Exploration of Antidiabetic and Diuretic Activities of *Lumnitzera racemosa* and *Eclipta alba* with Molecular Docking Study

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ABSTRACT

The goal of the current study was to look at the leaves of *Lumnitzera racemosa* (Family: Combretaceae) and aerial part of *Eclipta alba* (Family: Asteraceae) for its phytochemical constituents and selected pharmacological activities (diuretic and antidiabetic). Diuretic medications are used to treat hypertension. Diabetes and hypertension are two common disease of geriatric patients and our aim was to explore medicinal plants of Sundarbans to find therapeutic of these diseases. The presence of carbohydrates, glycosides, reducing sugar, tannins, flavonoids, alkaloids, proteins, gum, steroids, saponin, and acidic chemicals was shown by phytochemical study of *L. racemosa* and *E. alba* showed the existence of tannin, saponin, flavonoid, gum, alkaloid, steroid, and terpenoid. Both plants extract was fractionated depending on polarity using *n*-hexane (non-polar), ethyl acetate (medium-polar) and water (polar). In the diuretic activity assay using Swiss Albino mice, none of the fractions of *L. racemosa* and *E. alba* showed diuretic activity but *n*-hexane 500 mg/kg of *E. alba* exhibited little diuretic activity compared to the standard frusemide. Swiss Albino mice were used to assess the oral glucose tolerance test (OGTT) in order to measure antidiabetic activity. In OGTT, water fraction 500 mg/kg of *L. racemosa* and *E. alba* showed blood glucose lowering activity compared to the standard Glibenclamide. We had performed *in silico* study among reported eight (08) compounds of *L. racemosa* against 1V4S (human glucokinase) protein. In comparison to classic glibenclamide (-8.5 kcal/mol), myricitrin had a good docking score of -8.4 kcal/mol. Based on these findings, we hypothesized that *L. racemosa* and *E. alba* could be a possible source of therapeutic leads for hypertension and hyperglycemia.

Keywords: *Lumnitzera racemosa*, *Eclipta alba*, Diuretic activity, Antidiabetic activity, Molecular Docking Study.

INTRODUCTION

Medicinal plants are widely used all over the world for production of both traditional medicine and modern drugs as well as development of new drugs. According to estimates from the World Health Organization (WHO), 80% of people in developing nations, get their primary medical care from plant-based medications traditional medicines. The need for medicinal plants in Bangladesh for the manufacture of traditional medicines (ayurvedic, unani and homeopathic) is rising day by day. Bangladesh has a rich flora of medicinal plants with a number of important active principles that have been used traditionally against a wide variety of diseases. Due to unwanted side-effects of synthetic and semisynthetic drugs, attention has been focused on utilizing natural medicine. Many people of our country have been suffering from diabetes and hypertension. Drugs called diuretics are used to treat hypertension. People used allopathic medicines for their treatment of diabetes and high blood pressure and these medicines show many adverse effects. Our target will explore medicinal plants of Sundarban to

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find new herbal medicine for the management of hypertension and diabetes

Mangrove species *Lumnitzera racemosa* (Family: Combretaceae) is widely distributed in tropical and subtropical regions. *L. racemosa* is a small tree or evergreen shrub that can reach a height of 8 meters. It has numerous spreading branches that are covered with twigs that are red, reddish-brown, or greyish-black, and the bark is brown to greyish-black. Flowering season varies from region to region. Qualitative studies reveal that the extracts of leaves, stems, and bark contain sugars, terpenoids, tannins, steroids, phenols, flavonoids, glycosides, coumarins, alkaloids, essential oils, anthraquinones, and saponins. [1]. Previous studies reported that *L. racemosa* has antibacterial, antifungal, antifungal, antidiarrheal, hepatoprotective and cytotoxic activities [2].

Asteraceae family member *Eclipta alba* is referred to as Bhringaraja and False daisy. It is a little annual herb that is frequently utilized for a range of therapeutic uses. It has numerous branches and can be horizontal or vertical. The majority of the second branches emerge from massive, up to 7 mm long, grey roots. Due to repressed white hair, prominent nodes, and red color, the stem is round or flat and rough. Leaves: oblong, lanceolate, sub-entire, strigose, 2.0–6.2 cm long, 1.5–1.9 cm wide, opposite, sessile to sub-sessile, acute to subacute, and hair oppressed on both faces. All year long, flowers bloom. *E. alba* has analgesic, hepatoprotective, antidiabetic, hair growth promoting, antioxidant, cardiovascular, anti-microbial, anti-epilepsy, anticancer, antiulcer, anti-venom, neuropharmacological and antihyperlipidemic properties [3]. Medicinal plants from different regions possess different biological activities.

The purpose of this experiment was to explore the antidiabetic and diuretic properties of two commonly used medicinal plants from the Sundarbans, *L. racemosa* and *E. alba*, using a molecular docking analysis of isolated chemicals to determine their mechanism of action. The identification of a novel biological activity in these plants could serve as a foundation for the creation of important medicines with improved pharmacological characteristics (safety and efficacy). These studies enable us to pinpoint the active ingredients that give medical effects and assist raise public awareness of the need of cultivating and protecting these kinds of medicinal plants. In the perspective of developing countries like Bangladesh, research on indigenous medicinal plants has also great potential of contributing substantially to the economy of the country. To achieve sustainable development goals (SDGs) we need safe, effective, quality and affordable essential medicines for all. The government is working hard to bring back traditional cultures and wants to promote domestic medicine over imported medications since both are accessible and affordable, which eventually strengthens the national economy.

MATERIALS AND METHODS

Plant Material and Extraction

The aerial portions of *E. alba* and the leaves of *L. racemosa* were gathered from the Sundarbans. Adulteration of any kind was forbidden during collection. Researchers from Khulna University's Forestry and Wood Technology Faculty and the Bangladesh National Herbarium in Mirpur, Dhaka, Bangladesh, identified the plants. *E. alba*'s voucher specimen number is 46081 DACB. The harvested plant parts were chopped into tiny pieces and allow to dry for 21 days away from the sun. Following the final milling of the dried plant materials, 500 g of it were extracted using a 15-day maceration technique and 1200 mL of 80% ethanol. A filter removed the extracts. Using a rotary evaporator, the solvents were removed at room temperature. Water (polar), ethylacetate (medium polar), and n-hexane (non-polar) were utilized to fractionate the ethanolic extract. First the extract was mixed with 500 mL water and added 500 mL n-hexane with it. Then the mixture was taken in a separating funnel and shaken and stand for 30 minutes. n-hexane fraction was separated. Remaining water fraction was mixed with

500 mL ethylacetate and again taken in separating funnel and water and ethylacetate fraction were separated. A preliminary screening for phytochemical and pharmacological effects was performed on the crude extracts.

Animals

For the tests, Swiss-Albino mice of both sexes, aged 12–14 weeks and weighing 25–30gram body weight, were gathered from the animal resources department of the International Center for Diarrheal Disease Research, Bangladesh (ICDDR, B). The animals were fed standard meals (ICDDR, B prepared) and housed in normal polypropylene cages. Prior conducting the experiment, the animals were allowed to acclimate for a total of 14 days at the animal house located in the Pharmacy Discipline of Khulna University. Standard laboratory settings included a relative humidity of 55–60%, a room temperature of 25–20 C, and a 12-hour light–dark cycle.

