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# Evaluation of Antidiabetic and Antidyslipidemic Effects of Methanolic Extract of *Pentas Schimperiana* Leaf in Mice

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**Abstract**

Diabetes mellitus (DM) poses a significant global health challenge and is on the rise in Ethiopia. Current pharmaceutical treatments for DM are expensive, relatively ineffective, and associated with safety concerns, including difficulties in administration. Pentas schimperiana, a traditional herbal medicine used for treating DM in Ethiopia, offers a potential alternative. This study aimed to assess the antidiabetic and antidyslipidemic properties of Pentas schimperiana to address these challenges. Three doses (250mg/kg, 500mg/kg, and 1000mg/kg) of methanolic leaf extracts of Pentas schimperiana were administered to three groups of mice: a control group with normal glucose levels, a group with glucose-infused mice, and a group with alloxan-induced diabetic mice. Glucose levels, lipid levels, and body weight were measured before and after experimental treatment. We used mixed-design ANOVA for data analysis, with statistical significance at $p \leq 0.05$. After 14 days of treatment, all three doses of methanolic leaf extract of Pentas schimperiana significantly reduced blood glucose levels in diabetic mice compared to normal mice (PS 250mg/kg, $p<0.01$; PS 500mg/kg and 1000mg/kg, $p<0.001$). Additionally, we observed significant body-weight loss and improvement in dyslipidemic profiles in diabetic mice treated with the plant extract. The study suggests that methanolic leaf extract of Pentas schimperiana may serve as a potential treatment for DM in resource-limited settings. We recommended Further molecular studies to elucidate its mechanisms of action and potential clinical applications.

**Keywords:** Diabetes mellitus, Pentas schimperiana, antidiabetic, antidyslipidemic, Alloxan, albino mice

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**Introduction**

Diabetes Mellitus (DM) encompasses a spectrum of metabolic disorders characterized by elevated blood glucose levels arising from impaired insulin secretion, action, or both, posing a significant global health concern and precipitating macro and microvascular complications (18). The burden of DM in Sub-Saharan Africa is increased, where limited access to diagnosis and treatment exacerbates mortality and complications among affected individuals (15). Globally, the prevalence of DM is substantial, with estimates indicating a trajectory towards a further rise, with Ethiopia reporting a significant number of cases (12).

Addressing DM presents a multifaceted challenge for the scientific community, given the limitations and complications associated with current antidiabetic medications, including Sulphonylureas, Metformin, Acarbose, and Thiazolidinedione derivatives (17). Consequently, there is an urgent imperative to explore alternative, safer, and more efficacious therapeutic agents. Herbal extracts represent a promising avenue, with a substantial proportion of the global population relying on them for disease management (20).

Pentas schimperiana, a semi-woody shrub native to Ethiopia and Cameroon, holds promise as a traditional remedy for various ailments, including DM. In Ethiopian folk medicine, it is known as "Woinagrefet" and is utilized in managing epilepsy and DM through oral administration of its root bark powder (22). Phytochemical analysis has revealed the presence of bioactive compounds such as saponins, flavonoids, tannins, steroids, and phenolic compounds in the methanolic leaf extract of P. schimperiana. Moreover, experimental evidence has demonstrated its potential to lower blood glucose levels in alloxan-induced diabetic mice dose-dependently (9).

Historically, many modern antidiabetic medications have been derived from medicinal plants, underscoring the importance of exploring botanical sources for novel therapeutics. Plant-derived secondary metabolites, including flavonoids, alkaloids, and phenolic compounds, possess intrinsic glucose-lowering properties (19, 26, 28, 31, 36, 37). Given the pivotal role of oxidative stress in DM pathogenesis, antioxidants have emerged as a promising avenue for treatment and prevention (2, 21, 36). Previous investigations into P. schimperiana have highlighted its antioxidant activity, prompting further exploration of its potential therapeutic effects in DM (9).

Hence, this study seeks to rigorously evaluate the antidiabetic and antidyslipidemic properties of the methanolic leaf extract of P. schimperiana in alloxan-induced diabetic mice, aiming to contribute to the growing body of knowledge on natural remedies for DM management.

**Methods**

**Drugs, Chemicals, and Instruments**

In this study, we utilized the following drugs, chemicals, and instruments: Alloxan (Sigma Aldrich, Germany); absolute methanol from Nice Chemicals, India; glibenclamide from Julphar Pharmaceuticals, Ethiopia; normal saline from Addis Pharmaceutical Factory, Ethiopia; 40% glucose solution from Addis Pharmaceutical Factory, Ethiopia; PRODIGY® blood glucose meter and strips from Ok Biotech Co., Ltd., Taiwan; and Mindray BS-240 clinical chemistry analyzer from Shenzhen Mindray Bio-Medical Electronics Co., Ltd., China. All other chemicals employed were of analytical grade.

