ANTIDIABETIC ACTIVITY OF *Astercantha longifolia ROOT* EXTRACT IN STREPTOZOTOCIN INDUCED DIABETIC RATS

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Abstract

Diabetes is a life threatening disease now days irrespective of large number of modern medicines for its treatment. Still many people in rural belt of India rely on traditional healing practices and medicinal plants for their daily healthcare needs. The floral biodiversity of Chhattisgarh state of India provide traditional healers with an impressive pool of natural pharmacy practice. So scientific evidence of these medicines is required. Arurvedic system placed Astercantha longifolia plant in a special position as seethaveeryam, mathuravipaka and used for the treatment of diabetes. So antidiabetic potential of this plant evaluated on streptozotocin induced diabetes model. Preliminary phytochemical study reveals that aqueous extract of AL contains alkaloids, glycosides, sterols and flavonoids. Based on the importance of flavoids total phenolic content was determined by FC reagent method. This reveals that each mg of gallic acid is equivalent to one gram of the root powder, so there is satisfactory content of flavonoids. Aqueous extract of AL was administered at two different dose of 100mg/kg and 200mg/kg to diabetic rats caused significant ($P < 0.001$) reduction of blood glucose levels which was related to dose and duration of treatment. Maximum reduction was observed on day 21 (20.21% and 42.36%, respectively). AL 200 mg/kg exhibited maximum glucose lowering effect in diabetic rats comparable to that of glibenclamide 50.22% reduction in blood glucose levels at the end of the study when compared to diabetic control.

Keywords: Astercantha longifolia, treatment, diabetes model.

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1. Introduction

There is immense technological advancement in modern medicine for the treatment of various diseases. Now a day's diabetes is a major problem in human population irrespective of sex and age. There are various synthetic medicines available for the treatment of diabetes [1]. Still many people in rural belt of India rely on traditional healing practices and medicinal plants for their daily healthcare needs. The floral biodiversity of Chhattisgarh state of India provide traditional healers with an impressive pool of natural pharmacy practice. From which plants are selected as remedies, or as ingredients to prepare herbal medicines, with out scientific evidence. *Astercantha longifolia* is one of the plants used by traditional healers of Chhattisgarh for the treatment of various diseases [2]. Arurvedic system placed this plant in a special position as seethaveeryam, mathuravipaka and used for the treatment of premeham (diabetes) [3]. Scientific evidence has been established for the antidiabetic activity of leaf extracts. From literature survey it has been concluded that there no scientific evidence about antidiabetic activity of roots of *Asteracantha longifolia.* So a suitable plan has been designed for the evaluation of antidiabetic activity of aqueous extract of root.

2. Materials and Methods

2.1. Collection of Plant material

Astercantha longifolia root and whole plant was collected from the tribal belt of Pendra region of Chhattisgarh and Identified by botanist. A voucher specimen was stored in department of Pharmacognosy, GGV, Bilaspur. The roots collected dried under shade and grinded with the help of ball mill. The powder is stored in air tight container for further use.

2.2. Extraction of Plant Material

About 200 gm of dried root powder was extracted with water by maceration for 7 days with occasional shaking and addition of ethanol for preventing from fungal growth. The aqueous extracts were filtered and concentrated to dryness under reduced pressure. The resulting aqueous extract was freeze-dried, finally giving 9.24 g (i.e., 4.62% yield) of a greenish-brown, powdery crude aqueous extract of *Astercantha longifolia root.* The root extract was stored in air tight container for further use.

2.3. Preliminary Phytochemical Screening

Preliminary phytochemical screening was under taken with the aqueous extract as per the method of Kokate, 1994 [4]. This is followed by total phenolic contant.

2.4. Estimation of Total Phenolic Content

Total phenolic content was determined colorimetrically with Folin-Ciocaletu (FC) reagent method. 0.5 ml of 0.1% aqueous dilution of extract, 2.5 ml of freshly prepared 0.2M FC reagent and 2ml of sodium carbonate solution were mixed and kept in the dark for 30 min. Absorbance of the resulting solution was measured at 760 nm. Total phenolic content was expressed as mg of gallic acid equivalents (GAE) per gram of extracts, using a standard curve of gallic acid. Observation was carried out in triplicate [5].

2.5. Experimental animals

Adult male albino Wistar rats (180–200 g) obtained from Indian Institute of Chemical Biology (IICB), Kolkata, were maintained at the Animal House of Institute of Pharmacy, GGv, Bilaspur used for the study. Rats were housed in standard environmental conditions of temperature (22±2℃) and humidity (55±5℃) and a 12/12h light dark cycle. The animals had free access to standard laboratory diet and tap water ad libitum under hygienic conditions. Each experimental group had separate set of animals and care was taken to ensure that animals used for one response. Animals were habituated to laboratory conditions for at least 48-72 hours prior to experimental protocol to minimize if any of non-specific stress.

2.6. Acute oral Toxicity study

Acute oral toxicity was studied in a group of six animals with a single dose of 2000mg/kg as single dose as per OECD guideline 423. Animals were observed for 14 days for mortality and any physiological change.

