Asian Journal of Pharmaceutical and Health Sciences
www.ajphs.com

The antihyperglycemic and hypolipidemic effects of the aqueous extract of *Abrus precatorius* in alloxan-induced diabetes mellitus in albino rats

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ARTICLE HISTORY

Received: 22.01.2016

Accepted: 07.03.2016

Available online: 30.03.2016

Keywords:

Alloxan, Diabetes mellitus, Fasting blood glucose, Lipid profile

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ABSTRACT

Abrus precatorius (Fabaceae) is a plant well known for its use in traditional medicine in the management of diabetes mellitus. The whole plant was extracted via cold maceration using distilled water. DM was induced in rats using alloxan (150 mg/kg i.p). Diabetic rats were divided into 4 groups of 5 rats each. The first 2 groups received the extract (100 & 400 mg/kg) respectively, the 3rd, glibenclamide (5mg/kg), while the last received distilled water (2ml/kg), i.e the untreated diabetic rats. A fifth group comprised of non-diabetic rats given distilled water (2ml/kg); control rats. All administrations were done orally for 14 days. Fasting blood glucose was evaluated on days 0, 1, 7 and 14.Blood samples were also obtained and analysed for HDL, LDL, triglycerides and total cholesterol. *Abrus precatorius* produced significant ($p<0.05$) reduction in the fasting blood glucose of diabetic rats from day 1 through 14 by both doses in comparison with the untreated diabetic rats. This effect was most prominent on day 7 by the 400 mg/kg dose ($p<0.0001$) and compares favourably with glibenclamide ($p<0.0001$) which produced a similar blood glucose lowering effect. The HDL was significantly ($p<0.05$) increased following treatment with the 400 mg/kg dose in comparison with the untreated diabetic and control rats. Other parameters were not significantly affected on treatment with both doses of the extract.The plant possesses blood glucose lowering effect, with the added advantage of an increase in the HDL level and thus a useful remedy in the management of diabetes mellitus.

INTRODUCTION

Plants are directly used as medicine by a majority of cultures around the world, for example Africa, Chinese and Indian medicine. Medicinal plants are resources for new drugs.

The study of medicinal plants has created an essential area for further studies and research, and the discovery of new lead compounds for drug development [1].*Abrus precatorius* is a slender, perennial climber that twines around trees, shrubs, and hedges [2, 3]. It grows best in fairly dry regions at low elevations in tropical climates such as India, Sri Lanka, Thailand, tropical Africa and the West Indies. The seeds are used to treat diabetes and chronic nephritis. The plant is used in some traditional medicine to treat scratches and sores, and wounds caused by dogs, cats, and mice, and are also used with other ingredients to treat leucoderma. The plant is also traditionally used to treat tetanus, and to prevent rabies. The powdered seeds are used as oral contraceptives by various African tribes [2].

Diabetes mellitus, an anomaly in the blood glucose is divided into type 1, characterized by loss of the insulin producing beta cells of the islets of Langerhans in the pancreas, leading to insulin deficiency. This type can be further classified as immune-mediated or idiopathic [4]. And type 2 characterized by insulin resistance, which may be combined by relatively reduced insulin secretion [5]. Type 2 diabetes is the most common type. Hyperlipidemias are common in patients with diabetes and further increase the risk of ischemic heart disease, especially in type2 diabetes. Detection and control of hyperlipidemia can reduce myocardial infarction, coronary deaths, and overall mortality [6]. The use of herbal medicine is associated with lesser risk of side effects when compared with conventional medicines. This study therefore is important because it ascertains the

antidiabetic effect of the plant, identifies the various phytoconstituents present in *Abrus precatorius* probably responsible for this effect and also evaluates its effect on the lipid profile of alloxan-induced diabetes mellitus in rats. And hence give credence to its use in traditional medicine for the treatment of diabetes mellitus.

MATERIALS AND METHODS

Collection and preparation of plant material

The whole plant of *Abrus precatorius* was obtained from Isihor quarters, Benin City, Edo state, Nigeria in August, 2014. The plant was identified in the Department of Pharmacognosy, University of Benin, Nigeria where a herbarium specimen exists. A voucher specimen with no: FHI 157085 exists at the Forest Research Institute of Nigeria, Ibadan, Nigeria.

The whole plant was air dried for several days and then pulverized into powdered sample. 800 g of the powdered sample was extracted via cold maceration using 2 L of distilled water for 72hrs. The macerated extract was filtered with a filter cloth and the filtrate concentrated in an oven (< 40°C). The percentage extract yield was calculated. The extract was reconstituted prior to administration. The percentage yield gave 7.41%.