**Collection of Plants and Extraction of Crude Components**

After obtaining approval from the Debre Berhan University Institutional Review Board committee, comprised of Dr. Esubalaew Tesfahun, Dr. Amare Ayalew, and Dr. Gezahegn Degafe, we conducted the collection of fresh leaves of P. schimperiana from Ankober, North Shoa Zone of the Amhara region, Ethiopia, in May 2019. The plant specimen underwent taxonomic identification and authentication before being archived at the Herbarium of Addis Ababa University with voucher number ATA0001. Subsequently, 100 g of dried fresh leaves of the plant material were macerated and placed in stoppered conical flasks containing 500 mL of methanol for five days. After filtration, the filtrate underwent drying under reduced pressure using a rotary evaporator, producing 3 g of crude extract. The weighed crude extract was stored at -4 °C until its intended use.

**Animals for the Experiment**

We utilized healthy Swiss albino mice of both sexes, weighing between 25 and 30 grams, for this study. These mice were procured from the Ethiopian Public Health Institute in Addis Ababa, Ethiopia. Upon arrival, the animals were housed in standard cages and maintained under regulated conditions with a 12-hour light-dark cycle at the Department of Pharmacology, Addis Ababa University. Before the commencement of the experiment, the mice underwent a seven-day acclimatization period, during which they were provided with a standard diet and unrestricted access to water. The study was conducted following the guidelines for ethical care and use of laboratory animals (8).

**Assessment of Acute Oral Toxicity**

Following the guidelines provided by the Organization for Economic Cooperation and Development (OECD) 425, we conducted an acute oral toxicity study of P. schimperiana. Five female albino mice weighing 25 and 30 grams were selected for the study. Initially, a single mouse, after a 4-hour fasting period, received an oral dose of 2000 mg/kg of the leaf extract and was closely monitored for any behavioral or physical changes over the subsequent 24 hours. Subsequently, the remaining four female mice were administered the same dose, and rigorous observation continued for fourteen days to assess any signs of toxicity or mortality. Following this, the dose of the extract was escalated to 5000 mg/kg, and a similar procedure was carried out for an additional set of five female mice.

**Inducing Diabetes**

We induced Diabetes in the albino mice by administering Alloxan (2, 4, 5, 6-tetraoxypyrimidine; 5, 6-dioxyuracil). Before Alloxan administration, the mice underwent a sixteen-hour fasting period. A fresh Alloxan solution was prepared using normal saline (0.9%) and administered intraperitoneally at 150 mg/kg, following established protocols (9, 16, 25). Glucose levels were monitored in the mice 72 hours after Alloxan administration. Mice with fasting blood glucose levels exceeding 200 mg/dl were classified as diabetic mice and included in the study (4, 25, 32). Subsequently, diabetic mice were randomly assigned to their respective experimental groups.

**Experimental Procedures and Designs**

We established various test groups for the study, including normoglycemic, oral glucose loaded, single dose-treated diabetic, and repeated daily dose-treated diabetic groups, each comprising six mice. Within these groups, negative control mice received 10 ml/kg normal saline, while tested mice were administered PS extract at varying doses: 250 mg/kg body weight, 500 mg/kg body weight, and 1000 mg/kg body weight. Additionally, a positive control group was treated with glibenclamide at 5

mg/kg. For the repeated daily dose-treated diabetic models, six groups were formed, including five groups of diabetic mice and one group of normal mice. Diabetic control mice received 10 ml/kg NS, while tested mice were administered PS extract at doses of 250 mg/kg body weight, 500 mg/kg body weight, and 1000 mg/kg body weight. A positive control group received glibenclamide at 5 mg/kg, and a normal control group received 10 ml/kg NS. Plant extract doses were determined based on acute oral toxicity results, and administration occurred via oral gavage after dissolving in normal saline at a volume not exceeding 10 mL/kg body weight per individual animal (1).