Chemicals

We procured sodium nitropruside and sodium molibdate from Merck in Germany. We purchased sodium phosphate, sulphanilamide, and N 1-naphthyl ethylenediamine dihydrochloride from BDH, England, along with ethylene diamine tetraacetic acid (EDTA). Sigma-Aldrich (USA) provided the following chemicals: sodium hydroxide, hydrochloric acid, ferric chloride, potassium dichromate, sulfuric acid, sodium nitroprusside, n-hexane, ethyl acetate, and ethanol. Frusemide sodium and Glibenclamide, two common medications, were obtained from Beximco Pharmaceuticals Ltd. in Dhaka, Bangladesh.

Phytochemical tests

A preliminary phytochemical screening was performed on the *L. racemosa* and *E. alba* extract in order to identify the main functional groups [4]. Each test involved taking a 2% (w/v) solution of the extracts in ethanol. The various chemical groups in the extract were identified using the following techniques. Subsequently, the extract was employed for pharmacological examination.

Tests for Reducing Sugar

Benedict's test requires filling a test tube with 0.5 mL of the plant material's ethanol extract. The test tube is filled with 5 mL of Benedict's solution, brought to a boil for 5 minutes, and then allowed to cool naturally. There is reducing sugar present when a red cuprous oxide precipitate forms.

Fehling's Test (Standard Test): After boiling for a few minutes, 1 mL of a mixture of equal volumes of Fehling's solutions A and B is added to 2 mL of an aqueous extract of the plant material. When reducing sugar was present, a red or brick red precipitate happened.

Test for Combined Reducing Sugar: 1 mL of a plant extract in water is heated for 5 minutes together with 2 mL of diluted hydrochloric acid. The mixture is then cooled and neutralized with sodium hydroxide solution.

Tests for Tannins

Test for Ferric Chloride: Five milliliter solutions of every extract are placed in separate test tubes. Next, a 1 mL solution containing 5% ferric chloride is added. Presence of tannins is proven by a greenish-black precipitate.

Test for potassium dichromate: Five milliliter volumes of each extract are placed in separate test tubes. Next, a 10% potassium dichromate solution (one milliliter) is added. Yellow precipitate indicates the existence of tannins.

Test for Flavonoids

An ethanolic extract of the plant materials is combined with a small amount of strong hydrochloric acid in a few drops. The presence of flavonoids is indicated by the rapid development of a red tint.

Test for Saponins

Each extract is diluted to a volume of 20 mL with distilled water, and the mixture is shaken in a graduated cylinder for a period of 15 minutes. The existence of saponins is indicated by the formation of a 1 cm layer of foam.

Test for Gums

After taking 5 mL solutions of each extract, add

sulfuric acid and Molish reagent. When two liquids combine, a reddish-violet ring is formed, signifying the presence of carbohydrates and gum.

Test for Steroids

Libermann-Burchard test: Add 2 mL of Libermann-Burchard reagent to a 1 mL solution of ethanolic extract. The presence of steroids is indicated by a reddish-purple hue.

Test for sulfuric acid: 1 milliliter of a chloroform extract solution is obtained, and 1 milliliter of sulfuric acid is added. Steroids are indicated by the color red.

Test for Alkaloids

Mayer's test: It involves filling a test tube with 0.2 mL of diluted hydrochloric acid and 2 mL of the extract solution. Mayer's reagent (1 mL) is then added. The presence of alkaloids is shown by the formation of a yellow precipitate.

Dragendroff's test: It involves filling a test tube with 0.2 mL of diluted hydrochloric acid and 2 mL of each extract solution. Next, add 1 milliliter of Dragendroff's reagent. Alkaloids are indicated by an orange-brown precipitate.

Wagner's test: It involves filling a test tube with 0.2 mL of diluted hydrochloric acid and 2 mL of each extract solution. Wagner's reagent (iodine solution) is then added in a volume of 1 mL. The occurrence of alkaloids can be seen by a reddish-brown precipitate.

Hager's test: In a test tube, 2 mL of each extract solution and 0.2 mL of diluted hydrochloric acid are added. Hager's reagent, or 1 mL of picric acid solution, is then added. Precipitate with a yellowish tint suggests the presence of alkaloids.

Test for Terpenoids

2 mL sample of the ethanolic extract solution was heated for about 2 minutes after 2 mL of concentrated H2SO4 was added. The presence of terpenoids was suggested by a grayish appearance.

Test for Glycosides

Little amount of 10% (w/v) NaOH was added to an aliquot containing 3 mL of the aqueous extract solution. Next, a small amount of recently made 10% (w/v) sodium nitroprusside was added to the combination solution. The lack of glycosides was demonstrated by the formation of no blue, pink, or red colour.

Oral glucose tolerance tests (OGTT) for Antidiabetic Assay

Both plants' OGTT were conducted using a slightly modified version of the method Joy and Kuttan, 1999 had previously published [5. 6]. The mice were chosen by chance and placed into six (06) groups, labeled group-I, group-II and group-III, group-IV, group V, and group VI, with five (05) mice in each group. The experimental animals were maintained in a fasting state (having no food or drink except water for at least 10 hours but not greater than 16 hours). Every group was given a certain treatment, such as the test sample, the standard, and the control. The doses of the test sample, reference, and control drugs were adjusted to comply with precise weight values of each mouse. Every group was given a specific therapy. Each mouse was accurately weighed and the dosages of the test and control materials were adjusted before commencing any treatment. Glibenclamide (10 mg/kg bogy weight), control (1% tween-80 solution in water) and test sample (250 mg/kg and 500 mg/kg) were given orally via feeding needle at zero-hour doses of. All groups received treatment with a 10% glucose solution (2 gm/kg body weight) after 30 minutes. By taking blood samples from the tail vein approximately 30, 60, 90, 120, and 150 minutes after the glucose was administered, blood glucose loading was ascertained. Lastly, a glucometer was used to measure the blood glucose level [5, 6]. The unit of measurement for blood glucose was mmol/L. At 0.05, the threshold for statistical significance was established.

Diuretic Activity Assay

Both plants' diuretic activity was assessed in compliance with the guidelines provided by Mekonnen *et al.* (2010) and Mamun *et al.* (2003) [7, 8]. Using a random selection process, the mice were split up into six groups (group I,

group II, group III, group IV, group V, and group VI), each with five (05) mice. Prior the experiment, all the groups were given an 18-hour fast and water deprivation. As a control, animals in group I were given normal saline (15 mL/kg, p.o.), while animals in group II were given the conventional medication furosemide (5 mg/kg, p.o.) in normal saline. The test groups, which were groups III, IV, V, and VI, were given n-hexane at doses of 250 and 500 mg/kg along with a water fraction in typical saline. The animals were put in metabolic cages that were specifically made to segregate urine and feces as soon as they were dosed (5 per cage; ugo basile®, Italy). The animals were kept in their cages at (25.0±0.5) °C for the duration of the experiment, and food and water were not given for six hours after the mice were placed in them. During the 6-hour treatment period, the urinary output (Vo) was obtained every hour, and the urine was then held at -20 °C for additional examination. To compare the extracts' effects on urine excretion, the following parameters were computed. The ratio of total urination to total liquid delivered was used to compute urinary excretion, which was independent of animal weight (Formula-I). A drug's diuretic activity at a particular dose was measured using the ratio of urine excretion in the test group to urine excretion in the control group (Formula-II). The extract's diuretic action was compared to the standard medication in the test group (Formula-III) in order to determine its diuretic activity. Cl^- was determined titrimetrically, and the electrolyte content (Na^+ and K^+) of the urine sample was checked with a flame photometer. Using the proper techniques, pH, conductivity, and density were also ascertained.

$$\text{Urinary excretion} = \frac{\text{Total urinary output } (V_0)}{\text{Total liquid administered } (V_i)} \times 100 \ldots\ldots \text{(I)}$$

$$\text{Diuretic action} = \frac{\text{Urinary excretion of treatment groups}}{\text{Urinary excretion of control group}} \ldots\ldots\ldots \text{(II)}$$

$$\text{Diuretic activity} = \frac{\text{Diuretic action of test groups}}{\text{Diuretic action of standard group}} \ldots\ldots\ldots\ldots \text{(III)}$$

***In silico* Molecular Docking Study**

The "Vina Wizard" program in PyRx - Python Prescription 0.8 was used to execute molecular docking investigation between the receptors and the ligands (separately) [9]. First, the receptors and the ligands (independently) were put into the PyRx program with the proper compound—that is, macromolecule or ligand—declaration. Two distinct, optimized cube-sized boxes with precise measurements that line up with the receptors' main active site residues. PyMOL then paired the upgraded receptor with the docked ligands (separately). To ensure adequate visualization, the combined structures were seen using Discovery Studio. Images were saved in the optimal positions after the ligand interactions with amino acids were seen.

