

ANTIDIABETIC EFFECT OF METHALONIC LEAF EXTRACT OF *PTEROCARPUS ERINACEUS* IN STREPTOZOTOCIN INDUCED DIABETIC RATS

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Abstract: The plant *Pterocarpus erinaceus* has been widely utilized Phytotherapeutically for its anti-mycotic, anti-helminthics, anti-malaria, anti-gonadotropic activities. The anti-diabetic effect was investigated using the methanolic leaf extract of the plant on male albino rats by monitoring the Blood Glucose Concentration before and after the treatment. Phytochemical screening of the plant reveals the presence of Saponins, Tannins, Flavonoids, Alkaloids, Steroids, Carbohydrates and Cardiac glycosides. Acute toxicity study of the methanolic leaf extract of the plant was greater than 5000mg/kg when administered via the oral route. The fasting blood glucose concentration of the animals was determined at day 0 then at day 1, 4 and 7 post administration. At day 4 and day 7, there was significant ($p < 0.05$) reduction in the blood glucose concentration of the group that received 250mg/kg, 500 mg/kg, 1000mg/kg of the extract. There was no significant change in the weight of the animal across the group. The result indicates that the methanolic leaf extract of *Pterocarpus erinaceus* Poir has anti-diabetic activity as it reduces the blood glucose concentration from 330mg/dl to 117mg/dl. This study shows possible beneficial effects of *Pterocarpus erinaceus* in the management of diabetes.

Key words: Diabetes mellitus, *Streptozotocin*, *Blood glucose*, *Pterocarpus erinaceus* Diabetic rats.

I. INTRODUCTION

Diabetes mellitus is a complex, chronic disease. It is a condition characterized by an elevation of the level of glucose in the blood. Insulin, a hormone produced by the pancreas controls the blood glucose level by regulating the production and storage of glucose. In diabetes there may be a decrease in the body's ability to respond to insulin or a decrease in the insulin produced by the pancreas which leads to abnormalities in the metabolism of carbohydrates, proteins and fats. The resulting hyperglycemia may lead to acute metabolic complications including ketoacidosis and in the long term contribute to chronic micro-vascular complications (Yim et al., 2007).

Joseph and Jini, (2011) define diabetes mellitus as a heterogeneous metabolic disorder characterized by altered carbohydrate, lipid and protein metabolism which cause hyperglycemia resulting from insufficient insulin secretion, insulin action or both. It is classified basically into primary or Insulin Dependent Diabetes Mellitus (IDDM) and secondary or Non-Insulin Dependent Diabetes mellitus (NIDDM) with the secondary type accounting for barely 1 to 2%. (Chuhwak and Pam, 2007). Africa indigenous herbal medicine is widely used throughout the African continent, despite an apparent lack of scientific evidence for their quality, safety and efficacy (Amos et al., 2002). *Pterocarpus erinaceus* belongs to the family Fabaceae. The common names are Barwood, Bloodwood. The Hausa call it Maijinni. The ethanolic stem bark extract has been used to treat inflammatory pathologies like dermatitis, gastric ulcer, rheumatism. (Salawu et al., 2008). The plant also has anti-malarial activity and antimicrobial activity (Salawu et al., 2008). The heartwood is a source of a red dye, which is used for dyeing body, hair and cloth. The bark is used for tanning. The reddish bark exudates is known as kinotannic acid and is used in traditional medicine internally to treat diarrhea, dysentery, fever, gonorrhea, intestinal worms, and externally to treat eye complaints, ulcers and sores. Kinotannic acid has strong astringent property. Decoctions or infusions of the bark or roots is used for treating bronchial infections, toothache, menstruation complaints, anaemia, post-partum haemorrhage, ringworms, leprosy, wounds, tumors, antiemetic, purgative and tonic. Leaf decoctions are applied to treat syphilis. (Duvall, 2008). The leaves are used as febrifuge, the bark is used for tooth and mouth

troubles. The bark is used as astringent for treating severe diarrhea and dysentery. The grated root is mixed with tobacco and smoked in a pipe as a cough remedy. (Edward, 1999). The aqueous extract of *Pterocarpus erinaceus* stem bark has anti-inflammatory, analgesic and antioxidant activities (Anne, 2012). The anti-malaria (Karou et al., 2003), anti-helminthic (Maidou et al., 2005) and anti-gonadotropic (Benie et al., 2003) activities of the plant has also been investigated. The plant has both in-vitro and in-vivo anti-mycotic activity (Etuk et al., 2008). The plant is very popular among the Hausa speaking people of North-western Nigeria because of its medicinal properties. The stem bark decoction is taken orally for gastrointestinal upset (ICRAF, 1998). The phytochemical screening using standard procedures of analysis revealed the presence of Tannins, flavonoids, saponins, triterpenoids, steroids (Anne, 2012). The bark extracts showed in-vitro antibacterial and antifungal activities. The effectiveness of the bark as a wound-healing agent was due to the presence of phenolic compounds that have an effect on the complementary system (part of the immune system). There are claims by some herbal practitioners in Plateau state; Nigeria that the aqueous leaves extract of *Pterocarpus erinaceus* is effective in the management of diabetes mellitus. However, the scientific evidence to back the claim is lacking. In view of this, the aim of this study is to evaluate the antidiabetic effect of the methanolic leaf extract of *Pterocarpus erinaceus* on streptozotocin-induced diabetic rats.