**Glucose Measurement**

Blood samples were collected from the tail vein of each animal using an aseptic technique. According to the operating procedures, the blood glucose level was determined using the PRODIGY® blood glucose meter and strips (Ok Biotech Co., Ltd., Taiwan). The test was done in triplicate. For the hypoglycemic test, overnight fasted mice received their respective treatment, and BGL was measured at zero (just before treatment), one, two, four, and six hours after treatment (27, 33). Thirty minutes after each group's respective treatment (7, 33), 40 % w/v glucose solution was given orally to each animal at the dose of 2.5g/kg for glucose tolerance test (33). BGL was measured for each animal just before respective treatments (at zero minutes), and then at 30, 60, and 120 minutes after oral glucose loading (27, 33). For the single-dose test, overnight fasted animals received normal saline, P. schimperiana leaf extract, and glibenclamide according to their corresponding grouping. BGL was measured at zero hours (just before treatment); and at two, four, six, and eight hours after treatment (27, 33). In the antihyperglycemic activity study with a repeated daily dose, mice were treated regularly with normal saline, P. schimperiana leaf extract, and glibenclamide once per day for fourteen days per each grouping. Each overnight fasted (16 hours) animal's BGL was documented just before starting the treatment, on the first day of treatment ( 72 hours after alloxan administration) as a baseline, and on the seventh and fourteenth day of treatment (34).

**Assessment of Changes in Body Weight and Serum Lipid Levels**

The body weight of each animal was documented before treatment initiation, on the first day following 72 hours post-alloxan administration, and on the seventh and fourteenth days of respective treatments. On the 15th day, mice that had fasted overnight were euthanized with intraperitoneal (IP) administration of sodium pentobarbitone at a dose of 150mg/kg (34, 38). Subsequently, blood specimens were collected via cardiac puncture using a sterile gel tube. The blood samples were allowed to stand at room temperature for two hours before being centrifuged. The serum lipid profile, including high-density lipoprotein, total cholesterol, and triglycerides, was determined using the Mindray BS-240 clinical chemistry analyzer (Shenzhen Mindray Bio-Medical Electronics Co., Ltd, China).

**Data Analysis**

The data were recorded, summarized, and analyzed utilizing SPSS version 21 software (USA). Mean and standard deviation (SD) calculations were performed, and the variation between and within groups was assessed using mixed-design ANOVA. A p-value of $\leq 0.05$ was deemed indicative of statistical significance.

**Results**

**Acute Oral Toxicity**

The methanolic leaf extract of Pentas schimperiana (PS) demonstrated no mortality in mice at doses of 2000mg/kg and 5000mg/kg, with none exhibiting

signs of toxicity (including behavioral, neurological, or autonomic) within the initial 24 hours and throughout the 14-day study period. The LD50 value for the P. schimperiana leaf extract was higher than 5000mg/kg.

**Hypoglycemic Impact of P. schimperiana in Normoglycemic Mice**

Baseline fasting blood glucose levels did not differ significantly across all groups (Table 1). Among the three doses tested (PS 250 mg/kg, PS 500 mg/kg, and PS 1000 mg/kg), none demonstrated a statistically significant reduction in blood glucose levels compared to the negative control group. However, glibenclamide (5 mg/kg) notably decreased blood glucose levels at the 1st ($P<0.05$), 2nd ($P<0.01$), 4th, and 6th ($P<0.001$) hours compared to the negative control group.

Furthermore, glucose loading significantly reduced blood glucose levels at the 1st ($P<0.05$), 2nd, 4th, and 6th hours ($P<0.001$) compared to the PS 250 mg/kg-treated group; at the 2nd, 4th, and 6th hours ($P<0.001$) compared to the PS 500 mg/kg-treated group; and at the 1st ($P<0.05$), 2nd, 4th, and 6th hours ($P<0.001$) compared to the PS 1000 mg/kg-treated group.

No statistically significant difference in blood glucose levels was observed at all time points when comparing the treated groups with P. schimperiana leaf extract. At all time points, none of the three doses of the plant extract and normal saline significantly decreased blood glucose levels compared to the corresponding baseline level. However, glibenclamide significantly reduced blood glucose levels at the 1st ($P<0.01$), 2nd, 4th, and 6th ($P<0.001$) hours compared to the initial level.

**Table 1. Hypoglycemic Potential of Methanolic Leaf Extract from P. Schimperiana in Normoglycemic Mice.**

| | **Fasting Blood Glucose Level (mg/dl)** | | | | |
|---|---|---|---|---|---|
| Group | 0 hr | 1 hr | 2 hr | 4 hr | 6 hr |
| NS 10ml/kg | 94.16 ± 5.11 | 95.16 ± 4.07 | 88.66 ± 5.78 | 98.33 ± 4.92 | 89.33 ± 6.34 |
| PS 250mg/kg | 91.66 ± 3.32 | 95.66 ± 6.43 | 90.16 ± 6.30 | 99.66 ± 5.78 | 90.83 ± 6.4 |
| PS 500mg/kg | 90.83 ± 5.56 | 93.50 ± 3.39 | 92.16 ± 5.38 | 93.66 ± 5.78 | 92 ± 6.06 |
| PS 1000mg/kg | 94.16 ± 5.98 | 95.83 ± 6.14 | 92.33 ± 6.34 | 97.33 ± 3.82 | 90.33 ± 6.91 |
| GLC 5mg/kg | 94.83 ± 3.76 | 86.16 ± 3.31 $^{a*\ b*\ d*\ \beta\phi}$ | 73.83 ± 4.11 $^{a\phi\ b\psi\ c\psi\ d\psi\ \beta\psi}$ | 72.66 ± 3.38 $^{a\psi\ b\psi\ c\psi\ d\psi\ \beta\psi}$ | 62.16 ± 3.71 $^{a\psi\ b\psi c\psi\ d\psi\ \beta\psi}$ |

