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PHYTO-CONSTITUENT AND SCREENING OF ANTIHYPERGLYCEMIC ACTIVITY OF *ALANGIUM LAMARCKII* ROOTS EXTRACT

Ashutosh Mishra*, Rameshwar Ahirwar, Anita Shivhare Vijay Nigam, Pawan Bajpai, Sanjay Jain

Daksh Institute of Pharmaceutical Science, Chhatarpur (M.P.)

***Correspondence Info:**
Ashutosh Mishra
Daksh Institute of Pharmaceutical Science, Chhatarpur (M.P.)
Email:
researcharticlee78@gmail.com

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ABSTRACT

Diabetes is a serious complex condition which can affect the entire body. Diabetes requires daily self care and if complications develop, diabetes can have a significant impact on quality of life and can reduce life expectancy. While there is currently no cure for diabetes, you can live an enjoyable life by learning about the condition and effectively managing it. It is estimated that in 2010 there were globally 285 million people (approximately 6.4% of the adult population) suffering from this disease. The present study is undertaken to standardize the *Alangium lamarckii* and to evaluate its anti-diabetic activity with emphasize to its mechanism of action. The validation of anti-diabetic property is correlated with various biochemical parameters involved therein. The *in-vivo* antidiabetic activity of ethanolic extract of *Alangium lamarckii* root was tested by using an Alloxan induced diabetic rat. Acute toxicity study was carried out in rats. The procedure was followed by OECD 423 (Acute toxicity class method). 1/5th (400mg/kg) of the maximum safe dose (2000mg/kg) were selected for further study. Fasting blood sample were drawn from tail vein of rats at weekly intervals till the end of the study 1,7 and 14 days. On these days fasting blood glucose were collected and analysed for glucose. At the end of the study (14^{th} day) the ethanolic extract of *Alangium lamarckii* root (400 mg/kg p.o) treated diabetic groups showed statistically significant decrease in blood glucose similar to the standard drug glibenclamide (2mg/kg), which indicated block the alfa amyalase activity. The present study suggested that the isolation of active constituents from ethanolic extract of *Alangium lamarckii* root and characterize the compounds by using preliminary phytochemical studies.

Keywords: Introduction, Medicinal plant, Antihyperglycemic activity, Lamarckii roots extract

INTRODUCTION

Diabetes is a serious complex condition which can affect the entire body. Diabetes requires daily self care and if complications develop, diabetes can have a significant impact on quality of life and can reduce life expectancy. While there is currently no cure for diabetes, you can live an enjoyable life by learning

about the condition and effectively managing it. It is a fast growing global problem with huge social, health and economic consequences (Bastaki, 2005). It is estimated that in 2010 there were globally 285 million people (approximately 6.4% of the adult population) suffering from this disease. This number is estimated to increase to 430 million in the absence of better control or cure. An ageing population and obesity are two main reasons for the increase. Furthermore it has been shown that almost 50% of the putative diabetics are not diagnosed until 10 years after onset of the disease, hence the real prevalence of global diabetes must be astronomically high (Grover *et al.,* 2011). The term diabetes mellitus describes a metabolic disorder of multiple etiology characterized by chronic hyperglycaemia with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action, or both. Diabetes means that your blood sugar is too high. Your blood always has some sugar in it because the body needs sugar for energy to keep you going. But too much sugar in the blood is not good for your health. Diabetes is a disorder of metabolism the way our bodies use digested food for growth and energy (Olokoba, 2012). Most of the food we eat is broken down by the digestive juices into a simple sugar called glucose. Glucose is the main source of fuel for the body. Diabetes mellitus is a group of syndromes characterized by hyperglycemia. The World Health Organization (WHO) has designated diabetes mellitus as an epidemic, although it is a non-infectious disease. Nearly 2.3% of the world's population is estimated to be suffering from this disease, with an increasing trend of 4–5% every year.