II. METHODOLOGY

Collection of Animals

Twenty-five white male albino rats were collected from Animal House Department of the University of Jos and maintained on growers pettilizer feed till the time of experiment.

Collection and Extraction of Plant Material

Pterocarpus erinaceus leaves were collected from Jos North Local Government Area of Plateau State and identified by a taxonomist at the college of forestry Jos. The leaves were dried under the shade and the dried leaves were pounded using mortar and pestle. 180g of the powdered leaves was then weighed and then 70% v/v of methanol was used to extract the active principle of the plant via soxhlet extractor. The filtrate was placed in the oven to dry at a temperature of 50 degree centigrade. The plant yield 14.8g of dried extract.

PHYTOCHEMICAL SCREENING

The phytochemical screening was carried out according to protocols described by Trease and Evans (1985).

Acute Toxicity Study (LD₅₀ Determination)

The oral acute toxicity of the methanolic extract of *Pterocarpus erinaceus* was determined in rats as described by (Lorkes, 1983).

Experimental Animals

Twenty-five adult male healthy rats weighing between 100g and above were used for the study. The animals were distributed randomly into five groups of five animals each for streptozotocin-induced diabetic experiment.

Evaluation of Anti-Diabetic Activity

Diabetes was induced by intra-peritoneal injection of streptozotocin (65mg/kg) in citrate buffer of pH 4.5. After 72 hours the glucose levels were monitored with a glucometer for estimation of blood sugar level. The animals with blood sugar level of 170mg/dl and above (Lenzen, 2008) were considered diabetic. The normal blood glucose level in wistar white rat is between 85-132mg/dl (Kohn and Clifford, 2002). The diabetic animals were randomly distributed into five groups of five animals each and treated as follows:

Group I: Streptozotocin-induced diabetic animals but untreated to serve as negative control

Group II: Streptozotocin-induced diabetic animals treated with glibenclamide (5mg/kg) to serve as standard.

Group III: Streptozotocin-induced diabetic animals treated with *Pterocarpus erinaceus* leaf extract (250mg/kg)

Group IV: Streptozotocin-induced diabetic animals treated with *Pterocarpus erinaceus* leaf extract (500mg/kg)

Group V: Streptozotocin-induced diabetic animals treated with *Pterocarpus erinaceus* leaf extract (1000mg/kg).

Animals were fasted for 18 hours before administration of the extract and the standard drug (glibenclamide).

Blood Glucose Analysis

The tip of the rat tail was cut and the first drop of blood wiped off. The subsequent drops are used. The blood glucose concentration was determined at day 0, day 1, day 4 and day 7 of post administration of the extract. The weight of the animals was also taken at day 0, 1, 4 and 7 of post administration of the extract.

Analysis of Data

Data generated from the study were analyzed with the aid of statistical package for social sciences (SPSS) version 20.0. Mean weight and blood glucose concentration were generated for each study group across study period, also mean change and blood glucose concentration were generated for each group by subtracting the baseline value of each parameter from the specific measuring period. The mean change in weight and blood glucose concentration were compared for statistical significance in relation to normal group values using independent T-test set at $P < 0.05$ for significance.

III. RESULTS

Phytochemical Screening

The phytochemical screening of the extract reveals the presence of saponins, tannins, carbohydrates, alkaloids, cardiac glycosides. Flavonoids are present in abundance quantity while anthraquinone is absent.

Acute Toxicity Study

The acute toxicity study of the plant reveal that the methanolic leaf extract of *Pterocarpus erinaceus* is safe at a dose greater than 5000mg/kg as no death was recorded.

The result below shows that the methanolic leaf extract of *Pterocarpus erinaceus* pair reduce the blood concentration of the streptozotocin-induced diabetic male albino rats across the group.

TABLE: 1 Effect of the Extract on the Mean Blood Glucose Concentration on the Rats Across Study Groups Compared to Control Group

Group	Treatment	Dose mg/kg	Blood glucose concentration Day 0	Mg/dl Day 1	(Mean SEM) Day 4	Day 7
1	Extract	250	298.00±61.64	253.50±77.28	199.50±69.85*	158.50±49.56*
2	Extract	500	330.00±77.87	328.00±78.37	242.25±71.99	171.25±73.08*
3	Extract	1000	247.67±67.76	296.67±86.09	250.67±84.68	117.67±37.75*
4	Glibenclamide	5	379.50±53.70	382.75±52.14	347.75±39.61	239.25±16.96
5	Negative control		395.00±33.29	423.25±22.93	478.25±14.23	500.25±12.11

*Indicates significant difference ($P < 0.05$) in mean blood glucose concentration in study group compared to control group

The result in table 2, below shows that there was no significant change in the weight of the animals across the group.