Animals

Adult albino rats weighing 150-250 g were obtained from the animal house of the Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Benin, Nigeria. The animals were acclimatized for 14 days under standard environmental conditions on a regular feed (standard growers mash, Top feeds, Nigeria) and had access to water *ad libitum.*

Ethical approval was obtained from the ethical committee on the use of animals, Faculty of Pharmacy, University of Benin, Nigeria. The animals were also handled according to the standard protocols for the use of laboratory animals [7].

Drugs/Chemicals

Alloxan monohydrate (Qualikens, laboratory reagent grade, China), glibenclamide (Hovid Berhad, Västernorrland County Sweden), chloroform (Sigma Aldrich UK). Lipid profile kit (Randox, UK). All other chemicals used in the study were of analytical grade.

Phytochemical analysis

The aqueous extract of *Abrus precatorius* was subjected to preliminary phytochemical screening for various secondary plant metabolites. The methods of analysis employed were as described by [8].

Induction of experimental diabetes/Treatment

Diabetes was induced by intraperitoneal injection of 150 mg/kg body weight of alloxan monohydrate in overnight-fasted animals after acclimatization [9]. Diabetes was confirmed three days after in the alloxan treated animals showing fasting blood sugar (FBS) level greater than or equal to 200mg/dL (11.1mmol/L). Diabetes status determination was monitored on blood obtained from tail vein puncture and checked using a glucometer (Accucheck). Blood glucose levels greater than 200 mg/dl were used for research [10].A total of thirty five albino rats were divided into five groups of seven rats each. The animals were grouped as follows: Group 1 (reference/normal control rats) given distilled water daily, group 2: diabetic untreated rats (diabetic control) given distilled water daily. Groups 3, 4 and 5: diabetic rats administered glibenclamide (5mg/kg), the aqueous extract of *Abrus precatorious* 100 and 400 mg/kg body weight respectively.The different doses of the extract, drug and distilled water were administered to the rats orally using an orogastric tube on a daily basis for 14 days.

Determination of fasting blood glucose level

The blood glucose level of the alloxan induced diabetic rats was checked via blood samples obtained by cutting the tip of the tails to obtain blood from the tail vein. Blood glucose determination was done on day 0, 1, 7 and 14 following drugs/extract administration. On the 14th day, following blood glucose determination, the animals were sacrificed under chloroform anaesthesia with cervical dislocation and blood withdrawn for the lipid profile analysis.

Lipid profile analysis

Blood samples were collected using 2 ml and 5 ml syringes from the abdominal aorta and left ventricle of the heart. The blood samples were introduced into lithium heparinized bottles to avoid clotting of blood. The blood samples were centrifuged at 3,000g for 5 minutes and plasma samples obtained were thereafter used for the determination of lipid profile.

Statistical analysis

All data were expressed as mean ± SEM. Where applicable; the data were analyzed statistically by Student's t-test using Graph pad instat version 2.05a. P values less than 0.05 were considered as significant.

RESULTS

Phytochemical analysis

The results of the phytochemical screening are shown in table 1. It revealed the presence of the following secondary plant metabolites in the aqueous extract of *Abrus precatorius*: Simple

Table 1. : Preliminary phytochemical analysis of the aqueous extract of *Abrus precatorius*

Test	Result
Alkaloid	-
Anthraquinone	-
Tannins	+
Saponin	+
Reducing sugar	+
Flavonoids	+
Cardiac glycoside	+
Phenol	+
Simple sugar	+
Steroid	+

- : negative + : positive

Table 2. : Effect of *Abrus precatorius* and glibenclamide on lipid profile of alloxan induced diabetic rats

Group (mg/kg)	TC	HDL	TG	LDL
Control	41.3±0.9	21.5±1.7	42.5±2.1	11.6±1.6
UD	35.6±2.1	16.7±2.5	24.5±2.7	13.9±1.7
D+AP(100)	67.5±0.7	18.8±1.1[a]	39.5±1.4	15.0±0.7
D+AP (400)	38.0±1.0	28.9±0.7[a]	49.4±0.5	21.7±0.8
D+G(5)	28.8±0.6[a]	24.4±0.6[a]	46.5±1.7	16.5±0.5

n = 5 per group, Values are mean ± SEM
[a]p <0.05 significantly different from the untreated diabetic group.
Control: Normal rats administered distilled water
UD: Untreated diabetic group administered distilled water
D+ AP(100): Diabetic group treated with 100 mg/kg *Abrus precatorius*
D+ AP(400): Diabetic group treated with 400 mg/kg *Abrus precatorius*
D+ G (5): Diabetic group treated with 5 mg/kg glibenclamide

