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Antidiabetic Potential of *Ficus hispida* Fruit Extract in Alloxan-Induced Swiss Albino Diabetic Mice: In Vivo and Molecular Docking Study

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Highlights

- *Ficus hispida* fruit extract significantly reduced blood glucose in diabetic mice.
- MEFH improved lipid profile and liver and kidney biomarkers.
- GC-MS identified multiple bioactive phytochemicals in the fruit extract.
- Molecular docking demonstrated strong interactions with diabetes-related targets.
- ADMET analysis supported the drug-likeness and safety of major phytoconstituents.

ABSTRACT

Background and Aim: Diabetes mellitus is a major metabolic disorder characterized by persistent hyperglycemia and associated with severe complications affecting multiple organs. The limitations and adverse effects of currently available antidiabetic drugs have encouraged the exploration of medicinal plants as alternative therapeutic agents. The present study was designed to investigate the antidiabetic potential of the methanolic fruit extract of *Ficus hispida* (MEFH) in alloxan-induced diabetic mice and to support the findings through computational approaches.

Methods: Experimental diabetes was induced in Swiss albino mice by alloxan administration. The diabetic animals were treated orally with MEFH at doses of 200 and 400 mg/kg body weight, while glibenclamide was used as the standard drug. Blood glucose levels, body weight, serum biochemical markers, and lipid profile were evaluated. Preliminary phytochemical screening, molecular docking, pharmacokinetic analysis, and toxicity prediction were also performed to identify possible bioactive compounds and their mechanisms of action.

Results: MEFH treatment significantly reduced blood glucose levels and improved body weight and biochemical parameters in diabetic mice in a dose-dependent manner. The extract also restored altered lipid profile markers and exhibited no significant acute toxicity. In silico studies demonstrated favorable binding affinities of several phytochemicals against diabetes-related target proteins, with acceptable pharmacokinetic and toxicity profiles.

Conclusion: The combined in vivo and in silico findings suggest that *Ficus hispida* fruit extract possesses notable antidiabetic activity and may serve as a promising natural source for the development of novel antidiabetic agents. Further studies are required to isolate the active constituents and validate their clinical potential.

1| Introduction

Diabetes Mellitus (DM) is a persistent metabolic condition impacting individuals of all ages. It is defined by persistent hyperglycemia resulting from impairments in insulin secretion, insulin action, or a combination thereof. Prolonged hyperglycemia interferes with normal carbohydrate, lipid, and protein metabolism, resulting in systemic metabolic dysregulation [1]. Insufficient disease management may lead to the emergence of chronic, severe, and potentially incapacitating complications [2]. Currently, around 537 million individuals globally are afflicted with diabetes, a figure anticipated to increase to 643 million by 2030 and further to 783 million by 2045 [3]. Diabetes is notably more common in cities and wealthier countries, with rates of 10.8% and 10.4%, respectively [4]. In contrast, the condition is less common in rural areas and low-income countries, where the rates are 7.2% and 4.0%, respectively [5].

The growing prevalence of diabetes is a significant global health problem, mainly because of its harmful short- and long-term effects. These complications greatly increase illness and death rates [6]. Diabetes can lead to both immediate and long-term health problems. Immediate problems include diabetic ketoacidosis, hyperosmolar coma, hypoglycemia, and hyperglycemia. Long-term complications include retinopathy, nephropathy, neuropathy, and cardiovascular diseases; all of which arise from extended exposure to increased blood glucose levels and gradual vascular impairment [7].

Although significant advancements have been made in the creation of synthetic antidiabetic medications, a complete cure for diabetes mellitus remains unattainable. Modern medications primarily focus on managing blood sugar levels, rather than addressing the underlying causes of the disease. This limitation has spurred increasing interest in traditional medicinal plants as

potential alternative or supplementary treatments, largely due to the side effects, high costs, and limited accessibility of standard pharmaceuticals [8].

Medicinal plants are frequently studied for diabetes management, given the generally accepted safety of phytotherapy. Moreover, these plant-derived sources are distinguished by an abundance of bioactive phytoconstituents, including saponins, amino acids, polyphenols, alkaloids, flavonoids, phenolic compounds, and triterpenoids, which are believed to be pivotal to their antidiabetic efficacy [9]. Unlike synthetic medications, which frequently target a specific molecule, plant-based substances can have multiple therapeutic effects at the same time. These bioactive compounds facilitate the management of diabetes mellitus via various cellular mechanisms, such as the restoration of pancreatic β -cell function, modulation of carbohydrate-metabolizing enzymes, enhancement of peripheral glucose uptake, stimulation of muscle glycogen storage, and activation of insulin signaling pathways [10]. These multitargeted systems may yield enhanced treatment results while minimizing unwanted effects. Investigating medicinal plants for the identification of new antidiabetic agents with minimal adverse effects is of considerable scientific significance, as these plants have also demonstrated supplementary pharmacological properties, including anticancer and antioxidant activities; this may further assist in alleviating oxidative stress linked with diabetes and its related consequences [11, 12].

Ficus hispida L., a flowering plant in the Moraceae family, is known for its medicinal properties. Commonly referred to as the hairy fig or rough-leaved fig, this plant is found across a broad swath of territory. Its range includes Bangladesh, India, Nepal, Sri Lanka, Southeast Asia, and even Northern Australia. In Bangladesh, the plant goes by several names: Kakdumur, Khoksha-dumur, Dhungri, and Thoska. This tree, typically small to medium in stature, thrives in a variety of environments. The plant develops into a small to medium-sized tree, typically located along

riverbanks, forest edges, roadsides, and wastelands in tropical and subtropical areas. *F. hispida* is morphologically distinguished by its broad ovate leaves featuring a coarse and hairy texture, as well as cauliflorous fruits that develop directly on the stem and mature branches. Prior research on *F. hispida* and its related *Ficus* species has demonstrated the occurrence of various bioactive compounds. Several studies have reported the extraction of substances, including lupeol acetate, β -amyirin acetate, and ficushispimine-type alkaloids, from different plant components [13–15]. These chemical classes are acknowledged for their antioxidant, anti-inflammatory, antibacterial, and hypoglycemic activities [16, 17]. The existence of these bioactive compounds substantiates the historic therapeutic application of the plant.

In traditional practices, the entire plant of *F. hispida* is used in folk medicine. Its roots, fruits, leaves, bark, and latex are used in Ayurvedic and Unani medicine to treat leukoderma, bleeding, cough, asthma, skin problems, and digestive issues. Moreover, previous studies on its pharmacological properties have shown that *F. hispida* has antidiarrheal, antimicrobial, wound-healing, thrombolytic, and antinociceptive effects, which supports its potential for medical use, demonstrating its extensive therapeutic potential [18–20].

Specifically, the fruits of *F. hispida*, among various plant parts, harbor bioactive phytochemicals frequently associated with glucose regulation, and related *Ficus* species' fruits (e.g., *Ficus racemosa*) have shown antidiabetic properties in experimental models, thus supporting the relevance of fruit-based extracts in antidiabetic research [21]. Likewise, the fruits of *Ficus benghalensis* demonstrated hypoglycemic and hypolipidemic effects in diabetic rat models [22]. The antidiabetic effects of *Ficus religiosa* fruits have been documented via the modulation of carbohydrate-metabolizing enzymes and antioxidant mechanisms [23]. Furthermore, *Ficus carica* fruits demonstrate notable glucose-lowering effects, attributed to their rich polyphenol and

flavonoid content [24]. These findings jointly emphasize the importance of fruit-derived extracts from *Ficus* species in antidiabetic research and confirm the investigation of *F. hispida* fruits as a potential pharmaceutical resource.

Considering its rich phytochemical profile and its long history of use in traditional medicine, this study aimed to investigate the antidiabetic properties of the methanolic fruit extract of *F. hispida*, using a mouse model of diabetes induced by alloxan. This study aims to scientifically confirm the effectiveness of this intervention in lowering glucose levels and its role in improving metabolic problems related to diabetes.

2 | Methods

2.1 | Plant collection and preparation of crude extract

In May 2025, fresh fruits of *F. hispida* were harvested at Foy's Lake in Chittagong, Bangladesh. Professor Dr. Shaikh Bokhtear Uddin of the University of Chittagong in Bangladesh classified and authenticated the taxonomy of the plant material. A voucher specimen was retained for future reference (Accession No. 3647).

The harvested fruits were meticulously cleaned and thereafter dried in the shade at approximately 25 °C for a duration of two weeks. Initially, we pulverized the dried fruits (1 kg) using a mortar, followed by grinding them with a blender. We combined 500 g of the powdered substance with 2 L of 95% methanol and allowed it to rest at ambient temperature for three days. The maceration process was conducted two additional times with the residual plant material to ensure maximal extraction. The extracts were filtered using cotton and subsequently using Whatman filter paper. A rotary evaporator (RE200, Bibby Sterling, UK) was employed to concentrate the filtrate under

reduced pressure. The crude extract was stored in a glass vial at 4 °C until required for further research.

2.2 | Chemicals and reagents

All chemicals used in the study were of analytical grade and were procured from Sigma-Aldrich, Germany. Commercial reagent kits for the estimation of biochemical parameters and enzymes were obtained from Linear Chemicals (Barcelona, Spain). Glibenclamide was purchased from Advanced Chemical Industries (A.C.I.) Limited, Bangladesh.

2.3 | Experimental animals

We got 65 albino mice from Rajshahi University in Bangladesh. They were 8 to 10 weeks old and weighed between 25 to 35 grams. We kept the animals in standard laboratory cages for two weeks, with a temperature of 25 ± 1 °C and a 12-hour light/dark cycle [7]. During the experiment, the mice could eat as much standard pellet food and drink as they wanted. The University of Science and Technology Chittagong's (USTC) Institutional Animal Ethics Committee gave its approval for all animal experiments to be done in accordance with ethical guidelines.

2.4 | Statement of Ethics

All animal-based experimental methods were performed in compliance with ethical guidelines for the care and use of laboratory animals and received approval from the Ethical Review Committee of the University of Science and Technology Chittagong (USTC), Bangladesh (Approval No. USTMEBBC/17/08/25). All measures were implemented to mitigate animal suffering during the study duration. After giving alloxan to the mice, they were given a 2% glucose solution to drink to keep their blood sugar levels from dropping too low. At the end of the trial, the animals were put to sleep with chloroform anesthesia, which made sure they didn't suffer too much.

2.5 | Acute toxicity test

An acute oral toxicity study was performed with Swiss albino mice of both sexes, weighing 25 to 35 g (n = 35), following the guidelines established by the Organisation for Economic Co-operation and Development (OECD, 2008) [25]. Before the treatment, the animals were starved for 16 hours and then split into seven groups of five mice each. The control group received distilled water (2 mL/kg, orally), while the experimental groups received the methanolic extract orally at doses of 200, 400, 800, 1000, 2000, and 3000 mg/kg body weight to assess the extract's safety. After giving the animals the medicine, they were allowed to eat and drink as much as they wanted, but they were closely watched for signs of toxicity. During the first hour, we kept an eye on behavioral, autonomic, and neurological responses all the time. After 24 hours, they were checked again, and then every four hours after that. For 14 days in a row, the animals were watched every day to see if there were any delayed harmful effects and to keep track of any deaths that happened [26]. At the end of the observation period, blood samples were taken from mice treated with (200 and 400 mg/kg BW) for biochemical analysis to confirm dose-related biochemical responses relevant to the diabetic study. Serum was separated to check for markers of liver and kidney function, such as serum glutamate pyruvate transaminase (SGPT/ALT), serum glutamate oxaloacetate transaminase (SGOT/AST), alkaline phosphatase (ALP), and creatinine, using standard commercial diagnostic kits.

2.6 | Induction of experimental diabetes

Swiss albino mice were used for the induction of experimental diabetes. Diabetes was induced by intraperitoneal injection of alloxan monohydrate at a dose of 150 mg/kg body weight. Alloxan was freshly prepared in normal saline prior to administration. Blood glucose levels were evaluated

before alloxan injection and again after 72 hours to confirm the onset of diabetes [9, 27, 28]. Mice with fasting blood glucose levels exceeding 200 mg/dl were designated as diabetic and included in the study.

Subsequent to the diabetes diagnosis, the animals were randomly assigned to five groups, with six mice in each group. Group I served as the standard control and received normal saline. Group II consisted of diabetic mice treated with normal saline, functioning as the diabetic control group. Group III consisted of diabetic mice treated with the standard antidiabetic drug glibenclamide at a dosage of 5 mg/kg body weight. Groups IV and V consisted of diabetic mice that received the methanolic fruit extract of *F. hispida* (MEFH) at doses of 200 mg/kg and 400 mg/kg body weight, respectively. All treatments were administered daily over a period of 21 days. During the experiment, blood sugar levels and body weight were checked on a regular basis. After the treatment was over, the mice were killed and blood samples were taken to do biochemical tests like serum glucose, lipid profile, and markers of liver and kidney function.

2.7 | Phytochemical screening

The plant extracts were subjected to preliminary phytochemical screening using standard protocols [29, 30] to identify the presence or absence of primary and secondary metabolites such as carbohydrates, reducing sugars, fats and fixed oils, alkaloids, phenolics, flavonoids, saponins, tannins, tri-terpenoids and glycosides.

2.8 | GC-MS investigation

To determine the phytocompounds present in the methanolic extract of *Ficus Hispida* fruits Gas chromatography-mass spectroscopy (GC-MS) was utilized with Combined 8890 Gas Chromatography System (Agilent 19091S-433UI:2489267H, USA) coupled with Agilent 7010B

GC/TQ MS system, EI Ionization technique. With Column HP-5MS UI (5% phenyl methyl siloxane, 30 m $\times$ 250 μ m $\times$ 0.25 μ m). With a constant rate of 1.0 ml/min helium was used as the carrier gas. An injection volume of 1 μ l with a split ratio of 1:50 was used. The injector temperature was sustained at 250°C, Pressure:11.653 psi. The temperature program used maintained a temperature of 120°C for 2 min, increased the temperature to 320°C at a rate of 5°C /min, and maintained the temperature at 320°C for 5 min. With the total elution time being 27 min. 40–700 m/z was the mass range of the mass spectrometer's scan parameters and the electron impact ionization voltage was 70 eV. A computer-based mass spectra data bank was used to annotate the mass spectrum. Spectra peaks which were translated from separated components were identified by comparison with mass spectra chromatograms present in National Institute of Standards and Technology's (NIST), 2020 version GC–MS library software.

2.9 | In Silico Analysis

2.9.1 | The selection and preparation of target proteins

Four molecular targets related to diabetes were chosen for investigation of the antidiabetic potential of the phytochemicals identified from *F. hispida*, following their role in glucose metabolism and/or insulin signaling pathways [31]. This included peroxisome proliferator-activated receptor gamma (PPAR- γ ; PDB ID: 2ATH), insulin receptor kinase (IRK; PDB ID: 4IBM) [32], human α -amylase (PDB ID: 2QV4) [33], and dipeptidyl peptidase-IV (DPP-IV; PDB ID: 5KBY) [34]. The three-dimensional structures of the crystals were obtained from the Protein Data Bank (PDB).

