

In vivo evaluation of the antihyperglycemic potential of *Deverra scoparia* (Coss. & Durieu), a north African endemic species

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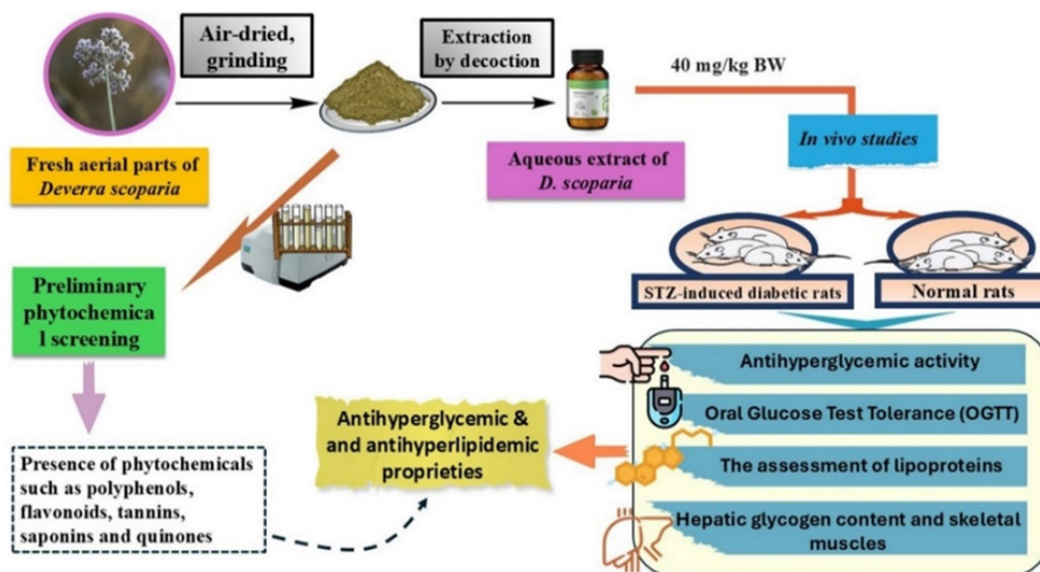


ABSTRACT

Deverra scoparia (*D. scoparia*) Coss. & Durieu is an endemic plant from North Africa that is commonly utilized in folk medicine for diabetes treatment. This study aimed to evaluate the in vivo antidiabetic effects of *D. scoparia* in both normal and STZ-induced diabetic rats. The investigation focused on the impact of an aqueous extract of *D. scoparia* administered at a dosage of 40 mg/kg on glycemia and lipid profiles in these rats. Additionally, the study included assessments of glycogen content in the liver and skeletal muscles (*EDL* and soleus), as well as a phytochemical analysis. Both single and repeated oral doses of the aqueous extract (40 mg/kg) resulted in a significant decrease in blood glucose, total cholesterol, and triglyceride levels in diabetic rats. Moreover, this extract improved glucose tolerance and enhanced hepatic glycogen content in the diabetic subjects. Notably, the plant exhibited a rich profile of certain phytochemicals, particularly phenolic acids and flavonoids. The findings of this study clearly indicate that the aqueous extract of *D. scoparia* possesses substantial antidiabetic activity.

Keywords: *Diabetes mellitus*, *hyperglycemia*, *D. scoparia*, *aqueous extract*.

GRAPHICAL ABSTRACT



INTRODUCTION

Diabetes mellitus is a condition where the pancreatic function is impaired, resulting in inadequate insulin secretion, or the body becomes unable to process the insulin it does create effectively [1]. Diabetes mellitus represents a major public health and economic challenge globally. While over 400 million people were living with the diagnosis in 2017, that number is probable to surge to 693 million by 2045 [2]. Poorly managed diabetes can result in severe complications, including nephropathy, retinopathy, neuropathy, and cardiovascular diseases [3]. While current management involves lifestyle changes alongside synthetic drugs and insulin, these pharmaceuticals often cause significant side effects like heart failure and osteoporosis [4]. This has driven a search for safer alternatives, specifically medicinal herbs which offer glucose regulation with fewer risks [5]. To date, over 1,200 plants have shown antidiabetic potential [6]. Among these is *Deverra scoparia* (locally known as "Guezzah" or "Tattacht"), a North African Apiaceae species used traditionally to treat various ailments, including diabetes [7-10]. As no animal-model studies currently exist regarding its efficacy, this study aims to evaluate the in vivo antidiabetic effects of *D. scoparia* in both healthy and streptozotocin-induced diabetic rats.

