Evaluation of Anti-hyperlipidemic and Hepato-renal Protective Role of Ethanolic Extract from *Mimosa pudica* Leaves on High Fat Induced Hyperlipidemic Rat Model

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Traditional herbal medicine is an ancient field of study that derives its inspiration from natural sources. For thousands of years, it has helped humans fight diseases and protect their health, energy, and ability to reproduce. This study examined the effects of a *Mimosa pudica* extract on the lipid profiles of rats with hyperlipidemia caused by a high-fat diet. Group 6, which was given a high fat dose of 900 mg/kg, had statistically significant outcomes ($p < 0.05$) in terms of Serum Glutamic Pyruvic Transaminase (SGPT) levels in relation to the liver function test. The results of the renal function test showed that group 6, which received doses of 900 mg/kg, had a statistically significant elevation in urea levels ($p < 0.05$). The analysis of creatinine levels in groups 4, 5, and 6 revealed statistically significant results ($p < 0.05$) for doses of 300, 600, and 900 mg/kg, respectively. Groups 5 and 6 had statistically significant results ($p<0.05$) for HDL and LDL. Groups 5 and 6 had statistically significant outcomes when administered dosages of 600 and 900 mg/kg, respectively. A statistically significant difference ($p < 0.05$) was seen in the triglyceride levels between groups 5 and 6. The total cholesterol levels in Group 6, which received a dose of 900 mg/kg, showed statistically significant results ($p < 0.05$). The main goal of this study is to find out what happens to the lipid profile of hyperlipidemic albino rats when they are given an ethanolic *M. pudica* extract. The current study in rat models showed that *M. pudica* had a hyperlipidemic impact, significantly increasing cholesterol levels. The bioactive constituents of the plant, such as tannins, flavonoids, and phenolics, are responsible for its cardioprotective properties. To gain a better understanding of the mechanisms of action of certain bioactive compounds, further analysis of *M. pudica*'s ethanolic extract is required.

1. INTRODUCTION

The liver, the largest glandular organ, controls the majority of an individual's physiological activities. The liver receives the entire volume of an individual's blood multiple times throughout the day. It has a vital function in human metabolism [1,2]. Excessive alcohol use, drug addiction, exposure to certain toxic compounds, or infection by viruses or parasites can induce an elevation in reactive oxygen species (ROS) activity, such as OH, H_2O_2, and O_2, which can lead to cellular damage in the liver [3]. The Centers for Disease Control and Prevention surveyed 1492 clinicians providing ambulatory treatment in non-government facilities, and found that hyperlipidemia ranks second only to hypertension among the top ten chronic conditions they see [4]. According to the research that has been conducted, the primary reason for hyperlipidemia is the excessive consumption of foods that are rich in fat [5]. The liver undergoes extensive metabolism of the most commonly used anti-hyperlipidemic drugs, such as Atorvastatin, Pravastatin, Fluvastatin, Simvastatin, Lovastatin, and Rosuvastatin, which results in very low bioavailability [6].

Statins function as reversible competitive inhibitors of the enzyme 3-hydroxy-3-methylglutaryl-co-A reductase (HMG-CoAR). By doing so, they reduce the production of cholesterol within cells. The ability of statins to penetrate the hepatocyte and inhibit HMG-CoAR dictates their pharmacological response [7]. Muscle problems, also known as statin-associated muscle symptoms (SAMS), are the most common side effect that limits the use of statins. Other side effects include diabetic mellitus (DM) and problems with the central nervous system [8]. In addition to the severe adverse effects, these artificial medications are expensive, and the patient may face financial challenges if they complete the whole course of treatment [9]. Therefore, it is essential to develop powerful antihyperlipidemic medications that have minimal negative side effects. Plants are crucial in the process of discovering and synthesising novel drugs. They are a valuable and plentiful source of naturally occurring therapeutic chemicals [10].

