Comprehensive Evaluation of Antidiabetic and Antihyperlipidemic Potential of *Coccinia grandis* Fruit Extracts in Streptozotocin-Induced Diabetic Rats

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ABSTRACT

Introduction: *Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia resulting from defects in insulin secretion, insulin action, or both. It is associated with serious complications including cardiovascular disease, neuropathy, nephropathy, and retinopathy. Conventional treatments, while effective, often have limitations and adverse effects, prompting interest in alternative therapies. Phytomedicine, derived from medicinal plants, offers a promising complementary approach due to its affordability, accessibility, and presence of bioactive compounds with antidiabetic properties. Increasing scientific evidence supports the potential of various plant extracts in modulating glucose metabolism and improving diabetic outcomes.*

Methods: *Ethanolic (EE-FCG), methanolic (ME-FCG), and chloroform (CE-FCG) extracts were prepared by reflux condensation and subjected to phytochemical screening and acute oral toxicity studies per OECD-423 guidelines. Diabetes was induced via STZ (30 mg/kg i.p.), and rats were treated with extracts at 100, 200, and 300 mg/kg orally for 21 days. Glibenclamide (5 mg/kg) served as the standard. Fasting blood glucose, serum lipid profile, and histopathology of pancreatic tissue were analyzed.*

Results: *EE-FCG and ME-FCG demonstrated significant, dose-dependent reductions in fasting blood glucose and serum lipid levels ($p < 0.001$) comparable to glibenclamide. CE-*

FCG showed moderate efficacy. All extracts were non-toxic up to 2000 mg/kg. Histopathology supported pancreatic β-cell protection in treated groups.

Conclusion: *Coccinia grandis fruit extracts exhibit potent antidiabetic and antihyperlipidemic effects, likely due to flavonoids, terpenoids, and phenolics. These findings support their traditional use and justify further clinical evaluation.*

Keywords: *Metabolic disorder, phytomedicine, antidiabetic, antihyperlipidemic, phytochemicals and streptozotocin etc.*