EXPERIMENTAL

Plant material: The aerial components of *D. scoparia* were sourced from Errachidia, Morocco (a semi-arid zone), during September 2020. The samples were subsequently air-dried at 45 °C. Taxonomic classification was confirmed, and the specimen was assigned voucher number DS 09/20 for storage at the Faculty of Sciences and Techniques herbarium.

Preparation of the aqueous extract: Following traditional Moroccan medicinal practices, an aqueous decoction was prepared by boiling 1 g of powdered *D. scoparia* aerial parts in 100 mL of distilled water for 10 minutes. Once cooled, the mixture was passed through a 0.2 µm Millipore filter (St Quentin en Yvelines, France) to remove all particulate matter [11]. The resulting extract was administered at a dosage of 40 mg/kg of body weight.

Preliminary phytochemical screening: A comprehensive preliminary phytochemical analysis of the *D. scoparia* aerial aqueous extract was performed to identify key bioactive constituents. Using established standard protocols [12], the analysis evaluated the presence of secondary metabolites, specifically polyphenols, flavonoids, tannins, saponins, quinones, sterols, terpenoids, and alkaloids.

Determination of total polyphenol, flavonoid and tannin contents: The total phenolic contents in *D. scoparia* aqueous extract were measured as it has been previously described [13]. The total phenolic compounds were quantified in micrograms of Gallic acid equivalent (GAE) per milliliter of sample, utilizing

the calibration curve equation; (where y represents absorbance and x denotes concentration in GAE µg/mL). The total flavonoid content was assessed using the method established by Kim *et al.*, [14]. Quantification of the total flavonoid content was performed in micrograms of Rutin equivalent (RE) per milliliter of sample, utilizing the calibration curve equation; (where Y represents absorbance and X denotes concentration, RE in µg/mL). The total tannin components were quantified using the method established by Broadhurst *et al.*, [15]. The total tannin constituents were quantified in micrograms of catechin equivalent (CE) per milliliter of sample, utilizing the calibration curve equation; (where y represents absorbance and x denotes concentration, CE in µg/mL).

Experimental animals: Adult male Wistar albino rats, with a body weight range of 120-220 g, were utilized for this study. The rats were housed individually in polyethylene cages under controlled laboratory conditions, with unrestricted access to food and water (*ad libitum*). Experimental protocols were approved by approved by the Institutional Animal Ethics Committee of the Faculty of Sciences and Techniques, Morocco (FST FSTE/2015), and conducted in accordance with international animal care and use guidelines.

Induction of diabetes: Following an overnight fast, Rats received intraperitoneal injections of streptozotocin (STZ; Sigma, St. Louis, MO) at 65 mg/kg in 0.1 M citrate buffer (pH 4.5). Controls were given distilled water. After three days, diabetic status was verified by plasma glucose >200 mg/dL [16].

Measurement of glycaemia: Both normal and STZ-diabetic rats were randomized into three groups (n = 5 per group). The control group received distilled water, while experimental groups were treated with either *D. scoparia* extract (40 mg/kg) or the reference drug, glibenclamide (5 mg/kg). For the acute study, glycemic levels were monitored for six hours following a single oral dose. In the sub-chronic study, treatments were administered daily via oral gavage for seven days, during which body weight and blood glucose were regularly recorded. For the Oral Glucose Tolerance Test (OGTT), rats were fasted for 12 hours before receiving a 2 g/kg glucose load. Blood glucose was then measured at 30, 60, 90, and 120 minutes post-administration. Total glycemic responses were quantified using the Area Under the Curve (AUC). All blood samples were collected via tail-tip puncture and analyzed using the glucose oxidase method with a Contour™ TS glucometer (Bayer Diabetes Care).

Effect on glycogen content: Upon completion of the experimental period, the liver, soleus, and extensor digitorum longus (EDL) muscles were excised to determine their glycogen content. Glycogen levels in these tissues were quantified using the direct anthrone reagent method described by Morris *et al.*, [1]. Final concentrations were calculated following the specific formula established by Carroll *et al.*, [18].

Determination of lipid profile: On the seventh day of the experiment, the rats were sedated, and blood samples were obtained via retro-orbital sinus puncture using a syringe for lipid profile analysis. Plasma total cholesterol (TC) and triglycerides (TGs) were evaluated using commercial kits (SGM Italic) [19].

