

In-Silico Evaluation of Bombax malabaricum Phytoconstituents: A Molecular Docking Approach to Novel Diuretic Agents

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Abstract

Bombax malabaricum flowers possess significant medicinal potential, particularly as a natural diuretic. Phytochemical analyses reveal that the flowers contain flavonoids, alkaloids, tannins, and phenolic compounds, which collectively contribute to a wide range of pharmacological activities, including diuretic, antioxidant, anti-inflammatory, antimicrobial, and hepatoprotective effects. These properties position Bombax malabaricum as a promising complementary or alternative option to conventional synthetic diuretics, especially in the management of conditions such as hypertension, heart failure, renal dysfunction, and fluid retention. Compared to standard diuretics, the plant's bioactive constituents may offer a less dose-dependent mechanism with potentially fewer adverse effects, including improved electrolyte balance. The synergistic actions of its phytoconstituents further enhance renal clearance and reduce oxidative stress, supporting overall physiological function. While traditional use of the plant is well-established, modern pharmacological validation underscores its relevance in contemporary therapeutic practices. Continued research and clinical trials are essential to standardize dosage, verify efficacy, and ensure safety. Overall, Bombax malabaricum represents a valuable natural candidate for developing safer and more holistic diuretic therapies.

Key Words: Bombax malabaricum, Phytochemical, flavonoids, diuretic

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Introduction to the Plant and Its Flower

Bombax malabaricum or the Malabar cotton tree (*Bombax malabaricum*) is a big and strong tree native to some portion of India and Southeast Asia. It is especially known by its vivid red flowers which blossom in groups, hence a remarkable presence in the places it is found as it is the tropical and subtropical environment. The tree is a member of the Malvaceae that also has kapok tree and grows in the forests where the soil is well drained. The use of the plant has been elaborate in traditional medicine because of the abundance of the bioactive compounds contained in the plant that makes up its therapeutic potential. The medicinal properties of the flowers of *Bombax malabaricum* are particularly where it has been used against several conditions including infections and even fluid retention.

Aim

The current research aims for preparing the ethanolic extract of the *Bombax malabaricum* and conducting in vitro antioxidant analysis and in silico molecular docking experiments to examine its diuretic action.

Objectives

To collect, authenticate and process the flowers of *Bombax malabaricum* for extraction. To prepare the ethanolic extract of *Bombax malabaricum* using standardized extraction techniques. To conduct preliminary phytochemical screening of the extract to identify key bioactive constituents. To evaluate the in vitro antioxidant activity of the ethanolic extract using established assays (e.g., DPPH, ABTS). To perform in silico molecular docking studies of selected phyto constituents against diuretic targets (e.g., carbonic anhydrase, Na^+/K^+ channels). To interpret the docking results in terms of binding affinity and interaction profiles for assessing potential diuretic activity. To correlate the antioxidant and docking findings with the known pharmacological properties of *Bombax malabaricum*.

MATERIALS AND METHODOLOGY

Reagents

DPPH - (1,1-diphenyl-2-picrylhydrazyl), Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), Fluorescein sodium salt, gallic acid, (+)-catechin, Folin Ciocalteu reagent, ascorbic acid standard and 2,2'-Azobis(2-methylpropionamidine) dihydrochloride (AAPH) were purchased. All chemicals and reagents were of analytical grade.

Plant material

The dried flowers of *Bombax malabaricum* were collected from hills area of sathyamangalam. The dried flowers of *Bombax malabaricum* were powdered with a steel blender passed through a 35-mm (42-mesh) sieve and were kept at -20°C in the dark until use.

Preparation of extracts

The extraction was performed individually with 500 mL water, 50% ethanol (ethanol:water, 50:50 v/v), or 80% acetone (acetone:water, 80:20, v/v) with slight revisions on each 50 g powered sample of the *Bombax malabaricum* flower. The samples were sonicated using a bath sonicator (20 min) and then steadily shaking (150 rpm) was carried out at 30°C at 24 h. This was followed by centrifugation of the extracts using an Eppendorf 5804R refrigerated bench top centrifuge at 10 000 g (10 min). This was centrifuged and the supernatant discarded and the pellet which remained was repeated by adding fresh solvents. This slurry was clarified and re-collected and evaporated to dryness under vacuum at 40°C to remove the solvents and then dried by a vacuum freeze-drier. The extracts were frozen at -20°C till the time of analysis. Extracts were dissolved in concentration required before the removal of the sample.

Determination of TPC

Folin Ciocalteu colorimetric procedure with little changes was used to measure TPC in extracts. In short, the prepared sample of the 10 mg extracts which had already been dissolved with 10 mL methanol/ water (5:5, v/v) solution and exactly 0.5 mL were thoroughly mixed with 2 mL ddH₂O and 0.5 mL of Folin | Ciocalteu reagent. The mixture was placed in a beaker and after 6 min, 5 mL of sodium carbonate (Na_2CO_3), 70 g/L was added, as well as 4 mL deionized water. The reaction mixture was incubated and allowed to rest at room temperature in a period of 90 min after which the absorbance may be read at 760 nm. Furthermore, there should be a sample that is made up of extract solution, but it should be treated the same way to cover any back ground occasioned by the use of methanol. The findings were amenable to comparison with the calibration curve of the gallic acid standard that had nine data points ranging between 0- 200 $\mu\text{g/mL}$. TPC was given in mg GAE/g sample.