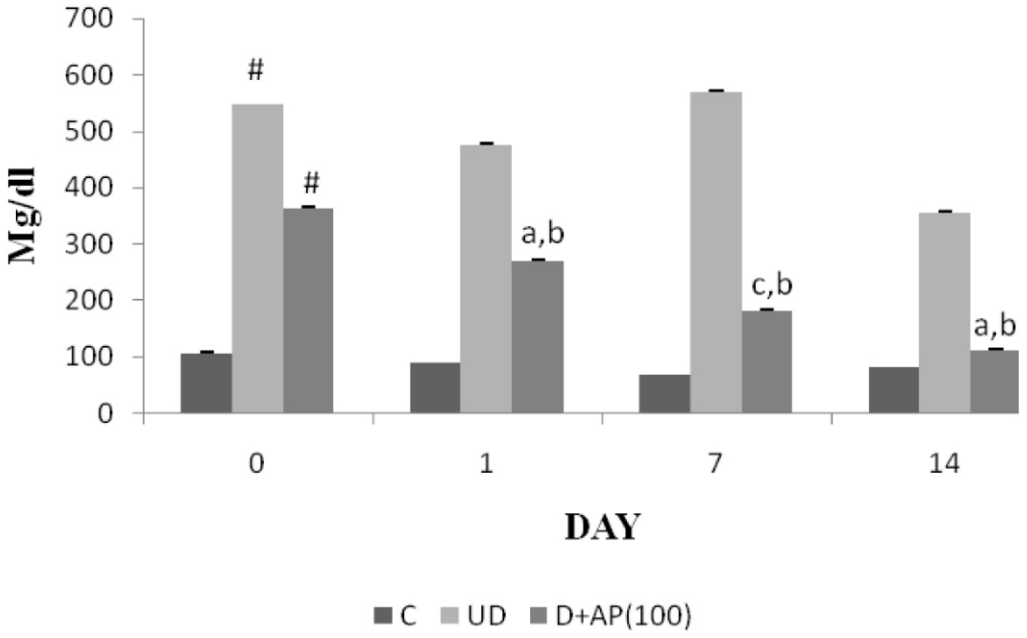

Fig 1. : The effect of *Abrus precatorius* (100 mg/kg) on the fasting blood glucose of diabetic rats.

n = 5 per group, values are mean blood glucose levels ± SEM.
[#]p<0.0001 significantly higher than the control, [a]p<0.05 and cp<0.0001 significantly lower than the untreated diabetic group, [b]p<0.05 significantly different from the preceeding day for D+AP(100).
C: Normal rats administered distilled water
UD: Untreated diabetic group administered distilled water
D+ AP (100): Diabetic group treated with 100 mg/kg *Abrus precatorius*

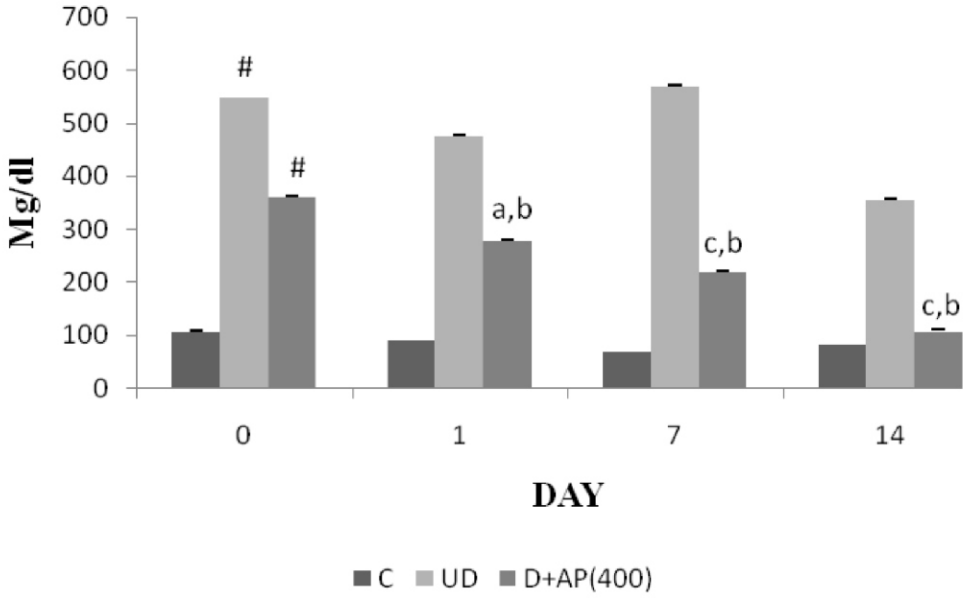

Fig 2. : The effect of *Abrus precatorius* (400 mg/kg) on the fasting blood glucose of diabetic rats.