The DPP-IV structure lacked several missing residues that were built with MODELLER integrated in UCSF ChimeraX [35, 36]. The structural integrity and stereochemical reliability were then assessed using the SAVES v6.1 server with PROCHECK, ERRAT and Verify3D analyses [37]. Protein structures were further prepared by removing co-crystallized ligands, water molecules, ions, and redundant chains using BIOVIA Discovery Studio Visualizer. Swiss-PdbViewer software was used to minimize the energy to generate stable conformations for docking with molecules [38].

2.9.2 | Ligand Preparation

Initially, 51 phytochemicals were considered, which had been identified by GC–MS analysis of *F. hispida*. Of these, 50 compounds were selected for the computational study on which their complete structural information was available. The molecular structures were obtained from the PubChem database for those that were available and were taken from the National Institute of Standards and Technology (NIST) database for those that were not available.

First, all ligands were imported into the PyRx platform and then the geometry was optimized using the Universal Force Field (UFF) built into PyRx. Conformational optimization of all molecules was performed using energy minimization before docking to obtain energetically favorable conformations and increase docking reliability.

2.9.3 | Molecular Docking Analysis:

AutoDock Vina embedded in PyRx was used for virtual screening and molecular docking studies [39]. The active site residues of both the target proteins were predicted by the use of the server named ProteinsPlus [40]. The corresponding binding pockets were centred on the grid boxes, and the boxes were centred to include all the key catalytic residues.

The native co-crystallized ligands were redocked into the binding sites to validate the docking protocol. The parameters used for docking were shown to be suitable as binding poses were found to be successfully superimposed with the experimental pose for each of the compounds. Docking simulations were then carried out with the exhaustiveness value of 32. The binding affinities are reported as docking scores (kcal/mol), and the values closer to negative indicate more favorable predicted protein–ligand interactions.

2.9.4 | Protein-Ligand Interaction analysis:

After docking, the top-ranked binding poses were identified by binding affinity score and visual inspection. Three-dimensional interaction analyses were carried out in PyMOL and two-dimensional interaction maps were produced with the help of BIOVIA Discovery Studio visualizer [41]. The interactions of amino acids residues, hydrogen bonds, hydrophobic interactions, π – π stacking interactions and van der Waals contacts were characterized, allowing the elucidation of the molecular basis of ligand binding.

2.9.5 | Pharmacokinetic and Drug-Likeness Evaluation

The SwissADME web server was used to evaluate the pharmacokinetic properties of the selected phytochemicals [42]. The parameters evaluated were related to the absorption, distribution, metabolism and excretion (ADME) of the drugs, such as gastrointestinal absorption, Lipophilicity (LogP), aqueous solubility, blood–brain barrier permeability, and Lipinski's rule of five. The drug-likeness was evaluated using the known medicinal chemistry filters with the aim of selecting compounds showing good oral bioavailability and pharmacokinetic properties.

2.9.6 | Toxicology Prediction:

The selected compounds were predicted for their toxicological properties using pkCSM platform.[43] Ames mutagenicity, hepatotoxicity, hERG I inhibition, hERG II inhibition, and acute oral toxicity (LD50) were key toxicity endpoints. Any compound with good pharmacokinetic properties and low predicted toxicity was regarded as a lead candidate for their potential as an antidiabetic agent.

2.10 | Statistical analysis

All experimental data were expressed as mean $\pm$ standard error of the mean (SEM). Statistical evaluation was carried out using one-way analysis of variance (ANOVA) followed by Dunnett's multiple comparison test employing IBM Statistical Package for the Social Sciences (SPSS, version 27). A p-value of less than 0.05 was considered statistically significant. Graphical presentation of the results was performed using GraphPad Prism software.

3 | Results

3.1 | Acute toxicity test

During a 14-day observational period, Swiss albino mice receiving oral doses of methanolic extract of *F. hispida*, ranging from 200 to 3000 mg/kg body weight, demonstrated no mortality or adverse effects. The treated animals did not show any unusual behavioral, autonomic, or neurological signs compared to the control group, and there were no delayed toxic effects during the study.

Serum samples for biochemical analysis were collected from animals given the extract at doses of 200 mg/kg and 400 mg/kg, selected due to the absence of significant toxicity noted during the dose-escalation phase. An assessment of hepatic and renal function markers revealed that serum levels of SGPT (ALT), SGOT (AST), ALP, and creatinine in the extract-treated groups were

within normal physiological ranges and comparable to those of the control group. The findings indicate that the methanolic extract of *F. hispida* did not cause any acute hepatic or renal injury at the designated concentrations as shown in Table 1.

TABLE 1 | Acute toxicity assessment of methanolic fruit extract of *F. hispida* (MEFH) on serum biochemical markers in mice

Experimental Group	SGPT (U/L)	SGOT (U/L)	ALP (U/L)	Creatinine (mg/dl)
Control	58.25 ± 32.35	213.55 ± 55.45	8.35 ± 2.85	0.05 ± 0.01
MEFH 200mg/kg	24.80 ± 10.20	221.70 ± 14.40	2.10 ± 1.60	0.09 ± 0.07
MEFH 400mg/kg	45.60 ± 26.79	206.67 ± 8.09	4.27 ± 1.47	0.12 ± 0.01

3.2 |Effect of *F. hispida* extract on Blood Glucose Level

Table 2 and Figure 1 show how the methanolic fruit extract of *F. hispida* (MEFH) affected the blood sugar levels of diabetic mice that had been given alloxan. When alloxan was given intraperitoneally, it caused a big rise ($p < 0.001$) in blood glucose levels in diabetic control mice (12.71 ± 1.06 mmol/L on day 3) compared to the normal control group (5.31 ± 0.66 mmol/L). During the whole study, mice with diabetic control had very high blood sugar levels.

Giving MEFH to diabetic mice caused their blood sugar levels to drop significantly and stay low for a long time. When MEFH was given to mice at a dose of 200 mg/kg BW, their blood glucose levels dropped significantly from day 7 onward (7.63 ± 0.69 mmol/L), and this drop stayed that way until day 21 (8.70 ± 0.37 mmol/L; $p < 0.01$) when compared to diabetic control mice (11.76

± 0.98 mmol/L). Similarly, the administration of MEFH at 400 mg/kg BW produced a more pronounced antihyperglycemic effect. Blood sugar levels dropped significantly starting on day 3 (7.50 ± 0.05 mmol/L; $p < 0.05$) and kept dropping until day 21 (7.10 ± 0.37 mmol/L; $p < 0.001$) when compared to diabetic control animals. At a dose of 400 mg/kg BW, MEFH lowered blood sugar levels in a way that was analogous to the conventional antidiabetic drug glibenclamide (5 mg/kg), which had a blood sugar level of 7.20 ± 0.66 mmol/L on day 21.

In diabetic mice that were given alloxan, MEFH had an antihyperglycemic effect that depended on the dose. The higher dose (400 mg/kg BW) caused blood glucose levels to drop more significantly and for a longer duration than the lower dose (200 mg/kg BW).

TABLE 2 | Impact of *F. hispida* extracts on blood glucose levels in alloxan-induced diabetic mice

Experimental Groups	Blood Glucose Level (mmol/L)				
	Day 0	Day 3	Day 7	Day 14	Day 21
Normal Control	5.00 ± 0.05	5.31 ± 0.66	5.86 ± 0.18	5.97 ± 0.24	5.43 ± 0.33
Diabetic Control	6.93 ± 0.31	$12.71 \pm 1.06^{\dagger\dagger\dagger}$	$11.55 \pm 0.71^{\dagger\dagger}$	$11.58 \pm 0.71^{\dagger\dagger}$	$11.76 \pm 0.98^{\dagger\dagger}$
Standard Glibenclamide 5mg/kg	6.60 ± 0.25	$7.73 \pm 2.61^*$	$7.67 \pm 0.33^{**}$	$8.06 \pm 0.37^{**}$	$7.20 \pm 0.66^{**}$
MEFH 200mg/kg	6.43 ± 0.52	7.50 ± 1.34	$7.63 \pm 0.69^{**}$	$8.80 \pm 0.11^{**}$	$8.70 \pm 0.37^{**}$

MEFH	5.13 ± 0.14	7.50 ± 0.05*	7.20 ±	7.60 ± 0.66**	7.10 ±
400mg/kg			0.61***		0.37***

Note: Values are presented as mean ± SEM, (n=6). †††p<0.001, ††p<0.01, †p<0.05 compared to normal control, ***p<0.001, **p<0.01, *p<0.05 compared to diabetic control.

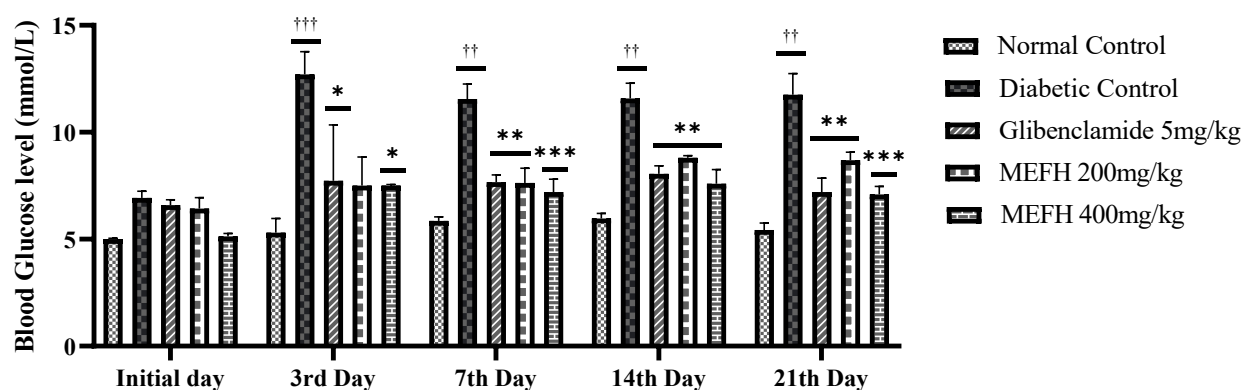


FIGURE 1 | Impact of *F. hispida* Extracts on Blood Glucose Concentrations in Alloxan-Induced Diabetic Mice.

3.3 | Effect of *F. hispida* Extract on Body Weight

The impact of methanolic fruit extract of *F. hispida* (MEFH) on body weight alterations in alloxan-induced diabetic mice is illustrated in Table 3 and Figure 2. Normal control mice demonstrated a progressive increase in body weight during the experimental period, while diabetic control mice displayed a notable deviation in body weight relative to the normal control group. In diabetic control mice, body weight significantly increased on day 7 (32.33 ± 0.88 g; $p < 0.05$), followed by a steady decline on day 14 (29.67 ± 0.88 g; $p < 0.01$) and day 21 (28.33 ± 0.88 g; $p < 0.01$). MEFH therapy markedly increased body weight in diabetic mice. The treatment of MEFH at 200 mg/kg body weight significantly increased body weight from day 7 (33.33 ± 0.88 g; $p < 0.01$) and this increase was maintained until day 21 (35.00 ± 0.57 g; $p < 0.05$) compared to diabetic control mice.

Similarly, the administration of MEFH at 400 mg/kg body weight produced a more pronounced effect on body weight. A significant increase in body weight was seen on day 7 (29.33 ± 0.88 g; $p < 0.001$), which further escalated on day 14 (35.67 ± 2.02 g; $p < 0.01$) and day 21 (31.33 ± 0.88 g; $p < 0.001$) relative to diabetic control mice. Glibenclamide (5 mg/kg BW), a common antidiabetic drug, significantly increased the weight of diabetic mice, showing effects similar to those seen in MEFH-treated subjects. MEFH therapy greatly reduced weight loss related to diabetes in alloxan-induced diabetic mice, and the effect was dose-dependent.

TABLE 3 | Impact of *F. hispida* extracts on the body weight of alloxan-induced diabetic mice

Experimental Groups	Body Weight (g)				
	Day 0	Day 3	Day 7	Day 14	Day 21
Normal Control	27.33 ± 1.20	27.00 ± 1.15	29.33 ± 0.57	28.67 ± 1.15	30.76 ± 1.52
Diabetic Control	29.67 ± 0.67	30.67 ± 0.67	$32.33 \pm 0.88^{\dagger}$	$29.67 \pm 0.88^{\dagger\dagger}$	$28.33 \pm 0.88^{\dagger\dagger}$
Standard Glibenclamide 5mg/kg	32.67 ± 1.76	35.67 ± 2.60	$36.00 \pm 1.15^{**}$	37.33 ± 0.88	$32.67 \pm 1.45^{**}$
MEFH 200mg/kg	30.33 ± 0.33	32.33 ± 1.33	$33.33 \pm 0.88^{**}$	35.67 ± 1.76	$35.00 \pm 0.57^{*}$
MEFH 400mg/kg	28.00 ± 1.00	27.00 ± 1.15	$29.33 \pm 0.88^{***}$	$35.67 \pm 2.02^{**}$	$31.33 \pm 0.88^{***}$

Note: Values are presented as mean $\pm$ SEM, (n=6). $^{\dagger\dagger\dagger}p < 0.001$, $^{\dagger\dagger}p < 0.01$, $^{\dagger}p < 0.05$ compared to normal control, $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$ compared to diabetic control.

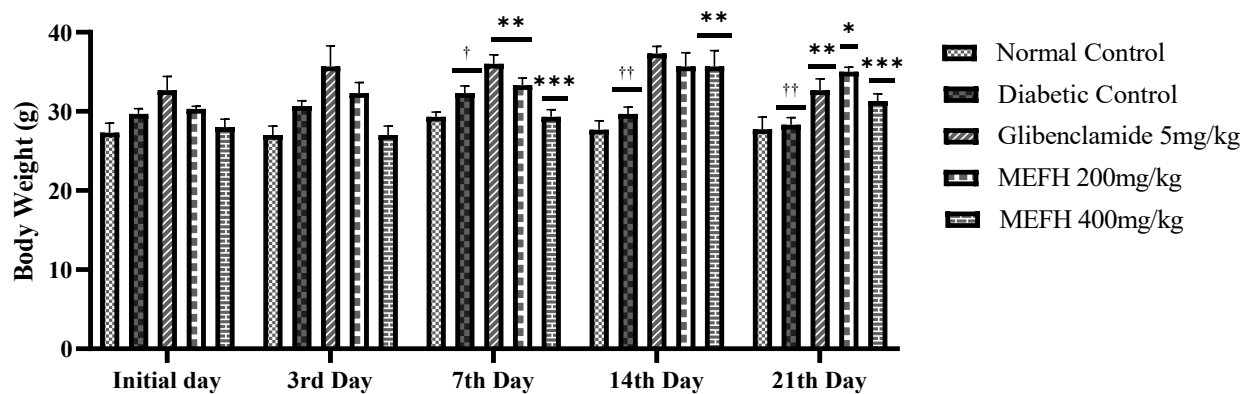


FIGURE 2 | Impact of *F. hispida* Extracts on Body Weight Variations in Alloxan-Induced Diabetic Mice.

