta-sitosterol 3-O-beta-D-galactopyranoside showed promise as a VGCC inhibitor, suggesting that a multifactorial mechanism underlies its observed antihypertensive effect.

Overall, the results of the molecular docking analyses revealed promising interactions between several bioactive compounds from HS and the selected antihypertensive targets. Kaempferol-3-O-rutinoside, beta-sitosterol 3-O-beta-D-galactopyranoside, and Rutin displayed notable binding affinities with phosphodiesterase 5A, angiotensin II T1 receptor, and angiotensin-converting enzyme 1, respectively. The formation of multiple hydrogen bonds and pi interactions suggests that these compounds are potential lead molecules. These findings provide molecular insights into the antihypertensive mechanisms of HS, which align with previous experimental observations [5-6, 10]. These findings also suggest a plausible molecular basis for the observed efficacy of *Hibiscus sabdariffa* in traditional medicine and could pave the way for the development of multitargeted natural therapeutics for hypertension management. The differences in docking scores of ≤1.0 kcal/mol fall within the margin of error for most docking algorithms. Therefore, compounds with similar scores were interpreted as having comparable predicted binding affinities, and emphasis was placed on groups of promising ligands rather than strict numerical rankings. It is important to acknowledge the limitations of in silico studies, including the simplifications and assumptions inherent in computational modeling. Experimental validation through *in vitro* and *in vivo* studies will be crucial to confirm these computational predictions. The complete docking scores and interaction details are presented in the Supplementary Tables.

3.2 Pharmacokinetic and Toxicological Evaluation

The top lead compounds (Fig. 14) presented varied pharmacokinetic profiles. Quercetin and cianidanol demonstrated high gastrointestinal absorption with no Lipinski violations, indicating favorable oral bioavailability. In contrast, glycosylated compounds such as Rutin and kaempferol-3-O-rutinoside were predicted to have poor absorption and multiple Lipinski violations due to their higher molecular weights and lower lipophilicity (Table 6). Toxicological analysis revealed that most of these compounds, including beta-sitosterol 3-O-beta-D-galactopyranoside, presented high safety margins with no significant cytochrome P450 (CYP450) enzyme inhibition. However, they were flagged for immunotoxicity, indicating potential risks to immune system function. Quercetin and gossypetin, despite their promising docking affinities, were identified as specific CYP450 inhibitors and flagged for potential carcinogenicity (Table 7).



Kaempferol-3-O-rutinoside Quercetin Gossypetin

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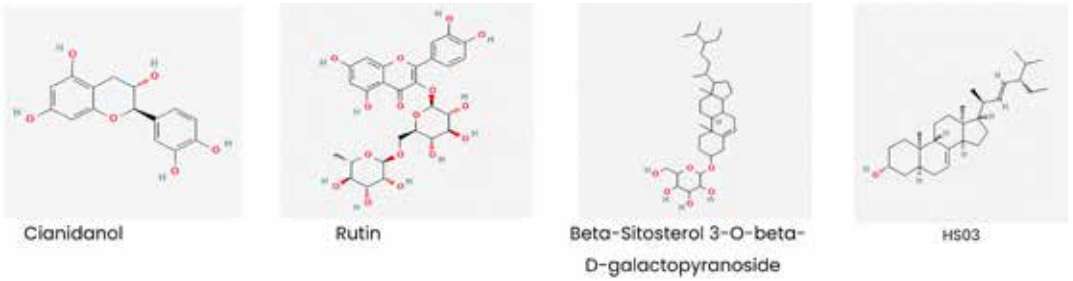

Figure 14: Chemical structure of the top-scoring compounds of Hibiscus sabdariffa against five selected antihypertensive targets

Table 6: Physicochemical Properties and Pharmacokinetics of top-scoring HS compounds

Compounds	Molecular Weight	Log Po/w	GI Absorption	Lipinski Violations	Bioavailability Score
Kaempferol-3-O-rutinoside	594.52 g/mol	-0.73	Low	3	0.17
HS03	412.69 g/mol	6.87	Low	1	0.55
Rutin	610.52 g/mol	-1.29	Low	3	0.17
Beta-Sitosterol 3-O-beta-D-galactopyranoside	576.85 g/mol	5.51	Low	1	0.55
Quercetin	302.24 g/mol	1.23	High	0	0.55
Gossypetin	318.24 g/mol	0.96	Low	1	0.55
Cianidanol	290.27 g/mol	0.85	High	0	0.55