Chemical compounds from medicinal plants may be helpful, say experts. Therefore, researchers are continually looking for new herbal medicines and other plant-derived therapies for a variety of disorders [4]. Traditional medicine employs herbal remedies, nutritional supplements, and alternative medicine in many nations. Most healthcare requirements rely on traditional, recently popular medications [11]. Medical plants

include several chemical components, which give them different pharmacological and therapeutic benefits. These include tanning agents, glycosides, alkaloids, saponins, polysaccharides, essential oils, terpenoids, resins, and plant lipids [12–14]. Genetically altering plants allows for precise chemical concentration control, resulting in the desired medical effect. Reverse genetics may boost alkaloids and other secondary metabolites [15]. Worldwide scientific advances have led to extensive study of the medicinal properties of plant species [16]. Plants are safe, powerful, and cheaper than synthetic drugs.

Mimosa pudica L. belonging to the family Mimosaceae found in tropical America and Australia, and India [17]. *M. pudica* L. is a creeping annual or perennial herb. It has been identified as lajjalu in Ayurveda and has been found to have antiasthmatic, aphrodisiac, analgesic, and antidepressant properties. *M. pudica* is known to possess sedative, emetic, and tonic properties, and has been used traditionally in the treatment of various ailments including alopecia, diarrhea, dysentery, insomnia, tumor, and various urogenital infections. Phytochemical studies on *M. pudica* have revealed the presence of alkaloids, non-protein amino acid (mimosine), flavonoids C-glycosides, sterols, terpenoids, tannins, and fatty acids [17,18]. In traditional folk agent that is utilized in the management of various diseases including cognitive dysfunction, demonstrated the improvement of memory in experimentally induced amnesia. The beneficial effects of *M. pudica* have been reported due to the presence of flavonoids, saponins, alkaloids, polyphenols, and steroids [19]. It has Wound healing activity, Regeneration of sciatic nerve, Antidepressant action, Anticonvulsant action, Hyperglycemic effect, Diuretic effect, Effect on uterine bleeding, Antifertility activity, Spasmogenetic potential, Antihepatotoxic and antioxidant potential, Antivenom activity, Antimicrobial properties and Aphrodisiac property [17,20,21,22].

The objective of our current investigation is to assess the hepatoprotective properties of *M. pudica*. This finding offers a cost-efficient substitute for statins, which are pricier and include certain undesirable consequences. The anti-hyperlipidemic properties of the *M. pudica* plant could potentially lead to the development of highly effective medications for the treatment of cardiovascular illnesses and associated ailments.

2. MATERIALS AND METHODS

2.1 Plant Collection and Extract Preparation

The specimens of *Mimosa pudica* leaves were collected from a Dhaka-based market. The National Herbarium of Bangladesh confirmed the validity of the material. Rinsing *Mimosa pudica* well with water was the initial step, followed by letting it dry naturally. Lastly, the dried leaves were ground into a fine powder of 700gm. The powder was soaked in 70% ethanol for fifteen days. Shaking, both intermittent and forceful, was also done. Filtration was then performed on the solution. A rotary evaporator was used to dry the collected filtrate under reduced pressure and temperature. In the end, the raw materials were tested for any relevant pharmaceuticals.

2.2 Drugs and Chemicals

Sigma-Aldrich, based in Germany, provided the ethanol. Healthcare Pharmaceutical Limited sent a free sample of Rosuvastatin, a commonly recommended drug for decreasing high cholesterol levels in the blood. We assessed the biochemical parameters using the Humalyzer 3000, a largely automated clinical chemistry analyzer. We bought the ingredients for the high-fat diet at a supermarket.

2.3 Experimental Animal Procurement, Nursing, and Grouping

The 90 male rats, ranging in weight from 120 to 150 grams, were procured from Jahangirnagar University in Savar, Dhaka. The specimens were maintained in a controlled environment that varied the temperature by three degrees Celsius, the relative humidity (65%) by five and a half percent, and the length of the day and night by twelve hours. Institution of Nutrition and Food Science (INFS) at University of Dhaka supplied this setting. They were given regular food and were permitted to drink water that had been cleansed. In order to see how the animals adapted, they were housed in this habitat for at least one week prior to the study. All procedures used in the experiments were in accordance with the standards established by the IEAC. Nine groups, each with ten rats, were created from a pool of ninety rats.