*Values are expressed as mean ± SD; n=6; [a] compared to the negative control, [b] compared to PS 250mg/kg, [c] compared to PS 500mg/kg, [d] compared to PS 1000mg/kg, and [β] compared to baseline blood glucose level; [*] P<0.05, [ϕ] P<0.01, [ψ] P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide.*

**The Antihyperglycemic Potential of P. schimperiana in Mice Orally Loaded with Glucose**

The baseline blood glucose levels showed no statistical differences among the groups (Table 2). PS doses of 1000mg/kg (P<0.001) and 500mg/kg (P<0.01) significantly reduced hyperglycemia in the 1st hour compared to the negative control. Glibenclamide (5 mg/kg) notably decreased hyperglycemia at the 1st (P<0.001) and 2nd (P<0.01) hours after glucose administration compared to the vehicle-treated group. No significant difference in blood glucose levels was observed among the three doses of the plant extract. Glibenclamide (5mg/kg) also significantly reduced hyperglycemia at the 1st and 2nd hours (P<0.001) compared to all three P. schimperiana leaf extract doses.

Thirty minutes after oral glucose administration, a statistically significant (P<0.001) increase in blood glucose levels was observed in all groups compared to baseline fasting levels, regardless of the treatment administered. Additionally, significant hyperglycemia was evident at 1 hour after oral glucose administration in the negative control and all P. schimperiana leaf extract-treated groups compared to their initial blood glucose levels. Blood glucose levels at the 1st and 2nd hours significantly decreased (P<0.001) compared to 30 minutes after oral glucose administration across all groups.

**Table 2. Effect of Methanolic Leaf Extract from P. Schimperiana on Oral Glucose Tolerance in Healthy Mice.**

| | **Fasting Blood glucose level (mg/kg)** | | | |
|---|---|---|---|---|
| Group | 0 min | 30 min | 60 min | 120 min |
| NS 10ml/kg | 89.83 ± 3.81 | 211.66 ± 10.80 $^{\beta\phi}$ | 165.66 ± 10.15 $^{\pi\phi}$ | 101.16 ± 3.65 $^{\pi\phi}$ |
| PS 250mg/kg | 90.33± 3.93 | 209 ± 12.88 $^{\beta\phi}$ | 151 ± 7.64 $^{\pi\phi}$ | 103.83 ± 4.26 $^{\pi\phi}$ |
| PS 500mg/kg | 88.33 ± 4.76 | 213 ± 7.53 $^{\beta\phi}$ | 146.66 ± 11.60 $^{\pi\phi a*}$ | 111.33 ± 14.06 $^{\pi\phi}$ |
| PS 1000mg/kg | 88 ± 5.54 | 208.16 ± 9.68 $^{\beta\phi}$ | 137 ± 6.69 $^{\pi\phi a\phi}$ | 103.50 ± 5.20 $^{\pi\phi}$ |
| GLC 5mg/kg | 88.50 ± 4.32 | 211.33 ± 14.71 $^{\beta\phi}$ | 90.33 ± 4.76 $^{\pi\phi a\phi b\phi c\phi d\phi}$ | 80.33 ± 4.67 $^{\pi\phi a* b\phi c\phi d\phi}$ |

*Values are expressed as mean ± SD; n=6; time refers to the time after oral glucose administration; [a] compared to the negative control, [b] compared to PS 250mg/kg, [c] compared to PS 500mg/kg, [d] compared to PS 1000mg/kg, and [β] compared to baseline blood glucose level, and [π] compared to the blood glucose level at 30 minutes; [*] P<0.01, [ϕ] P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide.*

**Antihyperglycemic Potential of a Single Dose of P. schimperiana Methanolic Leaf Extract in Diabetic Mice**

There were no significant reductions in blood glucose levels at the 2nd hour of treatment across all three doses of PS compared to the negative control group (Table 3). However, glibenclamide (5mg/kg) significantly decreased blood glucose levels at the 4th and 6th hours (P<0.05) and at the 8th hour (P<0.001) compared to the negative control. Mice treated with glibenclamide showed significantly decreased glucose levels at the 4th hour (P<0.05), at the 6th hour, and at the 8th hour (P<0.01) compared to those treated with PS extract at doses of 250mg/kg, 500mg/kg, and 1000mg/kg, respectively. Glibenclamide (5mg/kg) treated diabetic mice exhibited significantly reduced blood glucose levels at the 4th, 6th, and 8th hours compared to baseline levels.

