



Therapeutic Insights into *Mangifera indica*: Managing Diabetes and Hyperlipidemia in Alloxan-treated Rats

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Authors' contributions

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

Article Information

DOI: <https://doi.org/10.9734/jocamr/2025/v26i1617>

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: <https://www.sdiarticle5.com/review-history/129133>

Original Research Article

Received: 08/11/2024

Accepted: 10/01/2025

Published: 15/01/2025

ABSTRACT

People have used herbal medicine for healing since the dawn of human civilization. This study investigated the antidiabetic efficacy and lipid profile of *Mangifera indica*. We evaluated the antidiabetic activity through the alloxan-induced diabetic model. The 750 mg/kg dosage in group 6 yielded statistically significant findings ($p < 0.05$) regarding antidiabetic efficacy. The group receiving a 750 mg/kg dose exhibited statistically significant outcomes for total cholesterol, HDL, and triglycerides, with results of $197.40 \pm 6.28^*$, $52.21 \pm 4.47^*$, and $101.75 \pm 3.29^*$, respectively ($p < 0.05$).

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Cite as: Shawon, Somaya Jannat, Sabbir Khan, Sara Rahmat, Mafruza Khanam Prottasha, Md Navid Rahman Nafsan, Sadia Tasnim, and Israt Jahan Rasna. 2025. "Therapeutic Insights into *Mangifera Indica*: Managing Diabetes and Hyperlipidemia in Alloxan-Treated Rats". *Journal of Complementary and Alternative Medical Research* 26 (1):106-14. <https://doi.org/10.9734/jocamr/2025/v26i1617>.

However, no groups demonstrated statistically significant outcomes regarding LDL and triglyceride levels, despite a decrease in these parameters in the blood following the administration of the extract. No groups exhibited statistically significant outcomes regarding SGPT and SGOT; however, the levels of these two parameters decreased in the blood following the administration of the extract. In the kidney function test, urea and creatinine levels in group 6 demonstrated statistical significance ($p < 0.05$ at a dosage of 750 mg/kg, with values of 990.42 ± 4.82 and 11.70 ± 0.82 , respectively).

Keywords: *Mangifera indica*; HDL; LDL; diabetes; herbal medicine; triglyceride.

1. INTRODUCTION

Diabetes mellitus (DM) is a metabolic disorder marked by persistently elevated blood glucose levels (BGL) resulting from insufficient insulin action, synthesis, or both in target tissues. Persistent microvascular (retinopathy, neuropathy, and nephropathy) and macrovascular (coronary and peripheral artery diseases and stroke) problems, associated with chronic hyperglycemia, characterize all forms of diabetes mellitus. These issues result in organ damage and fatality, sometimes identified at an advanced stage or without sufficient medical supervision (Yikna & Yehualashet, 2021). The liver, the biggest glandular organ, oversees most physiological functions in the human body. The liver is the organ responsible for absorbing an individual's entire blood volume multiple times daily. It is essential for human metabolic processes (Himi et al., 2024). Excessive alcohol intake, substance dependence, exposure to some hazardous substances, or infection by viruses or parasites may result in increased levels of reactive oxygen species (ROS), including OH, H₂O₂, and O₂ (FM et al., 2023). This might result in hepatocellular damage. The Centers for Disease Control and Prevention performed a study involving 1,492 doctors providing ambulatory care in non-governmental institutions. The study indicated that hyperlipidemia ranks as the second most common chronic illness seen by these physicians, with hypertension being the only condition more often encountered (Baroi et al., 2023). The study's findings indicate that the predominant cause of hyperlipidemia is the excessive intake of high-fat meals (Zhang et al., 2021). The liver is essential for the metabolism of widely used anti-hyperlipidemic medications, including atorvastatin, pravastatin, fluvastatin, simvastatin, lovastatin, and rosuvastatin. As a result, the bioavailability of these medications is very low (Srinivasa et al., 2011). Statins may temporarily block the enzyme 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMG-

