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Hypoglycemic and Antihyperglycemic Effect of Stem Bark Extracts of *Parkia biglobosa* (Jacq) Benth

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Abstract

Hyperglycemia is defined as an excess of glucose in the blood. It is a clinical indicator of diabetes and happens when the body has too little insulin or when the body can't use insulin properly. Diabetes is constantly on rise, despite prevention efforts, and is a real public health problem. This study aims to evaluate the effect of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* on blood glucose. Phytochemical screening of these extracts was first carried out. Then, the effect of both extracts was tested in normoglycemic rats. Hyperglycemia was induced to normoglycemic rats by oral administration of glucose (4 g/kg), and rats were after treated with *Parkia biglobosa* extracts. Phytochemical screening revealed the presence of polyphenols, flavonoids, tannins, saponosides, alkaloids, sterols and polyterpenes in aqueous and hydroethanolic stem bark extracts of this plant. Results also showed that both extracts significantly reduce blood glucose 2 h after administration to normoglycemic rats. Moreover, these extracts were found to significantly reduce blood glucose 1 h after administration to hyperglycemic rats, with percentage reductions of 58.01; 60 and 60.27 % respectively in rats given aqueous extract (100 mg/kg), hydroethanolic extract (100 mg/kg) and glibenclamide (10 mg/kg), 1h 30 min after treatment.

Key words: *Parkia biglobosa*, diabetes, blood glucose, hypoglycemia, anti-hyperglycemia.

Introduction

Diabetes is a chronic disease that occurs when the pancreas does not produce enough insulin, or when the body is unable to effectively use the insulin it does produce. Insulin is, of course, a hormone that regulates the concentration of sugar in the blood. The consequence of insulin deficiency, or the body's inability to use this hormone effectively, is a sustained rise in blood glucose levels known as hyperglycemia. Hyperglycemia is a common effect of uncontrolled

diabetes that, over time, causes serious damage to many of the body's systems, particularly nerves and blood vessels (WHO, 2023).

Diabetes is constantly on the rise, despite prevention efforts, and constitutes a genuine public health problem. The WHO considers it one of the world's leading killers. According to the International Diabetes Federation, in 2022, 540 million people were living with diabetes (IDF, 2022), and this number is projected to reach 643 million by 2030, and 783 million by 2045 (IDF Diabetes Atlas, 2021).

Management of diabetes is currently based on hygienic-dietary rules (strict diets, regular physical activity) and pharmacological treatments. Treatments include insulin injections and many oral antidiabetic drugs such as biguanides and sulfonylureas (Holman and Turner, 1991). However, the high cost of these drugs puts them beyond the reach of many people. Moreover, treatment with sulfonylureas and biguanids is also associated with side effects and fail to significantly alter the course of diabetic complications (Bailey, 2008). In this situation, people are increasingly turning to traditional medicine for treatment.

Research on medicinal plants has gained considerable importance in recent decades and demand for the use of natural products in the treatment of diabetes is increasing worldwide. In Côte d'Ivoire, several plants used in traditional medicine to treat diabetes have been identified. Such is the case of *Parkia biglobosa* (Mimosaceae). In order to validate the traditional use of this plant as an antidiabetic, the present study was undertaken. Its aim was to evaluate the effect of *Parkia Biglobosa* stem bark extracts on blood glucose levels in normoglycemic and hyperglycemic rats.

Materials and methods

Plant material

The plant material consists of stem barks of *Parkia biglobosa* collected in the Gontougo region in the North-East of Côte d'Ivoire. Samples of this plant were transferred to the National Floristic Center of the Félix Houphouët-Boigny University of Abidjan where they were identified by comparison with authentic specimens registered under the number UCJ003173.

Animals

Wistar rats aged 10 to 12 weeks and weighing between 120 and 160 g were used in this study. These rats were bred at the animal house of the Normal Superior School of Abidjan. They were fed daily with pellets and had free access to water. All the animals were housed in plastic cages and kept at ambient temperature with 12 h of light during the day and 12 h of darkness in the

night. All experimental procedures have been examined and approved by the Ethical Committee of Health Sciences, Félix Houphouët-Boigny University of Abidjan.

Chemicals

Glibenclamide tablets (Daonil ® 5 mg; Sanofi-Aventis, France), a sulphonylurea hypoglycemic agent, was used as a reference antihyperglycemic. Pure anhydrous glucose (Cooper, France) was used to induce hyperglycaemia in rats. All other chemicals used were of analytical grade.