**Table 3. Antihyperglycemic Effect of a Single Dose of P. Schimperiana Methanolic Leaf Extract in Diabetic Mice.**

| | Fasting Blood glucose level (mg) | | | | |
|---|---|---|---|---|---|
| Group | 0 hr | 2 hr | 4 hr | 6 hr | 8 hr |
| NS 10ml/kg | 282.83 ± 29.73 | 292.5 ± 61.94 | 297 ± 44.45 | 275.5 ± 45.91 | 292.66 ± 42.87 |
| PS 250mg/kg | 290.16 ± 50.52 | 283.16 ± 50.52 | 293 ± 50.35 | 282.66 ± 50.49 | 265 ± 39.02 |
| PS 500mg/kg | 290 ± 39.61 | 277.33 ± 44.06 | 287.16 ± 44.27 | 276.5 ± 43.83 | 275.16 ± 43.20 |
| PS 1000mg/kg | 276.5 ± 48.94 | 267.16 ± 49.41 | 276.66 ± 50.16 | 267.5 ± 48.93 | 266.5 ± 48.93 |
| GLC 5mg/kg | 303.66 ± 50.10 | 295.83 ± 49.05 | 204.5 ± 38.83 $^{\beta\psi}$ a* b* c* | 184.83 ± 34.68 $^{\beta\psi}$ a* b$^{\phi}$ c* d* | 172 ± 30.89 $^{\beta\psi}$ a$^{\psi}$ b$^{\phi}$ c$^{\phi}$ d$^{\phi}$ |

*Values are expressed as mean ± SD; n=6; [a] compared to the negative control, [b] compared to PS 250mg/kg, [c] compared to PS 500mg/kg, [d] compared to PS 1000mg/kg, and [β] compared to baseline blood glucose level; [*] P<0.05, [ϕ] P<0.01, [ψ] P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide.*

**Antihyperglycemic Effect of Daily Repeated Doses of P. Schimperiana Methanolic Leaf Extract in Diabetic Mice**

As shown in Table 4, there were no significant differences in baseline blood glucose levels among all diabetic mice groups. However, the blood glucose levels in the diabetic groups were significantly higher than those in the control group at all time intervals (P<0.001). On the 7th day of treatment, PS 500mg/kg and PS 1000mg/kg significantly reduced blood glucose levels (P<0.05) compared to the diabetic control. All doses of the extract, PS 250mg/kg, PS 500mg/kg, and PS 1000mg/kg, significantly decreased blood glucose levels on the 14th day of treatment compared to the diabetic control (P<0.01, P<0.001, and P<0.001, respectively). A significant reduction in blood glucose levels was observed in the GLC-treated group on the 7th (P<0.01) and 14th (P<0.001) days of treatment compared to the diabetic control. However, there was no significant difference in blood glucose levels between the GLC-treated and plant extract-treated groups at all time points. Blood glucose levels in the plant extract-treated groups showed no statistically significant difference at all doses and time intervals. Both the GLC (5mg/kg) and P. schimperiana leaf extract-

treated groups significantly reduced blood glucose levels (P<0.001) on both the 7th and 14th days of treatment compared to their respective baseline levels.

**Table 4. Antihyperglycemic Effect of Daily Repeated Administration of P. schimperiana Methanolic Leaf Extract in Diabetic Mice.**

| | **Fasting Blood Glucose Level (mg/dl)** | | |
|---|---|---|---|
| Group | Baseline | 7th day | 14th day |
| Diabetic control | 282.83 ± 29.73 $^{n\,\psi}$ | 254.16 ± 28.88 | 246.83 ± 31.99 |
| PS 250mg/kg | 290.16 ± 50.52 $^{n\,\psi}$ | 219.83 ± 42.33 $^{\beta\,\psi}$ | 192.16 ± 13.01 $^{\beta\,\psi\,\alpha\,\phi}$ |
| PS 500mg/kg | 290 ± 39.61 $^{n\,\psi}$ | 199.66 ± 17.46 $^{\beta\,\psi\,\alpha^{*}}$ | 187.66 ± 13.79 $^{\beta\,\psi\,\alpha\,\psi}$ |
| PS 1000mg/kg | 276.5 ± 48.94 $^{n\,\psi}$ | 187.16 ± 9.34 $^{\beta\,\psi\,\alpha^{*}}$ | 179.33 ± 6.91 $^{\beta\,\psi\,\alpha\,\psi}$ |
| GLC 5mg/kg | 303.66 ± 50.10 $^{n\,\psi}$ | 185.16 ± 21.35 $^{\beta\,\psi\,\alpha\,\phi}$ | 160.5 ± 32.53 $^{\beta\,\psi\,\alpha\,\psi}$ |
| Normal control | 100.66 ± 5.71 | 81 ± 4.33 | 86.5 ± 5.35 |