3.4 | Effect of *F. hispida* Extract on Serum Glucose, Amylase, and Bilirubin Levels

The impact of methanolic fruit extract of *F. hispida* (MEFH) on serum biochemical markers in alloxan-induced diabetic mice is delineated in Table 4. As shown in Figure 3a, mice with diabetic control had a significant rise in serum glucose levels (279.67 ± 24.96 mg/dl; $p < 0.01$) compared to the normal control group (91.67 ± 2.02 mg/dl). Compared to diabetic control mice, treatment with MEFH at doses of 200 and 400 mg/kg BW significantly lowered blood glucose levels to 160.90 ± 39.49 mg/dl ($p < 0.05$) and 154.46 ± 26.38 mg/dl ($p < 0.01$), respectively. The glucose-lowering effect of MEFH at 400 mg/kg body weight was comparable to that of the standard medication glibenclamide (163.67 ± 13.97 mg/dl).

As shown in Figure 3b, the serum amylase levels in diabetic control mice (3441.20 ± 49.53 U/L; $p < 0.01$) were much higher than those in normal control mice (2050.00 ± 40.41 U/L). MEFH significantly reduced amylase levels in a dose-dependent manner, resulting in values of 2663.90 ± 18.91 U/L ($p < 0.01$) at 200 mg/kg BW and 1793.56 ± 68.65 U/L ($p < 0.001$) at 400 mg/kg BW.

The decline observed at the increased dosage was comparable to that of animals administered glibenclamide (2186.17 ± 27.29 U/L).

Figure 3c shows that the serum total bilirubin levels in diabetic control mice were significantly higher (0.51 ± 0.02 mg/dl; $p < 0.05$) than those in the normal control group (0.26 ± 0.01 mg/dl). Treatment with MEFH at doses of 200 and 400 mg/kg BW significantly reduced total bilirubin levels to 0.23 ± 0.01 mg/dl ($p < 0.01$) and 0.24 ± 0.01 mg/dl ($p < 0.001$), respectively.

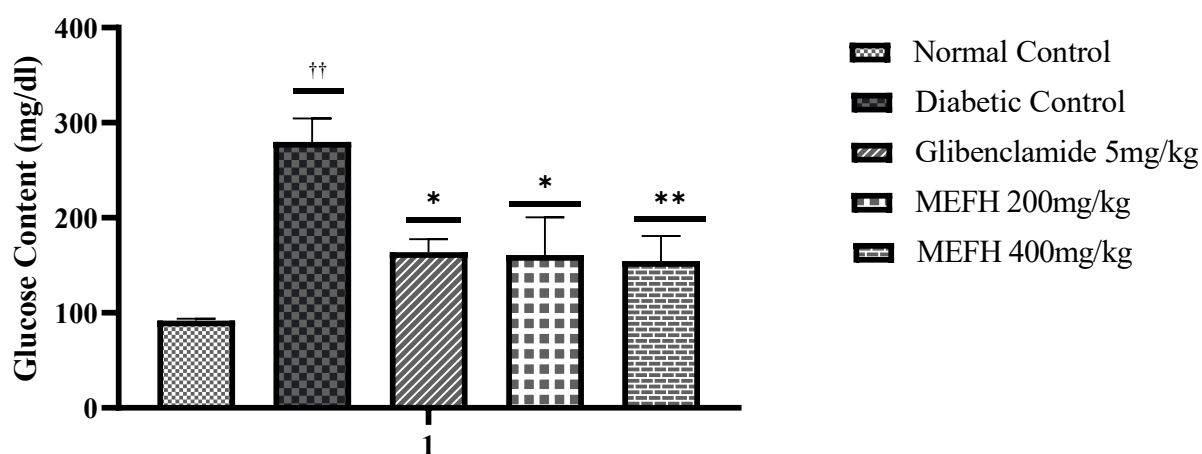
Direct bilirubin levels were significantly elevated in diabetic control mice (0.32 ± 0.01 mg/dl; $p < 0.05$) compared to normal control mice (0.18 ± 0.04 mg/dl), as depicted in Figure 3d. At 200 mg/kg BW, giving MEFH back to the body brought direct bilirubin levels back to 0.20 ± 0.01 mg/dl ($p < 0.05$), and at 400 mg/kg BW, they went back to 0.10 ± 0.02 mg/dl ($p < 0.05$). This is the same as what happened in the glibenclamide-treated control group.

TABLE 4 | The impact of *F. hispida* extracts on serum biochemical parameters.

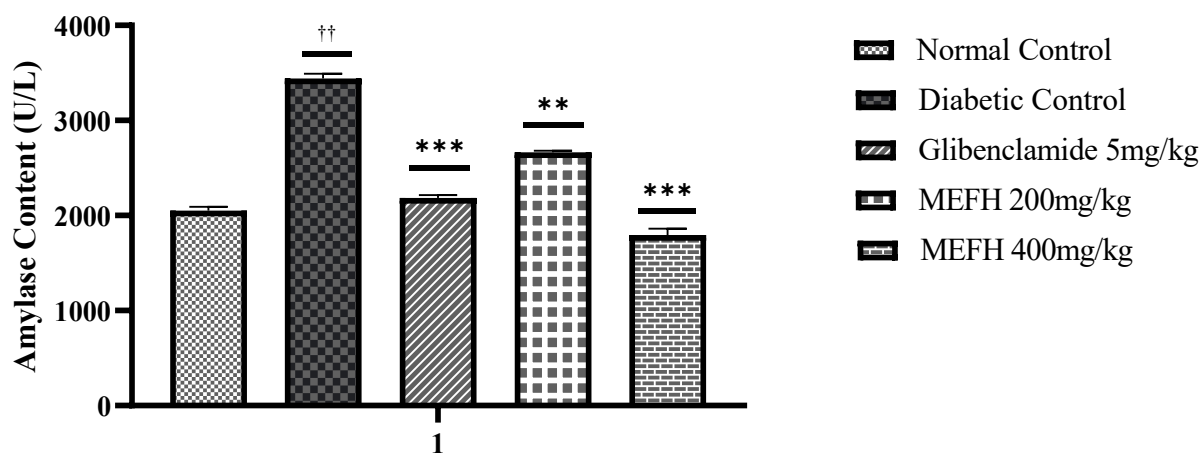
Experimental Groups	Glucose (mg/dl)	Amylase (U/L)	BILI T (mg/dl)	BILI D (mg/dl)
Normal Control	91.67 ± 2.02	2050.00 ± 40.41	0.26 ± 0.01	0.18 ± 0.04
Diabetic Control	$279.67 \pm 24.96^{\dagger\dagger}$	$3441.20 \pm 49.53^{\dagger\dagger}$	$0.51 \pm 0.02^{\dagger}$	$0.32 \pm 0.01^{\dagger}$
Standard Glibenclamide 5mg/kg	$163.67 \pm 13.97^*$	$2186.17 \pm 27.29^{***}$	$0.22 \pm 0.08^{**}$	$0.14 \pm 0.01^{**}$

MEFH 200mg/kg	160.90 ± 39.49*	2663.90 ± 18.91**	0.23 ± 0.01**	0.20 ± 0.01*
MEFH 400mg/kg	154.46 ± 26.38**	1793.56 ± 68.65***	0.24 ± 0.01***	0.10 ± 0.02*

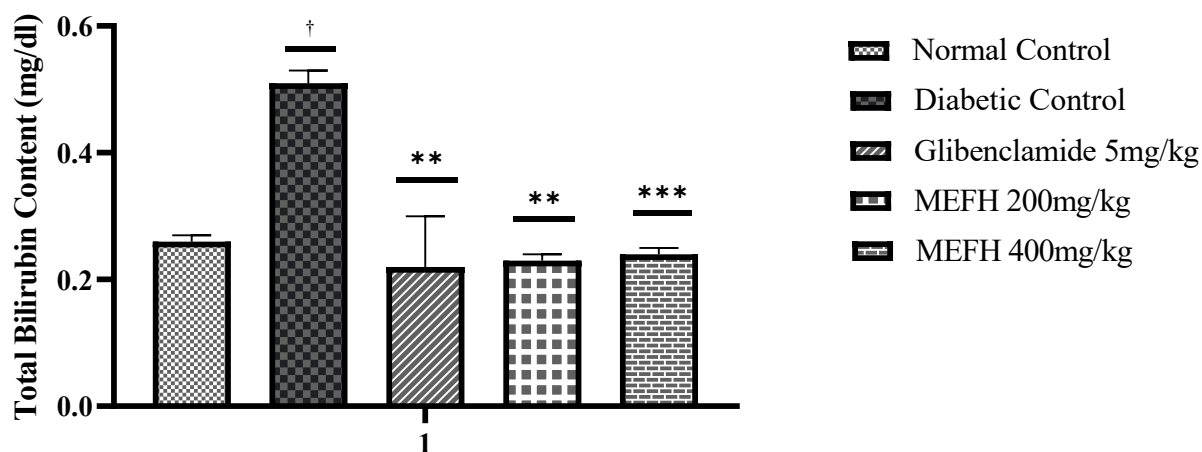
Note: Values are presented as mean ± SEM, (n=6). †††p<0.001, ††p<0.01, †p<0.05 compared to normal control, ***p<0.001, **p<0.01, *p<0.05 compared to diabetic control.



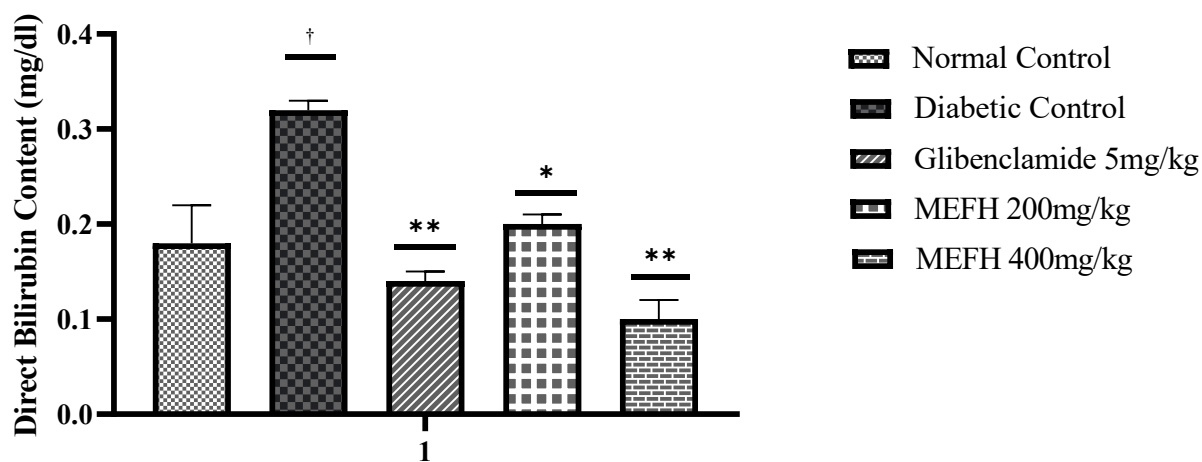
(a)



(b)



(c)



(d)

FIGURE 3 | The impact of *F. hispida* Extracts on Serum Glucose, Amylase, and Bilirubin Levels in Diabetic Mice.

3.5 | Effect of *F. hispida* Extract on Lipid Profile Parameters

The impact of methanolic fruit extract of *F. hispida* (MEFH) on lipid profile parameters in alloxan-induced diabetic mice is illustrated in Table 5 and Figure 4. Mice with diabetic control

demonstrated a marked increase in serum total cholesterol (TC; 277.37 ± 8.21 mg/dl; $p < 0.01$), triglycerides (TG; 199.33 ± 20.21 mg/dl; $p < 0.05$), and low-density lipoprotein (LDL; 129.19 ± 5.59 mg/dl; $p < 0.01$), alongside a significant decrease in high-density lipoprotein (HDL; 47.73 ± 2.52 mg/dl; $p < 0.01$) in comparison to the normal control group. Treatment with the standard antidiabetic agent glibenclamide (5 mg/kg BW) significantly improved the lipid profile in diabetic mice, evidenced by marked reductions in total cholesterol (TC) levels (142.17 ± 36.47 mg/dl; $p < 0.05$), triglycerides (TG) (156.17 ± 14.46 mg/dl; $p < 0.05$), and low-density lipoprotein (LDL) (49.83 ± 18.60 mg/dl; $p < 0.001$), along with a significant increase in high-density lipoprotein (HDL) levels (145.10 ± 37.00 mg/dl; $p < 0.001$), relative to diabetic control mice. Likewise, MEFH therapy resulted in considerable enhancements in lipid profile parameters in a dose-dependent manner. Compared to diabetic control mice, MEFH at 200 mg/kg body weight significantly lowered total cholesterol (TC) levels (137.46 ± 8.46 mg/dl; $p < 0.05$), triglycerides (TG) (142.53 ± 18.35 mg/dl; $p < 0.05$), and low-density lipoprotein (LDL) levels (40.87 ± 5.39 mg/dl; $p < 0.001$), while significantly raising high-density lipoprotein (HDL) levels (141.43 ± 5.71 mg/dl; $p < 0.01$). The effects were stronger when MEFH was at 400 mg/kg BW. It significantly lowered TC (123.76 ± 4.01 mg/dl; $p < 0.01$), TG (118.70 ± 11.75 mg/dl; $p < 0.01$), and LDL (31.43 ± 1.44 mg/dl; $p < 0.001$) levels, while significantly raising HDL levels (132.00 ± 9.44 mg/dl; $p < 0.01$). The lipid-modulating effects of MEFH at 400 mg/kg body weight were comparable to those of glibenclamide, a widely used medication for diabetes.

MEFH treatment significantly improved diabetes-related dyslipidemia in alloxan-induced diabetic mice.

TABLE 5 | The impact of *F. hispida* extracts on lipid profile parameters.

Experimental Groups	TC (mg/dl)	TG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Normal Control	92.50 ± 2.25	67.50 ± 2.63	95.30 ± 14.02	29.13 ± 1.42
Diabetic Control	277.37 ± 8.21 ^{††}	199.33 ± 20.21 [†]	47.73 ± 2.52 ^{††}	129.19 ± 5.59 ^{††}
Standard Glibenclamide 5mg/kg	142.17 ± 36.47 [*]	156.17 ± 14.46 [*]	145.10 ± 37.00 ^{***}	49.83 ± 18.60 ^{***}
MEFH 200mg/kg	137.46 ± 8.46 [*]	142.53 ± 18.35 [*]	141.43 ± 5.71 ^{**}	40.87 ± 5.39 ^{***}
MEFH 400mg/kg	123.76 ± 4.01 ^{**}	118.70 ± 11.75 ^{**}	132.00 ± 9.44 ^{**}	31.43 ± 1.44 ^{***}

Note: [†]P<0.05, ^{††}P<0.01, ^{†††}P<0.001 significant compared with Normal control, * P<0.05, ** P<0.01, ***

P<0.001 significant compared with Diabetic control. The one-way ANOVA and Dunnett's post hoc test were used to determine statistically significant differences between the groups. TC: Total cholesterol; TG: Triglyceride; HDL: High density lipoprotein; LDL: Low-density lipoprotein.