Statistical analysis: All values are reported as mean \pm S.E.M. Group comparisons were performed using two-way ANOVA followed by Bonferroni's post hoc test. Statistical analyses were conducted with GraphPad Prism 7, with $p < 0.05$ regarded as statistically significant. Glycogen content in liver, EDL, and Soleus muscles was analyzed using unpaired t-tests at a 95% confidence interval.

RESULTS AND DISCUSSION

Preliminary phytochemical screening of *D. scoparia*: Preliminary phytochemical analysis of the dried powder from the aerial parts of *D. scoparia* indicated that polyphenols, flavonoids, tannins, saponins, quinones, sterols, terpenoids, and alkaloids were detected (Table 1).

Table 1. Phytochemical screening concerning the aerial part of *D. scoparia*.

| Detected phytochemical group | Test | Results of the test |
|------------------------------|------------------------|---------------------|
| Polyphenols | Ferris chlorid test | (+) |
| Flavonoids | Shinoda test | (+) |
| Tannins | Ferric chlorid test | (+) |
| Saponins | Frothing test | (+) |
| Quinones | Sulfuric acid test | (+) |
| Sterols and terpenoids | Libermann_Buchard test | (+) |
| | Wagner's test | (-) |
| | Dragendorff's test | (-) |
| | Mayer's test | (-) |

Note: (+) presence, (-) absence

Quantification of total phenolic compounds, total flavonoid, and total tannins: The total phenolic constituents in the extract were quantified as 228.570 ± 7.150 mg of Gallic acid equivalent per gram of the extract. Flavonoids were determined to be comparable to 53.320 ± 1.710 mg of Rutin per gram of the extract. Tannins were quantified as 27.570 ± 1.33 mg of catechin equivalent per gram of the extract. The calibration curve utilized for quantifying the total phenolic, total flavonoid, and total tannin levels is illustrated in Fig. S1.

Antidiabetic activity: Single oral administration: The acute effects of *D. scoparia* aqueous extract on blood glucose levels are depicted in Fig. 1. While a 40 mg/kg dose had no measurable impact on the glycemia of healthy rats over a 6-hour period, it produced a significant antihyperglycemic effect in STZ-induced diabetic rats.

Specifically, blood glucose levels were significantly reduced as early as one hour post-administration ($p < 0.05$), with further significant decreases observed at 2, 4, and 6 hours post-treatment ($p < 0.0001$). A notable decrease in glycaemia was noticed in glibenclamide-treated normal animals after the first hour of treatment ($p < 0.05$), with this reduction becoming more pronounced at the end of six hours ($p < 0.0001$). Glibenclamide also resulted in significant decreases in glycemia in diabetic rats from the second hour to the final hour of therapy ($p < 0.0001$). The glycaemia of untreated diabetic and normal rats remained constant throughout six hours of therapy.

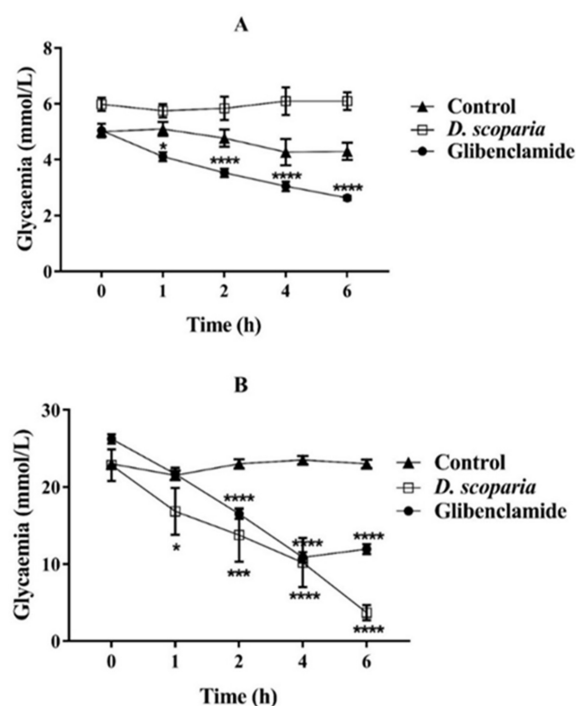


Fig. 1. Glycaemia over 6 h after a single oral administration of the aqueous extract of *D.scoparia* (40 mg/kg) in normal (A) and diabetic rats (B). Data are expressed as means \pm S.E.M., $n = 5$ rats per group. * $p < 0.05$, *** $p < 0.001$ and **** $p < 0.0001$ when compared to baseline values.

