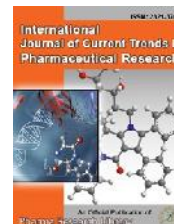




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Research Article

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Hypoglycaemic and Hypolipidemic Activity of Aqueous Extract of *Bauhinia Purpurea* Flower in Streptozotocin-Induced Diabetic Rats

Md. Sultan Ali, K. Suresh*, Y. Suresh, Vijaya Lakshmi

Safa College of Pharmacy, Kurnool, Andhra Pradesh, India-518218

ABSTRACT

The aqueous extract of *Bauhinia purpurea* flower (AEBPF) was tested for its hypoglycaemic and hypolipidemic activity by sub acute treatment method in streptozotocin-induced diabetic rats. Blood glucose levels were determined after oral administration of two concentrations of AEBPF (200 and 400 mg/kg) at 0th, 10th and 15th day and biochemical parameters like glycosylated haemoglobin, serum protein, serum creatinine and lipid profile were estimated after 14 days of sub acute study. The results revealed that in sub acute treated groups administration of a low dose of 200mg/kg reduced blood glucose level with less significance ($p < 0.05$) but a high dose of 400mg/kg reduced blood glucose significantly ($p < 0.01$). In addition, changes in body weight, glycosylated haemoglobin, serum protein, serum creatinine and serum lipid profile levels assessed in the extract treated diabetic rats were compared with standard control, diabetic control and normal animals in sub acute treated groups. Significant results were observed in the estimated parameters, thereby justifying the use of the plant in the indigenous system of medicine.

Keywords: Bauhinia purpurea flower, Hypoglycaemic activity, Hypolipidemic activity, Streptozotocin; Lipid profile.

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*Corresponding Author

K. Suresh
Safa College of Pharmacy,
Kurnool, Andhra Pradesh, India-518218
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1. Introduction

Bauhinia purpurea (Orchid tree, Camel's foot tree) is a plant belongs to a family fabaceae. It is a small to medium-sized deciduous fast-growing shrub or tree. The stem & bark of *Bauhinia purpurea* contains chemical constituents

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like carbohydrates, glycosides, Saponins, sterols and triterpenoids were present in aqueous extract [1]. Literature revealed that the plant having anti tumour activity, anti diabetic activity, anti inflammatory activities, analgesics, anti ulcer activity, antioxidant, anti hyperlipidemic activity, anti eosinophilic anthelmintic activity, nephroprotective activity immunomodulatory activity and hepatoprotective activity 2.

However, the hypoglycaemic and antidiabetic potentials of the flower part have not been scientifically evaluated despite the extensive use of the plant in the management of diabetes in traditional medicine. The present study was, therefore, designed to evaluate the dose- dependent hypoglycaemic and antidiabetic effects of the aqueous flower extract in normal, glucose fed hyperglycaemic and streptozotocin- induced diabetic rats.

2. Materials and Methods

Plant material

The flowers were collected during the month September to November and the plant was authenticated by a botanist. Then it was shade dried, powdered, weighed and stored in a clean, dry and air tight container.

Aqueous extract preparation

Powder was packed in a condenser and extracted with distilled water at 100°C by Soxhlet extractor. After extraction the residue was dried on water bath at 100°C to get a solid mass. The percentage yield of the aqueous extract of *Bauhinia purpurea* flower (AEBPF) was calculated [3]

Animals

Inbred adult wistar albino rats (150-200 g) of either sex were selected. The animals were maintained in a well-ventilated room with 12:12 hour light/dark cycle in polypropylene cages. Standard pellet fed and tap water was provided *ad libitum* through out experimentation period. Animals were acclimatized to laboratory conditions one week prior to initiation of experiments. Fasting refers to that the animals were deprived of food for 16 hours but were allowed to free access for water.

Determination of blood glucose levels

Blood was collected from tip of the tail vein and fasting blood glucose level (mg/dl) was measured using single touch glucometer (Ascensia ENTRUST, Bayer) based on glucose oxidase method.