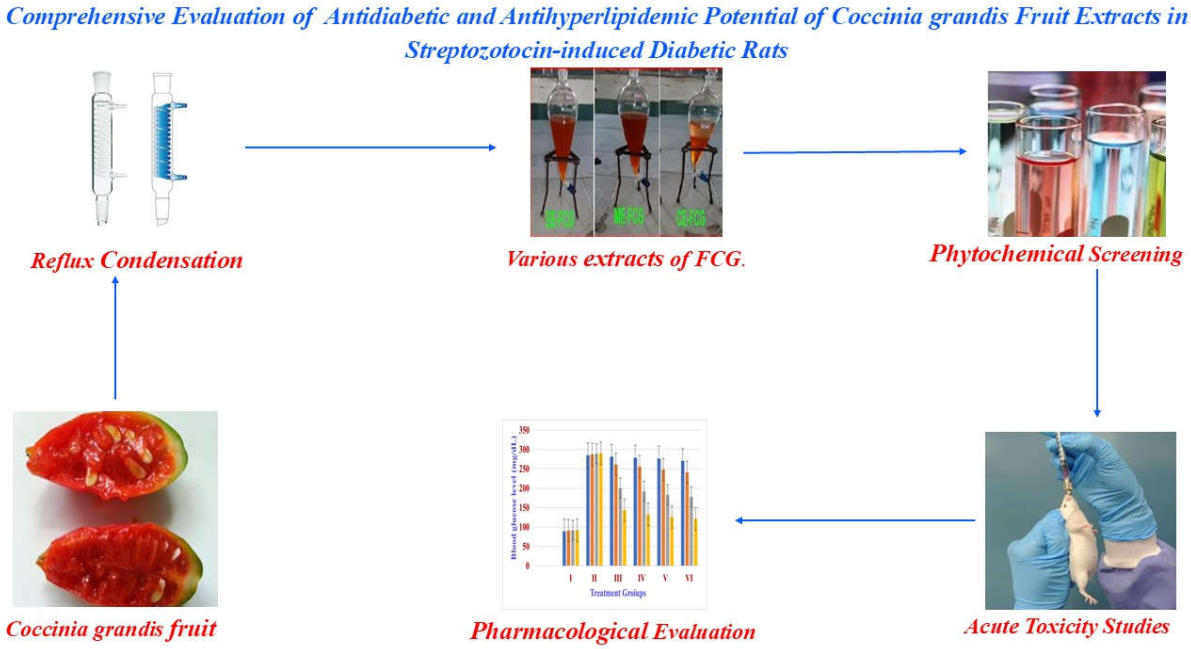

Fig-1: Graphical Abstract

1. INTRODUCTION

Nature has long served as a quintessential model of symbiosis, illustrating the intricate interdependence between biotic organisms and their abiotic environment. Among the countless endowments of the natural world, the plant kingdom holds a position of paramount importance in sustaining human existence, providing the fundamental necessities of life namely food, clothing, and shelter alongside a multitude of other invaluable resources. Beyond these primary contributions, nature constitutes an abundant repository of medicinal agents, many of which have been employed across civilizations for the treatment of human ailments. The corpus of ethnobotanical knowledge has evolved over thousands of years, shaped by empirical observation, experimentation, and the transmission of traditional wisdom across generations. As a result, contemporary healthcare systems have been profoundly enriched by these time-honored natural therapies. Given their heightened susceptibility to a broad spectrum of diseases and disorders, humans have consistently turned to the plant world for therapeutic intervention. Numerous phytochemicals have been isolated from various plant parts, and these bioactive constituents have been shown to possess a wide range of pharmacological and antimicrobial properties [1-3].

2. OVERVIEW OF *COCCINIA GRANDIS*

(L.) Voigt, commonly known as ivy gourd, is a fast-growing perennial climber belonging to the Cucurbitaceae family. Widely distributed across tropical Asia and Africa, it is traditionally used in various indigenous systems of medicine. The plant is valued for its rich phytochemical profile, including alkaloids, flavonoids, saponins, and tannins, which contribute to its therapeutic properties. It has demonstrated significant pharmacological activities such as antidiabetic, antioxidant, anti-inflammatory, and antimicrobial effects. The leaves and fruits are commonly consumed as vegetables and are believed to help regulate blood sugar levels, making the plant particularly beneficial for diabetic patients. Recent studies have further validated its traditional uses, highlighting its potential in modern phytotherapy. Owing to its wide-ranging health benefits, *Coccinia grandis* continues to be a subject of growing interest in ethnopharmacological research [4-6].

Fig-2: *Coccinia grandis* plant with ripen fruits.

3. MATERIALS AND METHOD

3.1. Drugs and Chemicals

The standard drug, Glibenclamide, was procured from a licensed local retail pharmacy and the all solvents and chemicals required for extraction and phytochemical screening, including ethanol, methanol, chloroform, dichloromethane, sodium chloride, and anhydrous magnesium sulphate, were of analytical grade. Streptozotocin was procured from Sigma-Aldrich's authorized distributor in India.

3.2. Experimental Animals

White male albino Wistar rats (200–250 g) were procured from the animal facility of C.L. Baid Metha College of Pharmacy, Chennai. The animals were acclimatized for seven days under standard laboratory conditions (25 ± 5°C, 12-hour light/dark cycle) with free access to food and water. They were housed in well-ventilated plastic cages and monitored to exclude any intercurrent infections. All experimental procedures were approved by the Institutional Animal Ethics Committee (IAEC) in accordance with CPCSEA guidelines (Approval No: IAEC/XXIX/06/2020).

3.3. Methodology for Extraction

Three separate samples, each containing 20 g of ripened *Coccinia grandis* fruit paste (prepared by mashing the fruits), were accurately weighed and transferred into three individual 250 mL round-bottomed flasks (RBFs). To the first flask, 50 mL of ethanol and 60 mL of dichloromethane were added; to the second, 50 mL of methanol and 60 mL of dichloromethane; and to the third, 50 mL of chloroform and 60 mL of dichloromethane. Each flask was fitted with a reflux condenser and heated on a steam bath for 5 minutes, with frequent agitation to facilitate extraction. Following reflux, the mixtures were filtered under suction, and the filtrates

were transferred to separating funnels. Each was washed three times with 150 mL portions of sodium chloride solution to remove impurities. The organic layers were then dried over anhydrous magnesium sulphate, filtered, and concentrated by evaporating the solvents under reduced pressure, avoiding heat application. The resulting extracts ethanolic extract (EE-FCG), methanolic extract (ME-FCG), and chloroform extract (CE-FCG) of *Coccinia grandis* fruits were stored under refrigeration until further phytochemical and pharmacological evaluation [7].

