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ANTDIABETIC ACTIVITY OF BARK EXTRACTS OF *PTEROCARPUS SANTALINUS* L. IN STREPTOZOTOCIN INDUCED DIABETIC RATS

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ABSTRACT
After induction of diabetes aqueous extract of aquous extract of bark of *Pterocarpus santalinus* has been given to the treated rats to establish the protective role. Different doses *Pterocarpus santalinus* were evaluated for hypoglycemic activity in normal and streptozocin diabetic rats. The oral administration of aquoues extract at a dosage of 0.7 g/kg body weight exhibited a significant antidiabetic activity in STZ diabetic rats, whereas in normal rats no hypoglycemic activity was observed.

INTRODUCTION

Diabetes mellitus is a chronic metabolic disease caused by an absolute or relative lack of insulin and or reduced insulin activity, which results in hyperglycemia and abnormalities in carbohydrate, protein and fat metabolism. Though different types of oral hypoglycemic agents are available along with insulin for the treatment of diabetes mellitus, there is a growing interest in herbal remedies due to the side effects associated with these therapeutic agents (Grover *et al.*, 2002). The investigation of anti-diabetic agents of plant origin which are used in traditional medicine is thus of great importance. The bark of *Pterocarpus santalinus* Linn. has been used in the indigenous systems of medicine in India for the treatment of several diseases. The plant is renowned for its characteristic timber of exquisite color, beauty and superlative technical qualities and ranks among finest luxury in Japan (Keshav Reddy and Srivasuki, 1990). The red wood yields a natural dye santalin, which is used in colouring pharmaceutical preparations and foodstuffs. The wood is used as an astringent and tonic for external application in inflammation; it is also used in treating headache, skin diseases, fever, boils, scorpion sting and to improve sight. The wood and fruit is used in treating diaphoretics, bilious infections and chronic dysentery (Krishnaveni and Srinivasa Rao, 2000). Critical review of the literature revealed that *Pterocarpus santalinus* L. has remained unexplored for many pharmacological activities claimed. The present investigation was undertaken to study the anti-diabetic effects of the aqueous bark extracts of *Pterocarpus santalinus* L. on streptozotocin (STZ)induced diabetic rats.

MATERIALS AND METHODS

Male wistar albino rats (weighing 160-200g) were procured from the Animal house, Indian Institute of Chemical Technology, Hyderabad under standard environmental conditions (12 hr light/dark cycles at 25-28 C, 60-80% relative humidity). They were fed with a standard diet (Hindustan Lever, India) allowed to acclimatize for 14 days before the procedure. The bark of *Pterocarpus santalinus* L. was collected from the hills near to the Nujivid. The dried bark of *Pterocarpus santalinus* L. was ground in to fine powder with auto-mix blender. Then the fine powder was suspended in equal amount of water and stirred intermittently and left overnight. The macerated pulp was then filtered through a coarse sieve and the filtrate was dried at reduced temperature. This dry mass (yield 185g/kg of powdered bark) served as aqueous extract of *Pterocarpus santalinus* L. for experimentation. Streptozotocin (STZ), purchased from Sigma Chemical Co. (Saint Louis, MO, USA) was dissolved in 0.1M ice-cold citrate buffer, pH 4.5, immediately before use. Six rats per group were administered STZ (60 mg/kg) by subcutaneous injection. After 48hr, fasting blood glucose levels as well as glycosuria were assessed to confirm the diabetic state. Only rats with a fasting blood glucose level of at least 250 mg/dL and positive urine glucose were considered diabetic and used in the experiment. Male Wistar albino rats weighing 180-250g were used. The animals were randomly divided into six groups of six animals each.

The rats in group II and V were given a daily oral dose of 0.35 g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark while group III and VI were given 0.7 g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark. All 6 groups of rats were sacrificed on last day (20^{th} day) of treatment by cervical dislocation and then blood and liver were collected for biochemical estimations. Body weights of all the animals

were recorded prior to the treatment and sacrifice. Blood glucose level was determined using a glucometer Accutrend GC^R (Boerhinger Mannheim, Germany) before and 48 h after STZ administration, for the confirmation of the diabetic state of animals. Glycated haemoglobin was estimated by the method of Eross *et al.*, (1984). The estimation of glycogen in tissues was carried out by the method of Kemp and Van Hejnigen (1954).

Group-I:	Normal rats
Group_II:	Normal rats treated with 0.35g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark.
Group-III:	Normal rats treated with 0.7 g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark.
Group-IV:	Diabetic untreated rats.
Group-V:	Diabetic treated rats treated with 0.35g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark.
Group-VI:	Diabetic treated rats treated with 0.7 g/kg body weight of aqueous extract of *Pterocarpus santalinus* bark.