Determination of the TFC

The TFC was measured using an aluminium colorimetric test. Different solutions of a standard solution of (+) -catechin at levels of 0, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100 mg/L were prepared by dissolving (+) -catechin in ddH₂O. A 1.0 mg/mL stock solution or standard (+)-catechin solutions (35 μL) were pipetted with 140 μL dd H₂O in 96 well plate. Once 10.5 μL of 5 percent sodium nitrite (NaNO_2) is added, it was incubated at 25°C for 5 min then 10.5 μL of 10 percent aluminium chloride

(AlCl₃) was added. 6 min later, it was allowed to reach 350 µL by adding 70 µL 1 M NaOH and ddH₂O. The absorbance was measured based on the ddH₂O as a blank at 490 nm. TFC was cited as mg catechin equivalents (CE)/g sample.

Determination of anthocyanin content

The acidic reaction which measures the anthocyanin content (AC) of *Bombax malabaricum* flower extracts was determined by the pH differential method. The findings were referred to as mg cyanidin-3-glucoside equivalents (CGE)/100 g of sample.

DPPH radical-scavenging activity

The powder of each extract was dissolved in 1 mL of methanol in ten milligrams. The above original stock solution (10 mg/mL) was diluted further serially with methanol to obtain a series of concentrations of 0-450 µg/mL. 50 µL of these stock solutions were thereafter added to 250 µL 0.005 percent methanol DPPH solution which was added in 96 well plates. The DPPH solution was made freshly per day. The decrease in absorbance was determined at 490 nm at an end point of 30 min of incubation at 25 °C in the dark in a 1420 multilabel counter. The purple solvent of DPPH became yellow due to the pairing of its single electron with a hydrogen atom originating in potential antioxidant. Methanol and ascorbic acid were set as the negative and positive control respectively. The percentage of DPPH free radical- quenching activity was determined from the formula:

$$\text{DPPH scavenging activity (\%)} = (1 - (A_1 - A_2)/A_0) * 100$$

where A₀ the absorbance of the methanol solution of DPPH but absent the extract, A₁ (DPPH + extract) absorbance and A₂ the absorbance of the extract alone. DPPH radical-scavenging activity can be presented as a 50 percent inhibition concentration (IC₅₀), i.e. a concentration of an extract or a pure compound needed by 50 percent of inhibition of DPPH radicals within the defined time frame. The less the value of IC₅₀, the greater the antioxidant activity. The repeatability of each sample was 6 times.

Statistical analysis

All data were reported as the means ± SD for at least three replications. Analysis of variance (ANOVA) and Pearson's correlation coefficients were performed to compare the data using SAS 8.0 software package.

In-Silico Molecular Docking Studies

Protein Preparation

The 3-D structure of the targeted protein Diuretics (PDB ID: 1Z9Y) i.e. the human carbonic anhydrase II in complex with furosemide sulfonamide inhibitor was obtained at the Protein Data Bank (PDB) (www.rcsb.org/pdb). In order to examine the structural characteristics as well as the active sites of these proteins, we have used CASTP (Computed Atlas of Surface Topography of Proteins) and PyMOL molecular visualization tool. Under this strategy, the surface properties of the proteins and possible places of interaction could be well studied. According to Lipinski rule of five, drug creation prospects necessitate the substance to be well-regarded candidates with respect to the accompanying properties: a molecular weight of less than zero to 500 g/mol, a log P partition coefficient of not more prominent than five, less than hundred hydrogen bond acceptors (principally N and O), not more than five hydrogen bond donors (OH and NH groups, by way of example) and not more than ten rotatable bonds. The necessities are needed in ensuring the proper oral bioavailability and pharmacokinetics of the medication candidates. Docking studies were thus conducted on substances which met this criterion and the affinity of these substances in binding into protein was assessed. We are hoping of identifying potential candidates through a rigorous screening process and be able to hit the defective diuretics proteins that are related to the diuresis and possibly originate new treatment methods.

Ligand Preparation

The GC-MS analysis carried out earlier by researcher identified the major phyto constituent that occurs inside both the species and this is the one that is causing the effect of diuretic activity and the 3D models of the compounds are draw using Pubchem database.

Molecular Import and Preparation

The file of the molecular structures was read and separated into the appropriate parts (ligands, cofactors, water and proteins) and these structures where then automatically prepared. MVD helped to locate the bonds, aromaticity, distribute charges and contribute explicit hydrogen. The cavities were located using the built-in cavity detector and thus the search space was able to be limited in the most interesting regions.

Docking

The molecular docking programs provide an accurate prediction of the interaction of a small molecule (ligands) with a target protein (enzyme or receptor). The software that we have used here is Molegro Virtual Docker. MVD scoring function of the docking bases on Piecewise Linear Potential (PLP). As shown in every ligand docking, the optimum orientation of the ligand protein complex was

evaluated and hydrogen bonds were also found and annotated. To inspect and analyze the ligand energy, MVD score, which is the linear combination of electrostatic interaction and hydrogen bonds, was used. Any valence checking and the addition of H atoms were carried out. The search algorithm conducted the differential evolution drooping process and made use of these cavities holes detected by the cavity detection algorithm to direct further the search, to that particular region, when the docking stimulation took place. The warnings caused by unlikely preparations and missing structural information (e.g. unknown residues) were considered and the errors were also tried to be corrected. The poses ranked at the top were used but reranking procedure was used to further enhance the docking accuracy.

Analysis

With the use of pose organizer, the poses returned by the docking engine were seen. Pose organizer was able to dynamically load poses to a docking run enabling the ligand to be browsed over thousands of ligands. Other energy terms and interactions were checked at the same time and more sophisticated reranking and binding affinity scores were computed.

The hydrogen bond and the electrostatic interaction were unrenewed on swapping between poses. The selected ligand acquired by MolDock score was compared with the standard drug. The ligands with highest MolDock score were assigned as the best ligands and a potential lead molecule in Diuretic activity.