Diabetes mellitus is a group of syndromes characterized by hyperglycemia, altered metabolism of lipids, carbohydrates and proteins, and an increased risk of complications from vascular disease Zimmet and Alberti, 2001). More than 400 species of plants have been reported to display hypoglycemic effects, but only a few of them have been investigated. Although many drugs are available to control the diabetes but has several adverse effects. *Alangium lamarckii* highly reputed plant in ayurvedic system of medicine for the treatment of various ailments (Itoh *et al.,* 2000). The plant was reported to have activities such as purgative, anti-asthmatic, antihelmintic, used in the treatment of headaches, antioxidant and anti-diabetic. This study was undertaken to investigate the anti-diabetic activity of extract of roots of *Alangium lamarckii* in alloxan induced diabetic rats. The present study is undertaken to standardize the *Alangium lamarckii* and to evaluate its anti-diabetic activity with emphasize to its mechanism of action. The validation of anti-diabetic property is correlated with various biochemical parameters involved therein.

MATERIALS AND METHODS

Plants materials Lamarckii roots extract were collected from the local market of Bhopal, (M.P.) during the month of May, 2023. The specimens were identified and authenticated by Dr. Zia ul Hassan, Assistant professor, Department of Botany, Saifia College of Science & Education, Bhopal and their herbarium was deposited. These collected specimens were chosen for the extraction process.

Preparation of methanolic extract

In a 2-liter size conical flask, 400 g of each powdered plant material was soaked in a litre of analytical grade methanol (Khandelwal, 2007). The flasks holding each plant material were routinely shaken, corked, and left to stand at room temperature for 48 hours. Filtration via Whatman filter paper No. 1 separated the menstruum in each case. The filtrates were then concentrated in a rotary evaporator at 50°C before being dried fully in a hot-air oven at 35°C. The concentrates were sealed and stored at 4°C until they were used in an in vitro bioassay.

Qualitative Phytochemical Screening

Test for Saponins

Test for Alkaloids

Test for Terpenoids

Test for Flavonoids

Test for Cardiac Glycosides

Test for Steroids

Test for Phenols

Evaluation of Anti-diabetic activity

Selection of animals

Male wistar albino rats of weighing about 150 –200 gm were used for the study. The animals were obtained the animal house from Sapiens lab Bhopal M.P College of Pharmacy, and were approved by Ethical Committee and approval no. is CPCSEA/PS/561.

Maintenance of animals

The animal house was well ventilated and animals had 15-20 ± 2 ºC. The animals were housed in large spacious hygienic cages during the course of the experimental period. The animals were fed with rat pellets feed supplied by M/s Hindustan Lever Limited, Bangalore, India and filtered water ad libitum. Animals were deprived of food & allowed free access to water.

Experimental grouping of animals

The experimental rats were divided into five groups of five animals in each group. The animals were fasted overnight before the experimental schedule began but allowed free access to tap water (Kumar *et al.,* 2011).

Group I: The rats received 2 ml/kg Normal Saline. Serve as normal controls.

Group II: Review Single dose of Alloxan monohydrate (150 mg/kg body weight) by intraperitoneally and served as Negative control.

Group III: The diabetic rat given orally roots extract at the dose of 200 mg/kg.

Group IV: The diabetic rat given orally plant extract at the dose of 400 mg/kg.

Group V: Review Glibenclamide 5mg/kg for 21 days & served as positive control.

Induction of diabetes

Alloxan monohydrate induced diabetes mellitus was induced in the normoglycaemic male albino rats (Kumar *et al.,* 2010). Animals were allowed to fast 24 hrs and were injected intraperitonially with freshly prepared alloxan monohydrate in Sterile Normal Saline in dose of 150 mg/kg body weight. Blood glucose was measured after 24 hr of alloxanisation and it was confirmed that the given dose was sufficient for inducing diabetes in the animals. The animals were maintained in the diabetic state over a period of 21 days. Rats showing fasting blood glucose levels (>250 mg/dl) were selected for

the study. Mortality rate of the animals were Nil.