Table 7: Toxicological and Metabolic Highlights of the top-scoring HS compounds

Compounds	Lethal Dose 50	CYP Inhibition	Notable Toxicity
Kaempferol-3-O-rutinoside	5000 mg/kg	None	Immunotoxicity
HS03	2000 mg/kg	None	Immunotoxicity
Rutin	5000 mg/kg	None	Immunotoxicity
Beta-Sitosterol 3-O-beta-D-galactopyranoside	8000 mg/kg	None	Immunotoxicity
Quercetin	159 mg/kg	CYP1A2, CYP3A4, CYP2D6 Inhibitor	Carcinogenicity, Immunotoxicity
Gossypetin	159 mg/kg	CYP1A2, CYP3A4, CYP2D6 Inhibitor	Carcinogenicity, Immunotoxicity
Cianidanol	10000 mg/kg	None	None

ADMET analysis revealed critical insights into the pharmacokinetic parameters of the bioactive compounds from *HS*. Despite their favorable binding affinities, lead compounds such as Rutin and kaempferol-3-O-rutinoside exhibited suboptimal gastrointestinal absorption, bioavailability, and multiple Lipinski violations. This could limit their efficacy *in vivo* by hindering their ability to reach therapeutic concentrations at their target site. While our results indicate potential bioavailability challenges for certain compounds, the literature suggests a more nuanced perspective. Ma et al. (2017) reported favorable gastrointestinal absorption permeability for kaempferol-3-O-rutinoside [36], and Ganeshpurkar and Saluja (2016) reported approximately 10% oral bioavailability for Rutin [37]. Rahman et al. (2021) further emphasized Rutin's robust biological activities [38]. These findings suggest that these compounds and their analogs may retain therapeutic potential despite challenges associated with their predicted bioavailability.

Interestingly, beta-sitosterol 3-O-beta-D-galactopyranoside and its analogs exhibited satisfactory bioavailability despite their poor gastrointestinal absorption. Strategies such as nanoparticle-based delivery, liposomal encapsulation, prodrug design, or cocrystals have been proposed to enhance the oral bioavailability of potential phytocompounds [39]. Incorporating such approaches may improve the translational potential of these promising compounds. In contrast, quercetin and its analogs demonstrated favorable pharmacokinetic properties, including high gastrointestinal absorption and bioavailability, making them strong candidates for further development as antihypertensive agents.

The low likelihood of CP450 enzyme inhibition for most compounds minimizes the risk of drug–drug interactions, a crucial factor in combination therapies. However, quercetin and its analogs were identified as potential CYP450 inhibitors, necessitating careful consideration in clinical applications.

Toxicological evaluation revealed generally acceptable acute toxicity profiles but raised concerns regarding potential immunotoxicity. These findings necessitate further investigation into their potential immunomodulatory effects and long-term safety. Quercetin and several of its analogs were identified as potential mutagens and carcinogens, highlighting the need for structural optimization to improve safety while preserving their therapeutic efficacy.

While this study provides a detailed assessment of individual bioactive compounds, the pharmacokinetic and safety profiles within the *Hibiscus sabdariffa* matrix may be influenced by complex synergistic interactions. These interactions warrant further investigation to fully understand the holistic therapeutic potential of this medicinal plant.

4. CONCLUSION

This in silico study provides evidence for the antihypertensive potential of HS through its ability to modulate multiple molecular targets associated with blood pressure regulation. The identification of key bioactive compounds, such as Rutin, kaempferol-3-O-rutinoside, and beta-sitosterol 3-O-beta-D-galactopyranoside, which have favorable binding affinities for critical enzymes and receptors, provides a robust foundation for the development of natural antihypertensive therapeutics. These findings underscore the potential for HS as a complementary or alternative therapeutic approach to conventional antihypertensive medications. While these results offer valuable molecular insights, further validation through *in vitro* and *in vivo* studies is essential to confirm their therapeutic efficacy and elucidate their detailed mechanisms of action. Moreover, addressing the pharmacokinetic challenges associated with certain compounds and optimizing their safety profiles will be crucial for the successful translation of these findings into clinical applications. By integrating computational methodologies with existing experimental evidence, this study represents a pivotal step forward in natural product-based drug discovery and cardiovascular research. Such interdisciplinary efforts hold promise for developing novel, multitargeted treatments for hypertension, ultimately improving patient outcomes and addressing the global burden of this condition.