Extract preparation

Stem barks of *Parkia biglobosa* were shade at room temperature for 3 weeks. The dried samples were later grounded to a powder using a grinder. Aqueous extract was prepared by homogenizing 100 g of the powder in 1 L of distilled water using a blender. After several homogenization cycles, the homogenates were successively filtered, twice on cotton and once on Whatman filter paper (3 mm). The filtrate was concentrated to dryness under reduced pressure at 30°C using a rotary evaporator (BÜCHI). The resultant extract constitutes the aqueous extracts (Zirihi *et al.*, 2003). Hydroethanolic extract was prepared following the same process using a mixture of ethanol and water (70:30, V/V).

Phytochemical screening

The Phytochemical screening was carried out to highlight the presence of certain secondary metabolites in the plant extracts. The method used is based on staining reactions in test tubes between specific reagents and the corresponding chemical families present in the extracts (Wagner and Blatt, 2001). The chemical groups researched were sterols, polyterpenes, flavonoids, tannins, quinone compounds, saponosides and alkaloids.

Acute toxicity

The acute toxicity of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* was performed according to Organization for Economic Cooperation and Development (OECD) guideline n° 423 (OECD, 2001). Two groups of three rats, fasted overnight, were each orally administered a single dose of 2000 mg/kg bw of stem bark aqueous or hydroethanolic extract of *Parkia biglobosa*. Another group of three rats received distilled water. After the treatment, food was withheld for further 3-4 hours. Animals were observed individually at least once during the first 30 min after treatment, regularly during the first 24 hours (with special attention during the first 4 hours), and daily thereafter for a period of 14 days in order to record clinical signs of toxicity.

Assessment of hypoglycemic activity

Thirty (30) rats were divided into six groups of five rats each and fasted for 14 h prior to experimentation. Blood sample was taken before treatment to determine initial blood glucose levels. Rats were then administered with the different products. Group 1 received distilled water (3 mL/kg b.w) orally. Group 2 received glibenclamide (10 mg/kg b.w.) orally. Groups 3 and 4 were orally administered with aqueous extract of *P. biglobosa* at doses of 100 and 200 mg/kg b.w. Finally, Groups 5 and 6 were orally administered with hydroethanolic extract of *P. biglobosa* at doses of 100 and 200 mg/kg b.w. After treatment, blood samples were taken every hour for 4 hours, and blood glucose levels were determined using an On Call Plus glucometer on whole blood taken from the tail of each animal.

Glucose tolerance test

Thirty-five (35) rats, divided into seven groups of five rats each, were fasted for 14 h overnight. During the experiment, an initial blood sample was taken before induction of hyperglycemia to determine the initial blood glucose level. Hyperglycemia was then induced in rats from groups 2, 3, 4, 5, 6 and 7 by oral administration of 4 g/kg bw glucose. Group 1 received distilled water. Sixty (60) min after glucose administration, blood glucose levels were again determined in all rats, which were immediately treated as follows:

- Groups 1 and 2 received distilled water (3 mL/kg b.w) orally;
- Group 3 received 10 mg/kg glibenclamide orally;
- Group 4 and 5 were orally administered with *P. biglobosa* aqueous extract at doses of 100 and 200 mg/kg b.w;
- Groups 6 and 7 were orally administered with hydroethanolic extract of *P. biglobosa* at doses of 100 and 200 mg/kg.

After treatment, blood samples were taken every 30 min for 120 min to determine blood glucose levels. Blood glucose levels were measured using an On Call Plus glucometer on whole blood taken from the tail of each animal.

Statistical analysis

Graph Pad InStat Prism 7.0 software (Microsoft U.S.A) was used for statistical analysis of the data. Results were expressed as mean with standard error on mean (MEAN \pm SEM). One-way analysis of variance (ANOVA ONE WAY) followed by Dunett's multiple comparison test, was used to determine differences between means. The difference was significant for a p-value of less than 5% ($P < 0.05$).

Results

Phytochemical screening

Phytochemical analysis revealed the presence of polyphenols, flavonoids, saponosides, alkaloids, sterols and polyterpenes in aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa*. Quinonic compounds are present in the hydroethanolic extract, but absent in the aqueous one. The same applies to gallic tannins, which were found in the hydroethanolic extract, but not in the aqueous extract. Catechic tannins, on the other hand, were present only in the aqueous extract (Table 1).