RESULTS AND DISCUSSION

Extraction Yield

The extraction yields of extractable compounds using different solvents are summarized in Table 1. Water (WE), 50% ethanol (EE50) and 80% acetone (AE80) extracts prepared from 50 g *Bombax malabaricum* flower gave a yield of 258.4, 287.2 and 198.9 mg extractable compounds (EC) per gram of dried flower, respectively.

Total Phenolic Content

Table 1 shows that all extracts of *Bombax malabaricum* flower exhibited noticeable amounts of phenolic compounds. The TPC of different extracts were assayed by the Folin and Ciocalteu method using gallic acid as a standard ($y = 0.0041x - 0.00412$, $R^2 = 0.9975$). The highest TPC of 138.12 ± 0.45 mg GAE/g was obtained in AE80, whereas the lowest TPC of 62.89 ± 1.15 mg gallic acid equivalents (GAE)/g was achieved in WE. The TPC of *Bombax malabaricum* flower extracts was much higher than that reported for cranberry ($153.7 \mu\text{mol/g}$, equivalently 27.94 mg GAE/g) and comparable with that of black teas (82.3–145.1 mg GAE/g).

Total Flavonoid Content

The TFC of different extracts from *Bombax malabaricum* flower was assayed by aluminum colorimetric assay using (+)-catechin as a standard ($y = 0.0035x + 0.00421$, $R^2 = 0.9927$). The data presented in Table 1 indicated that the highest flavonoid content of 92.11 ± 14.88 mg catechin equivalents (CE)/g was obtained in AE80, whereas the lowest flavonoid content of 41.81 ± 3.52 mg CE/g was obtained in WE. In the present study, it was noted that AE80, which was abundant in phenolics, was also abundant in flavonoids (which contribute to the TPC). In addition, the ratio of total flavonoid compounds to the total phenolic compounds (TFC/TPC) ranged from 58.22 to 71.47% (Table 1), showing that flavonoids were the major phenolic components in the extracts.

Anthocyanin Content

As shown in Table 1, no anthocyanin compounds were found in the AE80 and EE50. However, WE contained a rather low amount of anthocyanins of 79.21 ± 5.32 mg CGE/100 g. Generally, there are some anthocyanins present in fresh flowers of *Bombax malabaricum*. Niranjana and Gupta (1973) isolated two anthocyanidin glycosides, pelargonidin 5- β -D-glucopyranoside and cyanidin-7 methylether-3- β -glucopyranoside from the methanol hydrochloric acid (1%) extract of fresh *Bombax malabaricum* petals. However, because *Bombax malabaricum* has a short flowering time, the flowers after dropping are usually collected and air-dried for storage in traditional Chinese medicinal methods. Therefore, the low AC in the extracts of dried flowers from *Bombax malabaricum* might be ascribed to the thermal decomposition of anthocyanins during the air-dried treatment. All these results of TPC, TFC and AC mentioned earlier indicated that the extract yields and recovery of antioxidant compounds from the plant materials were strongly dependent on the nature of the extracting solvent. It is clear that different solvents used for the extraction of plant antioxidant compounds have different abilities due to the varied chemical characteristics and polarities of different antioxidant compounds. Polar solvents are frequently employed for the recovery of polyphenols from a plant matrix. The most suitable of these solvents are aqueous mixtures containing ethanol, methanol, acetone and ethyl acetate. Meanwhile, water is also a widely used solvent, which can extract the major components in vegetables and plants. In the present study, 80% acetone and 50% ethanol showed excellent antioxidant compound extraction yields from *Bombax malabaricum* flower, which was in agreement with the reports that aqueous ethanol and aqueous acetone had good recovery of antioxidant compounds from plant materials.

DPPH Radical-Scavenging Activity

DPPH radical-scavenging activity of nine different concentrations of extracts from *Bombax malabaricum* flower was investigated. With the increasing concentration of EE50 from 0 to 450 µg/mL, the scavenging rate of DPPH radical presented a dose-dependent manner ($y = 0.1751x + 8.1391$, $R^2 = 0.9753$). Furthermore, DPPH radical-scavenging activity was also linearly correlated with the concentrations of AE80 and ascorbic acid in the range of 0 to 150 µg/mL and WE in the range of 0 to 350 µg/mL, respectively. The corresponding correlation coefficients were 0.9801, 0.9895 and 0.9964 for AE80, ascorbic acid and WE, respectively. But the scavenging rate of DPPH radical presented a plateau after reaching 87% in the AE80 or ascorbic acid additional test.

Flower Extracts

Table 1: Extraction Yield and Antioxidant Content of *Bombax malabaricum*

Solvent	Extraction Yield (mg EC/g of dried flower)	Total Phenolic Content (TPC) (mg GAE/g)	Total Flavonoid Content (TFC) (mg CE/g)	Anthocyanin Content (mg CGE/100 g)	DPPH Radical-Scavenging Activity
Water (WE)	258.4	62.89 ± 1.15	41.81 ± 3.52	79.21 ± 5.32	86% at 350 µg/mL
50% Ethanol (EE50)	287.2	108.45 ± 0.89	67.33 ± 9.64	0 (No anthocyanins)	86% at 450 µg/mL
80% Acetone (AE80)	198.9	138.12 ± 0.45	92.11 ± 14.88	0 (No anthocyanins)	87% at 150 µg/mL

- Anthocyanins were present only in the water extract (WE), but no anthocyanins were found in AE80 and EE50.

- TPC was assayed using the Folin-Ciocalteu method with gallic acid as standard.
- TFC was determined using the aluminum colorimetric assay with (+)-catechin as standard.
- DPPH radical-scavenging activity was measured at various concentrations (0– 450 µg/mL), with the activity showing dose-dependent behavior and plateauing at 86% for AE80 and ascorbic acid.