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References

1. Carey RM, Moran AE, Whelton PK. Treatment of Hypertension. JAMA. 2022 Nov 8;328(18):1849.

2. Mohsen Ibrahim M. Hypertension in Developing Countries: A Major Challenge for the Future. Curr Hypertens Rep. 2018 May 1;20(5):38.

3. Oparil S, Acelajado MC, Bakris GL, Berlowitz DR, Cífková R, Dominiczak AF, et al. Hypertension. Nat Rev Dis Primers. 2018 Mar 22;4(1):18014.

4. Organization WH. Hypertension [Internet]. 2023. Available from: https://www.who.int/news-room/fact-sheets/detail/hypertension

5. Abdel-Rahman RF, Hessin AF, Abdelbaset M, Ogaly HA, Abd-Elsalam RM, Hassan SM. Antihypertensive Effects of Roselle-Olive Combination in L-NAME-Induced Hypertensive Rats. Oxid Med Cell Longev. 2017 Jan 22;2017(1).

6. Hopkins AL, Lamm MG, Funk JL, Ritenbaugh C. Hibiscus sabdariffa L. in the treatment of hypertension and hyperlipidemia: A comprehensive review of animal and human studies. Fitoterapia. 2013 Mar;85:84–94.

7. Da-Costa-Rocha I, Bonnlaender B, Sievers H, Pischel I, Heinrich M. Hibiscus sabdariffa L. – A phytochemical and pharmacological review. Food Chem. 2014 Dec;165:424–43.

8. Montalvo-González E, Villagrán Z, González-Torres S, Iñiguez-Muñoz L, Isiordia-Espinoza M, Ruvalcaba-Gómez J, et al. Physiological Effects and Human Health Benefits of Hibiscus sabdariffa: A Review of Clinical Trials. Pharmaceuticals. 2022 Apr 12;15(4):464.

9. Edo GI, Samuel PO, Jikah AN, Oloni GO, Ifejika MN, Oghenegueke O, et al. Proximate composition and health benefit of Roselle leaf (Hibiscus sabdariffa). Insight on food and health benefits. Food Chemistry Advances. 2023 Dec;3:100437.

10. Salem MA, Ezzat SM, Ahmed KA, Alseekh S, Fernie AR, Essam RM. A Comparative Study of the Antihypertensive and Cardioprotective Potentials of Hot and Cold Aqueous Extracts of Hibiscus sabdariffa L. in Relation to Their Metabolic Profiles. Front Pharmacol. 2022 Feb 23;13.

11. Jalalyazdi M, Ramezani J, Izadi-Moud A, Madani-Sani F, Shahlaei S, Ghiasi S. Effect of hibiscus sabdariffa on blood pressure in patients with stage 1 hypertension. J Adv Pharm Technol Res. 2019;10(3):107.

12. Ochoa D, Hercules A, Carmona M, Suveges D, Baker J, Malangone C, et al. The next-generation Open Targets Platform: reimagined, redesigned, rebuilt. Nucleic Acids Res. 2023 Jan 6;51(D1):D1353–9.

13. Berman HM. The Protein Data Bank. Nucleic Acids Res. 2000 Jan 1;28(1):235–42.

14. Kim S, Chen J, Cheng T, Gindulyte A, He J, He S, et al. PubChem 2025 update. Nucleic Acids Res. 2025 Jan 6;53(D1):D1516–25.

15. Tian W, Chen C, Lei X, Zhao J, Liang J. CASTp 3.0: computed atlas of surface topography of proteins. Nucleic Acids Res. 2018 Jul 2;46(W1):W363–7.

16. Daina A, Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep. 2017 Mar 3;7(1):42717.

17. Banerjee P, Kemmler E, Dunkel M, Preissner R. ProTox 3.0: a webserver for the prediction of toxicity of chemicals. Nucleic Acids Res. 2024 Jul 5;52(W1):W513–20.

18. Khan MAH, Imig JD. Antihypertensive Drugs. In: Reference Module in Biomedical Sciences. Elsevier; 2018.

19. Mahobiya A, Singh TU, Rungsung S, Kumar T, Chandrasekaran G, Parida S, et al. Kaempferol-induces vasorelaxation via endothelium-independent pathways in rat isolated pulmonary artery. Pharmacological Reports. 2018 Oct;70(5):863–74.