Table 1: Phytochemical compounds of stem barks extracts of *Parkia biglobosa*.

Chemical groups		Aqueous extract	Hydroethanolic extract
Sterols and polyterpenes		+	+
Polyphenols		+	+
Flavonoids		+	+
Tannins	Gallic	-	+
	Catechic	+	-
Quinones		-	+
Alkaloids		+	+
Saponosides		+	+

(+): Presence of chemical group; (-): Absence of chemical group.

Acute toxicity

Administration of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* at a single dose of 2000 mg/kg b.w. to rats produced a change in mobility, accelerated respiration and drowsiness after 30 min. The animals were weakened 1 h after administration of both extracts. However, no mortality was observed. Three (3) hours later, the rats returned to their normal behavior and fed properly. No mortality was observed after two weeks. The lethal dose 50 (LD₅₀) is therefore estimated to be greater than 2000 mg/kg b.w.

Effects of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* on the glycemia of normoglycemic rats

Figure 1 shows the time-course effect of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* and glibenclamide on blood glucose levels in normoglycemic rats. Blood glucose levels remained constant, from 0.854 ± 0.014 g/L at baseline (T0) to 0.858 ± 0.008 g/L at the end of the experiment (T240) in rats treated with distilled water. In contrast, in rats treated with *Parkia biglobosa* extracts and glibenclamide, a decrease in blood glucose levels from baseline was recorded during experimentation. This decrease was significant ($p < 0.05$) 2 h after the rats had received the different doses of *Parkia biglobosa* extracts and glibenclamide. At the end of the experiment, i.e. 4 h after treatment, the initial blood glucose level of 0.848 ± 0.01 g/L fell to 0.56 ± 0.05 g/L in rats treated with aqueous extract of *Parkia biglobosa* at a dose of 200 mg/kg bw, and to 0.55 ± 0.058 g/L in those treated with hydroethanolic extract at the same dose, i.e. a reduction of 33.01 and 33.57% respectively. In rats treated with extracts at a dose of 100 mg/kg bw, the reduction in blood glucose levels was highly significant. With the 100 mg/kg bw aqueous extract, blood glucose levels in treated rats fell from 0.83 ± 0.007 g/L to 0.496 ± 0.045 g/L, 4 h after administration, i.e. a reduction of 40.24%. Meanwhile, in rats treated with the hydroethanolic extract at 100 mg/kg bw, blood glucose levels fell from 0.85 ± 0.008 g/L to 0.498 ± 0.034 g/L, 4 h post-treatment, i.e. a reduction of 41.41%. The effect of both extracts at 100 mg/kg bw was comparable to that of glibenclamide at 10 mg/kg bw, which reduced blood glucose levels by 40.23%. The variation in blood glucose levels recorded in treated rats during the experiment is summarized in Table 2.

