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Evaluation of the antihyperglycemic and hypolipidemic potential of indigenous cow urine powder in streptozotocin-induced diabetic rats

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Abstract

Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia resulting from impaired insulin secretion, insulin action, or both. Growing interest in natural products has encouraged the investigation of traditional remedies as complementary approaches for diabetes management. The present study evaluated the antihyperglycemic and hypolipidemic potential of indigenous cow urine powder in streptozotocin-induced diabetic rats. Thirty-six healthy Wistar albino rats were randomly allocated into six groups ($n = 6$ per group). Experimental diabetes was induced by a single intraperitoneal injection of streptozotocin (50 mg/kg body weight). Following confirmation of diabetes, animals received oral treatment with either gliclazide or indigenous cow urine powder at doses of 100, 200, and 400 mg/day for 15 consecutive days. At the end of the treatment period, fasting blood glucose and serum lipid profile parameters, including total cholesterol, low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides, were determined. Statistical analysis was performed using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test. Streptozotocin administration produced marked hyperglycemia and dyslipidemia, as evidenced by significant increases in fasting blood glucose, total cholesterol, LDL-cholesterol, and triglycerides, together with a significant reduction in HDL-cholesterol ($p < 0.001$). Treatment with indigenous cow urine powder significantly improved all biochemical parameters in a dose-dependent manner. The 400 mg/day dose produced the greatest response, reducing fasting blood glucose from 447.66 ± 12.84 mg/dL in diabetic control rats to 133.50 ± 4.97 mg/dL, while also significantly improving the serum lipid profile. Although gliclazide demonstrated greater overall efficacy, the highest dose of indigenous cow urine powder exhibited substantial antihyperglycemic and hypolipidemic activity. These findings suggest that indigenous cow urine powder has promising potential as a complementary therapeutic candidate for the management of diabetes-associated metabolic disturbances. However, further mechanistic investigations, comprehensive toxicological evaluations, and well-designed clinical studies are required to establish its safety, efficacy, and clinical applicability.

Keywords: Diabetes mellitus, indigenous cow urine powder, streptozotocin, hyperglycemia, dyslipidemia, experimental diabetes, antioxidant activity

Introduction

Diabetes mellitus (DM) is one of the most prevalent endocrine and metabolic disorders worldwide and is a major public health concern. At the time of the writing of this note, IDF estimated that 589 million people were living with diabetes worldwide in 2025 and is expected to be 850 million by 2050. Type 2 diabetes mellitus accounts for about 90-95% of all cases and has been associated with obesity, physical inactivity, unhealthy dietary habits, population aging, and genetic predisposition (IDF, 2025; Zheng *et al.*, 2023) ^[1, 4]. Diabetes is an increasing health issue and has also become a major economic burden for society due to its morbidity and mortality as well as healthcare costs. Diabetes is characterized by impaired insulin secretion, insulin resistance, chronic inflammation, oxidative stress, and disturbances in carbohydrate, lipid, and protein metabolism. The persistent hyperglycemia is associated with microvascular and macrovascular diseases (diabetic nephropathy, retinopathy, neuropathy, and cardiovascular diseases) that can lead to poorer health and death rates and the loss of life and quality of life in patients with diabetes, and the diseases of chronic

inflammation and oxidative stress are reported to be associated with diabetes (Chatterjee *et al.*, 2022; American Diabetes Association, 2025) [3, 2]. Thus, the search for effective treatment to control glycemic control and avoid diabetes-associated metabolic disturbances is an important area of research in diabetes. Experimental diabetes induced by streptozotocin (STZ) is one of the most widely used animal models for evaluating the efficacy of antidiabetic agents. Streptozotocin selectively destroys pancreatic β -cells through DNA alkylation, nitric oxide production, and oxidative stress-mediated mechanisms, resulting in insulin deficiency and sustained hyperglycemia that closely resembles the pathological features of diabetes mellitus (Szkudelski, 2001) [5]. So, the STZ-induced diabetic rat model has been extensively employed in the clinical study of natural and synthetic antidiabetic drugs. Natural products are now being explored as both alternatives or complementary therapies for diabetes treatment by a large number of researchers because of their availability, affordability, and pharmacological advantages. In particular, cow urine and its derivatives are of great significance in traditional Ayurvedic medicine and have antioxidant, antimicrobial, immunomodulatory, hepatoprotective, and bioenhancing properties (Randhawa and Sharma, 2010; Randhawa, 2010) [7, 8]. In this way, cow urine-derived products may improve glucose homeostasis, enhance antioxidant defense, and ameliorate diabetes-induced metabolic abnormalities by reducing oxidative stress and metabolic disturbances. Cow urine-derived products have already been used to treat diabetes-related metabolic disorders in past studies. However, in the absence of evidence on the antidiabetic effects of indigenous cow urine powder, particularly on glucose and lipid metabolism, we have tried to study the antihyperglycemic and hypolipidemic potential of indigenous cow urine powder in streptozotocin-induced diabetic rats, to provide experimental evidence to support its potential as a complementary therapeutic intervention for diabetes management.