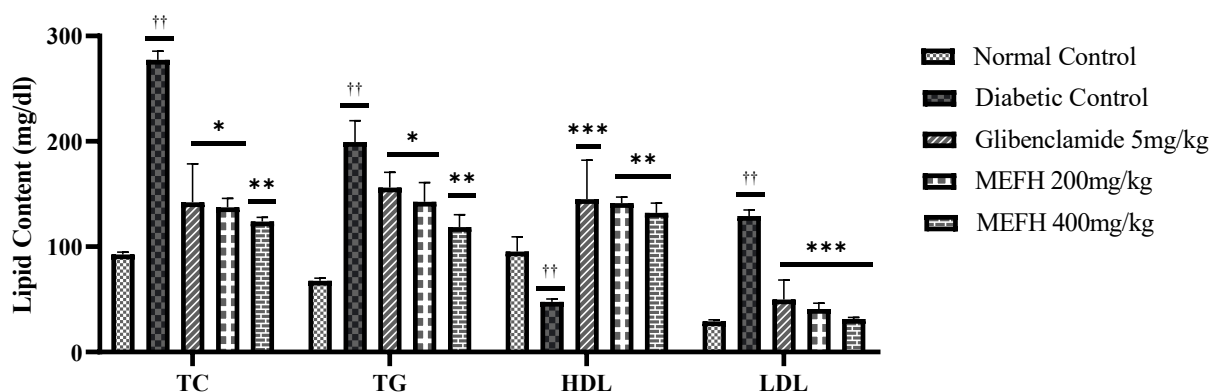


FIGURE 4 | The impact of *F. hispida* Extracts on Lipid Profile Parameters in Alloxan-Induced Diabetic Mice.

3.6 | Effect of *F. hispida* Extract on Liver and Kidney Function Markers

The impact of methanolic fruit extract of *F. hispida* (MEFH) on serum biochemical parameters associated with liver and kidney function in alloxan-induced diabetic mice is detailed in Table 6. As shown in Figure 5a, mice with diabetic control had a much higher level of serum glutamate pyruvate transaminase (SGPT; 61.63 ± 2.86 U/L; $p < 0.01$) than the normal control group (27.97 ± 0.81 U/L). Compared to diabetic control mice, treatment with MEFH at doses of 200 and 400 mg/kg BW significantly lowered SGPT levels to 37.00 ± 10.30 U/L ($p < 0.05$) and 21.87 ± 4.45 U/L ($p < 0.01$), respectively. The drop seen at the higher dose was similar to the one seen with the reference drug glibenclamide (36.77 ± 5.11 U/L).

As shown in Figure 5b, serum glutamate oxaloacetate transaminase (SGOT) levels were much higher in diabetic control mice (311.03 ± 41.89 U/L; $p < 0.01$) than in normal control mice (45.97 ± 1.94 U/L). Giving MEFH to the subjects caused a significant drop in SGOT levels that depended on the dose. At 200 mg/kg BW, the levels were 179.67 ± 45.41 U/L ($p < 0.01$), and at 400 mg/kg BW, they were 124.23 ± 1.17 U/L ($p < 0.001$). The effect of 400 mg/kg BW MEFH was similar to that of glibenclamide on mice (127.33 ± 9.12 U/L). Figure 5c shows that the alkaline phosphatase (ALP) levels in diabetic control mice were much higher (7.83 ± 1.19 U/L; $p < 0.05$) than in the normal control group (3.53 ± 0.17 U/L). MEFH treatment at doses of 200 and 400 mg/kg BW significantly reduced ALP levels to 2.03 ± 0.52 U/L ($p < 0.01$) and 2.60 ± 0.28 U/L ($p < 0.01$), respectively, compared to diabetic control mice.

As shown in Figure 5d, diabetic control mice had a much higher blood urea level (92.76 ± 9.86 mg/dl; $p < 0.01$) than normal control mice (31.93 ± 0.88 mg/dl). The administration of MEFH significantly reduced serum urea levels to 37.91 ± 0.55 mg/dl ($p < 0.05$) at 200 mg/kg BW and

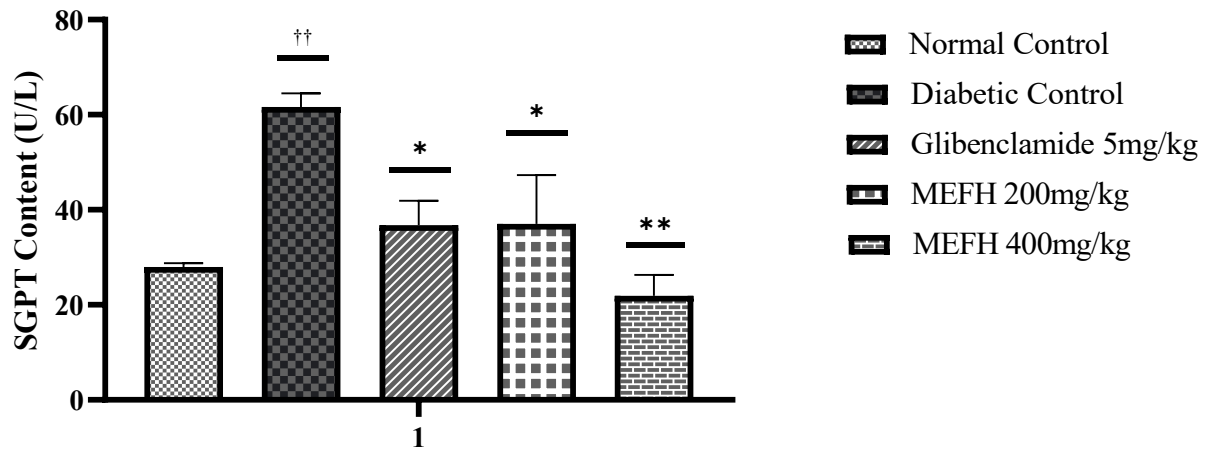
32.97 ± 2.27 mg/dl ($p < 0.05$) at 400 mg/kg BW, comparable to the glibenclamide-treated group (36.24 ± 5.44 mg/dl).

In a comparable manner, serum creatinine levels were significantly elevated in diabetic control mice (0.92 ± 0.31 mg/dl; $p < 0.01$) compared to the normal control group (0.25 ± 0.05 mg/dl), as illustrated in Figure 5e. At doses of 200 and 400 mg/kg BW, MEFH brought creatinine levels back to normal levels of 0.18 ± 0.01 mg/dl ($p < 0.01$) and 0.16 ± 0.03 mg/dl ($p < 0.001$), respectively. The effect of MEFH at the higher dose was close to that of the standard drug glibenclamide (0.33 ± 0.05 mg/dl).

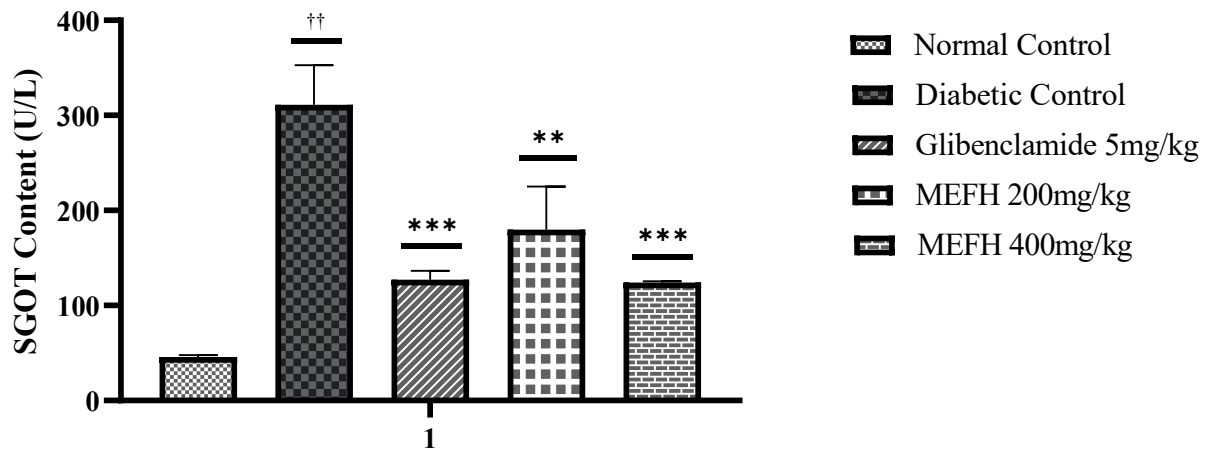
TABLE 6 | The impact of *F. hispida* extracts on serum biochemical parameters

Experimental Groups	SGPT (U/L)	SGOT (U/L)	ALP (U/L)	UREA (mg/dl)	CREA (mg/dl)
Normal Control	27.97 ± 0.81	45.97 ± 1.94	3.53 ± 0.17	31.93 ± 0.88	0.25 ± 0.05
Diabetic Control	61.63 ± 2.86 ^{††}	311.03 ± 41.89 ^{††}	7.83 ± 1.19 [†]	92.76 ± 9.86 ^{††}	0.92 ± 0.31 ^{††}
Standard Glibenclamide 5mg/kg	36.77 ± 5.11 [*]	127.33 ± 9.12 ^{***}	3.17 ± 0.67 ^{**}	36.24 ± 5.44 [*]	0.33 ± 0.05 [*]
MEFH 200mg/kg	37.00 ± 10.30 [*]	179.67 ± 45.41 ^{**}	2.03 ± 0.52 ^{**}	37.91 ± 0.55 [*]	0.18 ± 0.01 ^{**}
MEFH 400mg/kg	21.87 ± 4.45 ^{**}	124.23 ± 1.17 ^{***}	2.60 ± 0.28 ^{**}	32.97 ± 2.27 [*]	0.16 ± 0.03 ^{***}

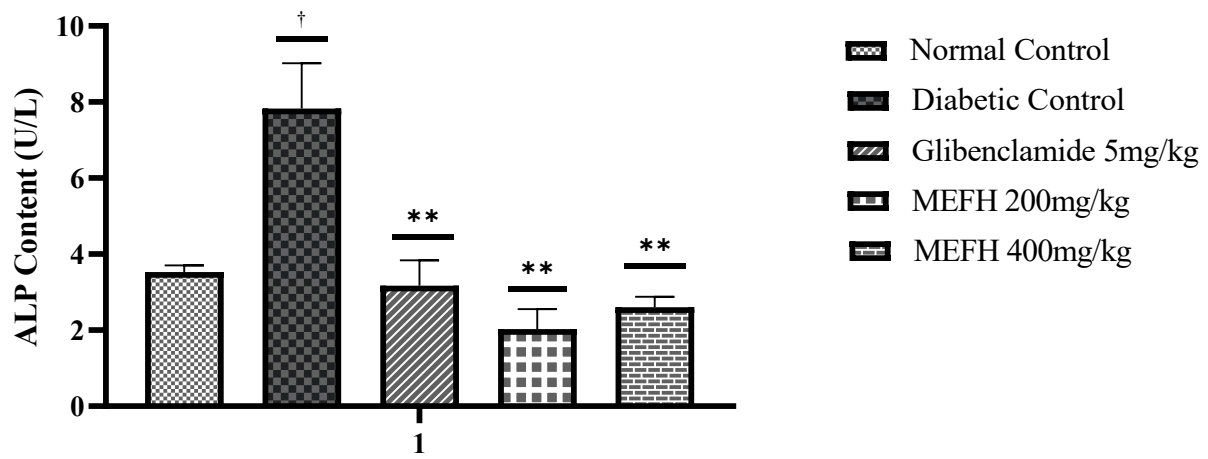
Values are presented as mean $\pm$ SEM, (n=6). $^{\dagger\dagger\dagger}p<0.001$, $^{\dagger\dagger}p<0.01$, $^{\dagger}p<0.05$ compared to normal control, $^{***}p<0.001$, $^{**}p<0.01$, $^{*}p<0.05$ compared to diabetic control.



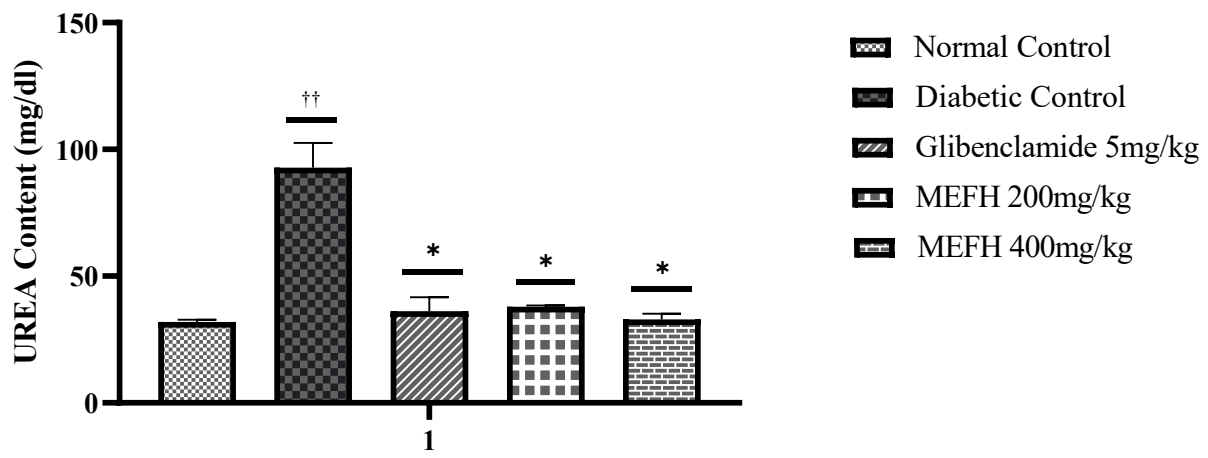
(a)



(b)



(c)



(d)

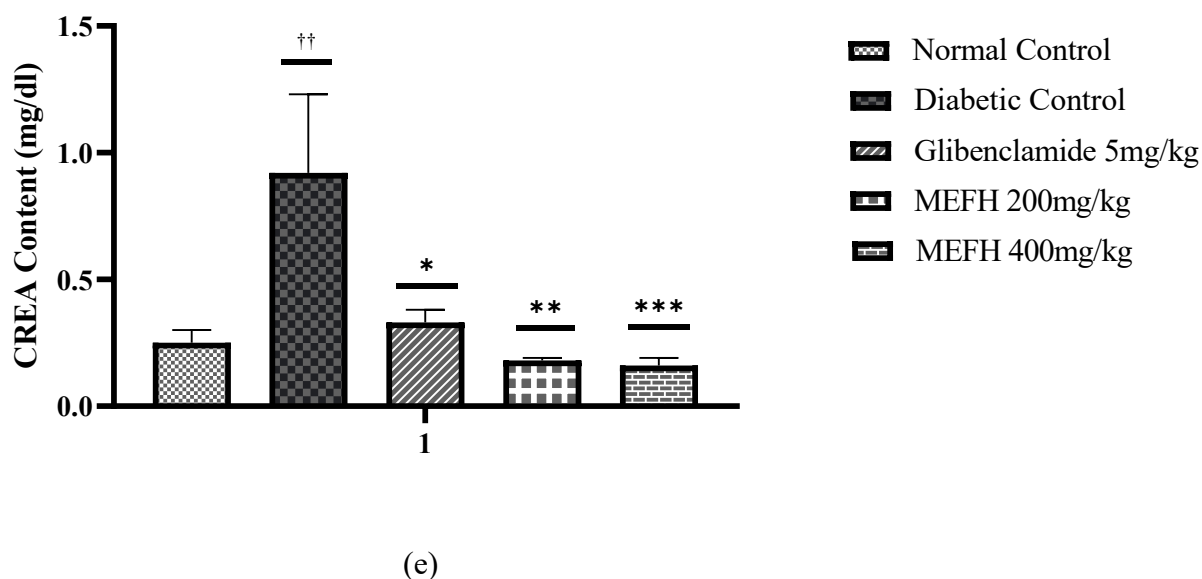


FIGURE 5 | Impact of *F. hispida* Extracts on Liver and Kidney Function Biomarkers in Diabetic Mice.