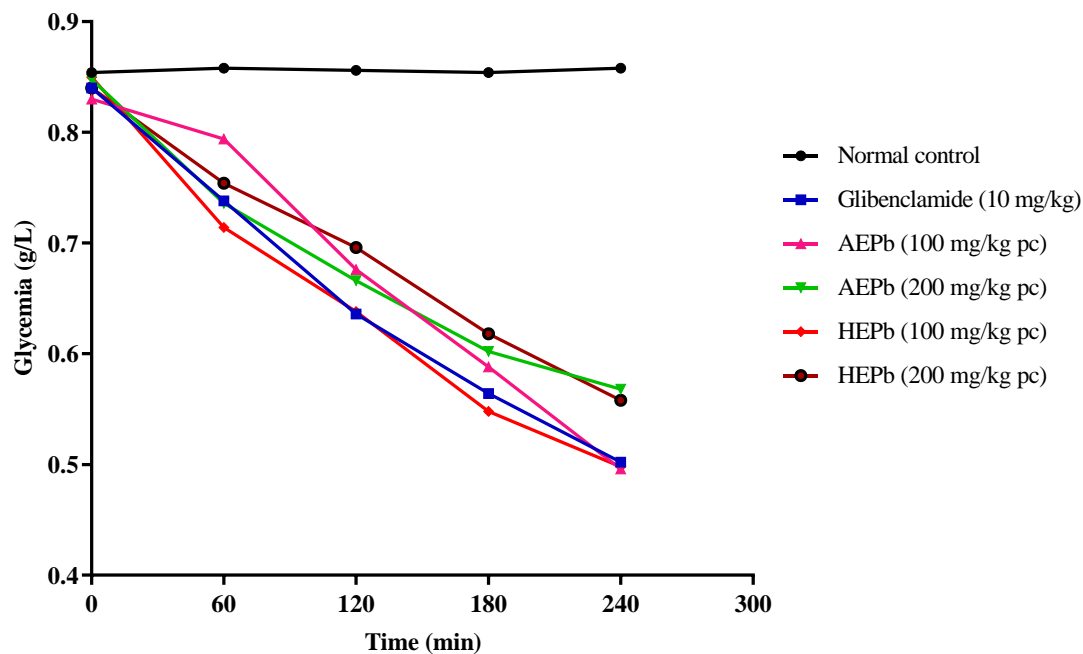


Figure 1: Time course of blood glucose levels in normoglycemic rats treated with stem bark extracts of *Parkia biglobosa* and glibenclamide

AEPb: aqueous extract of *P. biglobosa*; HEPb: ethanolic extract of *P. biglobosa*

Table 2: Changes in blood glucose levels in normoglycemic rats treated with stem bark extracts of *Parkia biglobosa* and glibenclamide

Treated rat groups	Change in blood glucose (%)				
	T0	T60	T120	T180	T240
Group 1 (distilled water)	0	+0.46	+0.27	0	+0.46
Group 2 (Glibenclamide 10 mg/kg)	0	-12.14	-24.28	-32.85	-40.23
Group 3 (AEPb 100 mg/kg)	0	-4.33	-18.55	-29.15	-40.24
Group 4 (AEPb 200 mg/kg)	0	-13.2	-21.46	-29	-33.01
Group 5 (HEPb 100 mg/kg)	0	-16	-24.94	-35.52	-41.41
Group 6 (HEPb 200 mg/kg)	0	-10.23	-17.14	-26.42	-33.57

(+): increase in blood glucose levels; (-): decrease in blood glucose level; AEPb: aqueous extract of *P. biglobosa*; HEPb: ethanolic extract of *P. biglobosa*; T0: time before administration of *P. biglobosa* extracts and glibenclamide. T60, T90, T120, T150: time (min) after administration of *P. biglobosa* extracts and glibenclamide.

Effects of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* on the glycemia of hyperglycemic rats

The time-course effect of aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* and glibenclamide on blood glucose levels in hyperglycemic rats is shown in Figure. 2. The mean basal blood glucose of the rats was 0.85 ± 0.003 g/L before glucose administration. One hour after oral administration of glucose at a dose of 4 g/kg bw, blood glucose levels increased very significantly ($p < 0.001$) in all rats, from 0.85 ± 0.003 to values ranging from 1.23 ± 0.03 to 1.46 ± 0.08 g/L. Treatment of the rats following the onset of hyperglycemia resulted in a rapid drop in blood glucose levels, after 30 min (T90), in rats given aqueous extract (100 and 200 mg/kg bw), hydroethanolic extract (100 mg/kg bw) and glibenclamide (10 mg/kg bw). But, this reduction was only significant ($p < 0.01$) in rats given hydroethanolic extract of *P. biglobosa* at 100 mg/kg bw; blood glucose levels in these rats fell from 1.45 ± 0.05 to 0.89 ± 0.04 g/L, i.e. a reduction of 38.62%. One hour after treatment (T120), blood glucose levels decreased very significantly in all treated rats compared to that of hyperglycemic rats, with values statistically equal to those of normoglycemic rats. At 90 min post-treatment (T150), blood glucose levels reached 0.55 ± 0.1 ; 0.62 ± 0.06 ; 0.58 ± 0.03 ; 0.65 ± 0.03 and 0.58 ± 0.03 g/L respectively in rats treated with aqueous extract at 100 and 200 mg/kg bw, hydroethanolic extract at 100 mg/kg

bw and glibenclamide at 10 mg/kg bw. These values correspond to reduction rates of 58.01; 51.93; 60.0; 47.15 and 60.27% respectively (Table 3).