Arrangement of protein molecules

The human glucokinase protein model, PDB ID 1V4S, was chosen and obtained from the protein bank (https://www.rcsb.org/) in order to conduct the molecular docking investigation. The Discovery studio 2020 client then processed the obtained protein 3D structures to eliminate all associated ligands and water molecules in order to prepare the environment for subsequently interacting with the chosen ligand molecules [10].

Setting up ligands

Glibenclamide (CID: 3488), a typical drug used in biological tests, has 3D structures that may be obtained from PubChem (https://pubchem.ncbi.nih.gov/). We choose eight (08) isolated chemicals from *L. racemosa* that have been reported [11]. Furthermore, we retrieved several compounds that were reported to be present in the *L. racemosa* from the same website, including methyl gallate (4) (CID: 7428), 3,4,3'-tri-O-methylellagic acid (3) (CID: 5281860), myricitrin (5) (CID: 5281673), stigmasterol (6) (CID: 5280794), kaempferol (7) (CID: 5280863), isoguaiacin (8) (CID: 10314441), and others. Chem3D was used to draw the other two chemicals, racelactone (1) and botulin (2). PyRx analyzed those in order to minimize energy use.

Site specific binding and visualization

In order to allow all of the chosen protein molecules to be available for binding with the conducted ligands, we made the decision to use the blind docking approach. PyRx and auto dock vina 4.2 were used for these procedures, and the grid dimensions for the 1V4S protein were x: y: z = 58.7568:76.6570:65.3039 [10].

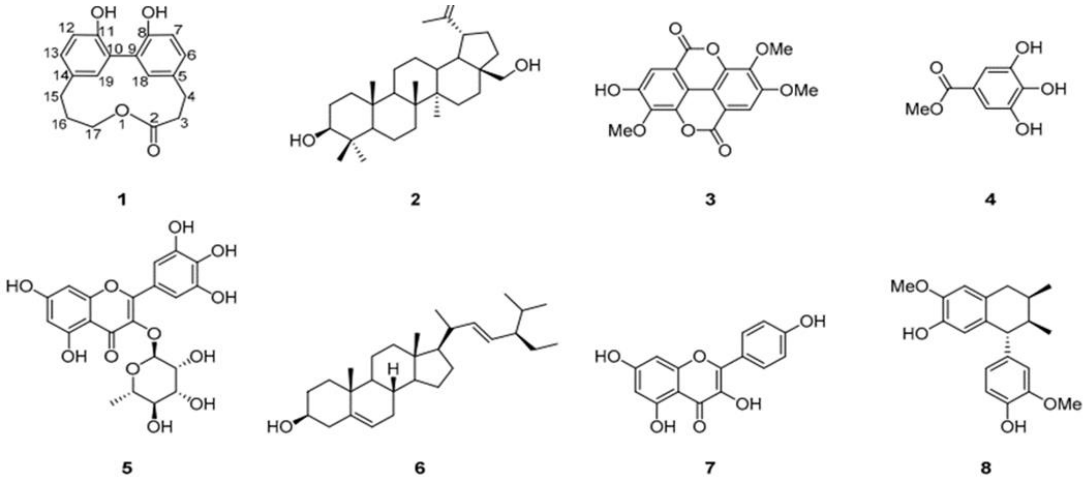


Figure 1: Reported compounds isolated from *L. racemose*

Analytical statistics

Using Excel and Prism, the Student's t-test was applied to identify significant differences between the test group and the control group.

RESULTS AND DISCUSSION

Results

Numerous secondary metabolites, including reducing sugars, flavonoids, phenolic compounds, tannins, proteins, glycosides, alkaloids, saponins, steroids, terpenoids, and acidic compounds, were found in the phytochemical analysis of the *L. racemosa* and *E. alba* extract. These metabolites may be responsible for some of the plant's therapeutic qualities (Table 1).

Table 1: Outcome of *L. racemosa* and *E. alba* extract Chemical Group Tests

Constituent	*L. racemosa*	*E. alba*
Phenolic Compound	+	+
Flavonoids	+	+
Carbohydrates	+	-
Reducing Sugar	+	-
Tannins	+	+
Saponin	-	+
Glycoside	+	-
Gums	-	+
Alkaloids	+	+
Protein	+	-
Acidic Compounds	+	+

Antidiabetic activity assay

The antidiabetic activity of the different fractions of *L. racemosa* and *E. alba* extract were examined using OGTT. Blood sugar levels declined in a dose-dependent way in the water fractions of the *L. racemosa* extract. 5.20 and 4.30

mMol/L at the dose of 500 mg/kg body weight in contrast to the reference drug Glibenclamide 5.06 and 4.1 mMol/L after 90 and 150 min (Table 2).

The amount of glucose in the blood dropped in a dose-dependent way in the water fractions of the *E. alba* extract. 5.32 and 5.46 mMol/L at the dose of 250 mg/kg body weight in contrast to the reference drug Glibenclamide 5.44 and 4.58 mMol/L after 90 and 150 min (Table 3).

Table 2: Oral Sugar Tolerance test result of *L. racemosa* extract

Administered dose & Test Groups	Mice Number	Mice Weight(gm)	Blood Sugar Level (mmol/L)			
			At Fasting State (At 0 min)	After the Administration of Glucose		
				At 30 min	At 90 min	At 150 min
Control	1	27	5.4	13.6	7.6	6.6
	2	25	6.4	16.0	8.2	5.8
	3	28	5.3	11.8	7.3	6.8
	4	26	5.5	12.3	8.6	6.2
	5	27	6.1	14.1	8.0	5.9
	Average		**5.74**	**13.56**	**7.94**	**6.26**
Positive control Glibenclamide (5 mg/kg)	1	26	4.7	10.8	6.1	5.2
	2	25	4.2	13.7	5.6	4.5
	3	25	5.6	9.4	3.8	3.0
	4	27	5.9	11.2	4.9	3.4
	5	27	4.8	10.2	6.3	4.1
	Average		**5.04**	**11.06**	**5.06**	**4.1**
***n*-hexane fraction of *L. racemosa* (250 mg/kg)**	1	25	5.3	10.1	5.6	5.2
	2	25	6.0	9.1	6.9	5.8
	3	27	5.7	10.5	5.8	4.9
	4	26	4.9	12.1	6.5	5.9
	5	28	5.2	11.5	6.2	6.0
	Average		**5.42**	**10.74**	**6.25**	**5.56**
***n*-hexane fraction of *L. racemosa* (500 mg/kg)**	1	25	4.8	10.0	6.5	4.6
	2	28	6.4	11.7	7.6	6.2
	3	26	5.9	12.1	7.9	6.7
	4	25	6.1	10.3	5.4	4.9
	5	27	5.2	11.2	6.3	5.9
	Average		**5.68**	**11.06**	**6.74**	**5.66**
H_2O fraction of *L. racemosa* (250 mg/kg)	1	26	5.7	10.3	6.5	5.7
	2	26	6.3	11.1	7.2	6.2
	3	27	5.9	12.5	7.9	6.9
	4	25	4.9	10.5	6.8	5.9
	5	28	6.2	12.2	5.9	4.9
	Average		**5.8**	**11.32**	**6.86**	**5.92**
H_2O fraction of *L. racemosa* (500 mg/kg	1	26	5.9	10.5	5.0	4.5
	2	28	6.7	12.1	5.4	4.2
	3	25	4.9	10.2	5.1	4.4
	4	27	5.2	11.5	5.2	4.3
	5	25	4.2	11.6	5.3	4.1
	Average		**5.38**	**11.18**	**5.20**	**4.30**