Materials and Methods

Ethical Approval

All animal experiments were conducted in accordance with the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India. The experimental protocol was reviewed and approved by the Institutional Animal Ethics Committee (IAEC), registered with CPCSEA (Registration No. 1321/AC/10/CPCSEA). Every effort was made to minimize animal suffering and to use the minimum number of animals necessary to achieve scientifically valid results, in compliance with internationally accepted ethical principles for the care and use of laboratory animals.

Experimental Animals

Thirty-six healthy adult Wistar albino rats of either sex (180-220 g) were obtained from a CPCSEA-registered breeding facility for the experiment. Animals were housed for a week prior to the beginning of the experiment, with standard laboratory conditions at $22 \pm 2^\circ\text{C}$ and 12 h light/12 h dark photoperiod and relative humidity of 50-60%. The animals were fed laboratory pellet food and drinking water ad libitum throughout the experiment.

Preparation of Indigenous Cow Urine Powder

Fresh morning urine was collected from clinically healthy indigenous cows in a standard management system. The

urine was collected in a controlled laboratory environment and dried to obtain powdered material. The dried powder was finely pulverized and stored in sterile airtight containers and kept dry and cool until further use in the experiment.

Induction of Experimental Diabetes

Experimental diabetes was induced with a single intraperitoneal injection of STZ at 50 mg/kg body weight that was freshly prepared in 0.1 M citrate buffer (pH 4.5). A glucometer was used for fasting blood glucose levels after 72 hours. Animals with fasting blood glucose greater than 300 mg/dL were considered diabetic and included in the study.

Experimental Design

After confirmation of diabetes, a group of 6 rats were randomly allocated to six different groups ($n = 6$): (1) Normal control (non-diabetic) (2) Diabetic control (3) Gliclazide (standard antidiabetic drug) (4) Diabetic rats treated with indigenous cow urine powder (100 mg/day) (5) Diabetic rats treated with indigenous cow urine powder (200 mg/day) (6) Diabetic rats treated with indigenous cow urine powder (400 mg/day). All treatments were administered orally once daily for 15 days.

Biochemical Analysis

Blood samples from overnight-fasted animals were collected under anesthesia for biochemical analysis at the end of the treatment period. The serum was separated by centrifugation and was analyzed for fasting blood glucose, total cholesterol, low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, and triglyceride concentrations using commercially available diagnostic kits according to the manufacturers' protocols.

Statistical Analysis

All data are presented as mean $\pm$ standard error of the mean (SEM). Statistical analyses were performed using SPSS software. Differences between experimental groups were assessed by ANOVA and Tukey's multiple comparison post hoc test. A value of $p < 0.05$ was considered statistically significant.

Results

Effect of Indigenous Cow Urine Powder on Glycemic and Lipid Profile

Streptozotocin treatment was effective in inducing diabetes, as fasting blood glucose levels in the diabetic group were significantly increased compared to the normal group (447.66 ± 12.84 vs 91.66 ± 3.12 mg/dL; $p < 0.001$). Oral administration of indigenous cow urine powder was found to reduce fasting blood glucose levels in a dose-dependent manner. The greatest reduction was observed in the treatment group of 400 mg/day (133.50 ± 4.97 mg/dL), while the standard drug gliclazide showed the lowest glucose level (125.25 ± 5.21 mg/dL). One-way ANOVA demonstrated a significant effect on blood glucose ($F = 215.7$, $p < 0.001$).

Experimental diabetes also showed significant dyslipidemia with higher serum total cholesterol, LDL-cholesterol, and triglyceride levels, and lower HDL-cholesterol in comparison with the normal control group ($p < 0.001$). The indigenous cow urine powder significantly improved all lipid profile parameters in a dose-dependent manner. The 400 mg/day treatment group was the most effective

treatment. The variance analysis revealed statistically significant differences in total cholesterol ($F = 38.4$, $p < 0.001$), LDL-cholesterol ($F = 52.6$, $p < 0.001$), HDL-cholesterol ($F = 18.2$, $p < 0.001$), and triglycerides ($F = 41.9$, $p < 0.001$).

In general, indigenous cow urine powder showed a dose-dependent antihyperglycemic and hypolipidemic activity in streptozotocin-induced diabetic rats, with 400 mg/day being the most effective dose among all groups.