n = 5 per group, values are mean blood glucose levels ± SEM.
[#]p<0.0001 significantly higher than the control, [a]p<0.05 and [c]p<0.0001 significantly lower than the untreated diabetic group, [b]p<0.05 significantly different from the preceeding day for D+AP (400).
C: Normal rats administered distilled water
UD: Untreated diabetic group administered distilled water
D+ AP(400): Diabetic group treated with 400 mg/kg Abrus precatorius

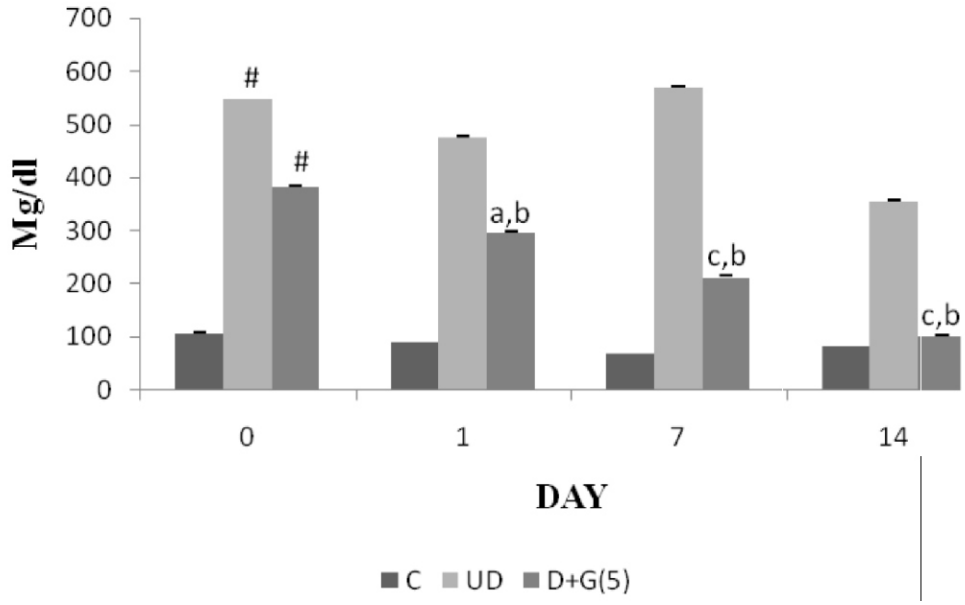

Fig 3. : The effect of Glibenclamide (5 mg/kg) on the fasting blood glucose of diabetic rats.

n = 5 per group, values are mean blood glucose levels ± SEM.
[#]p<0.0001 significantly higher than the control, [a]p<0.05 and [c]p<0.0001 significantly lower than the untreated diabetic group, [b]p<0.05 significantly different from the preceding day for D+G(5).
C: Normal rats administered distilled water
UD: Untreated diabetic group administered distilled water
D+ G (5): Diabetic group treated with 5 mg/kg glibenclamide

reducing sugar, saponins, tannins, flavonoids, cardiac glycosides, phenols, steroids.

The effect of the aqueous extract of *Abrus precatorius* on the fasting blood glucose of alloxan induced diabetic rats.

The results are presented in figures 1 to 3. In the diabetic rats treated with 100 and 400mg/kg of the aqueous extract of *Abrus precatorius*, there was a significant reduction in the blood glucose level ($p<0.05$) from day 1 following treatment to day 14 compared to the untreated diabetic rats, however the effect was most prominent day 14.

The reduction can be seen to be better with the higher dose of the extract.

There was also a significant reduction in the blood glucose levels of the diabetic rats treated with glibenclamide ($p<0.05$) compared to the untreated diabetic rats (fig 3).The effect of the extract can be said to compare well to that of the standard (glibenclamide), where significant reduction ($p<0.05$) in the blood glucose level of the diabetic rats was also observed. It can thus be said that *Abrus precatorius* has a similar antidiabetic effect as glibenclamide.

The effect of *Abrus precatorius* on the lipid profile of alloxan-induced diabetic rats.