Repeated oral administration: The effects of daily oral administration over a seven-day period are presented in Fig. 2. In healthy rats, *D. scoparia* (40 mg/kg) caused no significant glycemic changes. In contrast, the diabetic cohort treated with the extract displayed a significant reduction in glycemia by the second day, a trend that remained highly significant through days four and seven ($p < 0.0001$). Meanwhile, glibenclamide induced a moderate hypoglycemic response in normal rats by day four ($p < 0.01$), which intensified by day seven ($p < 0.0001$). In the diabetic group, glibenclamide treatment resulted in a consistently significant decrease in glucose levels from day two until the study's conclusion ($p < 0.0001$).

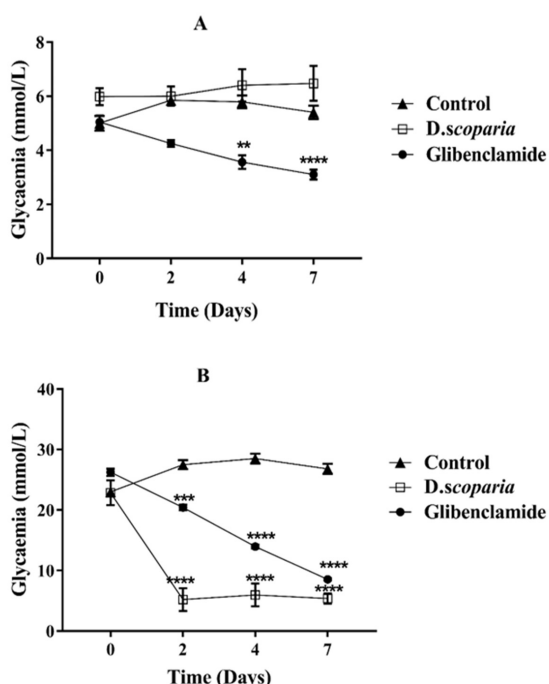


Fig. 2. Glycaemia over daily repeated oral administration of the aqueous extract of *D. scoparia* (40 mg/kg) for 7 days in normal (A) and diabetic rats (B). Data are expressed as means \pm S.E.M., n=5 rats per group. **p<0.01, ***p<0.001 and ****p<0.0001 when compared to baseline values.

Effect on fasting body weight: The impact of a seven-day treatment regimen on body weight is presented in Fig. 3. Daily administration of *D. scoparia*

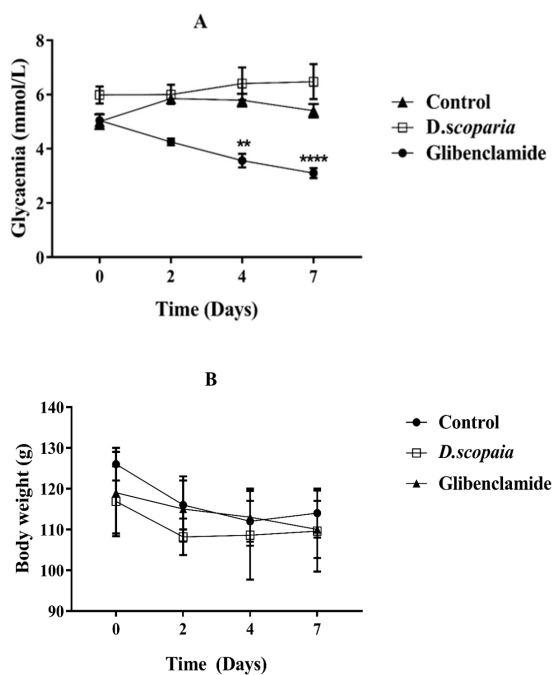


Fig. 3. Body weight variation after repeated oral administration of *D. scoparia* aqueous extract (40 mg/kg) for 7 days in normal (A) and diabetic rats (B). Data are expressed as mean \pm S.E.M. n=5.

aqueous extract (40 mg/kg) resulted in no significant weight fluctuations in either the healthy or STZ-diabetic cohorts. Similarly, no significant changes in body weight were recorded for the groups receiving glibenclamide.