Table 1. Composition of high fat diet

Food Ingredients	Composition
Lipid (50%)	Milk powder (10%)
	Ghee (30%)
	Mutton fat (40%)
	Coconut oil (10%)
	Butter (10%)
Carbohydrate (40%)	Boiled rice (40%)
	Smashed potato (40%)
	Boiled corn (20%)
Protein (10%)	Dry powdered prone (40%)
	Dry boiled mutton (20%)
	Cheese (20%)
	Egg (20%)

Table 2. Antihyperlipidemic grouping details

Group number	Group Status	Treatment specimen & Dose
1	Negative Control	Physiological Saline
2	HFD Control	High Fat Diet
3	High Fat Diet +RV_{10}	High Fat Diet + Rosuvastatin
4	High Fat Diet+ *M. pudica*	High Fat Diet+ MP_{300}
5	High Fat Diet+ *M. pudica*	High Fat Diet + MP_{600}
6	High Fat Diet+ *M. pudica*	High Fat Diet + MP_{900}
7	*M. pudica*	MP_{300}
8	*M. pudica*	MP_{600}
9	*M. pudica*	MP_{900}

HFD= High fat diet, RV= Rosuvastatin, MP= M. pudica.

Table 3. Application of treatment efficacy

Group Number	Group Specification	Treatment species	Dose treatment species (mg/kg)
1	Negative control	Physiological saline	10 ml/kg
2	High Fat	N/A	N/A
3	HF+RV$_{10}$	Rovast 10mg/kg	10
4	HF +MP$_{300}$	*M. pudica*	300
5	HF+MP$_{800}$	*M. pudica*	600
6	HF+MP$_{900}$	*M. pudica*	900
7	MP$_{300}$	*M. pudica*	300
8	MP$_{600}$	*M. pudica*	600
9	MP$_{900}$	*M. pudica*	900

HFD= High fat diet, RV= Rosuvastatin, MP= M. pudica

2.4 Experimental Design

For the purpose of studying its anti-hyperlipidemic activity, rats were weighed individually and then split into nine separate groups. Each group had 10 rats, and the distribution of the animals was determined by their weight. Table 1 displays the atorvastatin control group, which consists of rats given atorvastatin in conjunction with a high-fat diet. This was done because administering the drug alone would have been fatal for the animals. The presence or absence of a therapeutic treatment in this group of rats is indicated by the value of N/A.

2.5 High Fat Diet

The high-fat diet was modified based on the composition supplied by Levin and Dunn-Meynell [23]. The high fat diet is composed of 50% lipid, 40% carbohydrate, and 10% protein. The diet's composition is shown in Table 1. After mixing the ingredients thoroughly, the high fat diet was given to the rats to induce obesity for 10 weeks.

2.6 Evaluation of Anti-hyperlipidemic Activity

For this experiment, 90 rats were randomly picked and equally divided into fourteen groups.

Sacrifice and analysis of blood parameter: After 10 weeks, all rats belonged too different groups were sacrificed and blood samples were collected and taken in test tube and one drop of heparin was added in each test tube to counteract the blood clotting process. Next, bloods were taken in eppendorf tube and placed in centrifuge machine. Subsequently, the samples were centrifuge at 5000 rpm for 5 minutes. Afterwards, supernatant serum was collected carefully using micro-pipette. Finally these serums were used analyze different parameter by Humalyzer 3000.

Statistical analysis: Microsoft Excel was utilized to capture and evaluate all of the outcomes, or raw data, in numerical terms. Data was analyzed using descriptive statistics and reported as mean±SEM. To evaluate statistical significance, we used SPSS 16's "one-way Anova test" to examine inter-group heterogenicity based on different biological factors. The occurrences were deemed statistically significant because the 'p' value was less than 0.05 ($p < 0.5$).

3. RESULTS AND DISCUSSION

Traditional medicine and ethnomedicine, which examine the healing practices of different ethnic groups, have existed since the beginning of human civilization. Traditional medicine has a long history of using the therapeutic capabilities of the earth's natural resources. Traditional medicine in many nations and cultures is based on the use of herbs—plants or plant products—and plant extracts as major medicinal components. People have used traditional plant and herb extracts, along with isolated active components, as remedies for many years. This research used a rat model of high-fat-induced hyperlipidemia to evaluate how a *M. pudica* extract changed lipid profiles.