Study on diabetic rats:

Induction of diabetes

Adult inbred wistar albino rats (32 numbers) of either sex were over night fasted and received a freshly prepared solution of streptozotocin (STZ), [Sigma Chemical Co, St Louis, MO, USA], (45 mg/kg) in 0.1 M sodium citrate buffer, PH 4.5, injected intraperitoneally in a volume of 1 ml/kg. After injection the animals had free access to food and water and were given 5% glucose in their drinking water for the first 24 hours to counter any initial hypoglycemia. Normal rats (6 numbers) received 1ml citrate buffer as vehicle. The development of diabetes was confirmed after 48 hours of the streptozotocin injection. The animals with fasting blood glucose level more

than 200 mg/dl were selected for the experimentation. In the present study, glibenclamide (0.4 mg/kg) was used as the standard drug [4].

Subacute antidiabetic effect of test samples

The animals were divided into 5 groups. Group I consists of normoglycaemic rats and the remaining 4 groups consisted of 6 STZ induced diabetic rats. The mentioned groups were treated orally as follows: Group I -Normal control (0.5% CMC 5ml/kg), Group II- Disease control (0.5% CMC 5ml/kg), Group III-Standard control (glibenclamide 0.4mg/kg) Group IV and Group V- Test control (AEBPF of 200 and 400 mg/kg).

The above mentioned treatment schedule was followed for the respective group of animals for 14 days. Body weight changes were measured for overnight fasted animals on 0th, 10th and 15th day of study and the blood samples were collected at the same days to estimate blood glucose levels using glucometer [5]. At the end of the study, all the animals were sacrificed under light ether anaesthesia. The rats were sacrificed by decapitation and blood was collected by bleeding of carotid artery and serum was separated to study various biochemical parameters like glycosylated haemoglobin (Excel diagnostics pvt Ltd), serum protein (Beacon diagnostics Ltd), total cholesterol (Span diagnostics Ltd), HDL-cholesterol (Span diagnostics Ltd), triglycerides (Span diagnostics Ltd), LDL-cholesterol, VLDL-cholesterol [6, 7].

Statistical analysis

Values are presented as means \pm S.E.M. Statistical difference between the treatments and the controls were tested by one-way analysis of variance (ANOVA) followed by Dunnett's test using 7.5 version of SPSS computer software. The values were considered significant when $P < 0.05$.

3. Results and discussions

Sub-acute effect of AEBPF on body weight and blood glucose level in STZ induced

The AEBPF at oral dose level of 200mg/kg do not show significant improvement in the body weight of STZ induced diabetic rats up to the 10th day of treatment and showed a slight significance in the body weight improvement on 15th day ($P < 0.05$). An oral dose of 400mg/kg and standard drug glibenclamide (0.4mg/kg) shows significant ($P < 0.01$) improvement in the body weight of STZ induced diabetic rats on 10th day and 15th day of treatment. Treatment with AEBPF 200mg/kg showed less significant result ($P < 0.05$) in the reduction of the blood glucose level on 10th day compared to a high dose of 400mg/kg and standard ($P < 0.01$). Treatment with AEBPF of both doses and standard produced a significant ($P < 0.01$) drop in blood glucose level on 15th day of Sub acute study. Results are shown in Table 1.

Sub-acute effect of AEBPF on biochemical parameters in STZ induced diabetic rats

The diabetic control rats showed significant increase in the glycosylated haemoglobin (GHb%), total cholesterol(TC),