Table 3: Oral Sugar Tolerance Test result of *E. alba* extract

Administered dose & Test Group	Mice Number	Weight (gm) of mice	Blood Glucose Level (mmol/L)			
			At Fasting State (At 0 min)	After the Administration of Glucose		
				At 30 min	At 90 min	At 150 min
Control	1	28	5.5	12.5	8.2	5.9
	2	29	6.3	15.0	7.5	6.7
	3	28	5.5	12.9	8.5	5.8
	4	29	5.7	13.5	7.9	6.3
	5	27	6.3	14.7	8.3	6.2
	Average		**5.86**	**13.72**	**8.08**	**6.18**
Positive control Glibenclamide (5 mg/kg)	1	28	5.0	11.1	5.9	4.8
	2	29	4.7	12.2	5.5	4.6
	3	27	5.3	10.9	4.8	4.3
	4	27	5.5	11.5	5.2	4.5
	5	29	4.9	11.4	5.8	4.7
	Average		**5.08**	**11.42**	**5.44**	**4.58**
***n*-hexane fraction of *E. alba* (250 mg/kg)**	1	29	5.5	12.9	8.4	7.4
	2	28	5.6	13.2	8.8	6.9
	3	27	5.4	13.6	8.9	6.8
	4	28	5.9	13.4	9.2	7.2
	5	28	5.7	13.8	9.3	7.0
	Average		**5.62**	**13.38**	**8.92**	**7.06**
***n*-hexane fraction of *E. alba* (500 mg/kg)**	1	29	5.8	12.1	7.8	7.5
	2	28	5.9	11.9	7.7	7.9
	3	29	5.7	12.4	7.9	7.7
	4	29	5.8	12.6	8.4	7.8
	5	27	5.6	12.3	8.3	7.5
	Average		**5.76**	**12.26**	**8.02**	**7.68**
H_2O fraction of *E. alba* (250 mg/kg)	1	29	5.5	11.1	5.4	5.3
	2	29	6.1	11.4	5.3	5.4
	3	27	5.8	11.9	4.9	5.5
	4	29	5.9	11.5	5.1	5.2
	5	28	6.1	12.1	5.9	5.9
	Average		**5.88**	**11.60**	**5.32**	**5.46**
H_2O fraction of *E. alba* (500 mg/kg	1	29	6.1	10.6	5.6	5.6
	2	28	5.7	11.1	5.7	5.8
	3	28	5.9	10.8	5.9	5.4
	4	27	5.1	11.7	5.9	5.3
	5	29	5.2	11.9	5.5	5.8
	Average		**5.6**	**11.22**	**5.72**	**5.58**

Diuretic Activity Assay

Different fractions of *L. racemosa* and *E. alba* extract failed to produce an increase in urine volume elimination when compared to the normal dose of Frusemide (5 mg/kg-bw) in the mice used to evaluate its diuretic activity (Table 4, 5, 10 & 11). Only n-hexane fraction of *E. alba* showed mild increase in urine volume but lower than to the standard Frusemide 5 mg/kg-bw. However, electrolyte

analysis of urine samples the *L. racemosa* and *E. alba* extract showed a considerable concentration of potassium, sodium, and chloride ion in the sample that was comparable to control and standard (Table 6, 7, 8, 9, 12, 13, 14, 15).

Table 4: Urinary output of different fractions of *L. racemosa*

Groups	Urine volume (V_O) mL						Urinary excretion, (V_O/ V_i) × 100					
	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr
Group-I (Negative Control)	0.5	1.6	2.1	2.5	3	3.6	4.1	13.33	17.5	20.83	25	30
Group-II Frusemide (5 mg/kg)	3.9	7.1	9	10	11.3	13.7	32.5	59.17	75	87.5	94.17	114.7
Group-III (*n*-hex. 250 mg/kg)	0.5	1	2	2.25	3	3.25	4.17	8.33	16.67	18.75	25	27.08
Group-IV (*n*-hex. 500 mg/kg)	0.5	1.75	2	3	3.75	4	4.16	14.58	16.67	25	31.25	33.33
Group-V (Water 250 mg/kg)	0	.25	1.75	2	2	2.25	0	2.08	14.58	16.67	16.67	18.75
Group-VI (Water 500 mg/kg)	0	1	2.25	1.75	2	2.5	0	8.33	18.75	14.58	16.67	20.83

Table 5: Diuretic action and activity of different fractions of *L. racemosa*

Groups	Diuretic action (U_{ET}/ U_{EC})						Diuretic activity (D_{AT}/ D_{AU})					
	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr
Group-I (Negative Control)	-	-	-	-	-	-	-	-	-	-	-	-
Group-II Frusemide (5 mg/kg)	7.8	4.43	4.28	4.2	3.77	3.81	-	-	-	-	-	-
Group-III (*n*-hexane 250 mg/kg)	1	0.63	0.95	0.9	1	0.9	0.12	0.14	0..22	0.26	0.27	0.24
Group-IV (*n*-hexane 500 mg/kg)	1	1.09	0.95	1.2	1.25	1.11	0.13	0.27	0.22	0.3	0.33	0..29
Group-V (Water 250 mg/kg)	0	0.16	0.83	0.8	0.67	0.63	0	0.04	0.19	0.2	0.2	0.18
Group-VI (Water 500 mg/kg)	0	0.63	1.07	0.7	0.67	0.69	0	0.14	0.25	0.18	0.17	0.18

Table 6: Concentration of Na+ in different fractions of *L. racemosa*

Groups	Flame Intensity (FI) vs [Na^+]				Concentration of Na^+ (mEq/L) ± SEM
	Replication 1		Replication 2		
	Flame Intensity	[Na^+] (mEq/L)	Flame Intensity	[Na^+] (mEq/L)	
Control (Normal Saline)	0.60	181.90	0.60	181.90	181.90 ± 0
Frusemide (5 mg/kg)	0.90	279.91	0.91	283.18	281.55 ± 1.64
Fr. *n*-hexane (250 mg/kg)	0.68	208.04	0.72	221.18	214.61 ± 6.57
Fr. *n*-hexane (500 mg/kg)	0.74	227.64	0.80	247.24	237.44 ± 9.8
Fr. H_2O (250 mg/kg)	0.62	188.44	0.66	201.51	194.97± .653
Fr. H_2O (500 mg/kg)	0.66	201.51	0.68	208.04	204.77± 3.26