Statistical Evaluation

Statistical evaluation was done using one way analysis of variance (ANOVA) followed by Dunnet' T- test. Statistical significance was set at $p< 0.001$, $p< 0.01$, p.

RESULTS AND DISCUSSION

The glucose level was significantly ($p <0.01$) high in alloxan control rats compared with normal control. But the level of serum glucose was significantly ($p<0.01$) decreased in diabetic rats AL Eated with extract as compared with alloxan control rats. On repeated administration of the extract for 21days, a significant decrease in the glucose level was observed compared to diabetic control. There was no significant difference between normal control and rats ALEated only with extract. It was evident that unALEated diabetic rats has elevated blood glucose levels and that the roots extract were able to correct this metabolic deviation from the Diabetic Control significantly since there was no significant difference between Normal Control and Diabetic Control the extract has antihyperglycemic activity and no hypoglycemic activity.

The total protein content was significantly ($p<0.01$) decreased when compared to the Normal Control in diabetic rats and the level was restored to nearly normal after. There was no significant difference between Normal control and rats only AL Eated with extract.

The level of lipid profiles in Normal Control, Diabetic Control and experimental rats was depicted in Table no. 1-2. In alloxan induced diabetic rats, there was a significant ($p<0.01$) increase of Cholesterol, Triglycerides, Phospholipids, LDL and VLDL Cholesterol and significant decrease in HDL Cholesterol in serum compared to Normal Control. The plant extract used in the experimental study significantly ($p<0.01$) decrease the level of Cholesterol, Triglycerides, Phospholipids, LDL and VLDL Cholesterol and increase the level of HDL Cholesterol. This indicated the roots extract had favorable effect on lipid metabolism of diabetic rats.

Diabetes Mellitus is a multi factorial disease, which is characterized by hyperglycemia and lipoprotein abnormalities. Glucosede privation in diabetic condition increased the fat metabolism to supply energy to cells. The roots extract AL Eated rats has restored both glucose and fat metabolism. Dearrangement of glucose, fat and protein metabolism during diabetes, results into the development of hyperlipidemia. Significant lowering of Total Cholesterol and rise in HDL Cholesterol is a very desirable biochemical state for the prevention of AtherosALerosis and Ischemic conditions.

Table no. 3, depicts the activity of ALP, SGOT, SGPT, Alkaline Phosphate, Serum Glutamase Oxalo acetate Transaminase levels were found to be increased significantly ($p<0.01$) in alloxanAL Eated diabetic rats in compare to Normal Control. The extract significantly ($p<0.01$) decreased the elevated alkaline phosphate, SGOT and SGPT in alloxan AL Eated rats and the level significantly ($p<0.01$) restored to normal after AL Eatment. There was no significant difference between Normal Control and Diabetic Control.

In diabetic animals the change in the levels of serum enzymes are directly related to changes in the metabolism in which the enzymes are involved. Many workers have reported increase in transaminase activities in the liver and serum of diabetic animals. The increased activity of transaminases which are active in the absence of insulin because of increased availability of amino acids in diabetic are responsible for the increased gluconeogenesis and ketogenesis observed in diabetes. In the present study the roots extract significantly decreased the enzyme activities. Hence the improvements noticed in the levels of the enzymes are as a consequence of improvement in carbohydrates, fats and protein metabolism. The lowering the value of SGOT and SGPT from higher value after the AL Eated also indicated the revival of insulin secretion. The increased level of ALP in diabetic rats found to be significantly reversed by the fraction. SGOT and SGPT levels are indicators of liver function, hence, restoration of normal levels indicate normal function of liver.