In-Silico Molecular Docking Studies

The basis of MolDock lies in the new heuristic search algorithm which integrates cavity-prediction with a differential evolution. An adaptation of the docking scoring function MolDock, the docking scoring function is a modification of what is known as the piecewise linear potential (PLP) this new docking scoring function introduces new terms involving electrostatic and hydrogen bonding. One of the determining parameters in analysing the docking results is the MolDock Score. The ligand molecules were evaluated using MolDock score, rerank score and hydrogen bond interactions. The ligand that has the lowest MolDock score has a high affinity to the target. Table 2 reveals the docking results of the selected ligands against diuretic protein respectively.

Table 2: Ranking of Ligands and poses against human carbonic anhydrase II in complex with furosemide as sulfonamide inhibitor based on MolDock score

Protein: 1Z9Y

Ligand Number	Ligand Name	MolDock Score	Rerank Score	H - Bond
222284	Beta-Sitosterol	-146.996	-93.4745	-2.15592
5281647	Mangiferin	-113.213	-102.553	-6.47906
5994	Luteol	-106.232	-84.4066	-2.5
3440	Furosemide (Standard)	-107.171	-88.995	-6.44131

2D, 3D and Secondary Interaction between selected ligands and Protein

Beta-Sitosterol Vs 1Z9Y

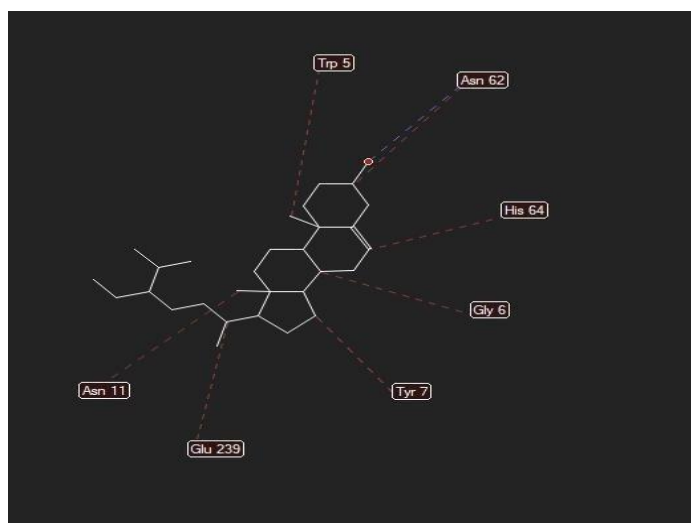


Figure 1: 2D interaction of Beta-Sitosterol with 1Z9Y Protein

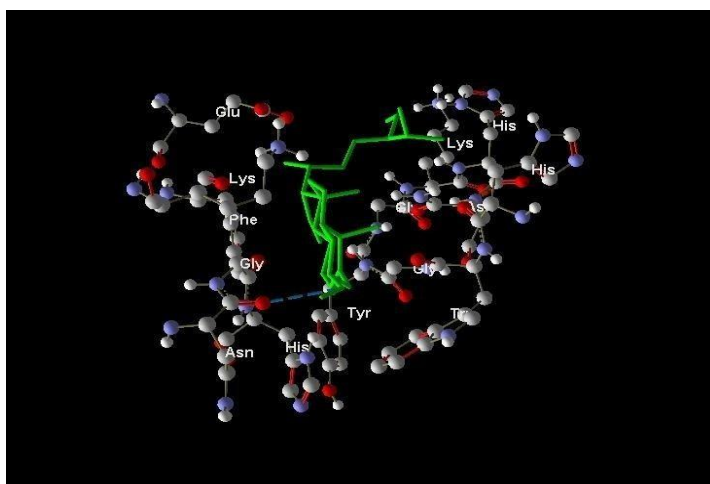


Figure 2: 3D interaction of Beta-Sitosterol with 1Z9Y Protein

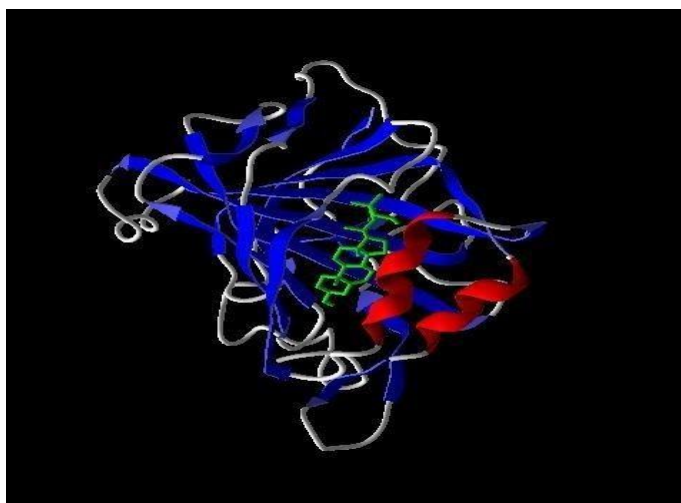


Figure 3: Secondary interaction of Beta-Sitosterol with 1Z9Y Protein

Mangiferin Vs 1Z9Y

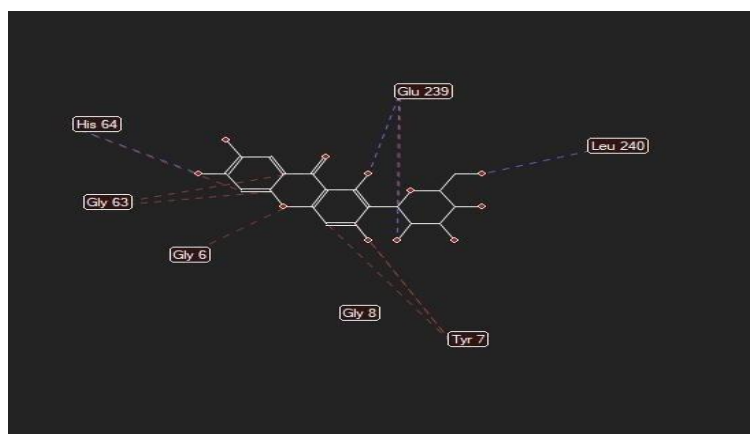


Figure 4: 2D interaction of Mangiferin with 1Z9Y Protein

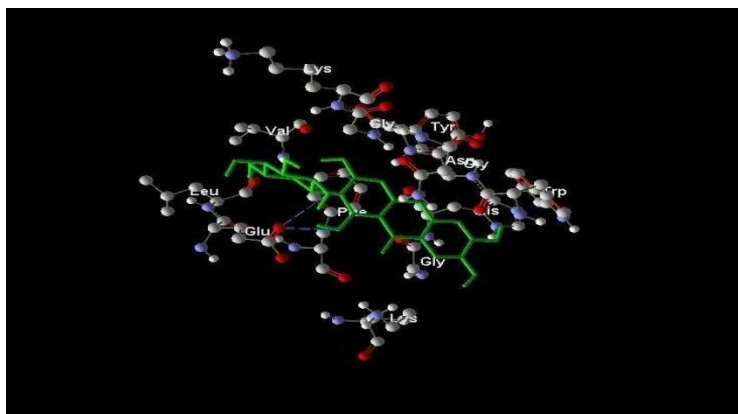


Figure 5: 3D interaction of Mangiferin with 1Z9Y Protein

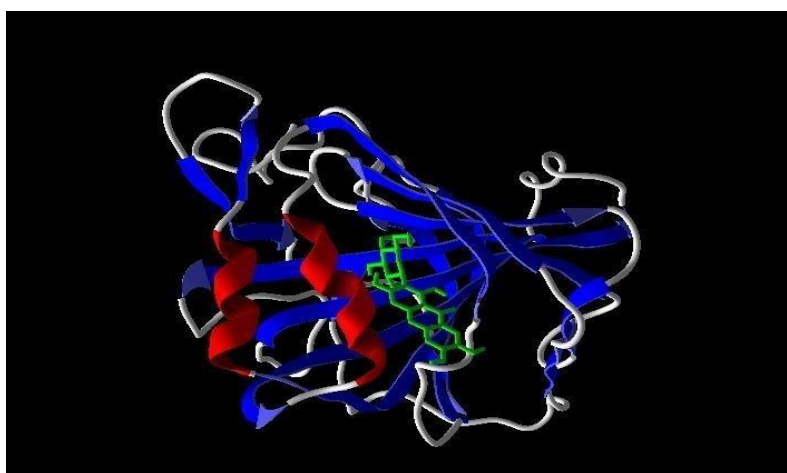
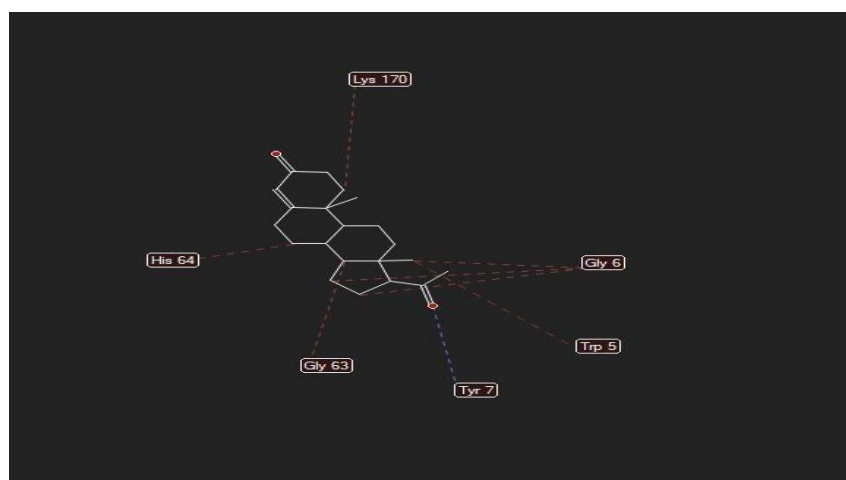
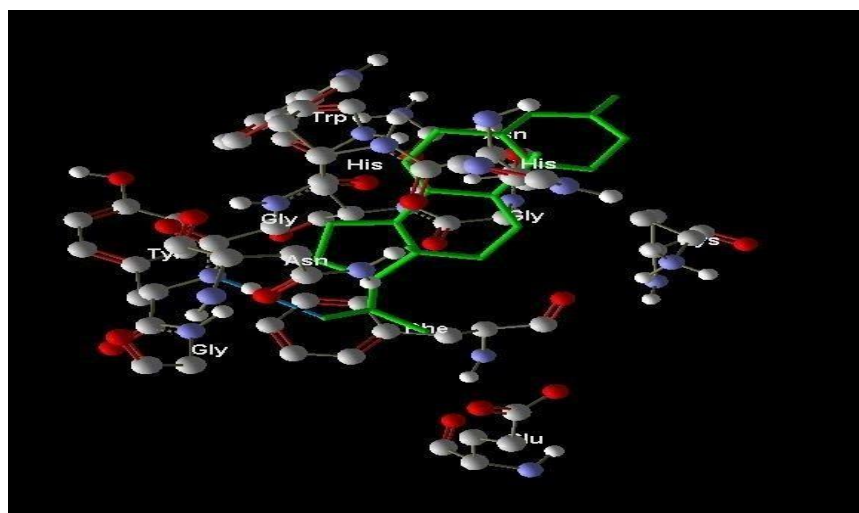