Table 7: Concentration of K^+ in different fractions of *L. racemosa*

Groups	Flame Intensity (FI) vs [K^+]				Concentration of K^+ (mEq/L) ± SEM
	Replication 1		Replication 2		
	Flame Intensity	[K^+] (mEq/L)	Flame Intensity	[K^+] (mEq/L)	
Control (Normal Saline)	0.35	19.65	0.35	19.65	19.65 ± 0
Frusemide (5 mg/kg)	0.55	42.79	0.56	43.95	43.37 ± 0.58
Fr. *n*-hexane (250 mg/kg)	0.45	31.21	0.46	32.37	31.79 ± 0.58
Fr. *n*-hexane (500 mg/kg)	0.48	34.68	0.46	32.37	33.52 ± 1.15
Fr. H_2O (250 mg/kg)	0.40	25.43	0.42	27.73	26.58± 1.15
Fr. H_2O (500 mg/kg)	0.42	27.73	0.45	31.21	29.47 ± 1.47

Table 8: Concentration of Cl^- in different fractions of *L. racemosa*

Groups	Replication 1		Replication 2		Average conc. of Cl^- (mEq/L) ± SEM
	Volume of Titre (mL)	[Cl^-] (mEq/L)	Volume of Titre (mL)	[Cl^-] (mEq/L)	
Control (Normal Saline)	0.086	90	0.086	90	90 ± 0
Frusemide (5 mg/kg)	0.097	117.5	0.099	122.5	120 ± 2.5
Fr. *n*-hexane (250 mg/kg)	0.089	104	0.092	105	104.5 ± 0.5
Fr. *n*-hexane (500 mg/kg)	0.092	111.5	0.094	114.5	113 ± 1.5
Fr. H_2O (250 mg/kg)	0.088	95	0.088	95	95 ± 0
Fr. H_2O (500 mg/kg)	0.090	100	0.093	101	100.5 ± 0.5

It is generally accepted that the Carbonic Anhydrase Inhibition (CAI) Index, K^+/Na^+ (Kaliuretic Index), Na^+/K^+ (Natriuretic Index), and Saluretic Index have a significant role in interpreting the mode of action of diuretic medications. The above parameters were also estimated as following-

Table 9: Effect of different treatment groups on Saluretic, Natriuretic, Kaluretic and CAI index of different fractions of *L. racemosa*

Groups	Cumulative ion Concentrations (mEq/L/6h)			Saluretic Index			Na^+ / K^+	K^+ / Na^+	CAI = {Cl^-/ (Na^++ K^+)}	Diuretic Index (DI)
	Na^+	K^+	Cl^-	Na^+	K^+	Cl^-				
Control (Normal Saline)	181.90	19.65	90	0	0	0	9.26	0.11	0.45	
Frusemide (5 mg/kg)	281.55	43.37	120	1.55	2.21	1.33	6.49	0.15	0.37	3.8
Fr. *n*-hexane (250 mg/kg)	214.61	31.79	104.5	1.14	1.58	1.15	6.75	0.14	0.42	0.90
Fr. *n*-hexane (500 mg/kg)	237.44	33.52	113	1.25	1.76	1.23	6.64	0.14	0.41	1.11
Fr. H_2O (250 mg/kg)	194.97	26.58	95	1.03	1.29	1.05	7.08	0.13	0.45	0.63
Fr. H_2O (500 mg/kg)	204.77	29.47	100.5	1.12	1.41	1.11	6.94	0.16	0.44	0.69

Diuretic index (DI) = Test group urine volume / Control group urine volume

Table 10: Urinary output of different fractions of *E. alba*

Groups	Urine Volume (V_o)						Urinary Excretion (V_o/V_i) ×100					
	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr
Group-I Control (0.9% NaCl)	1.5	1.7	2	2.1	2.2	2.2	12.5	14.17	16.7	17.5	18.3	18.3
Group-II Standard (Furosemide 5 mg/kg)	6.6	6.8	7.5	7.6	7.8	7.8	55	56.67	62.5	63.3	65	65
Group-V n-Hexane (250mg/kg)	4.2	4.3	4.4	4.5	4.6	4.6	35	35.83	36.7	37.5	38.3	38.3
Group-VI n-Hexane (500mg/kg)	5.7	5.9	6.2	6.4	6.5	6.5	47.5	49.17	51.7	53.3	54.2	54.2
Group-IX Water (250mg/kg)	2	2.2	2.4	2.5	2.5	2.6	16.7	18.3	20	20.83	20.83	21.7
Group-X Water (500mg/kg)	2	2.5	2.9	3	3	3.1	16.7	20.83	24.2	25	25	25.8

Table 11: Diuretic action and activity of different fractions of *E. alba*

Groups	Diuretic Action (U_{ET}/U_{EC})						Diuretic Activity (D_{AT}/D_{AU})					
	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr	1 hr	2 hr	3 hr	4 hr	5 hr	6 hr
Group-I Control (0.9% NaCl)	-	-	-	-	-	-	-	-	-	-	-	-
Group-II Standard (Furosemide 5mg/kg)	4.4	3.9	3.74	3.62	3.55	3.55	-	-	-	-	-	-
Group-V n-Hexane (250mg/kg)	2.8	2.53	2.2	2.14	2.09	2.09	0.64	0.65	0.59	0.59	.59	.59
Group-VI n-Hexane (500mg/kg)	3.67	3.47	3.1	3.05	2.96	2.96	0.85	0.89	0.83	0.84	.83	.83
Group-IX Water (250mg/kg)	1.34	1.29	1.2	1.19	1.14	1.19	0.30	0.33	0.32	0.33	.32	.34
Group-X Water (500mg/kg)	1.34	1.47	1.44	1.43	1.37	1.41	0.30	0.38	0.39	0.4	.39	0.4

Table 12: Concentration of Na^+ in different fractions of *E. alba*

Groups	Flame Intensity (FI) vs [Na^+]				Concentration of Na^+ (mEq/L) ± SEM
	Replication 1		Replication 2		
	Flame Intensity	[Na^+] (mEq/L)	Flame Intensity	[Na^+] (mEq/L)	
Control (Normal Saline)	0.62	182.70	0.61	183.30	183.00 ± 0
Frusemide (5 mg/kg)	0.91	280.88	0.92	284.32	282.60 ± 1.72
Fr. *n*-hexane (250 mg/kg)	0.68	209.21	0.71	220.34	214.76 ± 5.57
Fr. *n*-hexane (500 mg/kg)	0.75	229.86	0.78	246.45	238.16 ± 8.3
Fr. H_2O (250 mg/kg)	0.60	185.23	0.62	195.26	190.25 ± 5.02
Fr. H_2O (500 mg/kg)	0.63	193.64	0.61	201.37	197.51 ± 3.87

Table 13: Concentration of K^+ in different fractions of *E. alba*

Groups	Flame Intensity (FI) vs [K^+]				Concentration of K^+ (mEq/L) ± SEM
	Replication 1		Replication 2		
	Flame Intensity	[K^+] (mEq/L)	Flame Intensity	[K^+] (mEq/L)	
Control (Normal Saline)	0.33	18.79	0.34	18.48	18.64 ± 0
Frusemide (5 mg/kg)	0.56	43.85	0.56	43.90	43.88 ± 0.03
Fr. *n*-hexane (250 mg/kg)	0.44	30.33	0.45	31.64	30.99 ± 0.16
Fr. *n*-hexane (500 mg/kg)	0.46	32.34	0.45	31.88	32.11 ± 0.23
Fr. H_2O (250 mg/kg)	0.41	25.98	0.43	28.65	27.32 ± 1.34
Fr. H_2O (500 mg/kg)	0.43	28.34	0.44	30.86	29.60 ± 1.26