20. Ren J, Lu Y, Qian Y, Chen B, Wu T, Ji G. Recent progress regarding kaempferol for the treatment of various diseases (Review). Exp Ther Med. 2019 Aug 13;

21. Shahlehi S, Azizi A, Tengah A, Amdani SN, Petalcorin MIR. Anti-hypertensive vasodilatory action of Gynura procumbens mediated by kaempferol 3-O-rutinoside. F1000Res. 2020 Oct 12;9:1226.

22. Salehi B, Machin L, Monzote L, Sharifi-Rad J, Ezzat SM, Salem MA, et al. Therapeutic Potential of Quercetin: New Insights and Perspectives for Human Health. ACS Omega. 2020 May 26;5(20):11849–72.

23. Yang D, Wang T, Long M, Li P. Quercetin: Its Main Pharmacological Activity and Potential Application in Clinical Medicine. Oxid Med Cell Longev. 2020 Dec 30;2020:1–13.

24. Serban M, Sahebkar A, Zanchetti A, Mikhailidis DP, Howard G, Antal D, et al. Effects of Quercetin on Blood Pressure: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. J Am Heart Assoc. 2016 Jul 6;5(7).

25. Chen XQ, Hu T, Han Y, Huang W, Yuan HB, Zhang YT, et al. Preventive Effects of Catechins on Cardiovascular Disease. Molecules. 2016 Dec 21;21(12):1759.

26. Elbarbry F, Jones G, Ung A. Catechin Reduces Blood Pressure in Spontaneously Hypertensive Rats through Modulation of Arachidonic Acid Metabolism. Molecules. 2022 Dec 2;27(23):8432.

27. Mangels DR, Mohler ER. Catechins as Potential Mediators of Cardiovascular Health. Arterioscler Thromb Vasc Biol. 2017 May;37(5):757–63.

28. Aghababaei F, Hadidi M. Recent Advances in Potential Health Benefits of Quercetin. Pharmaceuticals. 2023 Jul 18;16(7):1020.

29. Susilowati A. Diuretic Effect of the Aqueous Extract of Green Tea Leaves. In: Proceedings of the Third International Conference on Sustainable Innovation 2019 – Health Science and Nursing (IcoSIHSN 2019). Paris, France: Atlantis Press; 2019.

30. Al-Dhabi NA, Arasu MV, Park CH, Park SU. An up-to-date review of rutin and its biological and pharmacological activities. EXCLI J [Internet]. 2015;14:59–63. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4614038/

31. Pandey J, Dev K, Chattopadhyay S, Kadan S, Sharma T, Maurya R, et al. β-Sitosterol-D-Glucopyranoside Mimics Estrogenic Properties and Stimulates Glucose Utilization in Skeletal Muscle Cells. Molecules. 2021 May 24;26(11):3129.

32. Bazyar H, Zare Javid A, Ahangarpour A, Zaman F, Hosseini SA, Zohoori V, et al. The effects of rutin supplement on blood pressure markers, some serum antioxidant enzymes, and quality of life in patients with type 2 diabetes mellitus compared with placebo. Front Nutr. 2023 Aug 3;10.

33. GANGA RAJU M, PREM PRASAD GOUD, SUVARCHALA REDDY NVL. ANTIHYPERTENSIVE EFFECT OF RUTIN: PHARMACOLOGICAL AND COMPUTATIONAL APPROACH. Asian Journal of Pharmaceutical and Clinical Research. 2019 Jun 14;87–92.

34. Oyagbemi AA, Bolaji-Alabi FB, Ajibade TO, Adejumobi OA, Ajani OS, Jarikre TA, et al. Novel antihypertensive action of rutin is mediated via inhibition of angiotensin converting enzyme/mineralocorticoid receptor/angiotensin 2 type 1 receptor (ATR1) signaling pathways in uninephrectomized hypertensive rats. J Food Biochem. 2020 Dec 21;44(12).

35. Ghaedi E, Foshati S, Ziaei R, Beigrezaei S, Kord-Varkaneh H, Ghavami A, et al. Effects of phytosterols supplementation on blood pressure: A systematic review and meta-analysis. Clinical Nutrition. 2020 Sep;39(9):2702–10.