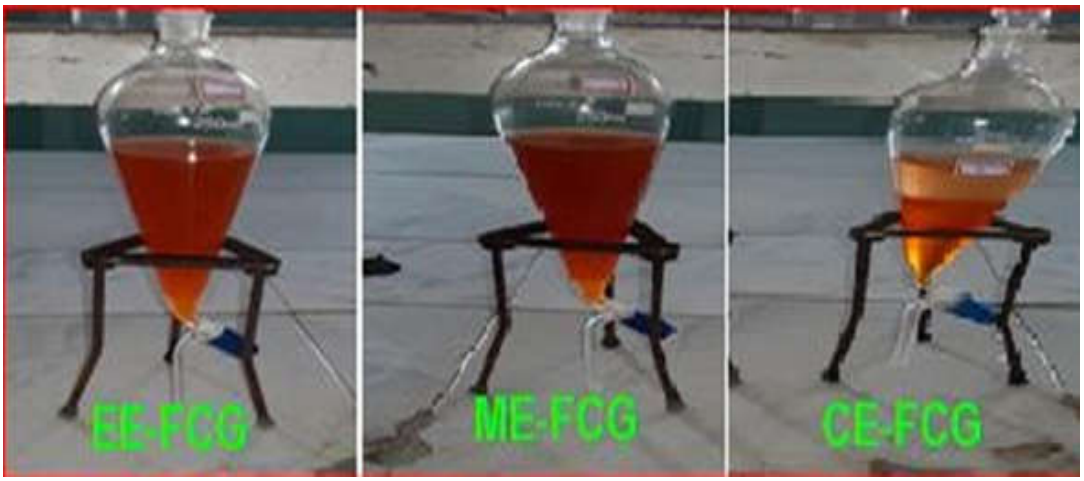

Fig-3: Various extracts of FCG.

3.4. Phytochemical Screening

Phytochemical screening of the various extracts of Coccinia grandis fruit was conducted using different qualitative confirmatory tests [8, 9].

3.5. Acute oral Toxicity Studies

In the present investigation, the acute oral toxicity of the ethanolic (EE-FCG), methanolic (ME-FCG), and chloroform (CE-FCG) extracts of *Coccinia grandis* fruit was assessed employing the acute toxic class method. This methodology utilized a sequential, stepwise protocol, wherein each phase involved administering the extract to cohorts of three Wistar rats. Prior to dosing, the animals were subjected to a fasting period of three to four hours, with unrestricted access to water. Subsequent to fasting, each animal was weighed, and the designated extract was administered orally at a single dose of 2000 mg/kg body weight. Post-administration, the subjects were individually monitored immediately and at least once within the first 30 minutes, followed by periodic observations over the initial 24-hour period, with heightened scrutiny during the first 4 hours. Daily monitoring was maintained for a total duration of 14 days to detect any manifestations of toxicity or mortality [10].

3.6. Evaluation of Antidiabetic Activity:

Diabetes mellitus was induced in the animals by a single intraperitoneal injection of streptozotocin (STZ), dissolved in 0.1 M cold sodium citrate buffer (pH 4.4), at a dose of 30 mg/kg body weight following an overnight fast. To prevent drug-induced hypoglycemia, the animals were provided with a 5% glucose solution overnight. After 72 hours, animals exhibiting fasting blood glucose levels exceeding 200 mg/dL along with glycosuria were classified as diabetic. Subsequently, the diabetic rats were randomly allocated into six groups, each consisting of six animals, for further experimental evaluation [11–13].

Experimental Groups:

A. Group I: Normal control (vehicle-treated)

B. Group II: Diabetic control (vehicle-treated)

C. Group III: Diabetic rats treated with E-FCG at 100 mg/kg orally (p. o.)

D. Group IV: Diabetic rats treated with E-FCG at 200 mg/kg p. o.

E. Group V: Diabetic rats treated with E-FCG at 300 mg/kg p. o.

F. Group VI: Diabetic rats treated with glibenclamide at 5 mg/kg p. o.

The vehicle, E-FCG, and glibenclamide were administered once daily for 21 consecutive days starting from the day of diabetes induction. Blood samples were obtained from the tip of the tail, and blood glucose levels were measured on days 0, 7, 14, and 21 using a glucometer with test strips. On the 21st day, blood was collected via retro-orbital puncture to determine serum cholesterol and triglyceride levels using an auto-analyzer. Fresh urine samples were collected on days 0 and 21 for qualitative estimation of glucose and ketone bodies using Keto-Diastix strips. Additionally, histopathological analysis of pancreatic tissue was conducted to assess cellular changes.