Table 14: Concentration of Cl^- in different fractions of *E. alba*

Groups	Replication 1		Replication 2		Average conc. of Cl^- (mEq/L) ± SEM
	Volume of Titre (mL)	[Cl^-] (mEq/L)	Volume of Titre (mL)	[Cl^-] (mEq/L)	
Control (Normal Saline)	0.087	91	0.085	89	90 ± 0
Frusemide (5 mg/kg)	0.096	116.6	0.099	122.8	119.7 ± 3.1
Fr. *n*-hexane (250 mg/kg)	0.088	101.16	0.089	101.98	101.57 ± 0.41
Fr. *n*-hexane (500 mg/kg)	0.091	109.76	0.093	111.81	110.79 ± 1.03
Fr. H_2O (250 mg/kg)	0.087	93.22	0.086	90.87	92.05 ± 1.18
Fr. H_2O (500 mg/kg)	0.091	100.2	0.093	101.7	100.5 ± 0.75

Table 15: Effect of different treatment groups on Saluretic, Natriuretic, Kaluretic and CAI index of different fractions of *E. alba*

Groups	Cumulative ion Concentrations (mEq/L/6h)			Saluretic Index			Na^+ / K^+	K^+ / Na^+	CAI = {Cl^-/ (Na^++ K^+)}	Diuretic Index (DI)
	Na^+	K^+	Cl^-	Na^+	K^+	Cl^-				
Control (Normal Saline)	183	18.64	90	0	0	0	9.82	0.10	0.45	
Frusemide (5 mg/kg)	282.60	43.88	119.7	1.54	2.35	1.33	6.44	0.16	0.37	3.55
Fr. *n*-hexane (250 mg/kg)	214.76	30.99	101.57	1.17	1.66	1.13	6.93	0.14	0.41	2.10
Fr. *n*-hexane (500 mg/kg)	238.16	32.11	110.79	1.30	1.72	1.23	7.42	0.13	0.49	2.95
Fr. H_2O (250 mg/kg)	190.25	27.32	92.05	1.03	1.47	1.02	6.96	0.14	0.42	1.18
Fr. H_2O (500 mg/kg)	197.51	29.60	100.5	1.07	1.59	1.12	6.67	0.15	0.44	1.41

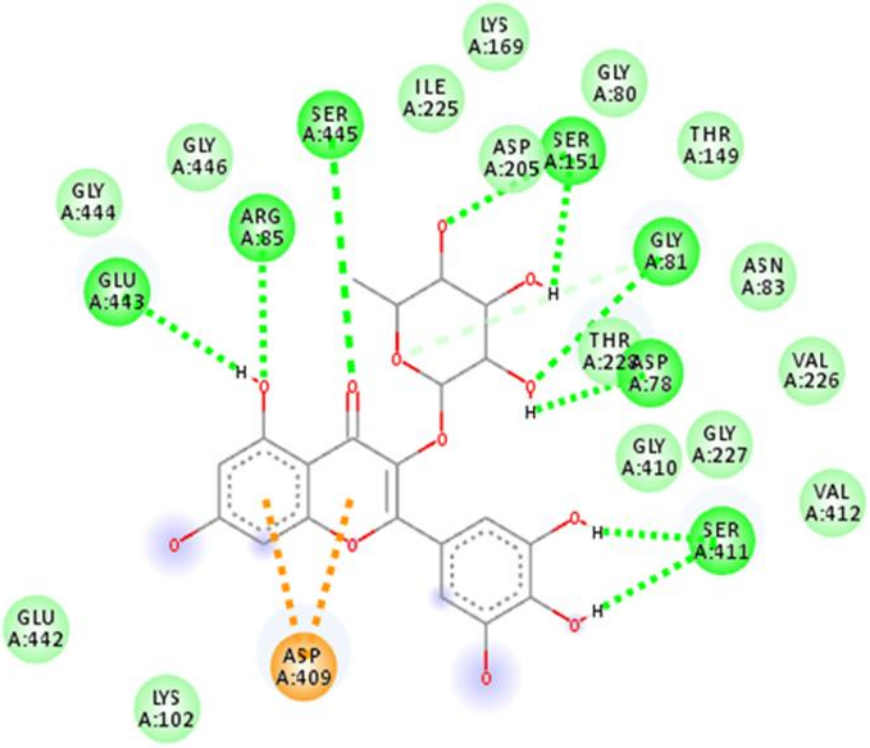
Diuretic index (DI) = Urine volume of test group/ Urine volume of control group

Molecular Docking Investigation

Molecular docking algorithms are frequently employed to forecast ligand energies and binding trends to proteins. The present study screened 08 compounds from *L. racemosa* against the diabetes-specific target protein Glucokinase (1V4S). According to the docking results, Myricitrin (**5**), Botulin (**2**), Kaempferol (**7**), and Racelactone (**1**) have the lowest docked binding energy compared to the standard Glibenclamide (Table 16).

Table 16: Docking score of reported eight (08) ligands from *L. racemosa* with 1V4S protein

Sl. No	Name of the ligands	Docking score (kcal/mol)
1	Racelactone (**1**)	-7.5
2	Botulin (**2**)	-7.9
3	3,4,3'-Tri-O-methylellagic acid (**3**)	-7.4
4	Methyl gallate (**4**)	-6.2
5	Myricitrin (**5**)	**-8.4**
6	Stigmasterol (**6**)	-7
7	Kaempferol (**7**)	-7.6
8	Isoguaiacin (**8**)	-7.3
9	Glibenclamide	**-8.5**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GLY A:81
ASN A:83
GLU A:443
THR A:228
ASP A:78
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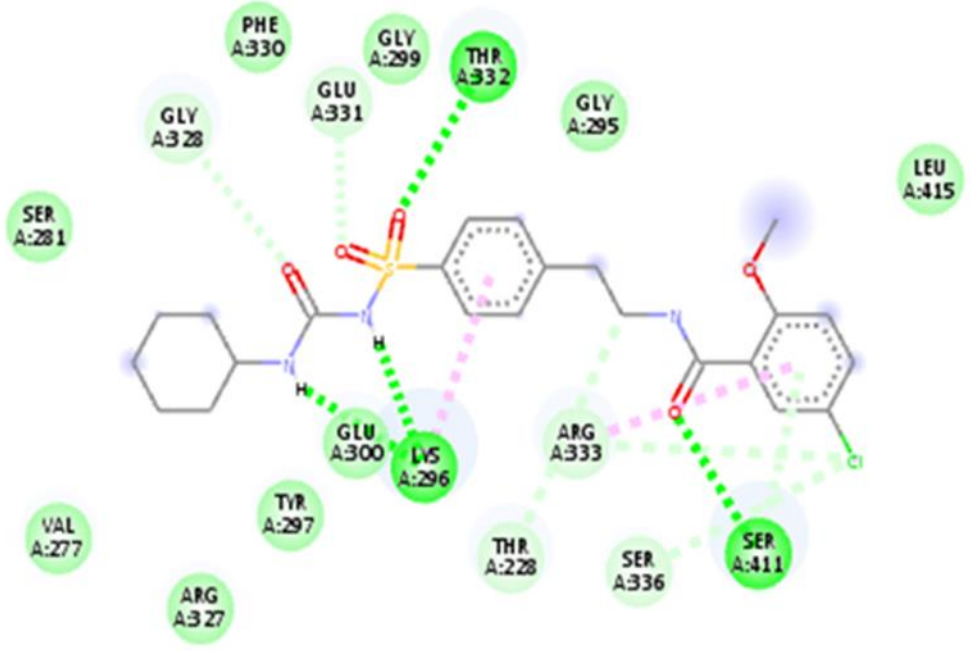
LYS A:102

(A)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LYS A:296
ARG A:333
TYR A:297
VAL A:277
THR A:228
SER A:336
SER A:411
ARG A:327

(B)

Figure 2: 2D interactions of (A) Myricitrin and (B) Glibenclamide with 1V4S protein

Table 17: Ligand binding to 1V4S protein amino acids

Ligands	**Amino acid interactions**
Myricitrin	Asp78, Asp205, Asp409, Lys102, Lys169, Glu442, Glu443, Gly80, Gly81, Gly410, Gly227, Gly444, Gly446, Arg85, Ser151, Ser445, Ser411, Ile225, Thr149, Thr228, Asn83, Val226, Val412.
Glibenclamide	Ser281, Ser336, Ser411, Gly295, Gly299, Gly328, Phe330, Glu300, Glu331, Thr228, Thr332, Leu415, Arg327, Arg333, Lys296, Tyr297, Val277.