*Values are expressed as mean ± SD; n=6; $^{\alpha}$ compared to diabetic control, $^{n}$ compared to normal control, $^{\beta}$ compared to baseline blood glucose level; $^{*}$ P<0.05, $^{\phi}$ P<0.01, $^{\psi}$ P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide.*

**Impact of Daily Repeated Administration of P. schimperiana Methanolic Leaf Extract on Body Weight in Diabetic Mice**

Alloxan-treated mice exhibited significant weight loss in the diabetic control group on the 7th and 14th days compared to the normal control (P<0.001) (Table 5). Administration of the plant extract at a dose of 1000mg/kg resulted in significant weight gain on the 7th day of treatment compared to the control group (P<0.05). However, 250mg/kg and 500mg/kg doses did not yield significant weight gain on the 7th day of treatment. All three doses of the plant extract showed considerable weight improvements on the 14th day of treatment compared to the diabetic control. At doses of 250mg/kg and 500mg/kg, the plant extract significantly reduced weight compared to the normal control group on the 7th and 14th days of treatment. Conversely, the weight of the glibenclamide-treated group significantly improved (P<0.001) on both the 7th and 14th days of treatment compared to the diabetic control group.

**Table 5. Effect of Daily Repeated Administration of P. Schimperiana Methanolic Leaf Extract on Body Weight in Diabetic Mice**

| Group | Body weight (gm.) | | | |
|---|---|---|---|---|
| | Before induction of Diabetes | Baseline | 7th day of treatment | 14th day of treatment |
| Diabetic control | 30.33 ± 4.17 | 25.33 ± 4.03 | 25.66 ± 3.26 $^{n\,\psi}$ | 21 ± 28.2 $^{\beta\,\phi\;\;n\,\psi}$ |
| PS 250mg/kg | 29 ± 3.22 | 24.33 ± 1.86 | 29 ± 1.26 $^{n*}$ | 25.66 ± 2.42 $^{n\,\psi\;\;\alpha*}$ |
| PS 500mg/kg | 31 ± 1.52 | 25.33 ± 3.03 | 27.66 ± 2.42 $^{n\,\phi}$ | 28.33 ± 2.5 $^{n*\;\alpha\,\psi}$ |
| PS 1000mg/kg | 29 ± 3.22 | 24.33 ± 2.84 | 30.33 ± 2.5 $^{\alpha*}$ | 28.66 ± 2.06 $^{\alpha\,\psi}$ |
| GLC 5mg/kg | 31 ± 3.52 | 25.33 ± 2.03 | 33.66 ± 1.86 $^{\alpha\,\psi}$ | 32.83 ± 1.47 $^{\alpha\,\psi}$ |
| Normal control | 29 ± 2.22 | 27.66 ± 2.16 | 34 ± 1.89 | 32.83 ± 2.13 |

*Values are expressed as mean ± SD; n=6; $^{\alpha}$ compared to diabetic control, $^{n}$ compared to normal control, $^{\beta}$ compared to baseline blood glucose level; $^{*}$ P<0.05, $^{\phi}$ P<0.01, $^{\psi}$ P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide*

**Impact of Daily Repeated Administration of P. schimperiana Methanolic Leaf Extract on Serum Lipid Levels in Diabetic Mice**

Total cholesterol (TC) and triglycerides (TG) were significantly elevated (P<0.001) in the diabetic control compared to the standard control (Table 6). Conversely, high-density lipoprotein (HDL) cholesterol was significantly reduced (P<0.001) in the diabetic control group compared to the normal control. After 14 days of administration, PS doses of 250mg/kg and 500mg/kg (P<0.05), as well as 1000mg/kg, demonstrated significantly increased HDL levels compared to the diabetic control (P<0.001). All doses of PS significantly reduced serum total cholesterol levels (P<0.001). At doses of 500mg/kg (P<0.05) and 1000mg/kg (P<0.01), PS extract decreased serum triglyceride levels compared to the diabetic control. GLC (5mg/kg) significantly reduced (P<0.001) serum total cholesterol and triglyceride levels while increasing (P<0.001) HDL cholesterol compared to the diabetic control. There were no statistically significant differences in the serum levels of TC, TG, and HDL-C when comparing each plant extract-treated group to each other. However, the GLC-treated group exhibited substantial (P<0.01) variations in HDL-C, TG, and TC serum levels compared to all doses of plant extract groups.