3.7 | Phytochemical screening

Using standard qualitative chemical tests, a preliminary phytochemical analysis of the methanolic fruit extract of *F. hispida* was done. The analysis found carbohydrates, reducing sugars, lipids and fixed oils, alkaloids, tannins, flavonoids, phenols and triterpenoids, but it did not find saponins or glycosides (Table 7).

The methanolic fruit extract of *F. hispida* contains flavonoids, phenols, tannins, alkaloids, and triterpenoids, which are all groups of phytochemicals that are known to lower blood sugar levels in plants. This is important because it shows that the extract has antidiabetic properties. Previous studies have shown that flavonoids, phenols, tannins, alkaloids, and triterpenoids can help lower blood sugar levels, which is how they work as antidiabetic agents[44–46]. Consequently, the antidiabetic effects of the methanolic fruit extract of *F. hispida* may result from the synergistic

and/or independent actions of these phytoconstituents on glucose metabolism and glycolytic enzyme activity.

TABLE 7 | Qualitative phytochemical analysis of methanolic fruit extract of *F. hispida*

Tests	Reagents	Inferences
Carbohydrates	Molish test	+
	Barfoed's test	+
Reducing Sugars	Fehling test	+
Fat and fixed oils	CuSO ₄ test	+
Alkaloids	Mayer's test	+
	Wagner's test	+
Saponins	Frothing test	-
Tannins	FeCl ₃ test	+
Flavonoids	Alkali test	+
	Lead acetate test	+
Phenols	Iodine test	+
	Ferric chloride test	+
Triterpenoids	Salkowskis test	+
Glycosides	Keller-killani test	-

3.8 | GC-MS investigation

The GC-MS study revealed 51 different phytochemicals. In the GC-MS analysis match factors had the range of 80 to 93, pointing toward a high spectral confidence in compound identification.

The retention times (RT) had a range of 8.07 to 26.67 minutes, and molecular weights where from 151.04 Da (pyrolo[3,2d]pyrimidin-2,4-dione) to 482.38 Da (b-amyrenonol acetate derivative), Showing visible structural diversity among the identified phytocompounds

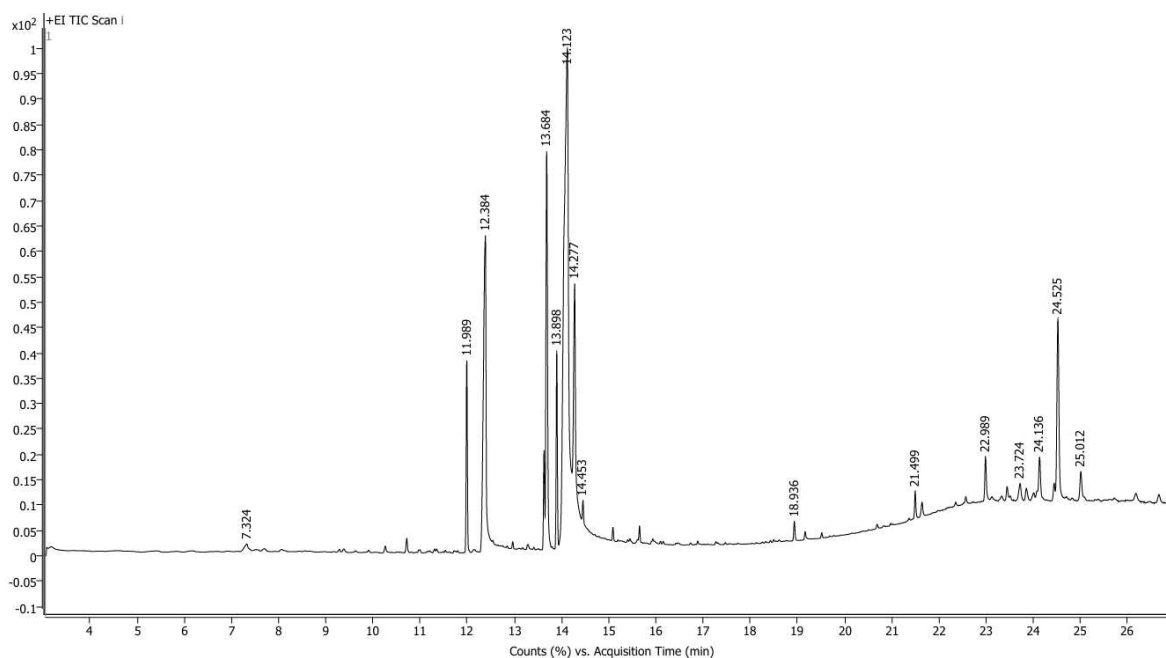


Fig 6. GC chromatogram of *Ficus Hispida* extract

TABLE 8 | . Table of phytocompounds of *Ficus Hispida* identified by GC-MS investigation.

Serial	RT	Compound Name	Match Factor	Formula	Base Peak MZ	MW	PubChem CID
1	8.07	Dodecanoic acid	84.58	C ₁₂ H ₂₄ O ₂	73.00	200.18	3893
2	8.92	2-p-Tolylpyridine	81.00	C ₁₂ H ₁₁ N	169.00	169.09	78219
3	9.03	Benzophenone	83.96	C ₁₃ H ₁₀ O	105.00	182.07	3102
4	9.18	Ethane, 1,2-dibromo-	82.54	C ₂ H ₄ Br ₂	109.10	185.87	7839
5	9.38	7-Oxabicyclo[4.1.0]heptan-3-ol, 6-(3-hydroxy-1-butenyl)-1,5,5-trimethyl-	86.73	C ₁₃ H ₂₂ O ₃	125.00	226.16	5363777

6	9.91	Methyl tetradecanoate	82.11	C15H30O2	74.00	242.23	31284
7	10.26	Tetradecanoic acid	89.72	C14H28O2	73.00	228.21	11005
8	10.72	5,5,8a-Trimethyl-3,5,6,7,8,8a-hexahydro-2H-chromene	80.89	C12H20O	124.00	180.15	580068
9	10.97	Pentadecanoic acid, methyl ester	87.20	C16H32O2	87.00	256.24	23518
10	11.19	3-Eicosyne	82.38	C20H38	95.00	278.30	549159
11	11.22	Ficusin	91.59	C11H6O3	186.00	186.03	6199
12	11.30	Pentadecanoic acid	87.11	C15H30O2	73.00	242.23	13849
13	11.35	Carbonic acid, eicosyl vinyl ester	87.54	C23H44O3	71.00	368.33	91693137
14	11.53	1-Hexadecanol	84.14	C16H34O	69.00	242.26	2682
15	11.72	Sulfurous acid, 2-ethylhexyl isohexyl ester	83.32	C14H30O3S	71.10	278.19	6420722
16	11.80	7-Hexadecenoic acid, methyl ester, (Z)-	86.75	C17H32O2	69.00	268.24	14029831
17	11.99	Hexadecanoic acid, methyl ester	92.15	C17H34O2	74.10	270.26	8181
18	12.14	1-Ethynylcyclododecanol	87.29	C14H24O	69.00	208.18	2828296
19	12.23	Benzenepropanoic acid, 3,5-bis(1,1-dimethylethyl)-4-hydroxy-, methyl ester	82.50	C18H28O3	277.00	292.20	62603
20	12.42	Phthalic acid, cyclobutyl isobutyl ester	82.43	C16H20O4	149.00	276.14	6424063
21	12.85	Cyclopropaneoctanoic acid, 2-hexyl-, methyl ester	83.63	C18H34O2	69.00	282.26	543331
22	12.96	Heptadecanoic acid, methyl ester	91.32	C18H36O2	87.00	284.27	15609
23	13.28	n-Hexadecanoic acid	83.87	C16H32O2	73.10	256.24	985
24	13.41	Bicyclo[9.3.1]pentadeca-3,7-dien-12-ol, 4,8,12,15,15-pentamethyl-, [1R-(1R*,3E,7E,11R*,12R*)]-	80.73	C20H34O	91.10	290.26	5377475
25	13.51	1-Dodecanol	85.07	C12H26O	83.00	186.20	8193
26	13.63	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	90.01	C19H34O2	67.00	294.26	5284421
27	13.68	9-Octadecenoic acid, methyl ester, (E)-	91.36	C19H36O2	67.00	296.27	5280590
28	13.90	Methyl stearate	93.26	C19H38O2	87.00	298.29	8201
29	14.21	Pyrolo[3,2-d]pyrimidin-2,4(1H,3H)-dione	87.98	C6H5N3O2	151.00	151.04	592097

30	14.28	Octadecanoic acid	87.59	C18H36O2	73.10	284.27	5281
31	15.20	5-Eicosene, (E)-	82.57	C20H40	69.10	280.31	5364600
32	15.40	Sulfurous acid, hexyl pentadecyl ester	87.28	C21H44O3S	71.00	376.30	6420414
33	15.45	cis-Methyl 11-eicosenoate	86.15	C21H40O2	69.00	324.30	5463047
34	15.65	Eicosanoic acid, methyl ester	90.47	C21H42O2	87.00	326.32	14259
35	16.10	2-Ethylbutyric acid, eicosyl ester	82.59	C26H52O2	117.00	396.40	...
36	16.15	Butyric acid, octadecyl ester	82.66	C22H44O2	89.00	340.33	...
37	16.74	Heptacosane	82.51	C27H56	71.00	380.44	11636
38	17.47	Phthalic acid, octyl 2-propylpentyl ester	87.30	C24H38O4	149.00	390.28	...
39	19.16	13-Docosenamide, (Z)-	81.22	C22H43NO	72.00	337.33	...
40	19.52	Squalene	91.93	C30H50	69.00	410.39	638072
41	21.50	Epilupeol; 20(29)-Lupen-3alpha-ol, acetate	81.05	C32H52O2	95.10	468.40	6427357
42	21.64	Cholesterol	81.79	C27H46O	91.00	386.36	5997
43	22.36	Campesterol	82.49	C28H48O	95.00	400.37	173183
44	22.57	Stigmasterol	90.09	C29H48O	95.00	412.37	5280794
45	22.99	.gamma.-Sitosterol	88.34	C29H50O	91.00	414.39	457801
46	22.99	.beta.-Sitosterol	80.69	C29H50O	91.10	414.39	222284
47	23.72	24-Norursa-3,12-diene	83.34	C29H46	218.30	394.36	...
48	24.14	24-Noroleana-3,12-diene	89.87	C29H46	203.10	394.36	15427754
49	24.44	9,19-Cyclolanost-24-en-3-ol, acetate, (3.beta.)-	81.73	C32H52O2	69.00	468.40	518616
50	25.01	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, acetate, (3.beta.,4.alpha.,5.alpha.)-	80.82	C32H52O2	93.00	468.40	537081
51	26.67	b-Amyrenonol, Ac derivative (isomer 1)	85.29	C32H50O3	135.00	482.38	...

3.8. | Docking study:

PPAR-gamma receptor molecular docking showed that all the docked ligands had significant binding affinities. stigmasterol (CID 5280794) has the highest docking score with -9.3 kcal/mol. It has one Lipinski violation which should be considered when evaluating its drug-likeness. Ficusin (CID 6199) exhibited docking score of -7.4 kcal/mol. Phthalic acid, Cyclobutyl isobutyl ester (CID 6423506) again provided docking score -7.2 kcal/mol indicating favorable binding. The docking score of Benzophenone (CID 3102) was -7.1 Kcal/mol. 1-Ethynylcyclododecanol (CID 2828296) had a docking score of -7.0 kcal/mol.

Benzophenone (CID 3102) completed Pi-Alkyl binding in amino acids : CYS A:285, LEU A:469, LEU A :465. It had also formed an interaction in Pi-Alkyl binding in HIS A:449.

Ficusin (CID 6199) when interacting with PPAR-gamma formed a hydrogen bond at SER A:289. It also took part in Pi interaction at PHE A:364, HIS A:449 and CYS A:285. Pi-alkyl interaction was seen in LEU A:465 and LEU A:469. Pi-Sulphur interaction was seen in MET A:364 other interactions at GLN A:286 and TYR A:473 were seen.