CoAR). This enzyme reduces cholesterol levels. This enables the reduction of cholesterol production inside the cells. Statins may penetrate hepatocytes and inhibit HMG-CoAR, which underlies their pharmacological actions (Schachter, 2005). Statin-associated muscle symptoms (SAMS), sometimes referred to as muscular complications, are the principal adverse effects that restrict the use of statins. Two other potentially harmful outcomes are the development of diabetes mellitus (DM) and problems involving the central nervous system (Thompson et al., 2016). These synthetic medications not only exhibit significant side effects, but they are also costly, possibly imposing financial burdens on patients who must persist in their use during the whole treatment regimen (Rupak et al., 2022). Consequently, it is essential to formulate highly efficacious antihyperlipidemic agents with low adverse effects. Plants are crucial in the identification and synthesis of innovative medicines (Islam et al., 2022). They function as a valuable and plentiful source of naturally occurring compounds for medicinal purposes. Experts in the field suggest that certain chemical compounds derived from medicinal plants have therapeutic capabilities. Consequently, researchers continually seek innovative herbal treatments and other plant-derived medications to successfully address diverse conditions (Baroi et al., 2023). Several countries globally have historically used traditional medicines as cures, originating from botanical sources, nutritional supplements, and alternative therapeutic practices. In recent years, the use of traditional medicine has markedly risen, with several individuals nationwide relying on it as a primary mode of healthcare (Chowdhury et al., 2024; Hossain et al., 2010; Gondi et al., 2015). Medicinal plants include a variety of chemical compounds, allowing them to produce a wide array of pharmacological and therapeutic effects. These compounds include many elements, such as tanning agents, glycosides, alkaloids, saponins, polysaccharides, essential oils, terpenoids, resins, and plant lipids

(Lima et al., 2023; Chowdhury et al., 2024; Saxena et al., 2013). Genetically modified plants enable precise regulation of chemical concentrations, ultimately achieving the intended medicinal effect. Reverse genetics has several potential applications, one of which is the enhancement of secondary metabolite production, including the synthesis of alkaloids (Lima et al., 2023). Global improvements in scientific study have resulted in an increased examination of the medicinal properties of plant species (Pracheta et al., 2011). People are increasingly favoring plants due to their intrinsic safety, robust pharmacological attributes, and economic advantages over manufactured medications.

The plant *Mangifera indica* L., cultivar Anwar Ratol, is renowned for its sweetness and is a member of the Anacardiaceae family. The genus *Mangifera* has 69 species, of which less than half provide edible fruits. Historically, people have used the bark of the mango plant to treat conditions such as diarrhea, cancer, diabetes, prostatitis, toothache, cough, and infections of the urinary system and skin. The stem bark serves as an emetic, diuretic, antibacterial, astringent, and hepatoprotective agent (Akram et al., 2014). Research indicates that the stem bark had anti-inflammatory and anti-amoebic effects, inhibited DNA damage and lipid peroxidation in rats, and displayed immunomodulatory and analgesic activities. Leaf extracts can protect the liver, stop ulcers from forming, lower cholesterol, fight free radicals, and kill both gram-positive and gram-negative pathogens (Mustapha et al., 2014; Nguyen et al., 2016). Extracts from the peel, stem bark, and leaves have hypoglycemic action in diabetic rats. Plant seeds have shown antimicrobial properties. Numerous substances extracted from the stem bark, leaves, and fruits include mangiferin, rhamnetin glycoside, quercetin, kaempferol O-glycoside, Indicoside A, Indicoside B, and manghophanal (Mustapha et al., 2014; Gupta, 2018).

This research examines the anti-diabetic properties and lipid profile of an ethanolic extract derived from *Mangifera indica*.

2. MATERIALS AND METHODS

Drugs, Chemicals, and Instruments: Ethanol and alloxan were procured from Sigma Aldrich in Germany. Healthcare Pharmaceutical Limited supplied us with a gratis sample of metformin, a widely used treatment for diabetes. The blood

serum analysis kits for many biomarkers were obtained from Plasmatic Laboratory Products Ltd. in the United Kingdom. This research used the Alere Inc. glucometer. We obtained it from Shahbag in Dhaka, Bangladesh. We evaluated the biochemical parameters with the Humalyzer 3000, a semiautomated clinical chemistry analyzer.