Oral glucose test tolerance: The OGTT results for both normal and diabetic rats are presented in Figs. 4 and 5. While baseline glycemia was uniform across all groups, *D. scoparia* (40 mg/kg) did not significantly alter blood glucose levels in normal rats over the 120-minute period. In contrast, glibenclamide (5 mg/kg) induced a significant reduction in healthy rats starting at 60 minutes (p<0.05), becoming more pronounced by the end of the test (p<0.01; Fig. 4A). In STZ-diabetic rats, neither the extract nor glibenclamide significantly shifted blood glucose concentrations in the direct time-course monitoring (Fig. 5A). However, the AUC analysis revealed distinct trends. In healthy rats, only glibenclamide significantly lowered the AUC (p<0.01), whereas *D. scoparia* had no effect (Fig. 4B). Conversely, in the diabetic cohort, both the aqueous extract and glibenclamide successfully reduced the total glycaemic response (AUC) compared to control animals (p<0.05 and p<0.01, respectively; Fig. 5B)

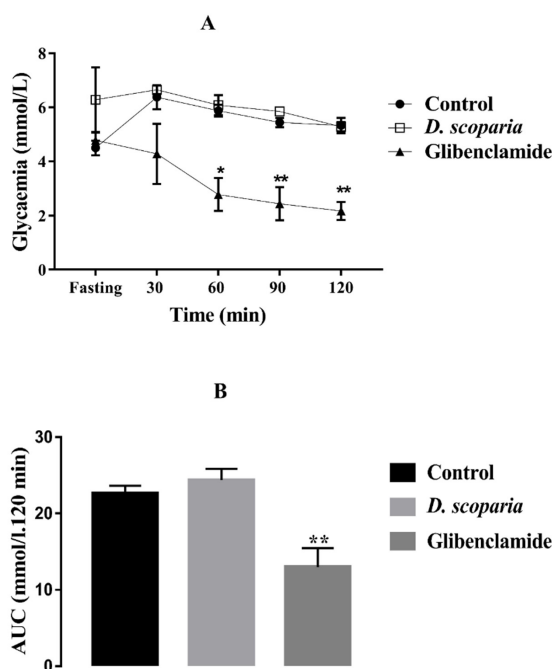


Fig. 4. Effect of *D. scoparia* aqueous extract (40 mg/kg) on oral glucose tolerance test in normal rats. (A): Line curves depict the changes in blood glucose response after 0, 30, 60, 90 and 120 min following administration of glucose (2 g/kg b.w.). Values are expressed as means \pm S.E.M; n=5. **p<0.05; ***p<0.01 and ****p<0.001 when compared to baseline values. (B): Area under the curve of the OGTT to compare treated groups with the control, **p<0.01.

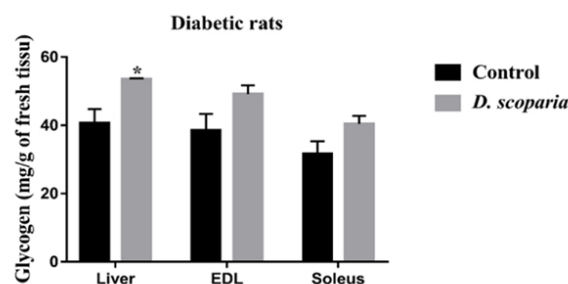
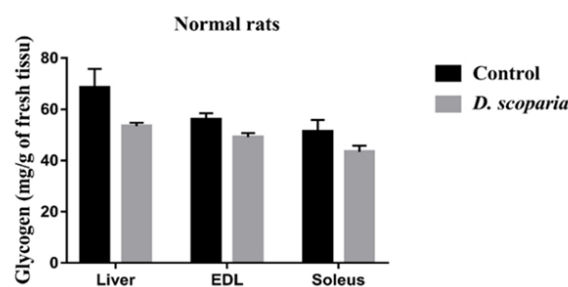
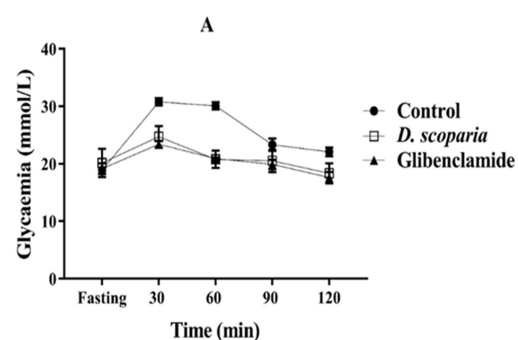


Fig. 5. Effect of *D. scoparia* aqueous extract (40 mg/kg) on oral glucose tolerance test in diabetic rats. (A): Line curves depict the changes in blood glucose response after 0, 30, 60, 90 and 120 min following administration of glucose (2 g/kg b.w.). Values are expressed as means \pm S.E.M; n=5. (B): Area under the curve of the OGTT to compare treated groups with the control. *p<0.05 and **p<0.01.