4. RESULTS AND DISCUSSION

4.1. Phytochemical Screening

Preliminary Phytochemical Screening of EE-FCG, ME-FCG, and CE-FCG (Ethanolic, Methanolic, and Chloroform extracts of *Coccinia grandis* fruits), showing the presence (+) or absence (–) of major phytochemical constituents:

Table-1: Preliminary Phytochemical Screening of EE-FCG, ME-FCG, and CE-FCG

Phytochemical Constituents	EE-FCG	ME-FCG	CE-FCG
Alkaloids	+	+	–
Flavonoids	+	+	+
Tannins	+	+	–
Saponins	+	+	–
Glycosides	+	+	–
Phenolic compounds	+	+	+
Steroids	–	–	+
Terpenoids	+	+	+
Carbohydrates	+	+	–
Proteins	–	+	–

Note: "+" indicates the presence of the constituent. **"–"** indicates its absence.

Findings and Summary:

Preliminary phytochemical screening of the ethanolic (EE-FCG), methanolic (ME-FCG), and chloroform (CE-FCG) extracts of *Coccinia grandis* fruits revealed the presence of various bioactive constituents. Both EE-FCG and ME-FCG tested positive for a wide range of phytochemicals including alkaloids, flavonoids, tannins, saponins, glycosides, phenolic compounds, terpenoids, and carbohydrates, indicating their rich phytochemical profile. In contrast, CE-FCG showed a more limited profile, with the presence of flavonoids, phenolic compounds, steroids, and terpenoids only. Notably, steroids were detected exclusively in the CE-FCG extract, while proteins were found only in ME-FCG. The absence of certain compounds, such as alkaloids and tannins in CE-FCG, may suggest a solvent-specific

extraction efficiency. These findings suggest that EE-FCG and ME-FCG are phytochemically more diverse and may hold greater therapeutic potential, warranting further pharmacological investigation.

4.2. Acute Oral Toxicity Studies

Table-2: For the dose selection by acute toxicity class method (OEC guide lines 423 of EE-FCG, ME-FCG and CE-FCG).

Sl. No.	Treatment group	Dose mg/kg	Sign of toxicity	Onset of toxicity	Duration
1.	EE- FCG	200	No	No	14 days
2.	ME-FCG	200	No	No	14 days
3.	CE-FCG	200	No	No	14 days

Findings and summary:

Acute oral toxicity studies were conducted in accordance with OECD Guideline 423 to evaluate the safety profile of the extracts at fixed dose levels for hazard classification purposes. The extracts were initially administered orally at a dose of 2000 mg/kg body weight, suspended in 1% carboxymethyl cellulose (CMC), and the animals were observed for 14 days for any signs of mortality or acute toxicity. Careful monitoring was carried out at least three times daily to assess effects on the central and autonomic nervous systems, motor activity, salivation, and other general indicators of toxicity. No signs of toxicity or mortality were observed at the tested dose, suggesting that the median lethal dose (LD_{50}) exceeds 2000 mg/kg body weight, placing the extracts in toxicity Class 5 (2000 mg/kg < LD_{50} < 2500 mg/kg). These findings indicate that all tested extracts are non-toxic at the administered dose and are well tolerated by experimental animals. Based on these results, a safe and effective dose of 200 mg/kg body weight was selected for further pharmacological evaluations.