Plant Collection and Extract Preparation:

Three distinct regions in Bangladesh were used to gather *Mangifera indica* plants: North Bengal, a hill-track area, and a low-land area. Following that, the subsequent step involved authentication and taxonomic identification. Bangladesh's National Herbarium kept the plant specimen in compliance with the relevant regulations. The leaves were dried in a shaded area for seven to ten days, then finely grind them. The powdered leaves were vigorously stirred for 96 hours while being soaked in a solution of 70% ethanol. After the soaking process, the extract was filtered, and the resulting liquid was collected. We concentrated it using a rotating evaporator. We collected and stored the dried extract stored in the refrigerator for future use.

Experimental Animal Handling: One hundred male Wistar rats, weighing between 125 and 200 grams, were acquired from the Pharmacy Department of Jahangirnagar University in Dhaka, Bangladesh. The rats were housed in a regulated environment at the Institute of Nutrition and Food Science, University of Dhaka, with a 12-hour light/dark cycle and a stable temperature of 25°C. We consistently supplied the participants with a standardized pellet diet and fresh water. The rats were placed at the facility to acclimatize before the trial began. The rat studies complied with the regulations established by the Institutional Animal Ethics Committee (IAEC). Ethical permission was obtained from the Department of Zoology at Dhaka University, under issue number 147/pharm.science.ewu. The researchers attended to and supervised the animals in compliance with the protocols established by the Swiss Academy of Medical Sciences (SAMS) and the Swiss Academy of Sciences (SCNAT).

Experimental Guidelines: The protocols outlined in the 2013 Declaration of Helsinki were strictly followed throughout the testing process. The research followed all of the "3R" guidelines, which are the cornerstones of Swiss and international laws governing the research on animals. The "R" prefix stands for "replacement,"

which may mean either an outright substitution (like using CGI instead of real animals) or a more nuanced change (like exchanging invertebrates for cells or tissue cultures) of the original organism. An animal model was used to ensure that the study was conducted thoroughly. Because of their unique pancreas and beta cells, rats were selected as study subjects for antidiabetic studies. Because of their bony backbones, mammals stand in stark contrast to invertebrates. The second "R" stands for "reduction," which refers to methods that either maximize the amount of data collected from each animal or minimize the number of animals required to collect enough data for study. The sample size estimate from the power analysis approach informed our selection of 10 rats for this investigation. We used this method to make sure everyone followed the rules. Reducing the pain and misery experienced by experimental animals is the third "R" in refinement. To make the rats more comfortable during surgery and lessen the pain from pinching, they were given a little isopropyl alcohol to massage on their tail tips before and after each blood glucose level test. In compliance with the 2013 revision to the Guidelines for the Euthanasia of Animals, the rats were given enough food throughout the experiment and then put to sleep in a painless manner at the end.

Experimental Design: We divided the rats into groups based on their body weight and subsequently tested them for antihyperglycemic action (Table 1). The rodents were categorized into groups according to their body weight, with 10 rats in each group. Table 1 illustrates the alloxan control group, consisting of rats that received only alloxan therapy. N/A indicates the absence of therapeutic treatment in this group.

Biological Sample Collection: Blood samples were collected by puncturing the rat's tail tip in order to assess blood glucose levels. For comparison's sake, when the animal was killed, its blood was quickly drawn from its heart and placed in a microcentrifuge tube. The fluid that forms on top was acquired by centrifuging the samples that were collected for 5 minutes at 5,000 rpm. Biochemical testing was made possible once the fluid was transferred to another microcentrifuge tube. In order to analyze the kidney and liver functions, the organs were quickly removed from the animal after sacrifice and washed extensively with a cold salt solution. In order to assess the rats' antihyperglycemic effect, we divided them into many groups based

on their weight (Table 1). Ten rats each group were used for the animals' weight analysis. Alloxan was the only treatment administered to the rats in the alloxan control group (Table 1). No therapeutic treatment is administered to this group when the indication is N/A.