Table 2: Effect of the Methanolic Leaf Extract of *Pterocarpus erinaceus* Poir. on the Weight of Streptozotocin-Induced Diabetic Male Albino Rats Across the Study Group.

Group	Treatment	Dose	Weight	(g)	(Mean± SEM)	
		mg/kg	Day 0	Day 1	Day 4	Day 7
1	Extract	250	151.00±37.17	157.57±37.98	154.73±39.19	157.40±39.95
2	Extract	500	193.65±46.67	185.28±41.90	178.90±39.06	184.23±39.93
3	Extract	1000	158.33±26.87	155.37±27.23	155.07±32.48	154.73±33.75
4	Glibenclamide	5	125.68±2.41	122.50±7.95	127.03±4.07	129.00±3.11
5	Negative control		127.75±4.41	127.65±6.56	117.28±7.90	106.20±6.93

No significant difference ($P > 0.05$) in mean weight in study group compared to control group, n=5

IV. Discussion

Diabetes mellitus is a fast growing metabolic disease in the world and as the knowledge of the multifactorial and heterogeneous nature of the disease increases so does the need for more challenging and appropriate therapies (Akah et al., 2002). In order to establish the scientific basis for the utility of *Pterocarpus erinaceus* in the treatment of diabetes, evaluation of the antidiabetic activity of the methanolic extract was carried out.

The result of the present study reveals that methanolic leaf extract of the plant has flavonoids in large amount, also alkaloids, saponins, tannins, cardiac glycosides, steroids, carbohydrates were all found to be present in the plant but anthraquinone is absent. The methanolic leaf extract of the plant shows decrease in blood glucose concentration on the animals across the group.

The blood glucose lowering activity of the plant might be due to its ability to restore the function of pancreatic tissues by causing an increase in insulin output or inhibit the intestinal absorption of glucose which is due to its phytochemical constituents (flavonoids, alkaloids, glycosides) as those constituents have been found to be frequently implicated as having anti-diabetic effect (Malviya et al., 2010). Flavonoids have been found also to cause pancreatic beta cells regranulation and thus enhance insulin release (Modak et al., 2007).

The anti-diabetic effect of the plant may be attributed to some of the plant constituents flavonoids, alkaloids, even as reported by previous literatures (Jung, et al., 2006). Also, Literature showed that saponins and flavonoids are good anti diabetic metabolites (Sharma et al., 2010).

Over the decades, an expanding body of evidence from epidemiological and laboratory studies have demonstrated that some plant as a whole or their identified ingredients with antioxidant properties have substantial protective effects on diabetes (Sabu and Kuttan, 1982), cardiovascular and renal disorders (Anderson et al., 2000) and several other human ailment (Lampe, 2003). However, antidiabetic effect of the extract largely depends on its antioxidant properties (Melai et al., 2011). Also, radical scavengers such as phenolics like flavonoids, alkaloids have been shown to be effective in preventing diabetes in animal models (Oberley, 1988), which is contain in this extract.

The activity of the plant may be due to its ability to restore the function of the pancreatic tissue by causing an increase in insulin output or inhibit the intestinal absorption of glucose or might also be due to its insulin secretagogue activity, (though this have not been proven scientifically) as reported by previous literature (Bhushan et al., 2010).

The acute toxicity study carried out reveals that the methanolic leaf extract of the plant is safe at a dose higher than 5000mg/kg, thus it can be said that the plant is safe to a high degree and least toxic and this agrees with the European chemical industry ecology and toxicology guideline which considers LD₅₀ above 2000mg/kg as likely to be non-toxic (IRAC, 2004). There is no significant change in weight observed in the diabetic animals treated with extract this shows that it might not have the obesity forming tendency which is one of the undesirable side effects associated with sulphonylureas. (Ojewale et al., 2013). Decrease in body weight in diabetic rat is as a result of excessive breakdown of tissue protein (Mishra et al., 2011). Treatment with extract improved body weight to a certain extent, indicating that extract has control over muscle wasting result from glycemic control. This suggests the antidiabetic effect of extract in diabetic rats.

In conclusion, the methanolic leaf extract of *Pterocarpus erinaceus* Poir showed significant blood glucose lowering activity across the group and also does not cause significant increase in weight of the animals. Further studies need to be carried out to determine the mechanism of action of the extract and which of the constituent(s) is largely responsible for the anti-diabetic effect.

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