4.3. Screening of Antidiabetic Activity

Table-3: Effect of EE-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21 in STZ induced diabetic rats

Group	Day 0	Day 7	Day 14	Day 21
I	89.2 ± 0.192	90.4 ± 0.834	91.6 ± 0.392	92.1 ± 0.214
II	285.1 ± 0.384	287.3 ± 0.149	289.2 ± 0.321	290.5 ± 0.118
III	281.4 ± 0.133	261.5 ± 0.023*	200.7 ± 0.155*	143.2 ± 0.098*
IV	278.7 ± 0.295*	255.9 ± 0.004*	192.4 ± 0.208**	132.5 ± 0.162**
V	276.9 ± 0.319**	248.3 ± 0.021**	183.3 ± 0.301***	125.6 ± 0.299***
VI	270.5 ± 0.377**	241.2 ± 0.065***	178.2 ± 0.441***	121.4 ± 0.351***

Note: I = Normal Control. II = Diabetic Control. III = EE-FCG (100 mg/kg). IV = EE-FCG (200 mg/kg). V = EE-FCG (300 mg/kg). VI = Glipalamide (5 mg/kg). Values are expressed as Mean ± SEM, n = 6. Significance levels: **p < 0.05, **p < 0.01, **p < 0.001 compared to Diabetic Control (Group II).

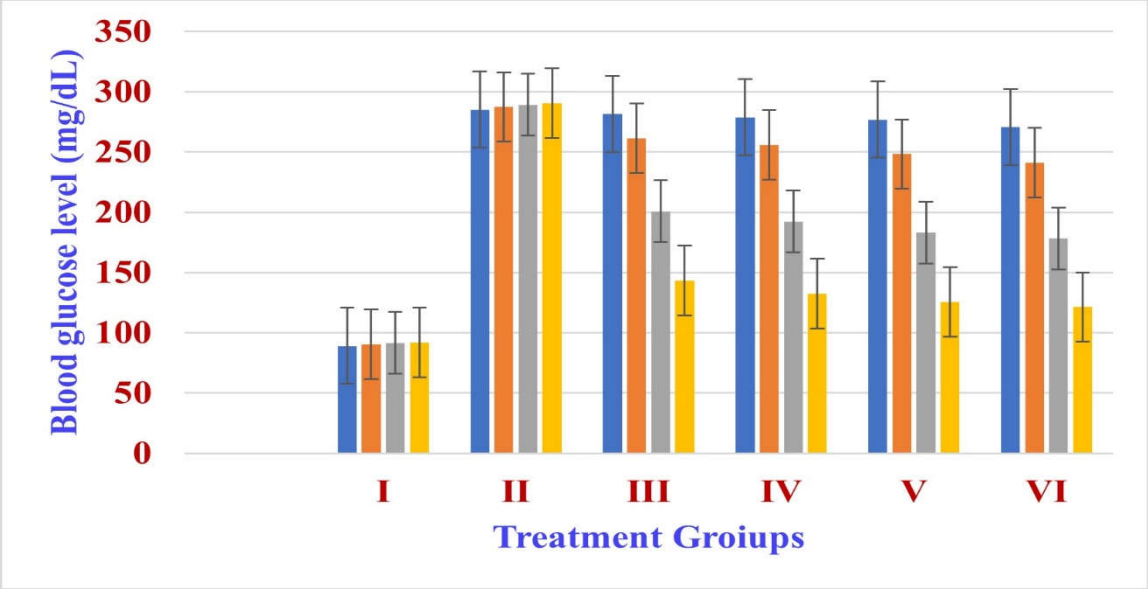

Fig-4: Effect of EE-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21

Table-4: Effect of ME-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21 in STZ induced diabetic rats

Group	Day 0	Day 7	Day 14	Day 21
I	91.1 ± 0.150	91.8 ± 0.921	92.4 ± 0.317	93.6 ± 0.239
II	284.3 ± 0.416	286.5 ± 0.121	288.7 ± 0.342	289.9 ± 0.107
III	278.9 ± 0.146	264.1 ± 0.034*	196.9 ± 0.173*	137.8 ± 0.102*
IV	276.0 ± 0.362*	259.0 ± 0.006*	189.5 ± 0.195**	129.2 ± 0.177**
V	274.7 ± 0.309**	250.4 ± 0.026**	180.1 ± 0.398***	121.8 ± 0.333***
VI	269.2 ± 0.369**	242.8 ± 0.083***	175.9 ± 0.522***	118.6 ± 0.362***

Note: I = Normal Control. II = Diabetic Control. III = ME-FCG (100 mg/kg). IV = ME-FCG (200 mg/kg). V = ME-FCG (300 mg/kg). VI = Glipalamide (5 mg/kg). Values are expressed as Mean ± SEM, n = 6. Significance levels: **p < 0.05, **p < 0.01, **p < 0.001 compared to Diabetic Control (Group II).