Estimation of Biochemical Parameters: By using a glucometer, the blood glucose level was ascertained. The Humaluzer 3000 was one of many tests administered, along with those for the lipid profile (HDL, LDL, Cholesterol, triglyceride), kidneys (Urea, Creatinine), and liver (SGPT and SGOT). We also tested liver and kidney samples for gluconeogenic and glycolytic enzyme activity

Statistical Analysis: Using the spreadsheet program MS Excel, we documented and examined all of our results, or raw data, in terms of numerical parameters. Descriptive statistics were applied to the collected data, and the results were presented as mean SD. We used the "One-way ANOVA test" in SPSS 16 to analyze inter-group heterogeneity according to several biological parameters in order to determine statistical significance. The events are deemed to have strong statistical significance since the 'p' value was lower than 0.05 ($p < 0.05$).

3. RESULTS AND DISCUSSION

Herbal medicine involves the utilization of medicinal plants for the prevention and treatment of illnesses, encompassing both traditional remedies prevalent in various cultures and the application of standardized and titrated herbal extracts. This study examined the antidiabetic effects and lipid profile associated with the herb *Mangifera indica* in a murine model. Diabetes represents a significant health challenge in the twenty-first century. Diabetes is a significant contributor to mortality, with macro- and microvascular complications leading to increased disability and substantial healthcare expenditures. The 750 mg/kg dosage in group 6 yielded statistically significant findings ($p < 0.05$) regarding antidiabetic efficacy (Fig. 1). Multiple investigations into plant extracts produced analogous results (Mandal et al., 2024; Ahmed et al., 2010). The group receiving a 750 mg/kg dose exhibited statistically significant outcomes for total cholesterol, HDL, and triglycerides, with results of $197.40 \pm 6.28^*$, $52.21 \pm 4.47^*$, and $101.75 \pm 3.29^*$, respectively ($p < 0.05$) (Table 2). However, no groups exhibited statistically significant outcomes regarding LDL and triglyceride levels, despite a decrease in these

Table 1. Anti-hyperglycemic Activity Analysis

Group number	Group Status	Treatment specimen	Dose of treatment specimen (mg/kg)	Group Abbreviation
1	Negative Control	Physiological Saline	10 mL/kg	N
2	Alloxan control	Alloxan	150 mg/kg	A
3	Alloxan + Metformin	Alloxan + Metformin	150 mg/kg + 100mg	A + M100
4	Alloxan + <i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract low dose	150 mg/kg + 250 mg/kg	A + MI ₂₅₀
5	Alloxan + <i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract medium dose	150 mg/kg + 500 mg/kg	A + MI ₅₀₀
6	Alloxan + <i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract high dose	150 mg/kg + 750 mg/kg	A+ MI ₇₅₀
7	Metformin	Metformin	100 mg/kg	M
8	<i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract low dose	250 mg/kg	MI ₂₅₀
9	<i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract medium dose	500 mg/kg	MI ₅₀₀
10	<i>Mangifera indica</i>	Alloxan + <i>Mangifera indica</i> extract high dose	750 mg/kg	MI ₇₅₀

Table 2. Lipid profile after administration of different dose of *Mangifera indica*

Groups	Total Cholesterol	HDL	LDL	Triglyceride	SGPT	SGOT	Urea	Creatinine
N	118.29± 7.20	84.27±4.27	22.53± 1.28	51.26±3.28	40.28± 5.29	36.77±2.28	38.46±4.28	0.81±0.073
A	213.45± 13.40	43.25±4.90	139.70± 4.77	117.50±8.97	97.53± 8.27	95.77± 8.43	102.53±7.29	2.74±0.84
A+M ₁₀₀	137± 10.28	70.16±5.29	65.28± 5.28	74.25±7.08	58.77± 6.82	51.51±7.32	54.57±6.29	1.34±0.81
A+MI ₂₅₀	208.46± 9.99	45.90±3.28	135.29± 7.73	114.93±6.28	95.28± 6.27	93.23±6.54	99.27±5.28	2.52±0.77
A+ MI ₅₀₀	203.40± 7.49	48.48±3.28	133.46± 5.28	108.08±7.24*	92.08± 5.82	89.23±4.77	94.76±6.28	2.23±0.63
A+ MI ₇₅₀	197.40± 6.28*	52.21±4.47*	131.46± 9.38	101.75±3.29*	87.77± 4.87	85.35±6.28	90.42±4.82*	1.70±0.82*
M ₁₀₀	114.28± 7.50	82.83±5.30	26.22± 4.28	54.28±3.50	33.50± 4.20	35.77±3.50	36.45±5.70	0.82±0.046
MI ₂₅₀	117.17± 6.28	85.19±4.62	22.88± 2.06	57.93±4.43	37.53± 4.70	37.93±4.28	32.90±4.38	0.77±0.049
MI ₅₀₀	114.50± 5.0	83.77±4.56	21.25± 1.09	51.28±3.28	38.14± 3.28	36.36±4.93	35.70±5.30	0.89±0.071
MI ₇₅₀	119.28± 6.29	86.22±4.56	23.40± 2.29	55.53±4.28	35.28± 2.28	37.56±4.22	36.73±4.20	0.76±0.068