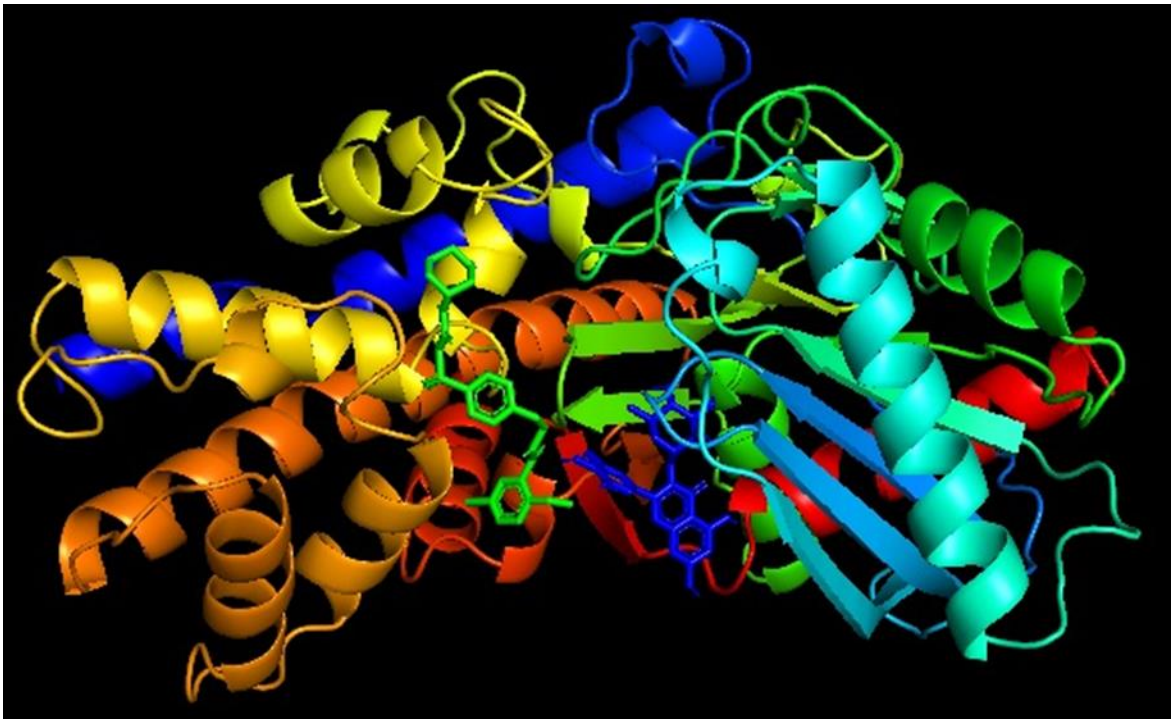

Figure 3: 3D interactions of Myricitrin (blue) and Glibenclamide (green) with 1V4S protein

DISCUSSION

Plants are blessings of mankind for its copious medicinal properties to defend against different diseases. Plants synthesize different secondary metabolites for its own protection from insects, microorganisms, diseases, and herbivorous mammals. These secondary metabolites perform momentous role on human body in various physiological disorders. According to WHO statistics, 80% of the global population either directly or indirectly uses medicinal plants to treat a variety of ailments. Preliminary phytochemical testing of the extracts of *L. racemosa* and *E. alba* suggested the presence of proteins, glycosides, alkaloids, terpenoids, flavonoids, phenolic compounds, tannins, and acidic compounds. The most

effective phytochemicals for medicinal use are flavonoids, alkaloids, tannins, and phenolic compounds. Polyphenolic products, including tannins, flavonoids, and phenolic acids, have already been proven to elicit a variety of biological reactions, including laxative, anthelmintic, antioxidant, and anti-inflammatory effects [12]. Terpenoids, flavonoids, and tannins are examples of phytochemicals that have analgesic properties [13, 14]. However, various phytochemicals existed in *L. racemosa* and *E. alba* extract helped us to perform different pharmacological tests.

The oral glucose tolerance test (OGTT) is the most reliable test for identifying borderline diabetes mellitus. The capability of the body to use glucose in the bloodstream is termed as glucose tolerance. As a result, the "glucose tolerance test" is a useful diagnostic tool for detecting diabetes mellitus, insulin resistance, diminished beta-cell activity, and occasionally acromegaly and reactive hypoglycemia [15]. Water fraction 500 mg/kg of *L. racemosa* extract gradually declines plasma glucose level in contrast to reference Glibenclamide. After 120 min, Water fraction of *L. racemosa* extract showed 4.58 mmol/L plasma sugar level at the dose of 500 mg/kg body weight compared to Glibenclamide 4.1 mmol/L. According to these results, Water fraction of *L. racemosa* extract exhibited hypoglycemic effect.

Diabetes is the ninth leading cause of death globally and one of the most common metabolic illnesses. Although there are reliable hypoglycemic agents for the control of diabetes, researchers are focusing upon multiple metabolic factors such as transporters, enzymes, and receptors in attempts to find a drug that is both more effective and less side effects. The enzyme glucokinase (GCK), which is mostly present in the liver and pancreatic beta cells, maintains blood glucose homeostasis [16]. One of the four hexokinase isozymes, glucokinase (GCK), converted glucose to glucose-6-phosphate in the hepatic and pancreatic cells. The rate at which glucose stimulates the synthesis of insulin is controlled by the perfect glucose sensor and rate-limiting enzyme GCK. Hence, the present *in silico* study is designed to determine the interaction between GCK and eight (08) reported compounds (ligands) of *L. racemosa*. In humans, pancreatic beta-cells and hepatoparenchymal cells create the majority of GCK [17]. In addition to aiding the liver in producing glycogen, GCK controls the pace at which insulin is produced by pancreatic cells in order to keep glucose homeostasis [18].

Molecular docking is a computer simulation approach that focuses on finding the ideal place of interaction between a chemical and a protein binding site. Using this method, the binding affinity is determined by predicting the ultimate orientation of the molecule within the binding site, which is chosen within the 3D coordinate space of the target. The significance and sensitivity of binding affinity values are determined by the largest negative value (greatest binding affinity or lowest binding energy), which represents the most valuable configuration of the complex formed when the associated ligand successfully binds with the target's active pockets [16]. According to binding affinity score Myricitrin (**5**) showed lowest binding score compared to standard Glibenclamide. Furthermore, by evaluating ligand binding modalities and orientation in the target protein 1V4S receptor pocket, the molecular docking model has been used to validate the anti-diabetic efficacy of Myricitrin (**5**). The ligand-receptor affinity was calculated using the binding free energy (G binding) value. The intermolecular energy, torsional free energy, and total internal energy were added to determine the binding free energy. As a result, the binding free energy was obtained and subtracted from the unbound system's energy. The optimal interaction site was found using the conformation with the lowest energy binding value.