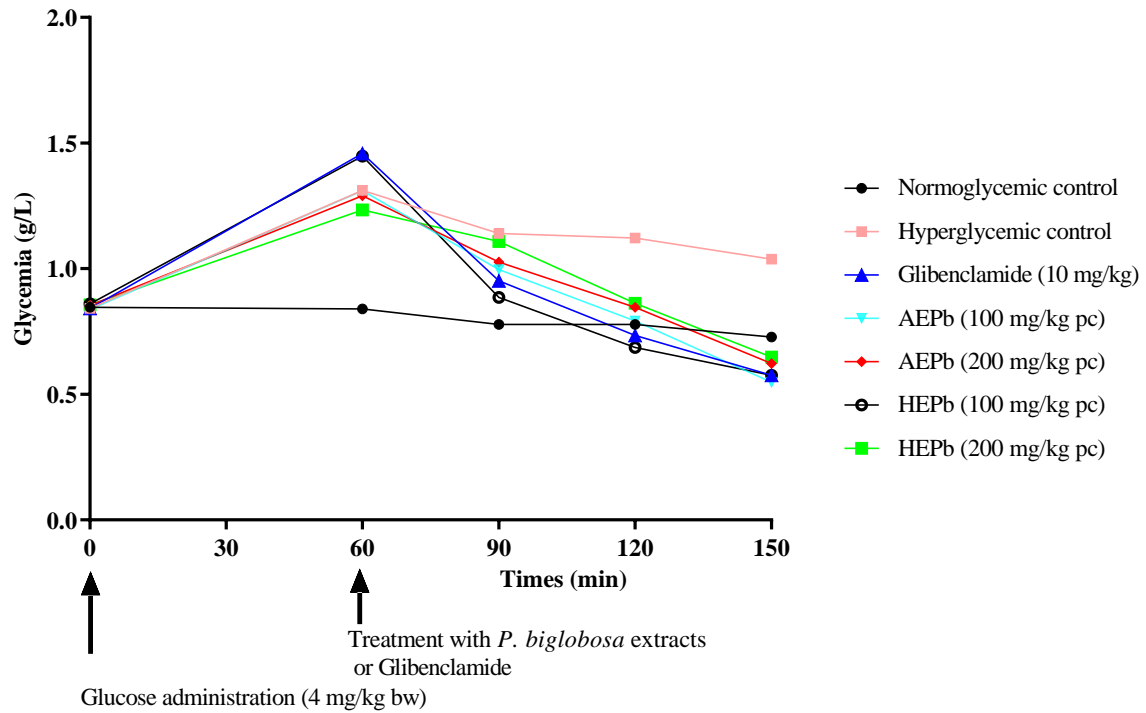


Figure 2: Time course of blood glucose levels in hyperglycemic rats treated with stem bark extracts of *Parkia biglobosa* and glibenclamide

AEPb: aqueous extract of *P. biglobosa*; HEPb: ethanolic extract of *P. biglobosa*

Table 3: Changes in blood glucose levels in hyperglycemic rats treated with stem bark extracts of *Parkia biglobosa* and glibenclamide

Treated rat groups	Change in blood glucose (%)				
	T0	T60	T90	T120	T150
Group 1 (normoglycemic)	0	-1.18	-8.24	-8.24	-14.12
Group 2 (Hyperglycemic control)	0	+55.95	-12,98	-14.5	-20.61
Group 3 (Glibenclamide 10 mg/kg)	0	+73.8	-34.93	-50	-60.27
Group 4 (AEPb 100 mg/kg)	0	+0.56	-23.66	-39.69	58.01
Group 5 (AEPb 200 mg/kg)	0	+51.76	-20.15	-34.10	-51.93
Group 6 (HEPb 100 mg/kg)	0	+68.6	-38.62	-52.41	-60
Group 7 (HEPb 200 mg/kg)	0	+43.02	-9.75	-30.08	-47.15

(+): increase in blood glucose levels; (-): decrease in blood glucose level; AEPb: aqueous extract of *P. biglobosa*; HEPb: ethanolic extract of *P. biglobosa*; T0: Time before glucose administration; T60: time

after induction of hyperglycemia; T90, T120, T150: time (min) after induction of hyperglycemia + treatment with stem bark extracts of *P. biglobosa* and glibenclamide.

Discussion

The phytochemical study revealed the presence of polyphenols, flavonoids, catechic and gallic tannins, quinone compounds, alkaloids, saponosides, sterols and polyterpenes in aqueous and hydroethanol stem bark extracts of *Parkia biglobosa*. These results are similar to those of Millogo-Kone *et al* (2006), who showed the presence of sterols, triterpenes, coumarins, flavones, anthraquinones, tannins, anthocyanins and saponins in stem bark of *P. biglobosa*.