Phthalic acid, Cyclobutyl isobutyl ester (CID 6423506) formed alkyl interactions at ARG A:228, LEU A:330, ILE A:326 and MET A:364. Pi interactions were seen at HIS A:449 and Pi-Sulfur at CYS A:285. Carbon-Hydrogen bond was present in CYS A:285

CYS A:285, and HIS A:449 were among the most common interacting amino acid residues in these ligands

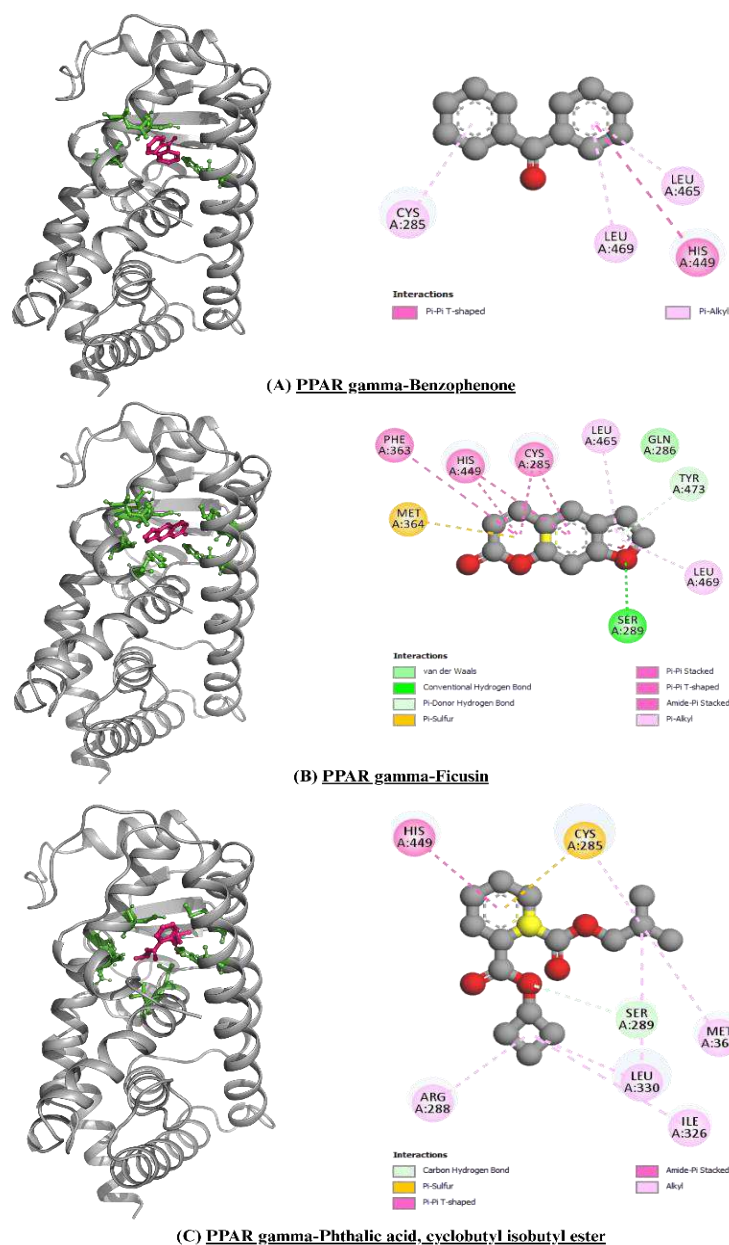


FIGURE 7 | Docking interaction (i) 3D-Interacting protein residues (green), Ligand (Pink) (ii) 2D diagram with residue interaction of (A)Benzophenone (B)Ficusin (C)Phthalic acid, cyclobutyl isobutyl ester.

24-Noroleana-3,12-diene (CID 15427754) with one Lipinski violation was observed to have the strongest docking score for the whole study, with -10.2 kcal/mol against Alpha-Amylase.

9,19-Cyclolanost-24-en-3-ol, acetate, (3.β.)- (CID 518616) was found to have one Lipinski violation with a docking score of -9.6 kcal/mol. The docking score of 1-Ethynylcyclododecanol (CID 2828296) was -7.4 kcal/mol and it had a desirable interaction with Alpha-Amylase. Benzophenone (CID 3102) displayed a docking score of -7.1 kcal/mol which is a reasonable docking score. The most common interacting residue was TYR A:62 among these ligands. Hydrogen bonding with Benzophenone (CID 3102) was seen at GLN A:62 with Pi-Pi stacking in TYR A:62 and TRP A:59. With 1-Ethynylcyclododecanol (CID 2828296) at ASP A:197 conventional hydrogen bonding was seen. And at TYR A:62 Pi-Alkyl bonding was visible

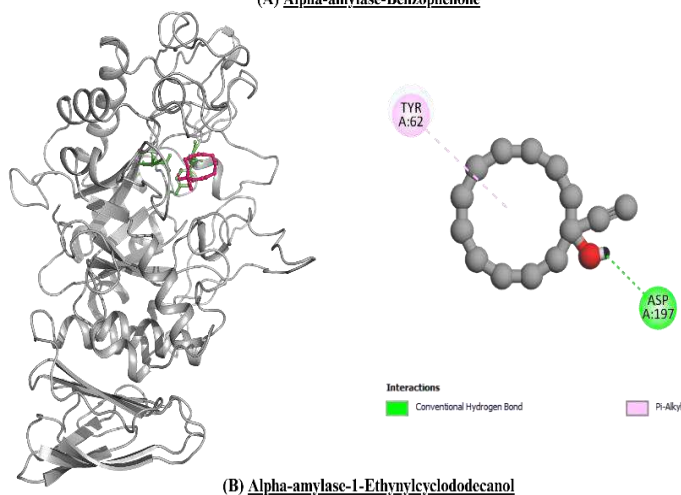
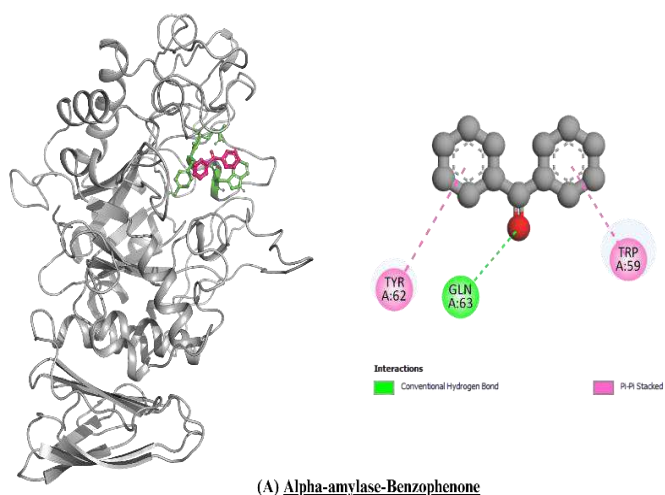


FIGURE 8 | Docking interaction (i) 3D-Interacting protein residues (Green), Ligand (Pink)
(ii) 2D diagram with residue interaction of (A)Benzophenone (B)1-Ethnylcyclododecanol.

The best interaction for Insulin receptor kinase with -8.4 kcal/mol was found with Stigmasterol (CID 5280794) which had one Lipinski violation.

Ficusin (CID 6199) showed a good docking score with a docking score of -7.3 kcal/mol. Benzophenone (CID 3102) exhibited a good docking score -7.1 kcal/mol.

The docking score for 2-p-Tolylpyridine (CID 78219) was -7.0 kcal/mol.

Benzophenone (CID 3102) formed Pi-Sigma bonds at MET A:1139 amino acid residue of insulin receptor kinase. Pi-Alkyl bonds were also formed at MET A:1153, VAL A:1010, ALA A:1028 amino acid residues of the target protein

Ficusin (CID 6199) formed a hydrogen bond with Insulin receptor kinase domain at MET A:1079. Pi-Alkyl bonds were formed at MET A:1139, LEU A:1002, ALA A:1028, MET A:1153 and VAL A:1010. Pi-Sigma bonds were also formed at VAL A:1010

The most common interacting amino acid residues with these ligands were MET A:1153, VAL A:1010, ALA A:1028, MET A:1139. These interactions were mostly Pi interactions.

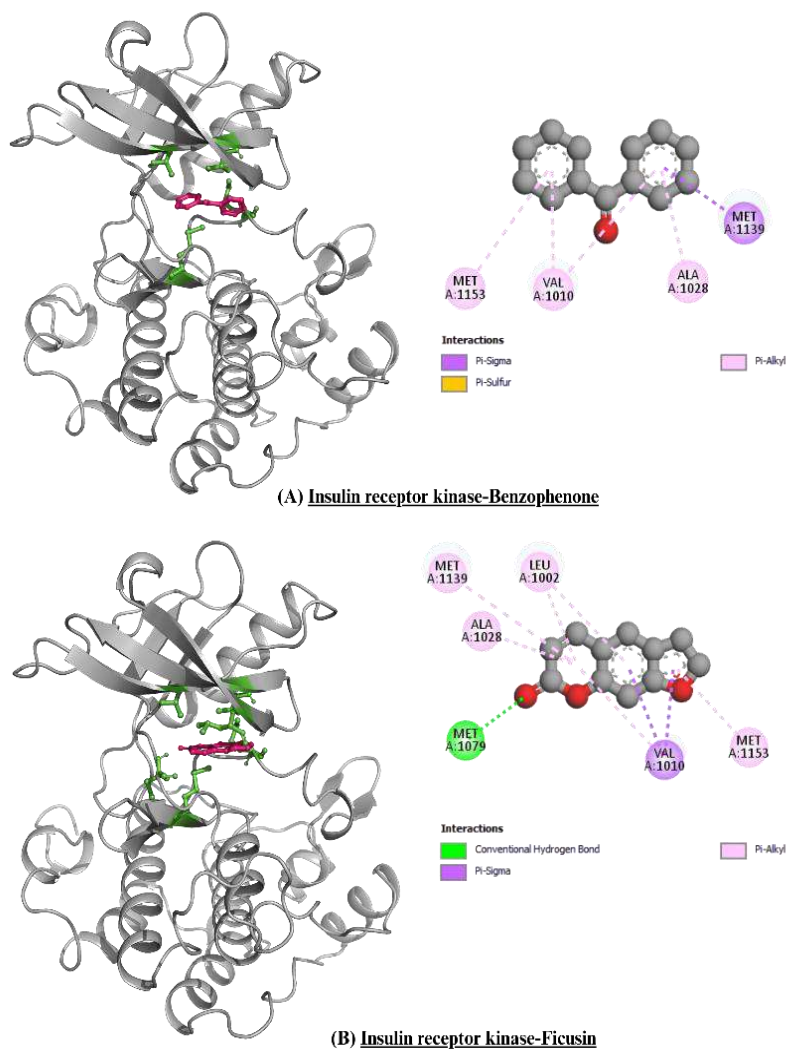


FIGURE 9 | Docking interaction (i) 3D-Interacting protein residues (green), Ligand (Pink) (ii) 2D diagram with residue interaction of (A)Benzophenone (B)Ficusin.

Benzophenone (CID 3102) exhibited the highest docking score against Dipeptidyl Peptidase IV (-7.3 kcal/mol), which further confirms the broad-spectrum binding potential of this phytochemical as observed consistently in all four targets in this study. The docking score of Ficusin (CID 6199) was found to be -7.1 kcal/mol, suggesting a favorable interaction. Phthalic acid, cyclobutyl

isobutyl ester (CID 6423506) also attained a docking score of -7.1 kcal/mol, which was the same as Ficusin's score and also indicated a favorable ligand interaction with this enzyme.

Benzophenone (CID 3102) formed Pi-interactions at TYR A:662 and TYR A:666.

(p-Tolyl)pyridine (CID 78219) formed important interactions in amino acid residues PHE A:357, VAL A:656, TYR A:662. Pi-Stacking was seen in TYR A:666. Carbon hydrogen bond was seen in HIS A:740.

Phthalic acid, cyclobutyl isobutyl ester (CID 6423506) formed a covalent hydrogen bond with the SER A:630 residue of DPP-IV which also took part in forming Pi interaction. Pi interaction was seen also in other amino acid residues such as TYR A:662 and TYR A:666. TYR A:666 also took part in Pi-Alkyl interaction with the ligand.

The interacting residues among ligands were TYR A:662, TYR A:666, PHE A:357, VAL A:656, HIS A:740, TYR A:631. Hydrogen bonding was seen by Phthalic acid, cyclobutyl isobutyl ester (CID 6423506) on SER A:630 of DPP-IV protein.

TYR A:666 and TYR A:662 were of the most common interacting residues between all these ligands

Among all the four protein targets Benzophenone (CID 3102) was found to be the most active phytochemical. It had good docking scores for PPAR-gamma (-7.1 kcal/mol), Alpha-Amylase (-7.1 kcal/mol), Insulin receptor kinase (-7.1 kcal/mol) and Dipeptidyl Peptidase IV (-7.3 kcal/mol). It is a good candidate for further study. Ficusin (CID 6199) also showed consistent binding to PPAR-gamma (-7.4 kcal/mol), Insulin receptor kinase (-7.3 kcal/mol) and DPP-IV (-7.1 kcal/mol) further supporting its potential as multi-target ligand candidate. 1-Ethynylcyclododecanol (CID

2828296) and 2-p-Tolylpyridine (CID 78219) also showed good binding energies against their respective targets.

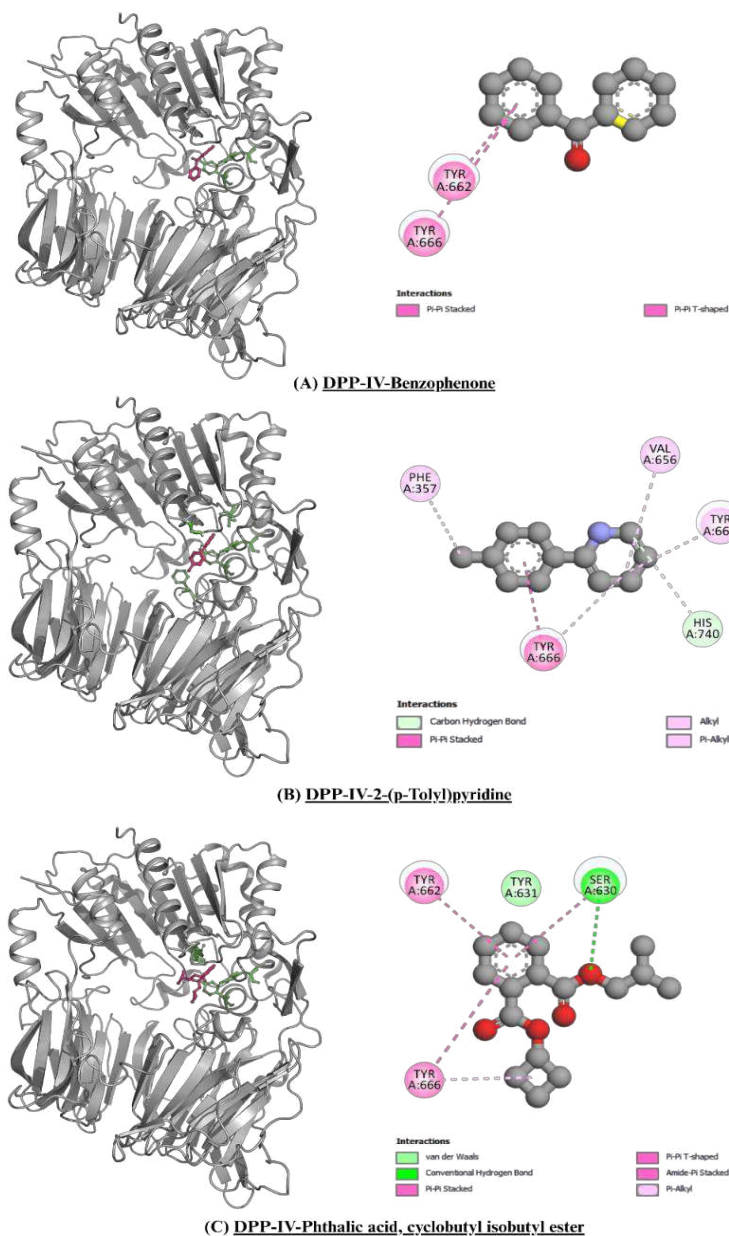


FIGURE 10 | Docking interaction (i) 3D-Interacting protein residues (green), Ligand (Pink) (ii) 2D diagram with residue interaction of (A)Benzophenone (B)2-p-tolypyridine (C)Phthalic acid, cyclobutyl isobutyl ester.