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 (Dunnett's test) compared to the control

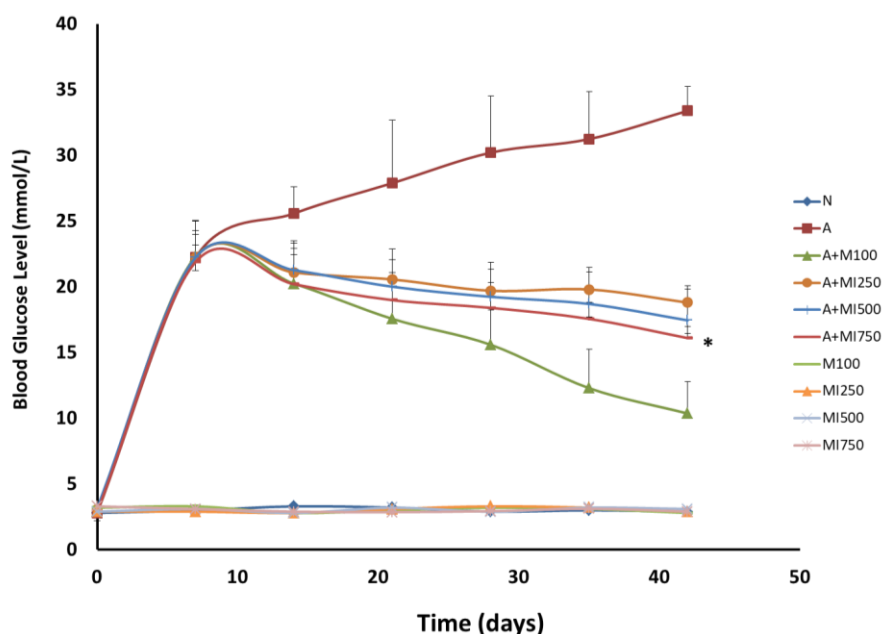


Fig. 1. Antidiabetic activity of different dose of *Mangifera indica*

parameters in the blood following the administration of the extract. Two studies on plant extracts produced comparable results (Bhowmik et al., 2024; Kumar et al., 2017). No groups exhibited statistically significant outcomes regarding SGPT and SGOT; however, the levels of these parameters decreased in the blood following the administration of the extract. In the kidney function test, urea and creatinine levels in group 6 exhibited statistically significant differences ($p < 0.05$) at a dosage of 750 mg/kg, with values of $90.42 \pm 4.82^*$ and $1.70 \pm 0.82^*$, respectively. Two studies on plant extracts produced comparable results (Osman et al., 2010; Mishra & Pancholi, 2013).

4. CONCLUSION

The ethanol extract of *Mangifera indica* demonstrates significant protective effects against diabetes, hypercholesterolemia, liver damage, and impaired kidney function. The extract exhibited a notable effect on the specified outcomes, indicating its possible therapeutic significance. Further research is required to isolate and identify the specific active compounds that contribute to its anti-diabetic and lipid-lowering effects. This may yield a better understanding of the mechanisms of action and facilitate the advancement of more effective treatments derived from this plant.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Ethical permission was obtained from the Department of Zoology at Dhaka University, under issue number 147/pharm.science.ewu.

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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