2.7. Experimental design

Antidiabetic activity was assessed in normal and streptozotocin-induced diabetic rats. Animals were fasted overnight for 16 h with free access to water throughout the duration of the experiment. Diabetes was induced by single intraperitoneal injection of 55 mg/kg of streptozotocin (STZ), dissolved in citrate buffer, pH 4.5. Control animals received only citrate buffer. Animals with fasting blood glucose above 250 mg/dl after 5 days of STZ injection were considered as diabetic and included in the study.

2.8. Evaluation of extract in streptozotocin-induced diabetic rats

Animals were divided into five groups randomly. Group I considered as Normal control received only distilled water, Group II as Diabetic control i.e diabetic rats received only distilled water, Group III as treated I i.e diabetic rats received 100mg/kg of extract, Group IV as treated II i.e diabetic rats treated with 200 mg/kg of extract, Group V: Diabetic Standard i.e diabetic rats received 10 mg/kg of glibenclamide. The freshly prepared extract solutions were orally administered daily for 21 days. Body weights and blood glucose analysis was done weekly on overnight fasted animals.

3. Result and Discussion

Preliminary phytochemical study reveals that aqueous extract of AL contains alkaloids, glycosides, sterols and Flavonoids. Based on the importance of flavoids total phenolic content was determined by FC reagent method. This reveals that each mg of gallic acid is equivalent to one gram of the root powder, so there is satisfactory content of flavonoids. Acute oral toxicity study was conducted as per the OECD guideline 423. Highest dose of 2000mg/kg of extract was treated. There is no observation of death or any marked physiological change. So dose of $1/10^{th}$ (200mg/kg) and $1/20^{th}$ (100mg/kg) was selected for study in streptozotocin induced rats. The effect of repeated oral administration of extract on blood glucose levels in STZ-diabetic rats is presented in Figure-1.

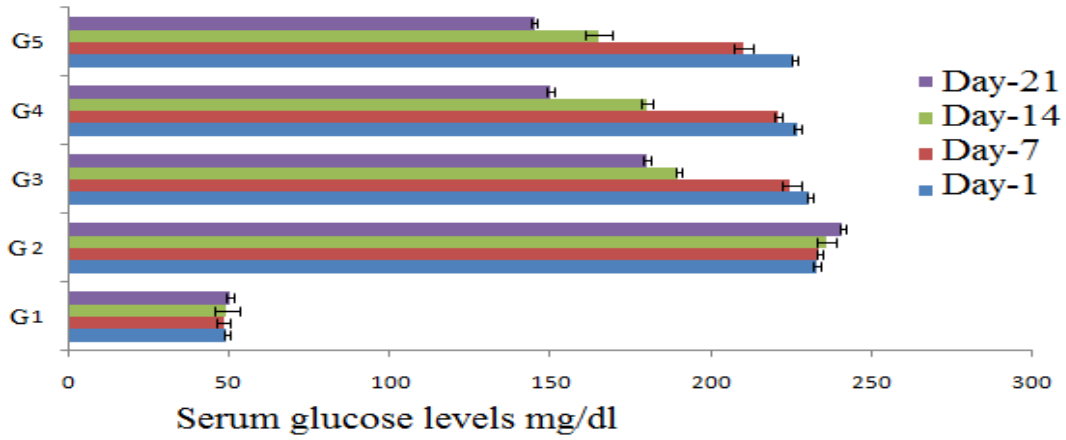

Figure-1: Effect of AL on blood glucose levels of diabetic rats, Each value is expressed as mean±S.E.M. (n = 6)

Aqueous extract of AL was administered at two different dose of 100mg/kg and 200mg/kg to diabetic rats caused significant ($P < 0.001$) reduction of blood glucose levels which was related to dose and duration of treatment. Maximum reduction was observed on day 21 (20.21% and 42.36%, respectively). AL 200 mg/kg exhibited maximum

glucose lowering effect in diabetic rats comparable to that of glibenclamide 50.22% reduction in blood glucose levels at the end of the study when compared to diabetic control. STZ produced significant loss in body weight as compared to normal animals during the study. Diabetic control continued to loose weight till 21st day while AL at the dose level of 200mg/kg and 100 mg/kg showed significant improvement. The hypoglycemic activity of AL was compared with glibenclamide, a standard hypoglycemic drug. The mode of action of STZ is, it induces hyperglycemia accompanied by hypoinsulinemia [6]. Oral administration of AL for 21 days caused a significant decrease in blood glucose levels. The possible mechanism by which AL acts as antidiabetic effect could be by potentiation of pancreatic secretion of insulin from existing β-cells of islets. Further diabetes is associated with, tissue damage mediated by free radicals by attacking membranes through peroxidation of unsaturated fatty acids [7]. Previous study by various researchers suggests that alteration in antioxidant enzyme status leads to extensive membrane damage and dysfunction [8]. Preliminary Phytochemical screening and total polyphenol estimation suggests that Al is rich with polyphenols. So antidiabetic activity of AL could be due to the antioxidant effect of flavonoids.

4. Conclusion

Aqueous extract of Al shows as a promising herbal antidiabetic agent, although this study is not enough to provide the possible mechanism of action. Although we have taken glebenclamide as standard drug and the antidiabetic activity of Al is comparable as that of glebenclamide still we can not conclude the mode of action of AL is similar to that of glebenclamide. So further study is required to establish its mechanism of action.

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