Figure 6: Secondary interaction of Mangiferin with 1Z9Y Protein

Luteol Vs 1Z9Y**Figure 7: 2D interaction of Luteol with 1Z9Y Protein****Figure 8: 3D interaction of Luteol with 1Z9Y Protein**

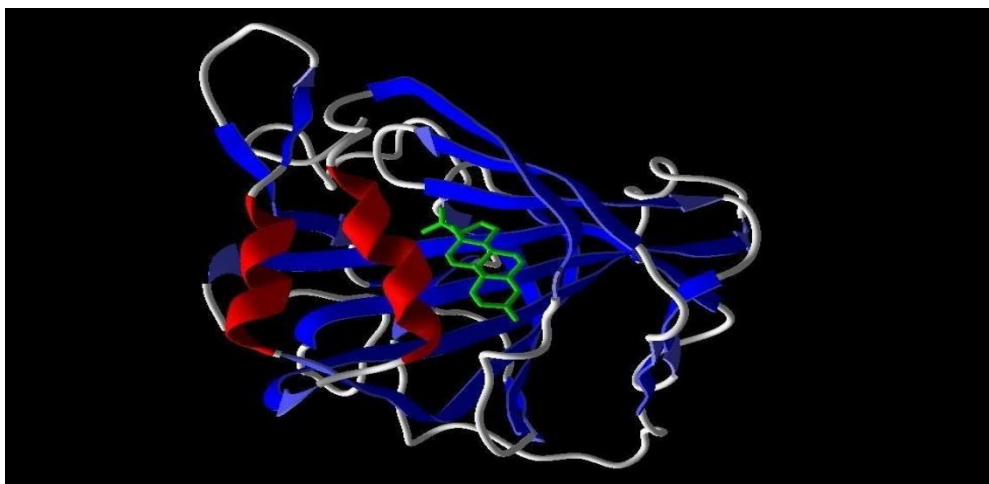


Figure 9: Secondary interaction of Luteol with 1Z9Y Protein

Furosemide Vs 1Z9Y

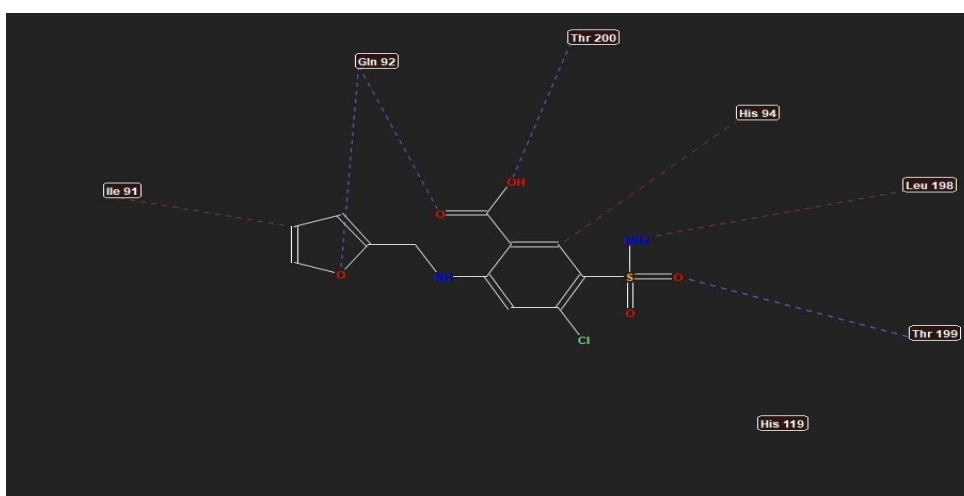


Figure 10: 2D interaction of Furosemide with 1Z9Y Protein

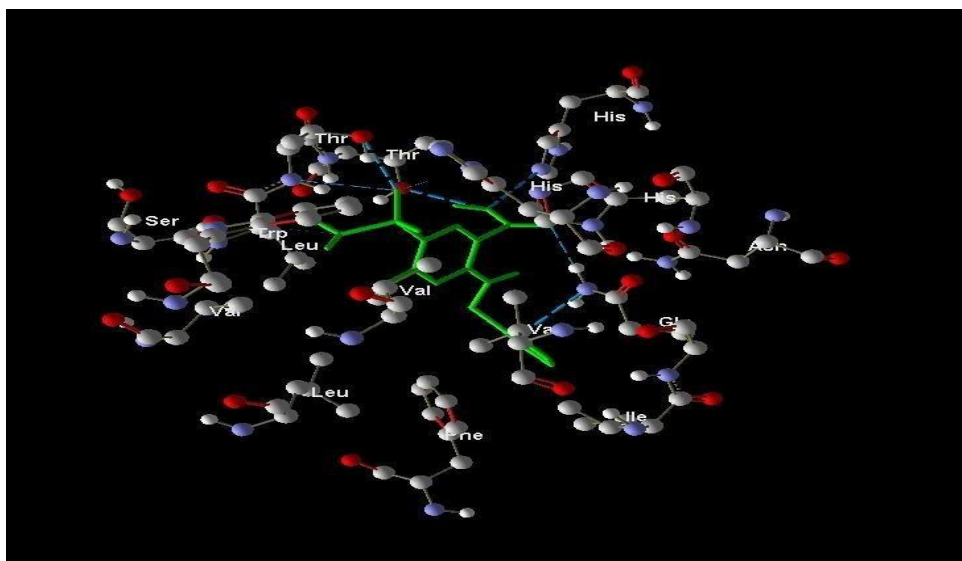


Figure 11: 3D interaction of Furosemide with 1Z9Y Protein

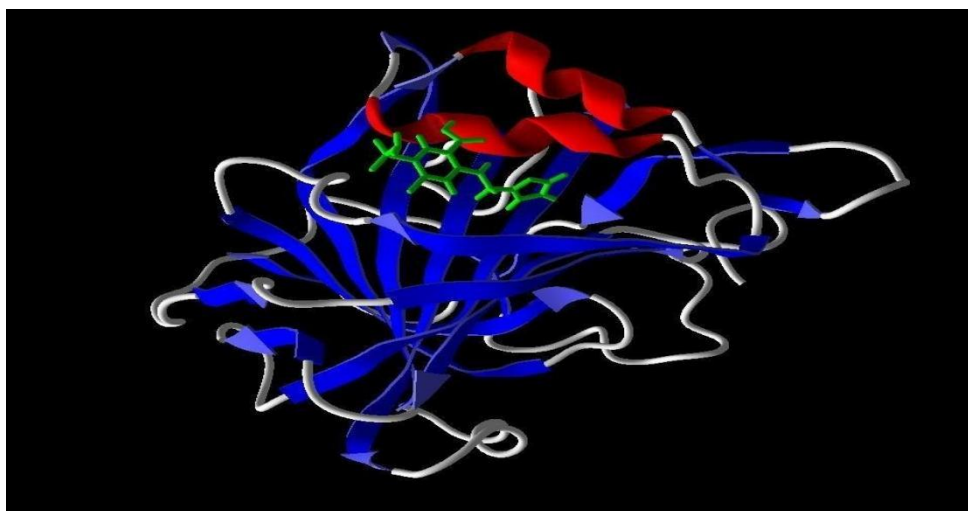


Figure 12: Secondary interaction of Furosemide with 1Z9Y Protein

The experiment explores the possibility of ligands to use diuretic action through in silico ligand docking to a target protein, a human carbonic anhydrase II (PDB ID: 1Z9Y) with bound furosemide. The Molegro Virtual Docker (MVD), an application that was employed to carry out molecular docking, was applied in the analysis, with the ligands being examined on their susceptibility of reacting with the active parts of the protein.

Beta-Sitosterol, Mangiferin and Luteol were the main ligands considered whereas Furosemide was the standard drug. Positions of the ligands were scored based on MolDock scores, which are better known as a scoring tool that explicitly captures electrostatic and hydrogen bonding interactions.

Key Findings:

The most positive MolDock whose binding scoring was -146.996 indicated the strongest binding between the ligands and Beta-Sitosterol. It depicted a good interaction with the target protein in form of hydrogen bond, electrostatic interaction (Figure 1, 2 and 3). This could imply that Beta-Sitosterol could also be an excellent prospect to be pursued in the course of diuretic treatment.

Mangiferin also displayed important interactions based on the score of -113.213 at MolDock (Figure 4, 5 and 6). Though it was not the best ranked ligand, had a good binding affinity and had a possible potential as a lead chemical molecule in therapeutic design.

The MolDock scores of Luteol and Furosemide were also -106.232 and -107.171 and were lesser than Beta-Sitosterol and Mangiferin which implies that they were less likely to bind to the protein of interest.