TABLE 9 | Docking energy/Docking score (kcal/mol) for phytochemicals against (A)PPAR-gamma (B)Human alpha amylase(C)Insulin kinase receptor (D)DDP-IV

Phytochemical	CID	Docking Score(kcal/mol)
(A)PPAR-gamma		
Stigmasterol	5280794	-9.3
Ficusin	6199	-7.4
Benzophenone	3102	-7.1
1-Ethynylcyclododecanol	2828296	-7
Phthalic acid, cyclobutyl isobutyl ester	6423506	-7.2
(B)Alpha-Amylase		
Benzophenone	3102	-7.1
1-Ethynylcyclododecanol	2828296	-7.4
24-Noroleana-3,12-diene	15427754	-10.2
9,19-Cyclolanost-24-en-3-ol, acetate, (3.beta.)-	518616	-9.6
(C)Insulin receptor kinase		
Benzophenone	3102	-7.1
Ficusin	6199	-7.3
2-p-Tolylpyridine	78219	-7
Stigmasterol	5280794	-8.4
(D)Dipeptidyl peptidase IV		

Benzophenone	3102	-7.3
Ficusin	78219	-7.1
Phthalic acid, cyclobutyl isobutyl ester	6423506	-7.1

3.9 | Pharmacokinetics

Among the docked 51 phytochemicals five of them: Benzophenone, 1-Ethynylcyclododecanol, Ficusin, 2-p-Tolylpyridine, and Phthalic acid cyclobutyl isobutyl ester showed promising docking energy with ADME and toxicity properties by evaluation on SwissADME and pkCSM where this phytochemicals exhibited high gastrointestinal absorption with consensus LogP values ranging from 2.12 to 3.72, pointing moderate to high lipophilicity.

Four of the five phytochemicals were classified as soluble, while 1-Ethynylcyclododecanol was moderately soluble, based on ESOL predictions. Hydrogen bond acceptors and donors were within drug-like ranges, and none of the phytochemical ADME and Toxicity profiles show any Lipinski rule-of-five violations.

Regarding cytochrome P450 interactions selective inhibition profiles were observed across CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4 isoforms, with no phytochemical inhibiting all five.

TABLE 10 | Druggable compounds selected from docking study by ADME and Toxicity analysis by Swiss-ADME and pkCSM server.

Compound name	Benzophenone	1-Ethynylcyclododecanol	Ficusin	2-p-Tolylpyridine	Phthalic acid, cyclobutyl isobutyl ester
ADME					
CID	3102	2828296	6199	78219	6423506
Formula	C13H10O	C14H24O	C11H6O3	C12H11N	C16H20O4
MW	182.22	208.34	186.16	169.22	276.33
H-bond acceptors	1	1	3	1	4
H-bond donors	0	1	0	0	0
Consensus Log P	2.93	3.72	2.12	2.85	3.32
ESOL Log S	-3.48	-4.27	-2.73	-3.39	-3.64
ESOL Class	Soluble	Moderately soluble	Soluble	Soluble	Soluble
GI absorption	High	High	High	High	High
BBB permeant	Yes	Yes	Yes	Yes	Yes

CYP1A2 inhibitor	Yes	No	Yes	Yes	Yes
CYP2C19 inhibitor	Yes	No	No	Yes	Yes
CYP2C9 inhibitor	No	Yes	No	No	Yes
CYP2D6 inhibitor	No	No	No	No	No
CYP3A4 inhibitor	No	No	No	No	No
Lipinski violations	0	0	0	0	0
Toxicity					
Oral Rat Acute Toxicity (LD50)	1.858 mol/kg	1.887 mol/kg	1.874 mol/kg	2.185 mol/kg	2.075 mol/kg
Ames test	No	No	No	No	No
Hepatotoxicity	No	No	No	No	No
hERG I inhibition	No	No	No	No	No
hERG II inhibition	No	No	No	No	No

In terms of toxicities. All these 5 phytochemicals (Benzophenone, 1-Ethynylcyclododecanol, Ficusin, 2-p-Tolylpyridine, and Phthalic acid cyclobutyl isobutyl ester) showed negative for the Ames test, hepatotoxicity, hERG I inhibition, and hERG II inhibition, reasoning a favorable safety profile with low mutagenic hepatotoxic and cardiotoxic risk.

4 | Discussion

The acute toxicity assessment demonstrated that the methanolic extract of *F. hispida* was well tolerated and deemed safe within the dosage range employed in this study. This study aimed to examine the antidiabetic properties of methanolic extracts of *F. hispida* in alloxan-induced diabetic mice. Alloxan is a recognized diabetogenic compound that specifically targets and damages pancreatic β -cells by producing reactive oxygen species (ROS), resulting in diminished insulin production and sustained hyperglycemia. The alloxan-induced diabetic model is commonly employed to evaluate the glucose-lowering effectiveness of prospective antidiabetic compounds [47]. Glibenclamide is frequently utilized as a benchmark antidiabetic agent in alloxan-induced mild diabetic animals to assess and compare the glucose-lowering effects of diverse bioactive substances [48].

The present investigation indicated that the diabetic control mice displayed markedly increased blood glucose levels, hence confirming the successful induction of diabetes. Treatment with MEFH at doses of 200 mg/kg and 400 mg/kg over a 21-day period resulted in a substantial decrease in blood glucose levels relative to the diabetic control group. The antihyperglycaemic effect was found to be dose-dependent; specifically, the 400 mg/kg dose elicited a glucose-lowering effect that was comparable to that of the established antidiabetic drug, glibenclamide.

The results indicate that the fruit extract derived from *F. hispida* exhibits noteworthy antidiabetic properties. The observed reduction in glucose levels could be ascribed to the presence of bioactive phytochemicals, including flavonoids, tannins, phenolic compounds, and triterpenoids; these compounds are recognized for their capacity to stimulate insulin secretion, facilitate peripheral glucose uptake, and impede the activity of carbohydrate-digesting enzymes [49, 50]. Comparable antidiabetic benefits have been observed in other species within the Moraceae family. For instance, *Ficus racemosa* demonstrated that methanol extracts substantially reduced blood glucose levels. This was achieved by enhancing insulin efficacy and inhibiting enzymes that degrade carbs in diabetic animals. This suggests a shared way that plant chemicals work in different *Ficus* species. [51].

Changes in body weight are an important indicator of metabolic health in people with diabetes. Diabetes is often linked to changes in body weight, which are caused by a lack of insulin, problems with how glucose is used, and increased breakdown of proteins [9]. In this study, mice with diabetes showed significant changes in body weight. In contrast, the groups treated with MEFH showed notable improvements, especially at the higher dose, indicating a return to metabolic balance.

Serum biochemical analysis indicated that diabetic control mice exhibited markedly elevated levels of serum glucose, amylase, and bilirubin, suggesting metabolic disruption and potential pancreatic and hepatic dysfunction linked to diabetes. The administration of MEFH markedly normalized these altered parameters in a dose-dependent manner, indicating a protective effect of the extract against diabetes-induced biochemical abnormalities. The protective effect of MEFH may be ascribed to bioactive phytochemicals, including flavonoids, phenolic compounds, tannins, and triterpenoids, which modulate glucose metabolism and enhance pancreatic and hepatic

function. Comparable normalization of serum glucose, amylase, and bilirubin has been documented with flavonoid-rich plant extracts like *Syzygium cumini*, *Bauhinia variegata*, *Hibiscus sabdariffa* and *Ficus racemosa* in diabetic models, indicating a common phytochemical-mediated mechanism[9, 52–54].

Diabetic dyslipidemia, a major metabolic issue, arises from long-term high blood sugar and problems with how insulin works. This condition then increases the risk of heart and blood vessel diseases. In this study, mice with diabetes showed significant disruptions in lipid balance. This was indicated by increased total cholesterol (TC), triglycerides (TG), and low-density lipoprotein (LDL) levels, along with decreased high-density lipoprotein (HDL) concentrations. These changes indicate that lipid transport is impaired, and the liver is producing more lipids, which is a common feature of diabetes. Notably, the methanolic fruit extract of *F. hispida* (MEFH) showed a dose-dependent ability to reverse these lipid abnormalities. The higher dose produced a lipid-modulating effect close to that of the standard drug. This effect of normalizing lipids suggests that MEFH might positively influence how lipids are processed and distributed. As a result, this could potentially reduce cardiovascular problems related to diabetes.[44, 55].

Diabetic mice exhibited marked elevations in liver and kidney function indicators, specifically SGPT, SGOT, ALP, urea, and creatinine. These alterations are indicative of diabetes-induced hepatic and renal dysfunction, a consequence of prolonged hyperglycemia. Conversely, the administration of the methanolic fruit extract of *F. hispida* (MEFH) led to a considerable reduction of these changes, with the effects being more obvious at higher dosages. The normalization of these biomarkers suggests that MEFH has a protective effect on the liver and kidneys in the context of diabetes. Furthermore, the lack of mortality or any noticeable negative effects observed during

the acute toxicity study strengthens the argument for MEFH's safety at the given doses, thereby supporting its potential for therapeutic use [56].

Diabetes mellitus presents a constellation of intricate metabolic disturbances, affecting glucose homeostasis, lipid profiles, and the functionality of various organs. The persistent hyperglycemia stemming from compromised insulin action initiates a cascade of events that disrupts the normal processing of lipids, thereby precipitating diabetic dyslipidemia. These lipid irregularities intensify metabolic strain, thereby precipitating functional deficits in essential organs, with the liver and kidneys being especially vulnerable. As a result, individuals with diabetes often present with elevated liver enzymes, specifically SGPT, SGOT, and ALP, alongside increased renal biomarkers such as urea and creatinine, which serve as indicators of hepatic and renal compromise. The robust correlation between excessive cholesterol levels and alterations in liver and kidney function underscores the pervasive nature of diabetes-related issues. Furthermore, when these markers normalize during diabetes treatment, it indicates improved metabolic regulation and the reinstatement of organ function [57, 58].

The initial phytochemical analysis of the methanolic fruit extract from *F. hispida* showed the presence of flavonoids, tannins, alkaloids, phenolic compounds, and triterpenoids. These compounds are widely known for their potential to help manage diabetes. These plant-based compounds help control blood sugar levels through several interconnected processes. Flavonoids and phenolic compounds help pancreatic β -cells release more insulin and improve insulin sensitivity in other tissues. This leads to better glucose uptake and use [59–61]. Tannins are known to inhibit important carbohydrate-digesting enzymes, such as α -amylase and α -glucosidase. This inhibition results in a decreased absorption of glucose in the intestines and a decrease in the elevation of blood sugar postprandially [62, 63]. Alkaloids could potentially improve glycaemic

control through the modulation of insulin signaling pathways and the augmentation of pancreatic function; conversely, triterpenoids exhibit insulin-mimetic characteristics and impede hepatic glucose production by promoting glycogen synthesis. The presence of diverse bioactive components within MEFH suggests a synergistic mode of action, wherein multiple metabolic targets are simultaneously influenced. Consequently, the antidiabetic effects observed with *F. hispida* methanolic fruit extract are probably attributable to the combined actions of these phytochemicals [64, 65]. While the results reveal considerable antidiabetic and metabolic benefits associated with MEFH, the study's limitations stem from the absence of molecular or histopathological examinations, thereby necessitating additional research to fully understand the underlying mechanisms.

In order to better understand the molecular mechanism underlying the observed antidiabetic effect, *in silico* molecular docking studies were carried out against four important therapeutic targets involved in diabetes mellitus, namely, PPAR- γ , insulin receptor kinase (IRK), α -amylase, and DPP-IV. The targets were chosen because they control different but interrelated pathways that are associated with glucose homeostasis. PPAR- γ and IRK are key factors associated with insulin sensitivity and insulin-mediated glucose uptake, while α -amylase and DPP-IV are associated with postprandial hyperglycemia and incretin changes, respectively. A methanolic extract of the fruit displayed several phytochemicals with good binding affinities towards these targets. Interestingly, Benzophenone and Ficusin exhibited the same interactions in several receptors, indicating a possible multitarget action. Combining these multiple targets would be especially useful in diabetes management, as simultaneous insulin signaling modulation, glucose utilization, and carbohydrate metabolism may yield better therapeutic results. Additionally, the good ADMET and toxicity results obtained for the lead compounds suggest their use as potent bioactive compounds

with vivo bioactivity. The docking and pharmacokinetic data together provide mechanistic evidence that supports the antihyperglycemic, antihyperlipidemic, and organ-protective activity of the *F. hispida* observed in the mice with alloxan-induced diabetes.

5 | Conclusion

The results of this study indicate that the methanolic fruit extract from *F. hispida* has substantial antidiabetic effects in mice with diabetes caused by alloxan. The extract's effect on blood glucose levels depended on the dose given. The highest dose worked as well as the standard drug, glibenclamide. Moreover, the extract treatment significantly improved the diabetes-related changes in body weight, fat levels, and indicators of liver and kidney function. This indicates an overall enhancement in metabolic equilibrium. The extract's safety is further supported by the absence of any observed toxic effects at the doses used. The observed effects of drugs are likely caused by active plant compounds, such as flavonoids, tannins, alkaloids, phenolic compounds, and triterpenoids. Moreover, the results suggest that *F. hispida* could be a promising natural option for managing diabetes and its related problems. Further research is needed to clarify the underlying mechanisms, isolate and characterize the bioactive constituents, identify the active components, and confirm its clinical usefulness, which will help determine its potential as a treatment.

Data availability statement

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper.

CRediT Author Contribution

F.A.N., and **P.A.J.** contributed to conceptualization and methodology development. **S.M.**, **M.I.S.J.**, **S.M.**, **T.H.F.**, and **S.C** performed data curation, investigation, and conducted the formal analysis. **P.A.J.**, **M.D.**, and **M.I** were responsible for funding acquisition and project administration. **M.D.**, **S.C.**, **I.B.W.**, and **P.A.J** provided resources and supervision. **F.A.N.**, **P.A.J.**, **S.M.** and **M.I.S.J.** prepared visualization and data presentation. **F.A.N.**, **P.A.J.**, **S.M.**, **E.K.**, **T.H.F.**, and **M.I.S.J.** wrote the original draft of the manuscript. All authors reviewed, edited, and approved the final version of the manuscript.