Fig.6. Effect of one week administration with the *D. scoparia* aqueous extract (40 mg/kg) on liver, EDL, and soleus glycogen content in normal diabetic rats. *p<0.05 when compared with control.

Effect on glycogen content: The effects of *D. scoparia* on tissue-specific glycogen storage are presented in Fig. 6. As expected, the STZ-diabetic rats showed a marked reduction in glycogen levels within the liver and skeletal muscles (soleus and EDL) compared to healthy controls. Treatment with the *D. scoparia* aqueous extract (40 mg/kg) significantly restored liver glycogen levels in the diabetic cohort (p<0.05).

However, no statistically significant changes were detected in the glycogen content of the EDL or soleus muscles following treatment. Furthermore, the extract did not alter glycogen storage in the liver or muscles of normal rats.

Antihyperlipidemic activity: The effects of repeated *D. scoparia* aqueous extract administration on plasma lipid profiles are summarized in Table 2. In the diabetic cohort, treatment with the extract (40 mg/kg) led to a significant reduction in both total cholesterol (TC) and triglycerides (TGs) (p<0.05 and p<0.01, respectively), though no effects were reported in healthy rats.

Table 2. Plasma total cholesterol, triglycerides and HDL-c levels over 7 days after repeated oral administration of *D. scoparia* aqueous extract (40 mg/kg) in normal and diabetic rats

| Experimental groups | Total cholesterol (TC) | | Triglycerides (TGs) | | HDL-c | | |
|---------------------|-------------------------------|-------------------|---------------------|---------------------|---------------------|-------------------|-----------------------|
| | 0d | 7d | 0d | 7d | 0d | 7d | |
| Normal groups | Control (distilled water) | 2.740 \pm 0.043 | 2.578 \pm 0.067 | 1.357 \pm 0.064 | 1.389 \pm 0.027 | 0.833 \pm 0.031 | 0.899 \pm 0.022 |
| | <i>D. scoparia</i> (40 mg/kg) | 1.342 \pm 0.249 | 1.225 \pm 0.140 | 1.021 \pm 0.228 | 1.154 \pm 0.124 | 0.430 \pm 0.141 | 0.416 \pm 0.147 |
| | Glibenclamide (5 mg/kg) | 2.390 \pm 0.075 | 2.280 \pm 0.095 | 1.376 \pm 0.048 | 0.870 \pm 0.050** | 1.024 \pm 0.049 | 1.331 \pm 0.091** |
| Diabetic groups | Control (distilled water) | 3.604 \pm 0.120 | 3.162 \pm 0.037 | 1.775 \pm 0.200 | 1.320 \pm 0.170 | 0.952 \pm 0.026 | 0.864 \pm 0.024 |
| | <i>D. scoparia</i> (40 mg/kg) | 2.290 \pm 0.132 | 1.321 \pm 0.149* | 2.540 \pm 0.347** | 1.472 \pm 0.24 | 0.351 \pm 0.059 | 0.316 \pm 0.039 |
| | Glibenclamide (5 mg/kg) | 3.440 \pm 0.493 | 2.518 \pm 0.062* | 1.508 \pm 0.134 | 1.823 \pm 0.050 | 0.823 \pm 0.050 | 1.508 \pm 0.134**** |

Note: Data are expressed as means \pm S.E.M., n=5 rats per group. *p<0.05; **p<0.01 and ****p<0.0001 when compared to baseline values.

Glibenclamide (5 mg/kg) also exhibited lipid-lowering effects, significantly reducing TGs in normal rats ($p < 0.01$) and moderately lowering TC in diabetic rats ($p < 0.05$). Notably, the control groups receiving distilled water showed no alterations in lipid markers throughout the study.