Table 1: Effect of roots extract of *Alangium lamarckii* on Serum Cholesterol, Triglycerides, Phospholipids, of control and experimental rats

Groups	Cholesterol (mg/dl)	Triglyceride (mg/dl)	Phospholipids (mg/dl)
Normal control (2ml normal saline)	180.2±1.15	127.49±0.94	9.27±0.35
Diabetic control (150mg/kg Alloxan & 2ml NS)	238.7±2.9	152.38±2.5	12.27±0.55
Diabetic rat given AL (200 mg/kg)	215.2±1.4**	142.88±1.2*	10.93±0.36*
Diabetic rat given AL (400 mg/kg)	190.6±0.99*	130.38±0.91**	8.68±0.41**
Diabetic rat given GLB (10 mg/kg)	185.08±1.79**	126.24±1.85**	11.02±0.13**

Table 2: Effect of roots extract of *Alangium lamarckii* on serum HDL, LDL and VLDL of control and experimental rats

Groups	HDL (mg/dl)	LDL (mg/dl)	VLDL(mg/dl)
Normal control (2ml normal saline)	53.28±1.8	101.42±1.6	25.5±1.5
Diabeticcontrol(150mg/kg Alloxan & 2ml NS)	33.71±1.2	164.56±1.8	30.48±1.4
Diabetic rat given AL (200 mg/kg)	37.09±1.6*	148.61±0.8**	25.58±1.8**
Diabetic rat given AL (400 mg/kg)	45.09±1.8**	119.37± 1.5**	26.08±2.5*
Diabetic rat given GLB (10 mg/kg)	48.21±1.18**	110.62±1.1**	24.25±2.6**

Table 3: Effect of roots extract of *Alangium lamarckii* on ALP, SGOT and SGPT of control and experimental rats

Groups	ALP(IU/L)	SGOT (IU/L)	SGPT(IU/L)
Normal control (2ml normal saline)	285.26±2.63	34.16±0.80	41.76±0.49
Diabetic control(150mg/kg Alloxan & 2ml NS)	552.76±1.71**	71.52±0.52**	88.52±1.88**
Diabetic rat given AL (200 mg/kg)	371.54±1.16**	62.36±0.89**	73.00±1.58**
Diabetic rat given AL (400 mg/kg)	283.60±2.26ns	35.12±2.88ns	40.35±1..54ns
Diabetic rat given GLB (10 mg/kg)	358.12±0.82**	56.37±1.60**	68.53±1.42**

CONCLUSION

The roots extraction has been performed by sequential extraction method The roots *of Alangium lamarckii using* the solvent with increasing polarity order (petroleum ether, ethyl acetate and ethanol) and the active extract was tested by *in vitro* antidiabetic screening method. The *in vitro* antidiabetic study have been performed based on the α-amylase inhibition assay. Each extracts were tested for α- amylase inhibition and the extract with minimum IC_{50} have been undergone phytochemical screening. The preliminary phytochemical tests was performed to identify the active phytochemicals present in the ethanolic extract of *Alangium lamarckii* and showed the presence of phenols, flavanoids, alkaloids, glycosides, saponins and terpenoids. The *in-vivo* antidiabetic activity of Ethanolic extract of *Alangium lamarckii* root was tested by using an Alloxan induced diabetic rat. Acute toxicity study was carried out in rats. The procedure was followed by OECD 423 (Acute toxicity class method). $1/5^{th}$ (400mg/kg) of the maximum safe dose (2000mg/kg) were selected for further study. Fasting blood sample were drawn from tail vein of rats at weekly intervals till the end of the study 1, 7 and 14 days. On these days fasting blood glucose were collected and analysed for glucose. At the end of the study (14^{th} day) the ethanolic extract of *Alangium lamarckii* root (400 mg/kg p.o) treated diabetic groups showed statistically significant decrease in blood glucose similar to the standard drug glibenclamide (2mg/kg), which indicated block the alfa amyalase activity. The present study suggested that the isolation of active constituents from ethanolic extract of *Alangium lamarckii* root and characterize the compounds by using preliminary phytochemical studies.

DECLARATION OF INTEREST

The authors declare no conflicts of interests. The authors alone are responsible for the content and writing of this article.

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