Group 6 ingested 900 mg/kg of fat and had significantly higher SGPT levels ($p < 0.05$). Even with a large dosage, SGOT levels did not decrease appreciably. However, there was a similar dose-dependent decrease in the SGOT level. The other groups did not provide any statistically meaningful findings. Two further studies found the same findings [24,25]. The renal function test findings indicated that group 6,

Table 4. Lipid profile of *M. pudica*

Groups	SGPT	SGOT	Creatinine	Urea	TC	HDL	LDL	TG
NC	33.67±3.26	38.21±4.23	0.53±0.24	37.42±4.10	125.24±6.90	84.74±4.90	38.42±3.38	47.29±6.18
HFD	86.96±7.24	89.30±7.52	2.89±0.83	102.36±9.36	229.46±12.41	41.46±3.30	146.39±13.49	114.79±13.26
HFD+ RV$_{10}$	58.24±5.73	62.46±5.23	1.46±0.63	57.57±8.26	161.46±11.93	65.53±5.53	80.22±10.53	66.43±8.93
HFD +MP$_{300}$	85.75±6.91	89.10±4.91	2.32±0.64*	100.46±8.62	226.20±8.78	44.26±5.59	142.20±13.43	110.21±6.97
HFD+MP$_{600}$	82.21±4.81	87.54±5.36	2.01±0.79*	97.39±7.70	221.34±9.91	49.39±6.90*	136.79±12.21*	104.36±7.47*
HFD+MP$_{900}$	80.42±3.75*	86.86±6.29	1.89±0.63*	93.10±6.61*	215.53±7.07*	54.26±8.29*	131.29±14.79*	96.24±6.90*
MP$_{300}$	35.22±2.16	37.30±3.31	0.63±0.37	35.50±3.63	127.73±6.24	82.82±4.39	39.59±4.57	49.21±7.93
MP$_{600}$	31.67±4.26	35.50±4.02	0.68±0.52	38.26±4.67	121.93±5.73	84.63±5.53	36.63±5.57	44.22±6.50
MP$_{900}$	35.57±3.26	36.31±3.40	0.54±0.83	32.29±3.17	126.22±6.78	83.19±2.93	33.27±6.21	47.70±6.22

Note: The results were expressed in Mean±SEM (standard mean error) *p< 0.05, **p< 0.01, and ***p< 0.001 were considered as statistically significant. The statistical analysis followed by one-way analysis of variance compared to the control. HFD=High fat diet, RV= Rosuvastatin, MP= M. pudica, TC=Total Cholesterol, TG= Triglyceride, HDL= High Density Lipoprotein, LDL= Low density lipoprotein, SGPT = Serum Glutamic Pyruvic Transaminase, SGOT= Serum Glutamic Oxaloacetic Transaminase

which got doses of 900 mg/kg, had substantially higher levels of urea. Creatinine levels in groups 4, 5, and 6 were statistically significant ($p<0.05$) at doses of 300, 600, and 900 mg/kg, respectively. The other groups did not provide any statistically meaningful findings. The results of two separate investigations on the problem were identical [26,27]. Groups 5 and 6 showed statistically significant results ($p<0.05$) for HDL and LDL. Groups 5 and 6 showed statistically significant results when given doses of 600 and 900 mg/kg. Triglyceride levels differed significantly ($p < 0.05$) between groups 5 and 6. Group 6 got a 900 mg/kg dose, resulting in statistically significant total cholesterol levels ($p<0.05$). The other groups did not provide any statistically meaningful findings. The findings were consistent in two more studies [28,29].

4. CONCLUSION

The primary focus of this study was on the hepatoprotective properties of an ethanolic *M. pudica* leaves extract. According to the findings of this research, an ethanol extract from the *M. pudica* plant may provide protection against excessive cholesterol, liver damage, and decreased kidney function. We need additional research to identify the components of the whole extract responsible for the anti-hyperlipidemia and diabetic benefits. Once we identify the active chemicals, we can proceed with a thorough examination.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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