triglycerides (TG), low density lipoproteins (LDL), very low density lipoproteins (VLDL), serum creatinine and a significant decrease in serum total protein and high density lipoprotein (HDL) levels when compared to normal control rats. GHb % level in AEBPF (200mg and 400mg/kg) treated diabetic rats decreased less significantly ($P<0.05$) compared to that in rats treated with standard ($P<0.01$). The serum total protein levels in AEBPF treated diabetic rats with doses of 200mg and 400mg/kg showed significant [$(P<0.05)$ & $(P<0.01)$] increase respectively. Glibenclamide (0.4mg/kg) also showed significant ($P<0.01$) results. Serum total cholesterol levels of diabetic animals treated with both the doses of AEBPF (200 and 400mg/kg) and standard showed significant ($p<0.01$) decrease in cholesterol level when compared to disease control group. Diabetic rats treated with standard and AEBPF of dose 200mg/kg showed less significance ($P<0.05$) in the reduction of serum HDL levels compared to that of those treated with AEBPF of dose 400 mg/kg ($P<0.01$). Triglycerides and LDL levels of diabetic rats treated with AEBPF of 200mg/kg were reduced less significantly ($P<0.05$) compared to that of rats treated with AEBPF of 400mg/kg and standard ($P<0.01$). In diabetic rats treated with AEBPF (200 and 400mg/kg) and standard, VLDL and serum creatinine levels were decreased significantly ($P<0.05$) compared to that of disease control group. Results are shown in Table 2.

Discussion

The present paper discussed about the antidiabetic effect of the aqueous extract of *Bauhinia purpurea* flower on streptozotocin induced diabetic rats. In the sub-acute study, induction of diabetes with STZ is associated with the characteristic loss of body weight, which is due to increased muscle wasting [8] and due to loss of proteins [9]. Diabetic rats treated with the AEBPF showed an increase in body weight as compared to the diabetic control, which may be due to its protective effect in controlling muscle wasting i.e. reversal of gluconeogenesis. Glibenclamide treatment brought down the sugar levels from the first day of the treatment. AEBPF (200 & 400mg/kg) treatment produces significant reduction in

blood glucose levels from 10th day of treatment and a steady decrease was observed there after.

Increased non-enzymatic and autooxidative glycosylation is one of the possible mechanisms linking hyperglycaemia and vascular complications of diabetes. In the present study diabetic rats had shown higher levels of HbA_{1c} compared to those in normal rats indicating their poor glycaemic control [10]. Treatment with AEBPF, showed a significant decrease in HbA_{1c} levels in diabetic rats. This property provides a practical and objective means of assessing average blood glucose levels over a time frame of about 2 months and has proven to be a very useful adjunct to self monitoring of blood glucose (SMBG) [11]. Excessive break down of body protein in conjunction with either inadequate supply or defective utilization observed in uncontrolled diabetes may be accompanied by hypoalbuminemia [12]. AEBPF seems to resort this effect due to the hypoglycaemic status [13].

Myocardial infarction, caused by atherosclerosis of the coronary arteries, is the most common cause of death in diabetics. Hypercholesterolemia and hypertriglyceridemia have been reported to occur in diabetic rats [14,15,16]. It is well known that the level of glycaemic control is the major determinant of serum level of very low density lipoprotein (VLDL). Several investigations demonstrated that near normalization of the blood glucose level resulted in significant reductions in levels of plasma cholesterol, triglycerides, free fatty acids and plasma protein. In the present study elevated serum total cholesterol, triglycerides, LDL-levels, VLDL-levels and reduced HDL levels were observed in STZ-induced diabetic rats. AEBPF (200mg/kg and 400 mg/kg) treatment in diabetic animals produced beneficial improvement in the lipid profile which showed a hypolipidemic effect in diabetic rats. Increased serum creatinine in diabetic rats indicates cardiac muscular damage. Elevated concentrations of serum creatinine were recovered by the treatment with AEBPF of both concentrations & standard glibenclamide suggesting their cardio protective effect. [17]

Table 1: Effect of sub acute treatment of aqueous extract of *Bauhinia purpurea* flower (AEBPF) on body weight changes and blood glucose level in STZ induced diabetic rats