Figure 2 showed the 2D interaction between glucokinase 1V4S protein with Myricitrin (**5**) and Glibenclamide. Compared to other ligands, Myricitrin (**5**) showed strong binding with glucokinase in terms of binding energy (-8.4 kcal/mol) compared to Glibenclamide (-8.5 kcal/mol). Myricitrin (**5**) formed

seven hydrogen bonding interactions with the amino acid residues of ARG85, ASP78, GLY81, SER151, SER411, GER445 and GLU443 (Figure 2). These kinds of 3D interactions were found in docking studies, which proved that particular ligand has a high affinity for the target protein (Figure 3).

Diuretics enhance the excretion of urine and optimize blood volume that finally regulate blood pressure. They primarily act on various nephron segments, increasing urine output. Diuretics may also accelerate the excretion of electrolytes. Diuretics are prescribed for treating a number of conditions, including kidney disease, influenza, water poisoning, hypertension, liver cirrhosis, and heart failure. A prior study found that a good diuretic activity is indicated by a diuretic index (DI) value greater than 1.50. On the other hand, the DI values between 0.72 and 0.99 and 1.00 and 1.50, respectively, show mild and moderate diuretic activity. There is no diuretic activity when the diuretic index value is less than 0.72 [19]. The extract displayed good diuretic activity as seen by the *n*-hexane fraction of *E. alba* extract, which showed DI 2.10 and 2.95 at the doses of 250 and 500 mg/kg while typical furosemide showed DI 3.55 (Table:15).

The plant extract's diuretic efficacy was dose- and time-dependent, suggesting that this impact is real, intrinsic, and potentially receptor-mediated [20]. For the management of hypertension, peripheral edema, ascites, and congestive heart failure, renal excretion of electrolytes is equally important as renal excretion of water [20]. The extract increased diuresis, which was evident in the electrolyte excretion as well. In a dose-dependent way, it markedly enhanced the excretion of urine electrolytes (Na^+, K^+, and Cl^-). Despite this, all fractions of the extract from *L. racemosa* and *E. alba* demonstrated higher K^+ excretion than the control. The ratio of Na^+/K^+ can be used to calculate the natriuretic activity (also known as the aldosterone secretory index) of the plant extract. Values higher than 2.0 suggest an effective natriuretic effect, whereas ratios higher than 10.0 indicate a potassium-sparing effect [21]. Between a score of six and seven, all fractions of the *L. racemosa* and *E. alba* extract showed strong natriuretic effects.

By inhibiting the Na^+-K^+-$2Cl^-$ symporter in the thick ascending limb of the loop of Henle, stimulating the production of renal prostaglandins, and inhibiting the carbonic anhydrase enzyme in the proximal convoluted tubule (PCT), loop diuretics increase the urinary flow rate and urinary excretion of Na^+, K^+, and Cl^- [22-24]. Since hypokalemia is one of the main side effects of loop and thiazide diuretics, oral K^+ supplements or potassium-sparing diuretics that lower urine K^+ excretion may be required [24-26]. The extract's ability to inhibit carbonic anhydrase is measured by the ratio of $Cl^-/ [Na^+ + K^+]$. It is possible to rule out carbonic anhydrase inhibition with values between 1.0 and 0.8. One can presume that there is enzyme inhibitory activity when the ratio decreases [21]. The carbonic anhydrase inhibitory indices of all fractions of *L. racemosa* and *E. alba* extract as well as normal Frusemide were less than 0.5 (Table 9 & 15). Accordingly, our study suggests that these plants may work as loop diuretics by inhibiting the carbonic anhydrase enzyme in the renal tubules, which raises urine pH.

CONCLUSION

Because medicines are essential to having a healthy population, which drives and sustains the economy, the significance of medicinal plants to the economy of developing nations like Bangladesh remains crucial and strategic. Maximum medicinal plants are abundantly indigenous to our country and we can use them at very cheap rates to produced different dosage forms that will boost our national economy. Pharmaceutical products based on medicinal plants strengthen the healthcare delivery system by providing the majority of the population with essential medications at reasonable costs. Moreover, not only medical benefits, implementation of this program will help boost revenue thereby contributing to wholesome national economy as well. Diuretics

decrease blood volume by enhancing urine volume and control blood pressure. Diuretic activity is directly related to kidney and other parts of urinary system. *L. racemosa* and *E. alba* have no traditional uses related to kidney and other parts of urinary system. So, in this experiment, both plants exhibited little increase in urine volume. Myricitrin (**5**) exhibited good docking score -8.4 kcal/mol against 1V4S protein that reflect good antidiabetic activity. So, Myricitrin (**5**) may be the lead compound for the future antidiabetic drug development. The results of this study will contribute to the increasing economic worth of medicinal plants, which developing nations must capitalize on to enhance their economic and healthcare systems.

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استكشاف الأنشطة المضادة لمرض السكر ومدرة للبول لنباتي Lumnitzera racemosa و Eclipta alba من خلال دراسة الالتحام الجزيئي

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ملخص

كان الهدف من الدراسة الحالية هو النظر في أوراق Lumnitzera Racemosa العائلة: Combretaceae والجزء الهوائي من Eclipta alba العائلة: (Asteraceaeلمكوناتها الكيميائية النباتية والأنشطة الدوائية المختارة (مدر للبول ومضاد لمرض السكر). تستخدم الأدوية المدرة للبول لعلاج ارتفاع ضغط الدم. يعد مرض السكري وارتفاع ضغط الدم من الأمراض الشائعة لدى مرضى الشيخوخة وكان هدفنا هو استكشاف النباتات الطبية في سوندربانس لإيجاد علاج لهذه الأمراض. أظهرت الدراسة الكيميائية النباتية لنبات L. Racemosa و E. alba وجود الكربوهيدرات والجليكوسيدات والسكر المختزل والعفص والفلافونويد والقلويدات والبروتينات والصمغ والمنشطات والسابونين والمواد الكيميائية الحمضية. الصمغ، قلويد، الستيرويد، وتيربينويد. تمت تجزئة مستخلص النباتين اعتمادًا على القطبية باستخدام ن-هكسان (غير قطبي)، وخلات الإيثيل (قطبي متوسط) والماء (قطبي). في اختبار نشاط مدر البول باستخدام فئران ألبينو سويسرية، لم يُظهر أي من أجزاء L. Racemosa وE. alba نشاطًا مدرًا للبول، لكن n-hexane 500 mg/kg من E. alba أظهر نشاطًا مدرًا للبول قليلًا مقارنة بالفروسيميد القياسي. تم استخدام الفئران البيضاء السويسرية لتقييم اختبار تحمل الجلوكوز عن طريق الفم ((OGTTمن أجل قياس النشاط المضاد لمرض السكر. في OGTT، أظهر جزء الماء 500 ملغم/كغم من L. Racemosa وE. alba نشاطًا لخفض نسبة الجلوكوز في الدم مقارنةً بـ Glibenclamide القياسي. لقد أجرينا دراسة سيليكو بين ثمانية (08) مركبات من L. Racemosa ضد بروتين 1) V4Sالجلوكوكيناز البشري). بالمقارنة مع جليبينكلاميد الكلاسيكي (-8.5 كيلو كالوري/مول)، كان للميريسيترين درجة إرساء جيدة تبلغ -8.4 كيلو كالوري/مول. بناءً على هذه النتائج، افترضنا أن L. Racemosa وE. alba يمكن أن يكونا مصدرًا محتملاً للعلاجات العلاجية لارتفاع ضغط الدم وارتفاع السكر في الدم.

الكلمات الدالة: Lumnitzera Racemosa، Eclipta alba، نشاط مدر للبول، نشاط مضاد لمرض السكر، دراسة الالتحام الجزيئي.

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