References

- [1] Alam S, Sarker MMR, Sultana TN, et al. Antidiabetic phytochemicals from medicinal plants: prospective candidates for new drug discovery and development. *Frontiers in endocrinology* 2022; 13: 800714.
- [2] Prandi FR, Lecis D, Illuminato F, et al. Epigenetic modifications and non-coding RNA in diabetes-mellitus-induced coronary artery disease: pathophysiological link and new therapeutic frontiers. *International Journal of Molecular Sciences* 2022; 23: 4589.
- [3] Tariq S, Mirza MR, Choudhary MI, et al. Prediction of type 2 diabetes at pre-diabetes stage by mass spectrometry: a preliminary study. *International Journal of Peptide Research and Therapeutics* 2022; 28: 111.

- [4] Yedjou CG, Grigsby J, Mbemi A, et al. The management of diabetes mellitus using medicinal plants and vitamins. *International journal of molecular sciences* 2023; 24: 9085.
- [5] Saeedi P, Petersohn I, Salpea P, et al. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the International Diabetes Federation Diabetes Atlas. *Diabetes research and clinical practice* 2019; 157: 107843.
- [6] Madan K, Paliwal S, Sharma S, et al. Metabolic syndrome: the constellation of co-morbidities, a global threat. *Endocrine, Metabolic & Immune Disorders-Drug Targets (Formerly Current Drug Targets-Immune, Endocrine & Metabolic Disorders)* 2023; 23: 1491–1504.
- [7] Arabnozari H, Shaki F, Saberi Najjar A, et al. The effect of Polygonum hyrcanicum Rech. f. hydroalcoholic extract on oxidative stress and nephropathy in alloxan-induced diabetic mice. *Scientific Reports* 2024; 14: 18117.
- [8] Pangavhane R, Pache M. Phytochemicals in The Management of Diabetes Mellitus: A Comprehensive Review. *International Journal of Scientific Research and Technology*.
- [9] Akter S, Ali H, Shati AA, et al. Antidiabetic activity of methanolic extract of Hibiscus sabdariffa Linn. fruit in alloxan-induced Swiss albino diabetic mice. *Open Agriculture* 2024; 9: 20220243.
- [10] Shehadeh MB, Suaifan GA, Abu-Odeh A. Plants secondary metabolites as blood glucose-lowering molecules. *Molecules* 2021; 26: 4333.
- [11] Fazeli-Nasab B, Sayyed RZ, Sobhanizadeh A. In Silico Molecular Docking Analysis of α -Pinene: An Antioxidant and Anticancer Drug Obtained from Myrtus communis. *International Journal of Cancer Management*; 14.
- [12] Trasca DM, Dop D, Stoica G-A, et al. The Antidiabetic Activity of Wild-Growing and Cultivated Medicinal Plants Used in Romania for Diabetes Mellitus Management: A Phytochemical and Pharmacological Review. *Pharmaceuticals* 2025; 18: 1035.
- [13] DK A, AS M, Hegde K, et al. A systematic review on pharmacological actions of Ficus hispida. *Int J Pharm Sci Rev Res* 2022; 144–7.
- [14] Ali M, Chaudhary N. Ficus hispida Linn.: A review of its pharmacognostic and ethnomedicinal properties. *Pharmacognosy reviews* 2011; 5: 96.
- [15] Jia X-Y, Wu Y-M, Li J-Y, et al. Alkaloid constituents of Ficus hispida and their antiinflammatory activity. *Natural Products and Bioprospecting* 2020; 10: 45–49.
- [16] Dalimunthe A, Carensia Gunawan M, Dhiya Utari Z, et al. In-depth analysis of lupeol: delving into the diverse pharmacological profile. *Frontiers in Pharmacology* 2024; 15: 1461478.
- [17] Viet TD, Anh LH, Xuan TD, et al. The Pharmaceutical Potential of α - and β -Amyrins. *Nutraceuticals* 2025; 5: 21.
- [18] Cheng J, Zhang B, Zhu W, et al. Traditional uses, phytochemistry, and pharmacology of Ficus hispida Lf: A review. *Journal of ethnopharmacology* 2020; 248: 112204.

- [19] Shahriar M, Islam S, Parvin S, et al. Thrombolytic activity and antimicrobial properties of *Ficus hispida*. *Journal of Scientific Research* 2013; 5: 393–397.
- [20] Jahan T, Shahreen S, Banik J, et al. Antinociceptive activity studies with methanol extracts of *Ficus hispida* Lf leaves and fruits in Swiss albino mice. *Advances in Natural and Applied Sciences* 2011; 5: 131–135.
- [21] Jahan N, Siddiquee NH, Riva RAK, et al. Mechanistic insight into the antidiabetic effects of *Ficus hispida* fruits: Inhibition of intestinal glucose absorption and pancreatic beta-cell apoptosis. *PLoS One* 2025; 20: e0337465.
- [22] Madiwalar VS, Dwivedi PS, Patil A, et al. *Ficus benghalensis* promotes the glucose uptake- Evidence with in silico and in vitro. *Journal of Diabetes & Metabolic Disorders* 2022; 21: 429–438.
- [23] Gaur R, Chauhan A, Kanta C. A critical review of antioxidant potential and pharmacological applications of important *Ficus* species. *Journal of Herbmmed Pharmacology* 2024; 13: 537–549.
- [24] Choi H, Choi J, Jang Y, et al. Anti-Obesity and Antidiabetic Effects of Fig (*Ficus carica* L.) Fermented Extract Using *Lactobacillus plantarum* BT-LP-01. *Applied Sciences* 2024; 14: 6412.
- [25] Jonsson M, Jestoi M, Nathanail AV, et al. Application of OECD Guideline 423 in assessing the acute oral toxicity of moniliformin. *Food and chemical toxicology* 2013; 53: 27–32.
- [26] El Hilaly J, Israili ZH, Lyoussi B. Acute and chronic toxicological studies of *Ajuga iva* in experimental animals. *Journal of ethnopharmacology* 2004; 91: 43–50.
- [27] Alam A, Jain NP. STUDY OF ANTI-DIABETIC ACTION OF NYMPHAEA NOUCHALI FLOWER EXTRACT IN ALLOXAN-INDUCED DIABETES IN RATS.
- [28] Rahman S, Jan G, Jan FG, et al. Phytochemical screening and antidiabetic, antihyperlipidemic, and antioxidant effects of *Leptopus cordifolius* Decne. in diabetic mice. *Frontiers in Pharmacology* 2021; 12: 643242.
- [29] Shaikh JR, Patil M. Qualitative tests for preliminary phytochemical screening: An overview. *International journal of chemical studies* 2020; 8: 603–608.
- [30] Patil MJ, Godbole SA. Phytochemical Screening and Quantitative Analysis of Total Alkaloid, Phenolic, Flavonoid and Tannin Content Using Spectrophotometric Assays in *Trichosanthes bracteata* Lam. *The Journal of Plant Science Research* 2024; 40: 317–325.
- [31] Mirza AZ, Althagafi II, Shamshad H. Role of PPAR receptor in different diseases and their ligands: Physiological importance and clinical implications. *European journal of medicinal chemistry* 2019; 166: 502–513.
- [32] Hsu C, Shih H, Chia Y, et al. Rutin potentiates insulin receptor kinase to enhance insulin-dependent glucose transporter 4 translocation. *Molecular Nutrition & Food Research* 2014; 58: 1168–1176.
- [33] Akshatha JV, SantoshKumar HS, Prakash HS, et al. In silico docking studies of α -amylase inhibitors from the anti-diabetic plant *Leucas ciliata* Benth. and an endophyte, *Streptomyces longisporoflavus*. *3 Biotech* 2021; 11: 51.

- [34] Biswas A, Paul T, Hossain MI, et al. Computational identification of a marine derived dual inhibitor for type 2 diabetes mellitus using integrated in silico approaches. *Scientific Reports*.
- [35] Šali A, Blundell TL. Comparative protein modelling by satisfaction of spatial restraints. *Journal of molecular biology* 1993; 234: 779–815.
- [36] Meng EC, Goddard TD, Pettersen EF, et al. UCSF ChimeraX: Tools for structure building and analysis. *Protein Science* 2023; 32: e4792.
- [37] Pontius J, Richelle J, Wodak SJ. Deviations from standard atomic volumes as a quality measure for protein crystal structures. *Journal of molecular biology* 1996; 264: 121–136.
- [38] Guex N, Peitsch MC. SWISS-MODEL and the Swiss-Pdb Viewer: an environment for comparative protein modeling. *electrophoresis* 1997; 18: 2714–2723.
- [39] Trott O, Olson A. Software news and update AutoDock Vina: Improving the speed and accuracy of docking with a new scoring function. *Effic Optim Multithreading* 2009; 31: 455–461.
- [40] Ehrt C, Schulze T, Graef J, et al. Proteins plus: a publicly available resource for protein structure mining. *Nucleic acids research* 2025; 53: W478–W484.
- [41] Schrödinger L. The PyMOL Molecular Graphics System, Version 1.8. (*No Title*).
- [42] Daina A, Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Scientific reports* 2017; 7: 42717.
- [43] Pires DE, Blundell TL, Ascher DB. pkCSM: predicting small-molecule pharmacokinetic and toxicity properties using graph-based signatures. *Journal of medicinal chemistry* 2015; 58: 4066–4072.
- [44] Khadka S, Vaal T, Gurung S, et al. Exploring the Antidiabetic and Lipid-Lowering Properties of Nepali Wild Turmeric (*Curcuma aromatica*): A Potential Natural Remedy. *Food Safety and Health*.
- [45] Sayem A, Isam H, Shil RC, et al. Phytochemicals from anti-diabetic medicinal plants: A comprehensive review of glycemic control mechanisms.
- [46] Pandey K, Paul A, Kumar M, et al. Phytochemical profiling and anti-diabetic study of hydroalcoholic leaf extract of *Swietenia macrophylla* King. *Biochemistry and Biophysics Reports* 2025; 43: 102200.
- [47] Ahmed MS, Hasan MM, Alam MS, et al. Evaluation of the antidiabetic potential of *Acmella Oleracea* (L): selection of optimal plant part and solvent extract via in vitro assays and their validation in alloxan-induced mice model. *3 Biotech* 2025; 15: 423.
- [48] Nigam V, Nambiar V. Therapeutic potential of *Aegle marmelos* (L.) Correa leaves as an antioxidant and anti-diabetic agent: A review. *International Journal of Pharma Sciences and Research* 2015; 6: 611–621.
- [49] Nguyen NH, Pham THT, Nguyen NTT, et al. Herbal Medicine in Diabetes Treatment: An Updated Strategy With Flavonoid Compounds in Preclinical and Clinical Studies. *Chemistry & Biodiversity* 2025; e02806.

- [50] Widodo A, Sulastri E, Ihwan I, et al. Antidiabetic Activity, Phytochemical Analysis, and Acute Oral Toxicity Test of Combined Ethanolic Extract of *Syzygium polyanthum* and *Muntingia calabura* Leaves. *The Scientific World Journal* 2024; 2024: 3607396.
- [51] Sharma S, Choudhary M, Sharma O, et al. Mechanistic insights into antidiabetic potential of *Ficus virens* against multi organ specific diabetic targets: molecular docking, MDS, MM-GBSA analysis. *Computational Biology and Chemistry* 2024; 113: 108185.
- [52] Bhuyan ZA, Rokeya B, Masum N, et al. Antidiabetic effect of *Syzygium cumini* L. seed on type 2 diabetic rats. *Dhaka University Journal of Biological Sciences* 2010; 19: 157–164.
- [53] Abdel-Halim AH, Fyiad A, Aboulthana WM, et al. Assessment of the anti-diabetic effect of *Bauhinia variegata* gold nano-extract against streptozotocin induced diabetes mellitus in rats. *J Appl Pharm Sci* 2020; 10: 077–091.
- [54] Ahmed F, Hudeda S, Urooj A. Antihyperglycemic activity of *Ficus racemosa* bark extract in type 2 diabetic individuals. *Journal of diabetes* 2011; 3: 318–319.
- [55] Paul TR, Monowar M, Rahman AS, et al. Antidiabetic, Hypolipidemic and Hepatoprotective Potential of Edible Leaves Extract from the Plant *Chenopodium album* (Linn) in Streptozotocin-Induced Diabetic Mice. *Am J Chem Pharm* 2025; 4: 29–36.
- [56] Adekemi FE, Folake JK, Omowumi FP. Antidiabetic effects of aqueous leaf extract of *Vernonia amygdalina* on serum liver markers in streptozotocin-induced diabetic albino Rats: a new data to support its Anti-diabetic effect. *Clinical Phytoscience* 2024; 10: 13.
- [57] Hajam YA, Kumar R, Reshi MS, et al. Administration of *Costus igneus* Nak leaf extract improves diabetic-induced impairment in hepatorenal functions in male albino rats. *Journal of King Saud University-Science* 2022; 34: 101911.
- [58] Tehseen I, Haq T, Ilahi I, et al. Antidiabetic and hepato-renal protective effects of medicinal plants in STZ induced diabetic rats. *Brazilian Journal of Biology* 2024; 84: e260189.
- [59] Gallardo-Villanueva P, Fernández-Marcelo T, Villamayor L, et al. Synergistic effect of a flavonoid-rich cocoa–carob blend and metformin in preserving pancreatic beta cells in Zucker diabetic fatty rats. *Nutrients* 2024; 16: 273.
- [60] Firdous SM, Irfan Z, Giri S, et al. Unravelling the role of natural herbs for the management of diabetes mellitus. *Diabetology & Metabolic Syndrome* 2025; 17: 398.
- [61] Bouyahya A, Balahbib A, Khalid A, et al. Clinical applications and mechanism insights of natural flavonoids against type 2 diabetes mellitus. *Heliyon*; 10.
- [62] Li K, Yao F, Du J, et al. Persimmon tannin decreased the glycemic response through decreasing the digestibility of starch and inhibiting α -amylase, α -glucosidase, and intestinal glucose uptake. *Journal of Agricultural and Food Chemistry* 2018; 66: 1629–1637.
- [63] Alam S, Dhar A, Hasan M, et al. Antidiabetic potential of commonly available fruit plants in Bangladesh: updates on prospective phytochemicals and their reported MoAs. *Molecules* 2022; 27: 8709.

- [64] Chahrour JA, Abdel Baki Z, El Badan D, et al. Herbal Medicines in the Management of Diabetes Mellitus: Plants, Bioactive Compounds, and Mechanisms of Action. *Biomolecules* 2025; 15: 1674.
- [65] Praparatana R, Maliyam P, Barrows LR, et al. Flavonoids and Phenols, the Potential Anti-Diabetic Compounds from Bauhinia strychnifolia Craib. Stem. *Molecules* 2022; 27: 2393.