Group	Treatment	Dose ¹ (Kg Body Weight)	Parameter gm & mg/dl	Day of measurement		
				0 Day	10 th Day	15 th Day
I	Control (0.5% SCMC)	5 ml	Body weight Blood glucose	193.45 ± 1.24 76.37 ± 1.6	197.42 ± 1.2 78.33 ± 2.5	211.5 ± 1.5 83.5 ± 5.5
II	Disease control (STZ)	45mg	Body weight Blood glucose	211.24 ± 0.99 246.83 ± 4.1	163.89 ± 1.4** 269.0 ± 6.9**	152.5 ± 0.7** 289.3 ± 5.3**
III	Standard (Glibenclamide+STZ)	0.4mg	Body weight Blood glucose	182.13 ± 2.64 234.0 ± 5.2	189.47 ± 3.2* 165.30 ± 4.5**	190.9 ± 1.5** 109.60 ± 3.2**
IV	Test I (AEBPF+STZ)	200mg	Body weight Blood glucose	194.62 ± 4.3 221.43 ± 4.3	169.6 ± 5.4 ^{ns} 182.16 ± 3.78*	172.20 ± 4.2* 134.16 ± 4.7**
V	Test II (AEBPF+STZ)	400mg	Body weight Blood glucose	188.92 ± 1.7 224.36 ± 3.1	179.60 ± 1.3* 171.33 ± 4.6**	185.60 ± 4.1** 167.17 ± 5.3**

Group II is compared with Group I. Groups III, IV, V are compared with group II. ** $P<0.01$, * $P<0.05$, ns- non significant

Table 2A: Effect of sub acute treatment of aqueous extract of *Bauhinia purpurea* flower (AEBPF) on various biochemical parameters in STZ induced diabetic rats

Group	Treatment	Dose ⁻¹ (Kg Body Weight)	Glycosy-lated haemoglobin (GHb %)	Serum total protein (mg/dl)	Total Cholesterol (mg/dl)	HDL (mg/dl)
I	Control (0.5% SCMC)	5 ml	7.317 ±0.75	6.405±0.14	119.3 ± 5.4	42.99 ±2.6
II	Disease control (STZ)	45mg	12.850±0.39**	3.81±0.13**	163.4±3.61**	29.37 ±1.9**
III	Standard (Glibenclamide+STZ)	0.4mg	7.80±0.65*	5.80±0.25**	122.7±3.8**	35.75 ±3.0*
IV	Test I (AEBPF+STZ)	200mg	8.45 ±0.50*	5.18±0.54*	125.3 ±3.46**	34.74 ±3.3*
V	Test II (AEBPF+STZ)	400mg	8.32 ±0.59*	5.28±0.5**	143.8 ±4.16**	39.03 ±2.4**

Table 2B: Effect of sub acute treatment of aqueous extract of *Bauhinia purpurea* flower (AEBPF) on various biochemical parameters in STZ induced diabetic rats

Group	Treatment	Dose ⁻¹ (Kg Body Weight)	Trigly cerides (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)	Creatinine (mg/dl)
I	Control (0.5% SCMC)	5 ml	141.0 ± 3.5	49.52 ±5.0	29.40 ±0.69	0.71 ±0.04
II	Disease control (STZ)	45mg	186.0 ± 3.9**	104.50 ±5.3**	38.61±0.77**	1.70±0.05**
III	Standard (Glibenclamide+STZ)	0.4mg	154.0 ± 2.9**	52.54 ±5.2**	33.41 ±0.60*	1.22 ±0.04*
IV	Test I (AEBPF+STZ)	200mg	167.6 ± 3.2*	85.36 ±3.5*	32.72 ±0.64*	1.34 ±0.03*
V	Test II (AEBPF+STZ)	400mg	151.6 ± 3.0**	56.87 ±3.0**	31.44 ±0.60*	1.28 ±0.04*

Group II is compared with group I and Group III, IV and V are compared with Group II. **P<0.01 *P<0.05

4. Conclusion

From this we can state that aqueous extract of *Bauhinia purpurea* flower has beneficial effects on blood glucose levels as well as improving hyperlipidaemia due to diabetes through its hypolipidemic action. Further studies are required to establish the hypoglycaemic activity of *Bauhinia purpurea* flower in terms of molecular mechanism(s) involved in the activity.

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