RESULTS AND DISCUSSION

After sacrificing the rats which were given treatment with *Pterocarpus santalinus* aqueous extract for 20days different parameters mentioned in materials and methods were analyzed. After the treatment, body weight was measured; blood is collected and used for measuring glucose, glycated hemoglobin and glycogen content. Table 1 depicts the changes in the body weights of rats and fasting glucose levels after treatment in all 6 groups of rats. The concentration of glucose in the control sample is 82 mg /dL. Due to the induction of diabetes it has raised to the 265 mg/dL. The increased concentration of plant extract brought the glucose level to 101 mg/dL. Similarly there is a decrease in the body weight by 28g in diabetic animal whereas the weight gain was noticed by 20 g in normal rats. The treatment of bark aqueous extract brought the loss to 10g in the body weight. In control sample which is treated with plant extract maintained the equal weight of animal in comparision with control. Glycated hemoglobin is an indicator of the progression of diabetes. Therefore glycated hemoglobin was measured in the control and diabetic samples. In the control sample the percentage of glycosylated hemoglobin was observed to be 7.15. This induction of diabetes in the rats brought enhancement in the glycosylated hemoglobin (Table 2) from 7.15 to 12.4. This increase of glycosylated hemoglobin serves as a marker to know the induction of diabetes. The treatment of aqueous extract of PS in diabetic rats maintained the original levels of glycosylated hemoglobin equal to that of control ones. Since there is a relation between glucose content and glycogen metabolism attempts have been made to estimate the amount of glycogen in liver tissue per gram of wet tissue. In control sample 11.56 mg of glycogen (glucose equivalents) was observed. In diabetic rat sample the content was reduced to 8.3 mg. This decrease in the glycogen content could be due to utilization of glucose. The PS aqueous extract treatment brought the normal levels of glycogen in diabetic rats (Table 3). The treatment of PS aqueous extract to control ones could not influence the glycogen content.

Table 1: Levels of glucose and changes of body weights of normal, normal treated, diabetic and diabetic treated rats

Group	Glucose mg/dl	Change in Body weight
I	82± 8.1	+20.1±5.1
II	80±8.1	+20.1±4.1
III	78±7.4	+20.2±5.2
IV	265±10.1	-28.0±8.2
V	125±10.1	-18.5±7.1
VI	101±10.2	-10.±7.1

Our results suggest that the aqueous bark extracts of *Pterocarpus santalinus* have dose-dependent anti-diabetic activities on STZ-induced diabetes. The metabolic disturbances were corrected after the bark extracts were administered for 2 weeks, as shown by the normalisation of fasting blood glucose levels, glycolated Hb, glycogen content and weight gain by diabetic-treated rats. The bark extracts appeared to have greater potency than reference drug in reducing the body weight, food and water intakes but was equipotent in blood sugar reductions.

The mechanisms by which STZ brings about its diabetic state include selective destruction of pancreatic insulin secreting beta cells, which make cells less active (Junod *et al.,* 1969; Jacot and Assal, 1989) and lead to poor glucose utilisation by tissues (Marles and Farnsworth, 1995). The aqueous bark extracts of *Pterocarpus santalinus* significantly reduced the high fasting glucose levels in stz-induced diabetic rats. This suggests that the extracts may possess an insulin like effect on peripheral tissues by either promoting glucose uptake and metabolism, by inhibiting hepatic gluconeogenesis (Ali *et al.,* 1993; Gray *et al.,* 2000) or absorption of glucose into the muscles and adipose tissues (Kamanyi *et al.,* 1994), by the stimulation of a regeneration process and revitalization of the remaining beta cells (Shanmugasundaram *et al.,* 1990; Rokeya *et al.,* 1999; Bolkent *et al.,* 2000). Thus the aqueous bark extracts of *Pterocarpus santalinus* possess anti-diabetic properties. However, we suggest that further work should be

Table 2: Effect of PS aqueous extract on the levels of glycated hemoglobin in different experimental animals

Groups	Glycated hemoglobin
I	7.15±0.6
II	7.16±0.7
III	7.18±0.6
IV	12.4±0.8
V	6.5±0.5
VI	7.13±0.6

Table 3: Effect of PS aqueous extract on the glycogen content of different experimental animals

Groups	Glycogen(mg)
I	11.56±0.9
II	11.45± 0.8
III	11.49±0.8
IV	8.3 ±0.5
V	9. 8±0.8
VI	11.25±0.9

carried out at molecular level to find out the absolute mechanism of action of the bark of *Pterocarpus santalinus* in experimental diabetes.

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