**Discussion**

This research suggests that the extract from the leaves of Pentas schimperiana might help treat diabetes mellitus. The extract was found to be non-toxic in acute oral toxicity tests, with an LD50 of over 5000 mg/kg, and it also significantly reduced blood glucose levels in diabetic albino mice. Additionally, it increased protective HDL cholesterol levels and decreased undesirable lipid profiles. The extract from P. schimperiana leaves appeared to support weight gain as well.

Diabetes mellitus (DM) is a metabolic disorder that results from impaired carbohydrate, lipid, and protein metabolism. It can be caused by either insufficient insulin secretion or insulin action. The global scientific community is continuously seeking safe and effective antidiabetic drugs. Studies on new medications derived from traditional medicinal plants, which are more accessible and do not require complex pharmaceutical processes, remain a vital area of research. The LD50 of the methanolic leaf extract

of P. schimperiana exceeds 5000 mg/kg, as previously reported in another study (9).

**Table 6. Effect of Daily Repeated Administration of P. Schimperiana Methanolic Leaf Extract on Serum Lipid Levels in Diabetic Mice**

| | **Serum Lipid Level (mg/dl)** | | |
|---|---|---|---|
| Group | HDL-C | TC | TG |
| Diabetic control | 26.66 ± 2.16 $^{n\psi}$ | 184.33 ± 3.32 $^{n\psi}$ | 149.83 ± 7.16 $^{n\psi}$ |
| PS 250mg/kg | 32.5 ± 1.87 $^{n\psi\alpha*}$ | 160.66 ± 2.16 $^{n\psi\ \alpha\psi}$ | 142.16 ± 4.44 $^{n\psi}$ |
| PS 500mg/kg | 32.5 ± 1.87 $^{n\psi\alpha*}$ | 164.5 ± 13.09 $^{n\psi\alpha\psi}$ | 140.66 ± 3.5 $^{n\psi\alpha*}$ |
| PS 1000mg/kg | 35.83 ± 2.48 $^{n\psi\alpha\psi}$ | 160 ± 4.04 $^{n\psi\ \alpha\psi}$ | 137.16 ± 2.85 $^{n\psi\alpha\phi}$ |
| GLC 5mg/kg | 43.66 ± 5.08 $^{\alpha\psi\ b\phi\ c\phi\ d\phi}$ | 90 ± 2.09 $^{\alpha\psi\ b\phi\ c\phi\ d\phi}$ | 64.5 ± 4.54 $^{\alpha\psi\ b\phi\ c\phi\ d\phi}$ |
| Normal control | 45.16 ± 3.18 | 77.33 ± 3.72 | 60.33 ± 3.72 |

*Values are expressed as mean ± SD; n=6; $^{\alpha}$ compared to diabetic control, $^{b}$ compared to PS 250mg/kg, $^{c}$ compared to PS 500mg/kg, $^{d}$ compared to PS 1000mg/kg, and $^{n}$ compared to normal control; $^{*}$ P<0.05, $^{\phi}$ P<0.01, $^{\psi}$ P<0.001; PS, Pentas schimperiana leaf extract; NS, normal saline; GLC, glibenclamide; TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein.*

The research study employed Alloxan, a compound often used to induce Diabetes in rodents (10, 35), to investigate its effects on mice. The results showed that administering 150 mg/kg of Alloxan resulted in sustained hyperglycemia in the mice, which aligns with previous studies (9). Furthermore, the study found that Alloxan caused hyperglycemia with minimal fluctuations in blood glucose levels over two weeks, similar to the diabetic control mice (Table 4). The mechanisms underlying pancreatic β cell dysfunction due to Alloxan involve DNA fragmentation and damage and the generation of superoxide radicals and reactive oxygen species (35).