Due to their perceived safety, efficacy, and high patient-acceptability, herbal therapies are increasingly integrated into diabetes management strategies. This investigation focused on evaluating the antidiabetic potential of the *D. scoparia* aqueous extract in both healthy and streptozotocin-induced diabetic rat models. Streptozotocin (STZ) remains a gold-standard screening tool for assessing the therapeutic properties of medicinal plants in experimental diabetes research [22]. Our experimental results confirm that streptozotocin induction successfully produced a severe diabetic state characterized by hyperglycemia, glucose intolerance, and metabolic dysfunction (muscle proteolysis and adipose tissue lipolysis). The therapeutic efficacy of glibenclamide in reversing these parameters validates the model's integrity. Notably, a single dose of *D. scoparia* aqueous extract (40 mg/kg) achieved a highly significant reduction in fasting glycemia in diabetic rats ($p < 0.0001$). Prolonged administration further established its potent antihyperglycemic properties. Given that STZ creates an insulin-deficient state, these effects may stem from stimulated insulin secretion from remaining β -cells.

The seven-day treatment period highlighted the extract's safety profile: blood glucose levels in healthy rats remained stable, demonstrating a lack of hypoglycemic risk under normoglycemic conditions. In contrast, diabetic rats showed a progressive, highly significant decline in glycemia. Intriguingly, at the tested doses, *D. scoparia* exhibited a more potent glucose-lowering effect than the standard drug glibenclamide, particularly on days 2, 4, and 7. While this suggests superior potency, further molecular assays are required to elucidate the specific pathways involved. Finally, although the OGTT remains a sensitive diagnostic tool, 120-minute post-load measurements may not fully capture the complexity of glucose processing [21]. The glucose AUC provides a more comprehensive assessment of glucose tolerance than single-point measurements, as it captures the total glycemic excursion following a glucose challenge [21, 22]. In this study, the AUC was significantly lower in diabetic rats treated with *D. scoparia* A.E. (40 mg/kg) compared to untreated diabetic controls ($p < 0.05$). This improved glucose tolerance likely results from enhanced insulin availability, optimized insulin function, or a reduction in intestinal glucose absorption [23]. Glycogen serves as the primary storage form of glucose in the liver and skeletal muscles, with its synthesis acting as a direct indicator of insulin signaling activity [24]. Consequently, insulin deficiency in diabetes typically leads to a significant depletion of hepatic glycogen stores [25].

Our findings confirmed this reduction in untreated diabetic rats across the liver, EDL, and soleus muscles. However, administration of *D. scoparia* aqueous extract significantly restored hepatic glycogen levels ($p < 0.05$). This suggests that the extract may counteract diabetic glycogen depletion by stimulating glycogenesis or inhibiting glycogenolysis. Interestingly, the lack of significant glycogen recovery in the EDL and soleus muscles might be attributed to the specific dosage used or the relatively short duration of the study. Beyond carbohydrate metabolism, the insulin deficiency inherent in diabetes also disrupts lipid profiles promoting the accumulation of total cholesterol and triglycerides which elevates the risk of cardiovascular complications [26, 27]. The experimental data demonstrated that *D. scoparia* aqueous extract (40 mg/kg) significantly lowered total cholesterol ($p < 0.05$) and triglycerides ($p < 0.001$) in diabetic rats. This antihyperlipidemic activity likely stems from improved glycemic control, enhanced insulin secretion, and a reduction in the mobilization of fatty acids from adipose tissue. To date, research on the bioactive profile of *D. scoparia* remains limited; however, our phytochemical screening identified a diverse array of secondary metabolites, including polyphenols, flavonoids, tannins, saponins, alkaloids, and terpenoids. Quantitative analysis confirmed substantial levels of phenolic compounds. These findings align with previous literature suggesting that natural phenols, tannins, and saponins mitigate diabetes by stimulating pancreatic insulin release, inhibiting hepatic glucose output, and regulating lipid metabolism [28-31].

CONCLUSIONS

The present investigation demonstrates that the aqueous extract of *D. scoparia* possesses significant *in vivo* antidiabetic efficacy. The observed antihyperglycemic and antihyperlipidemic effects are likely mediated by improved glucose tolerance, enhanced hepatic glucose uptake, and the inhibition of peripheral lipid mobilization. Our results provide compelling evidence validating the traditional use of *D. scoparia* in diabetes treatment.

AUTHOR CONTRIBUTIONS

AA: designed the research, analyzed the data, wrote the manuscript draft; MA: performed the experiments and analyzed the data; ME: supervised and revised the manuscript. All authors approved the final version of the manuscript.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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