The acute oral toxicity study estimated the lethal dose 50 (LD₅₀) of aqueous and hydroethanolic stem bark extracts of *P. biglobosa* at over 2000 mg/kg b.w. These extracts are therefore classified in category 5 of the globally harmonized system of classification of chemicals, which characterizes substances of low toxicity (OECD, 2001). This result corroborates those of Builder *et al.* (2012), who showed that the LD₅₀ of aqueous and alcoholic stem bark extracts of *P. biglobosa* was greater than 5000 mg/kg by the oral route.

The study of the effect of aqueous and hydroethanolic stem bark extracts of *P. biglobosa* on blood glucose shows that baseline blood glucose values in Wistar rats used range from 0.83 ± 0.01 to 0.854 ± 0.014 g/L. These values are consistent with normoglycemic Wistar rats, as reported by Giknis and Clifford (2008) and Diatta *et al.* (2017). Four (4) hours after treatment of rats with extracts of *P. biglobosa* and glibenclamide, baseline blood glucose levels was reduced by more than 30%. At dose of 100 mg/kg b.w, these extracts, like glibenclamide at 10 mg/kg b.w, reduced baseline blood glucose levels by over 40%. These results indicate that extracts of *P. biglobosa* have hypoglycemic properties. However, the reduction of blood glucose is not dose-dependent, as the effect of these plant extracts, which at 100 mg/kg bw is comparable to that of glibenclamide (10 mg/kg bw), is greater than that of the 200 mg/kg b.w. dose.

In the study of the effect of *P. biglobosa* extracts on induced hyperglycemia, oral administration of glucose at 4 g/kg bw resulted in a highly significant increase in blood glucose levels in rats. Treatment of hyperglycemic rats resulted in a rapid fall in blood glucose levels after 30 min in rats treated with aqueous extract at 100 and 200 mg/kg, and in those treated with hydroethanolic extract at 100 mg/kg and glibenclamide at 10 mg/kg. At 90 min post-treatment, hyperglycemia was reduced by almost 60% in rats treated with *P. biglobosa* extract at 100 mg/kg bw, as well as in those treated with glibenclamide at 10 mg/kg bw. This reduction in hyperglycemia could

be explained by glucose depletion due to the effect of plant extracts. These results are in agreement with those of Odetola *et al.* (2006), who showed that the methanolic extract of fermented *P. biglobosa* seeds administered at a dose of 6 g/kg reduced blood glucose levels by 64.4% in rats made diabetic by alloxane. The reduction of hyperglycemia by *P. biglobosa* extracts is not dose-dependent, as at 100 mg/kg bw, their effect is more significant than at 200 mg/kg bw. This is confirmed by Ibrahim *et al.* (2016), who revealed that the 150 mg/kg bw dose of the butanol fraction of *P. biglobosa* leaves is more effective against induced type 2 diabetes in rats compared with the 300 mg/kg dose.

Hypoglycemia and reduction in hyperglycemia observed in rats treated with aqueous and hydroethanolic stem bark extracts of *P. biglobosa* could be explained by stimulation of insulin secretion by the pancreas (Jackson and Bressler, 1981). The results also showed that hypoglycemic and antihyperglycemic effect of *P. biglobosa* extracts at 100 mg/kg bw were similar to those of glibenclamide, the reference hypoglycemic agent used in the present study. This suggests that these extracts may act via the same mechanism as this reference substance which passes through its direct action on GLUT 2 receptors (glucose transporter in the cell) or stimulation of residual β cells of the pancreas to allow the storage of glucose (Hue, 1987). Furthermore, the hypoglycemic and antihyperglycemic effects of these extracts could be attributed to the phenolic compounds, notably flavonoids, and other active compounds present. Many studies have in fact described the effect of flavonoid-rich plant extracts on blood glucose (Ragunathan and Sulochana, 1994; Fang *et al.* 2008; Araujo-León *et al.*, 2023).

Conclusion

This study showed that aqueous and hydroethanolic stem bark extracts of *Parkia biglobosa* are able to induce hypoglycemia in normoglycemic rats and reduce hyperglycemia in rats made hyperglycemic. The hypoglycemic and antihyperglycemic activity of these extracts is not dose-dependent, since their effect is greater at low doses than at high ones. This low-dose effect is also comparable to that of glibenclamide, the standard hypoglycemic drug.

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