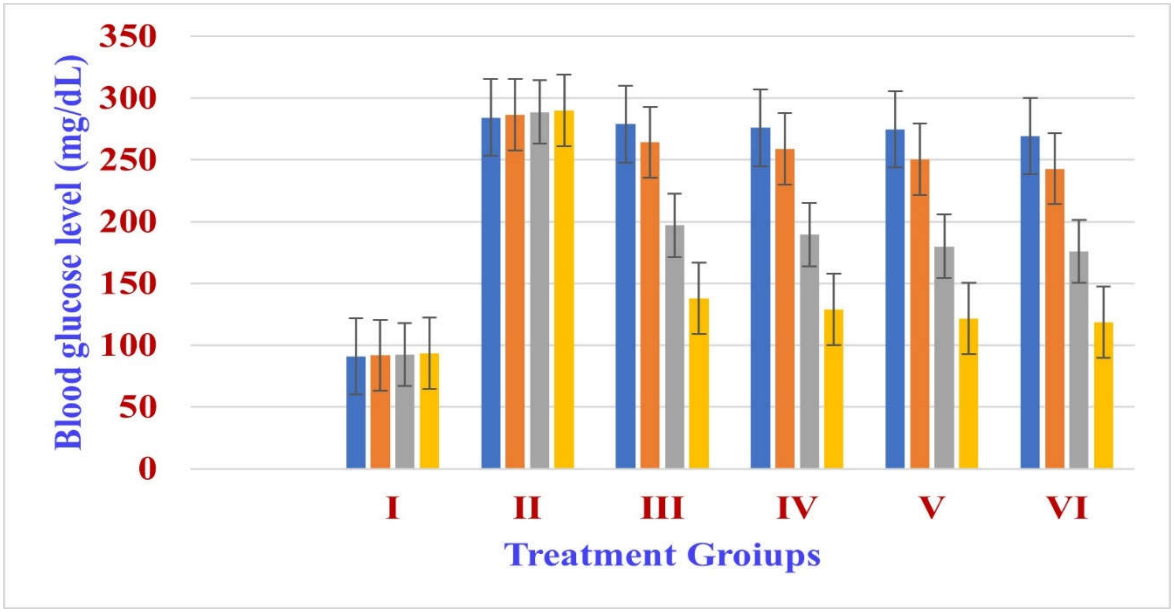

Fig-5: Effect of ME-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21

Table-4: Effect of CE-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21 in STZ induced diabetic rats

Group	Day 0	Day 7	Day 14	Day 21
I	88.6 ± 0.166	89.5 ± 0.886	90.9 ± 0.401	91.3 ± 0.207
II	282.2 ± 0.399	284.1 ± 0.110	285.9 ± 0.290	287.6 ± 0.098
III	279.8 ± 0.123	262.7 ± 0.020*	197.5 ± 0.166*	139.6 ± 0.093*
IV	277.3 ± 0.339*	256.8 ± 0.005*	190.3 ± 0.198**	130.9 ± 0.168**
V	275.6 ± 0.296**	247.9 ± 0.019**	181.8 ± 0.367***	124.2 ± 0.315***
VI	271.4 ± 0.357**	240.3 ± 0.071***	176.1 ± 0.503***	120.3 ± 0.342***

Note: I = Normal Control. II = Diabetic Control. III = CE-FCG (100 mg/kg). IV = CE-FCG (200 mg/kg). V = CE-FCG (300 mg/kg). VI = Glipalamide (5 mg/kg). Values are expressed as Mean ± SEM, n = 6. Significance levels: **p < 0.05, **p < 0.01, **p < 0.001 compared to Diabetic Control (Group II).