Table 1: Effect of Indigenous Cow Urine Powder on Blood Glucose and Lipid Profile in Streptozotocin-Induced Diabetic Rats (Mean $\pm$ SEM)

Parameter	Normal Control	Diabetic Control	Gliclazide	CUP 100 mg	CUP 200 mg	CUP 400 mg
Glucose (mg/dL)	91.66 $\pm$ 3.12	447.66 $\pm$ 12.84 ^a	125.25 $\pm$ 5.21 ^b	174.75 $\pm$ 7.42 ^b	163.25 $\pm$ 6.88 ^b	133.50 $\pm$ 4.97 ^b
Total Cholesterol (mg/dL)	81.65 $\pm$ 2.55	126.37 $\pm$ 4.22 ^a	67.72 $\pm$ 2.18 ^b	95.55 $\pm$ 3.14 ^b	93.70 $\pm$ 2.84 ^b	90.82 $\pm$ 2.56 ^b
LDL-Cholesterol (mg/dL)	28.11 $\pm$ 1.46	86.87 $\pm$ 3.95 ^a	16.17 $\pm$ 0.84 ^b	51.15 $\pm$ 2.27 ^b	47.22 $\pm$ 1.95 ^b	40.85 $\pm$ 1.74 ^b
HDL-Cholesterol (mg/dL)	53.53 $\pm$ 1.94	39.50 $\pm$ 1.62 ^a	51.55 $\pm$ 1.88 ^b	44.40 $\pm$ 1.71 ^b	46.47 $\pm$ 1.52 ^b	49.97 $\pm$ 1.69 ^b
Triglycerides (mg/dL)	77.21 $\pm$ 2.75	188.67 $\pm$ 5.87 ^a	95.35 $\pm$ 3.25 ^b	134.55 $\pm$ 4.52 ^b	132.70 $\pm$ 4.08 ^b	126.47 $\pm$ 3.76 ^b

Values are expressed as mean $\pm$ SEM (n = 6).

^a Significantly different from the normal control group ($p < 0.05$). ^b Significantly different from the diabetic control group ($p < 0.05$).

Discussion

We observed that indigenous cow urine powder has a dose-dependent antihyperglycemic and hypolipidemic effect in STZ-induced diabetic rats. As expected, the STZ therapy induced high levels of hyperglycemia and dyslipidemia from selective destruction of pancreatic β -cells and lack of insulin as was predicted by Szkudelski (Szkudelski, 2001) [5] on the STZ-induced diabetic disease. The indigenous cow urine powder significantly reduced fasting blood glucose levels and lipid profile and the 400 mg/day dose was the most effective. While we do not believe indigenous cow urine powder was better than the test drug as it was still in the range of 300 mg/day, the dose-dependent improvement indicates its biological significance. The results are in line with previous studies showing the antihyperglycemic activity of cow urine-derived products in experimental diabetes (Gururaja *et al.*, 2011) [9]. Diabetes-induced dyslipidemia (diabetes-related high cholesterol, LDL-cholesterol, and triglycerides combined with reduced HDL-cholesterol) is a major risk factor for cardiovascular diseases. The significant increase in lipid profiles achieved in the present study suggests that indigenous cow urine powder could be beneficial in lipid metabolism, possibly by enhancing insulin sensitivity and hepatic lipid synthesis, as is the case for a number of natural antioxidant technologies (Ceriello and Prattichizzo, 2023) [6]. The positive effect of cow urine-derived products may be due to the antioxidant properties of cow urine produced from the cow. Oxidative stress is a key contributing factor to β -cell dysfunction, insulin resistance, and diabetic complications. Cow urine preparations have been shown to possess antioxidant and free radical-scavenging activities that may improve the cellular antioxidant defenses and reduce oxidative damage (Randhawa and Sharma, 2010; Pant *et al.*, 2024) [7, 8, 10]. The exact bioactive ingredients and molecular pathways involved are yet to be elucidated. However, the present study is limited by the absence of insulin estimation, oxidative stress biomarkers, histopathological, and molecular studies. Therefore, further studies that are mechanistic, toxicological, and long-term in nature, and clinical trials should be conducted to prove the safety, mechanism, and therapeutic potential of indigenous cow urine powder. In summary, indigenous cow urine powder has shown antihyperglycemic and hypolipidemic potential, and may be a complementary therapy for diabetes. But its clinical applicability should be established through rigorous preclinical and clinical studies.

Conclusion

The indigenous cow urine powder has a significant dose-dependent antihyperglycemic and hypolipidemic activity in

streptozotocin-induced diabetic rats. The treatment with indigenous cow urine powder significantly improved fasting blood glucose and serum lipid levels, and the 400 mg/day dose showed the highest therapeutic response of the tested drug. Although the standard antidiabetic drug gliclazide had superior safety and efficacy, indigenous cow urine powder might be a useful treatment for diabetes-associated metabolic problems as well. However, further mechanistic investigations, detailed toxicological studies, and clinical trials on it in a randomized setting should be conducted in order to establish its safety, efficacy, and clinical utility before it is promoted for use in humans.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this manuscript.

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