The study revealed no statistically significant differences in baseline blood glucose levels (BGL) across all groups of experimental animals. However, a substantial decrease in BGL was observed in all groups after treatment with the respective dose of methanolic leaf extract and GLC (5mg/kg), indicating the changes in BGL. The methanolic leaf extract of P. schimperiana did not show significant hypoglycemic activity at all dose levels in normoglycemic mice (Table 1). In contrast, the extract at doses of 1000mg/kg and 500mg/kg revealed significant antihyperglycemic activity after a single dose of the plant extract in oral glucose-loaded mice. Moreover, repeated doses of the plant extract showed substantial antihyperglycemic activity and body weight increment in diabetic mice (Table 4, and Table 5). The extract at a dose of 1000mg/kg showed a relatively higher reduction in BGL, indicating that the antihyperglycemic activity of the plant extract was dose-dependent.

Many medicinal plants' presence of phytochemicals, such as alkaloids, phenolic compounds, flavonoids, and terpenoids, is believed to contribute to their antidiabetic activity (19, 26, 28, 31, 36, 37). According to recent reports, flavonoids exhibit β cell regenerating and insulinogenic effects (19, 26), which may explain the blood-glucose-lowering effect of P. schimperiana's methanolic leaf extract.

The exact mechanism by which P. schimperiana exerts its antidiabetic effects remains unclear, but it is thought to facilitate insulin secretion and enhance glucose uptake in peripheral tissues (9, 30). Further molecular studies are necessary to confirm the mechanism underlying the observed antihyperglycemic effects. The study confirmed that alloxan-induced Diabetes is associated with significant weight loss in experimental animals, and the P. schimperiana methanolic leaf extract-treated mice in this study experienced a reversal of this weight loss (Table 5). Type 2 diabetes is characterized by increased glycogenolysis, lipolysis, gluconeogenesis, and muscle wasting (11, 13), leading to tissue protein loss. The significant weight gain observed in the P. schimperiana methanolic leaf extract-treated mice suggests that the plant material may have antihyperglycemic effects.

Dyslipidemia is a severe complication commonly associated with Diabetes, characterized by low levels of HDL-C, high serum TG and TC (6, 11). This condition results from increased lipolysis and VLDL secretion due to hormone-sensitive lipase stimulation (5, 6). Additionally, insulin shortage caused by beta-cell dysfunction leads to reduced activity of lipoprotein lipase, which in turn decreases VLDL and chylomicron clearance (14). Elevated triglyceride levels can also stimulate the enzymatic activity of cholesteryl ester transfer protein, resulting in increased triglyceride content in both HDL and LDL; this can lead to the hydrolysis of triglyceride-enriched LDL particles, which can be broken down by lipoprotein lipase or hepatic lipase, forming dense LDL particles. However, HDL particles enriched with triglycerides are more prone to breaking down, which can disrupt the cascade (6). The study found that vehicle-treated diabetic control mice significantly increased serum TG and TC and substantially decreased HDL-C levels (Table 6). Treatment with P. schimperiana methanolic leaf extract for two weeks significantly reduced the serum TG and TC and increased HDL-C levels dose-dependently. However, the molecular mechanism underlying the leaf extract's antidyslipidemic activity is unclear, and whether it works by controlling hyperglycemia or directly affecting lipid metabolism is unknown.

**Conclusion**

The study results indicate that the methanolic leaf extract of P. schimperiana exhibits significant antihyperglycemic and antidyslipidemic activity in alloxan-induced mice. Additionally, the plant extract improves oral glucose tolerance and body weight. These findings support the traditional use of the plant for treating Diabetes mellitus. We recommended further research to explore the mechanisms of action of the plant's ingredients.

**Abbreviations and Acronyms**

ANOVA stands for Analysis of Variance; BGL for Blood Glucose Level; DM for Diabetes Mellitus; GLC for Glibenclamide; HDL for High-Density Lipoprotein; HDL-C for High-Density Lipoprotein Cholesterol; IDF for International Diabetes Federation; IP for Intraperitoneal; LD50 for Median Lethal Dose; NS for Normal Saline; OECD for Organization for Economic Cooperation and Development; PS for Pentas Schimperiana; SD for Standard Deviation; SPSS for Statistical Software Package for Social Science; TC for Total Cholesterol; TG for Triglyceride; VLDL for Very Low-Density Lipoprotein.

**Declarations**

**Availability of Data**

Complete raw data is available and can be accessed upon reasonable request.

**Ethical Approval**

The Institutional Review Board of Debre Berhan University granted the experiment's ethical

approval under protocol number P004. The study followed the Guideline for the Care and Use of Laboratory Animals [38].

**Competing Interests**

The authors have declared that there is no conflict of interest.

**Authors' Contributions**

The authors have made significant contributions to the concept and design, data collection, and the analysis and interpretation of data; participated in drafting the article or critically revising it for critical intellectual content; concurred on the journal for submission; provided final approval of the version to be published; and are prepared to take responsibility for all aspects of the work.

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**Consent for Publication**

Not applicable

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