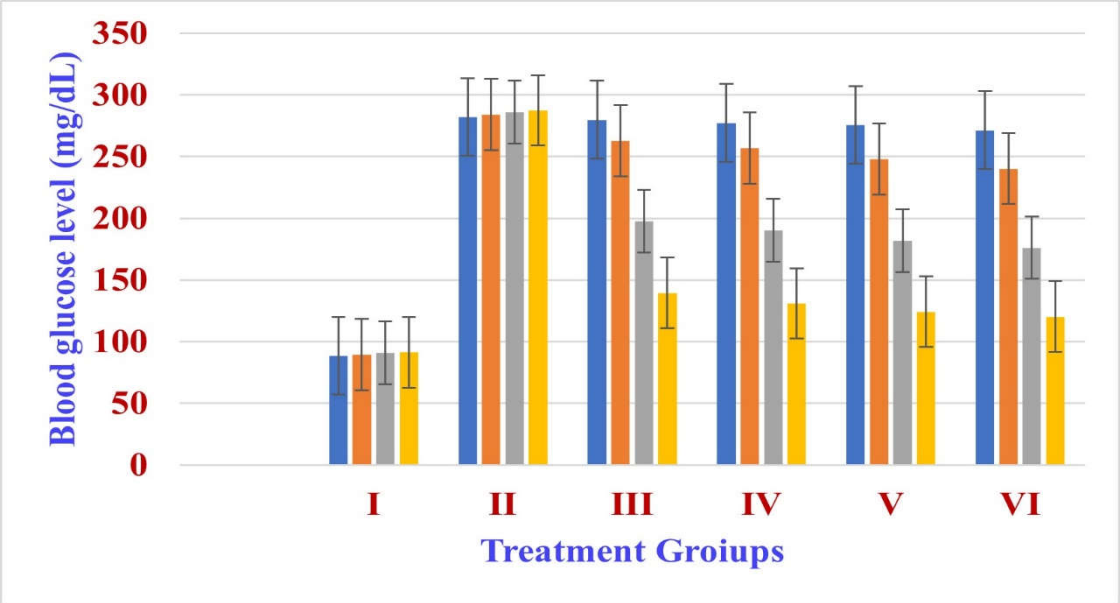

Fig-6: Effect of CE-FCG on blood glucose level (mg/dL) at Day 0, 7, 14, and 21

4.5. Screening of Antihyperlipidemic activity

Table-5: Effects of Test Extracts on lipid profile in rats.

Group	TCH (mg/dL)	TGS (mg/dL)	HDL–C (mg/dL)	LDL–C (mg/dL)	VLDL–C (mg/dL)
I	62.9 ± 1.12	66.5 ± 1.03	21.9 ± 0.87	26.1 ± 0.95	12.5 ± 0.48
II	87.1 ± 1.45	126.1 ± 1.68	24.1 ± 0.91	39.3 ± 1.02	27.1 ± 0.59
III	70.1 ± 1.24*	97.9 ± 1.32*	23.1 ± 0.89ns	31.4 ± 1.01*	17.8 ± 0.53*
IV	74.9 ± 1.38*	99.4 ± 1.44*	23.8 ± 0.93ns	28.9 ± 0.97**	18.8 ± 0.56*
V	73.1 ± 1.30*	98.6 ± 1.29*	23.4 ± 0.90ns	27.5 ± 0.92**	18.1 ± 0.51*
VI	57.9 ± 1.10***	70.2 ± 1.11***	22.9 ± 0.85ns	27.1 ± 0.90**	14.4 ± 0.49*

Notes: Values are expressed as Mean ± SEM, n = 6 per group. TCH: Total Cholesterol, TGS: Triglycerides, HDL–C: High-Density Lipoprotein Cholesterol, LDL–C: Low-Density

Lipoprotein Cholesterol, VLDL–C: Very Low-Density Lipoprotein Cholesterol. Statistical significance compared to Diabetic Control (Group II): *p < 0.05, **p < 0.01, ***p < 0.001*. Group-I: Normal. Group-II: Diabetic Control. Group-III: EE-FCG. Group-IV: MF-FCG. Group-V: CE-FCG. Group-VI: Lovastatin (Treated animals).