The detailed discussion of these ligands indicates that Beta-Sitosterol and Mangiferin can be regarded as the candidates that can be studied as a part of the diuretic drug research in the future. Their docking to human carbonic anhydrase II is found to be ideal in targeting carbonic anhydrase II which plays a major role in the action of diuretics.

DISCUSSION

Similar to the findings of the current thesis, there is a strong therapeutic potential of *Bombax malabaricum* flowers especially in regards to their diuretic properties. The quest into their phytochemical constituents, traditional use in medicinal practice and the relative performance to well-known synthetic diuretics, creates a strong framework around which they may consider adopting them in modern phytotherapeutics.

The use of diuretics in the medical field is not a new development especially in the treatment of diseases associated with excess fluid especially High blood pressure, Cardiac disease, liver cirrhosis and chronic renal diseases. The three diuretics classes traditionally used thiazides, loop diuretics, potassium-sparing agents and carbonic anhydrase inhibitors and have their effects concentrated on the specific nephron segments. They are however limited in use due to the presence of important side effects like electrolyte imbalance, dehydration and renal dysfunction.

That is why the study of the possibilities of plant-based alternatives, such as the *Bombax malabaricum*, becomes exciting, not to mention the lack of side effects typically produced by plants (compare the side effects of plants and their interconnected multi- compound bioactivity).

The combination of the flavonoids, alkaloid, tannins, phenolic compounds and glycosides have been witnessed to be the basis of pharmacological action of *Bombax malabaricum* on its flowers. Quercetin and kaempferol are flavonoids well known to have antioxidant and diuretic activity, they increase renal perfusion and increase natriuresis. Residual renal clearance is boosted by the alkaloids, i.e. arbutin and others well-known as possessing an antimicrobial and gentle diuretic effect, but which also have a pronounced effect on urinary tract infection. Tannins confer astringent, anti- inflammatory properties and are associated with fluid balance as well as to the health of the kidneys. Their synergistic effect is probably the cause of mild yet persistent effect of the diuresis exhibited in this study.