The results are presented in table 2. A decrease in the low density lipoproteins of the diabetic treated rats compared to the untreated diabetic rats was observed but this effect was not significant. There was also no significant difference in the triglyceride/total cholesterol levels of the diabetic rats treated with both doses of *Abrus precatorius* when compared to the untreated diabetic rats, although glibenclamide produced a significant reduction ($p<0.05$) in the total cholesterol. However a significant increase in high density lipoprotein of the treated diabetic compared to the untreated diabetic rats was observed by both doses ($p<0.05$), a similar occurrence with glibenclamide.

DISCUSSION

The antihyperglycemic action of the extract tested in this study could be attributed to the presence of tannins, cardiac glycosides, flavonoids and saponins which have been shown to be hypoglycemic. Terpenoids have also been shown to decrease blood sugar level in animal studies [11]. Previous studies, such as on the leaves of *Psidium guajava,* rich in flavanoids lowered blood glucose in humans [12]. In the elderly patients with hyperglycemia, saponins was noted to reduce serum glucose and finally another carried out on ginseng and its saponins lowered blood glucose in alloxan treated genetically diabetic and normal mice [12]. Hence the presence of saponins and flavanoids in this extract could have been responsible for the blood glucose lowering effect seen in alloxan-induced diabetic rats.

The untreated diabetic rats had significantly higher fasting blood glucose levels than the normal rats that received distilled water. This higher blood glucose level (>200mg/dl) confirmed the induction of diabetes by alloxan. Alloxan induces formation of superoxide radicals, which usually results in massive increase in cytosolic calcium levels. This heightened calcium level triggers the destruction of the pancreatic beta cells and ultimately negatively affects insulin synthesis and secretion [13]. The lowering effect of blood glucose levels in alloxan-induced diabetic rats by the aqueous whole plant extract of *Abrus precatorius* indicates that this plant possess some antihyperglycemic activity. The aqueous extract of *Abrus precatorius* lowered blood glucose to normal by the 14th day and did it as effectively as glibenclamide. Comparisons were made using glibenclamide which is a sulphonylureas. Sulphonylureas reduce blood glucose by increasing insulin secretion from pancreatic beta cells in patients with residual beta cell function [14]. This means that they are active in mild alloxan-induced diabetes and are inactive in intense alloxan-induced diabetes [15].

In our study the diabetic rats that received the extract showed normalization of blood glucose levels compared to untreated by the 14th day. This could be due to the possibility that some beta cells were still intact and were therefore stimulated to synthesize and release insulin by the extract. In addition, the extract may have shown antihyperglycemic action by enhancing tissue glucose utilization, decreasing the rate of carbohydrate absorption into the portal hepatic circulation, increased glucose transport and uptake, increased glycogen storage, and modulation of insulin secretion. Shane, [16] reported similar mode of action in his study on examination of hypoglycemic potential of the aqueous leaf extract of *Gymnema sylvestre*. This is also the mode of action of alpha glycosidase inhibitor such as acarbose and miglitol [17].

Though lipids play many important roles in the body, they can lead to cardiovascular disease when there is an abnormality in the blood level in an individual. Insulin deficiency has been shown to lead to a number of metabolic alterations in the animals viz a vis high blood glucose levels and increased cholesterol concentration [18]. Hyperlipidemia is one of the recognized complications of diabetes mellitus characterized by elevated levels of cholesterol, triacylglycerol and changes in lipoprotein composition. In severe diabetic conditions, the kidneys ability to remove wastes products such as urea and creatinine is compromised.

Results of the lipid profile show that the extract had no significant hypolipidemic activity on the normal and diabetic rats, though the HDL of the diabetic rats were significantly ($p<0.05$) increased by *Abrus precatorius*. This is definitely an advantage for the diabetic patient, as HDL level is an important tool in assessing the extent of cardiovascular risk in the diabetic patient. [6]. A high level always indicates a lower risk of CVS complications.From these findings, the extract of *Abrus precatorius* is an important alternative to orthodox drugs in the management of DM and possibly has various medicinal uses in addition to its antidiabetic property.

CONCLUSION

In conclusion, the aqueous extract of *Abrus precatorius* used in herbal medicine practice showed significant antidiabetic activity. The plant extract lowered the fasting blood glucose levels at both doses tested and was as effective as glibenclamide. The extract also increased high density lipoprotein of diabetic rats. This study confirms the use of *Abrus precatorius* in diabetes mellitus and further studies on exact mode of action and isolation of chemical constituent responsible for its effect is recommended. Conservation of this plant is also advocated.

Acknowledgement

Our profound gratitude goes to Mr Ibeh and Mr Phillip Obarisiagbon for their contributions towards this research.

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