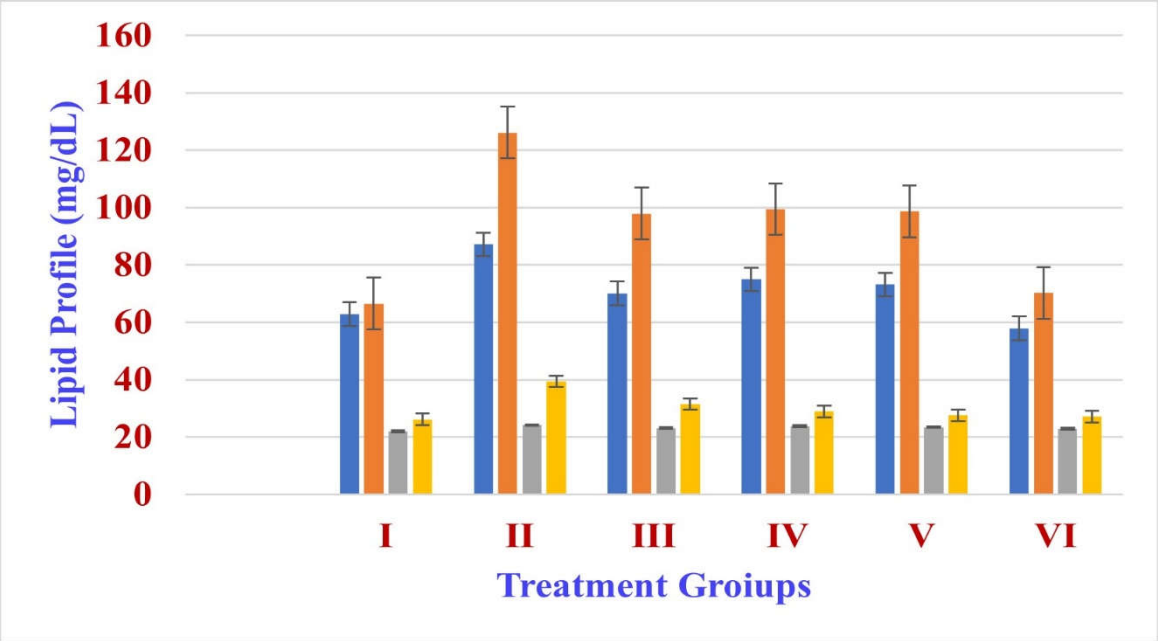

Fig-7: Effects of Test Extracts on lipid profile in rats.

Findings and Summary

In the present study, the antidiabetic activity of ethanolic (EE-FCG), methanolic (ME-FCG), and chloroform (CE-FCG) extracts of *Coccinia grandis* fruits was evaluated in STZ-induced diabetic Wistar rats. A progressive and dose-dependent reduction in fasting blood glucose levels was observed across the treatment groups. Significant reductions (**p < 0.05 to ***p < 0.001) were evident from Day 7 through Day 21 in all extract-treated groups when compared to the diabetic control. Among the extracts, EE-FCG and ME-FCG at 300 mg/kg exhibited near-comparable efficacy to the standard drug, glibenclamide (5 mg/kg), in reducing blood glucose levels by Day 21. These findings indicate that the extracts possess potent antihyperglycemic activity, likely attributed to the presence of bioactive phytoconstituents such as flavonoids, terpenoids, and phenolic compounds.

Additionally, the extracts demonstrated significant antihyperlipidemic effects in diabetic rats. Treatment with EE-FCG, ME-FCG, and CE-FCG notably reduced total cholesterol (TCH), triglycerides (TGS), low-density lipoprotein cholesterol (LDL-C), and very low-density lipoprotein cholesterol (VLDL-C) levels, while moderately improving or maintaining high-density lipoprotein cholesterol (HDL-C). The hypolipidemic efficacy was particularly pronounced in the ME-FCG and CE-FCG groups, comparable to that of the standard drug, lovastatin. The results suggest that *Coccinia grandis* extracts not only aid in glycemic control but also help in the management of diabetes-associated dyslipidemia, making them promising candidates for further pharmacological and clinical investigations.

5. CONCLUSION

The present investigation demonstrates that ethanolic (EE-FCG), methanolic (ME-FCG), and chloroform (CE-FCG) extracts of *Coccinia grandis* fruits possess significant antidiabetic and antihyperlipidemic properties in STZ-induced diabetic rats. Among these, EE-FCG and ME-FCG showed pronounced efficacy in reducing blood glucose and lipid levels, comparable to standard drugs like glibenclamide and lovastatin. The phytochemical richness, particularly of flavonoids, phenolics, and terpenoids, may contribute to these therapeutic effects. All extracts were found to be safe up to 2000 mg/kg in acute toxicity studies. These findings support the traditional use of *Coccinia grandis* in diabetes management and warrant further studies for isolating and characterizing the active constituents.

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