Notably, the thesis points out to the synergistic actions of these phytoconstituents. Instead of functioning as an agent in a single pharmacological axis, the synergetic bioactive agents regulate oxidative stress, lower the inflammation and affect sodium handling in the kidney. This combined action could possibly correlate with less frequent occurrence of the loss of electrolytes, which tends to be a disadvantage of most of the synthetic diuretics, when *Bombax malabaricum* preparations are used. Proportionate distribution of potassium preserving and sodium excretion agents implies more physiologically balanced fluid management, which, in turn, may decrease the occurrence of complications in the form of hypokalemia.

Ethnopharmacological significance of *Bombax malabaricum* increases its arguments in medicine. Traditionally applied in parts of India and Southeast Asia where it has been used to treat many afflictions, especially inflammatory and fluid- retention conditions. It has been used medically because of its bright red flowers which are rich in bioactive compounds. Its therapeutic profile is further offended by presence of mangiferin, an antioxidant and anti-inflammatory agent. Its hepatoprotective, anti-bacterial and wound-healing capabilities have been confirmed by previous researchers, thus making the current study greater and adding evidence that the capabilities of the plant are not limited on diuresis.

An analytical attitude also shows that the diuretic effects of *Bombax malabaricum* are comparable to those other better-known common herbal diuretics namely *Taraxacum officinale* (dandelion), *Camellia sinensis* (green tea), *Arctostaphylos uva-ursi* (uva ursi) and *Hibiscus sabdariffa* (hibiscus).

These plants have similar phytoconstituents such as flavonoids and phenolic compounds and are referred to have diuretic and anti-hypertensive activities. But what really stands out about *Bombax malabaricum* is the wider spectrum of its pharmacologic effects: in addition to its antimicrobial and hepatoprotective effect, it also has diuresis. Such biodual or even tritriple pharmacodynamic effect makes it an original representative of the integrated therapy especially in the cases of the patients with a complex clinical picture such as liver-kidney syndrome or cardio-renal pathologies.

Also the topic of natural vs. synthetic diuretics opens up a possibility for additional discussion. Although synthetic drugs are fast in their effects and can be quantified by their potency, this is the limitation since they are not adaptive to different patient profiles. Plant-based treatments, however, including *Bombax malabaricum* should be considered more adaptive and comprehensive and, more commonly, slower to take hold and longer to administer fully, on the other hand. With long term diseases, such as high blood pressure and over-hydration disorders, however, it can be more beneficial to intervene gradually and without causing side effects, which would lead to positive outcomes in the long term.

In the diseases that are characterized by fluid overload, the relevance of antioxidant and anti-inflammatory activity is also placed in context as highlighted in the thesis. Chronic inflammation is common in cases of renal and cardiovascular disease and it aggravates oxidative stress as well as undermines the functioning of the affected organs. *Bomax malabaricum* contains antioxidants and therefore, in addition to its symptomatic improvement due to diuresis, *Bombax* hinders antioxidative and inflammatory damages of renal and hepatic organs at the upstream level. This dualism serves to place the flower extracts beyond playing just a therapeutic role, but possibly a preventative role in high risk subjects.

Medicinally, the results of the current work support additional standardization and clinical confirmation of *Bombax malabaricum* -based preparations, as viewed of through the lens of pharmaceutical development. Poor standardization of dosage and pharmacokinetics profiles of the herbal medicine is also one of the major constraints towards the adoption of the herbal medicine. The future directions thus ought to encompass in vivo diuretic, dose response curve, bioavailability and long-term safety tests.

Additionally, comparative research with the first-line synthetic diuretics at the level of randomized clinical trials has the potential to define its actual usefulness in the common practice.

The scientific rigor is also provided in the thesis by discussing the techniques of solvent extraction. It is established with ample records that methanolic and ethanolic extractions show greater concentration of phenolics and flavonoids both of which are the two most likely agents behind the diuretic and antioxidant effects. The decision in using these solvents in extraction has made the study compatible with procedures confirmed by several phytopharmacological researches and this aspect portrays the validity of the study.

Finally, this piece of work gives an insight on what the future of the world of integrative medicine may be like with the combination of age old botanical expertise with the current pharmacological authentication. Along with the growing efficacy level of resistance to synthetic drugs, not only in the case of antimicrobial actions, but also regarding cardiovascular effects, the trend towards nature-based constituents, with their multi-targeting qualities and milder therapeutic properties is catching on. As depicted in this study, *Bombax malabaricum* stands in a good position to be incorporated into this paradigm shift.

CONCLUSION

The research paper highlights the intense medicinal value of *Bombax malabaricum* flowers concerning its uses as a natural diuretic. The flowers contain bioactive phytochemicals, specifically flavonoids, alkaloid, tannins and phenolic compounds with a diverse pharmacological activity, i.e., diuretic, antioxidants, anti-inflammatory, antimicrobial and hepatoprotective effects. These facts render the plant to be a useful substitute or complement to the standard diuretics, especially when used to treat the above mentioned conditions of hypertension, heart failures, renal dysfunction and fluid retention.

In comparison to the synthetic diuretics, *Bombax malabaricum* has a less dose-dependent mechanism of action with (possibly) less side effects (such as a more balanced electrolyte balance). Its purpose in traditional herbal medicine is now confirmed by modern phytochemical analysis in hopes of its inclusion in modern therapeutic plans. Its phytoconstituents in combination have a synergistic effect on increasing renal clearance, decrease oxidative stress and a better functioning overall.

To sum up, *Bombax malabaricum* is one of the most promising plant-based alternatives to treating health issues related to diuretics. More pharmacology research and clinical trials should be applied to standardize the dosage, prove the efficiency of the drug and provide safety. As the world turns a greater interest towards phytotherapy, the plant is considered to be of immense value in the formulation of safer, effective and holistic natural medicines.

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