

## Antidiabetic Activity of Hydroethanolic Leaf Extract of *Ixora macrothyrsa* (Tejism. & Binn.) on Streptozotocin Induced Diabetic Rats



### Biochemistry

**KEYWORDS :** Hydroethanolic, *Ixora macrothyrsa* (IM), Streptozotocin.

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### ABSTRACT

*In the present work antidiabetic activity of *Ixora macrothyrsa* was evaluated. The hydroethanolic leaf extract showed significant decrease in the levels of Glucose, Urea, Creatinine, Total cholesterol, Triglyceride, LDL, VLDL and increase in the level of HDL in Streptozotocin induced diabetic rats at a dose of 200 and 400 mg/kg body weight when administered orally for 30 days. This implies that the Phytochemicals present in the leaf extract might exert a potential in controlling lipid profile, blood glucose, Urea, Creatinine levels and lipid profile in the body.*

### Introduction

Diabetes is a complex and multifarious group of disorders characterized by hyperglycaemia that has reached epidemic proportions in the present century<sup>1</sup>. Insulin stimulates muscle and fat cells to remove glucose from the blood and stimulates the liver to metabolize glucose, causing the blood sugar level to decrease to the normal levels<sup>2</sup>. Management of diabetes without any side effects is still a challenge to the medicinal community. There is a continuous search for alternative drugs. Therefore, it is prudent to look for options in herbal medicine for diabetes<sup>3</sup>. Furthermore, after the recommendation made by WHO on diabetes mellitus, investigation on hypoglycaemic agents from medicinal plants have become more important<sup>4</sup>.

*Ixora coccinea* Linn is a small shrub which is cultivated throughout India. Its roots & flowers are used for the treatment of dysentery, dysmenorrhoea, leucorrhoea, haemoptysis, catarrhal bronchitis, hiccups, nausea, loss of appetite, chronic ulcers and externally for sores and eczema. Its leaves are used for the treatment of diarrhoea<sup>5</sup>.

In view of this, the current study was designed to evaluate the antidiabetic activity of *Ixora macrothyrsa* on Streptozotocin induced diabetic rats.

### Materials and methods

#### a) Plant collection and extraction

IM was collected from Coimbatore, Tamil Nadu. They were identified and certified by the Taxonomist, Botanical Survey of India (BSI), Coimbatore, TamilNadu, India. The shade dried leaf was ground to coarse powder. The coarse powder was extracted using 50% ethanol. The extracts were condensed to dryness using rotatory evaporator and crystals was used for the study.

#### b) Procurement of animals

Wistar strain male albino rats weighing about 120 – 150 g purchased from the scientific suppliers were used for the study. The animal house was well ventilated day and night in normal room temperature. The animals were housed in large spacious hygienic cages during the course of the experimental period. The animals were fed with standard rat pellet and adequate supply of water.

#### c) Acute Toxicity Study

The Fixed Dose Procedure (FDP), proposed in 1984 by the British Toxicology Society, is a method to assess a substance's acute oral toxicity. In this procedure, animals were fasted for 4 hours with free access to water only. The test substances were given at a fixed dose level of 1000 mg/kg (considered to be as the upper limit dose) to five female rats and was observed for 3 days. The objective was to identify a dose that produces clear signs of toxicity but no mortality. When the first animal was dosed with the upper limit dose and survived, the second animal receives the same dose. When a total of three animals had been dosed with the limit dose and no deaths have occurred, then three ani-

mals of the other sex were tested at the limit dose level. When there was no lethality, the test was terminated. The larger therapeutic index of the plant extract can be obtained when there is no death of rats even at a maximum dose of 1000 mg/kg body weight. Tests were repeated until it showed no lethality<sup>6</sup>.

#### d) Induction of Diabetes

Severe diabetes was induced in overnight fasting male rats by a single intraperitoneal injection of 50 mg/kg Streptozotocin in a volume 1 mg/ kg body weight dissolved in cold citrate buffer (PH-4.5). Hyperglycemia was confirmed by measuring glucose 72 hrs after the Streptozotocin shot and 7 days after injection, confirming a high glucose level. Rats with permanent high fasting blood glucose level >300mg/dl were included in the experiments<sup>7</sup>.

#### e) Experimental design

The experimental rats were divided into V groups of 6 animals each.

**Group I :** Normal control rats received standard pellet and water for 30 days.

**Group II :** Streptozotocin was injected intraperitoneally for 30 days served as a diabetic control

**Group III :** Streptozotocin induced diabetic rats were administered with 150 mg/kg body weight of Glibenclamide standard drug orally for 30 days.

**Group IV :** Streptozotocin induced diabetic rats were administered with hydroethanolic extract of IM at a concentration of 200 mg/kg body weight, orally for 30 days.

**Group V :** Streptozotocin induced diabetic rats were administered with hydroethanolic extract of IM at a concentration of 400 mg/kg body weight, orally for 30 days.

After the experimental regimen, the animals were fasted overnight and sacrificed by cervical dislocation under mild anaesthesia. Blood was collected through cardiac puncture and serum was separated by centrifugation at 2500 rpm for 15 minutes.

#### f) Biochemical analysis

Biochemical analysis in serum such as blood glucose was estimated according to the method of Trinder<sup>8</sup>. Urea was measured by the method of Wybenga<sup>9</sup>. Creatinine was measured the method of Slot and Scand<sup>10</sup>. Total cholesterol, LDL, HDL, VLDL levels was estimated according to Castelli<sup>11</sup>. As Triglyceride levels was estimated by the method of Philip and Mayne<sup>12</sup>.

#### g) Statistical analysis

Data obtained was expressed as mean±SD. Statistical analysis was performed by using the method of distribution statistics (standard descriptive analysis) and analysis of means (Student

t test) using R - Statistical Computing and Graphical Tools (formerly AT & T, Lucent technology). A probability of  $P < 0.05$  was considered significant.

## Results and Discussion

### Blood glucose, Urea and Creatinine

It is evident from the Table-1 that there was a significant decrease in blood glucose, Urea and Creatinine levels in Group-III, IV and V when compared with the diabetic control Group-II.

**Table-1 Effect of *Ixora macrothyrsa* on blood glucose, Urea and Creatinine levels of control and experimental rats**

Groups	Blood glucose (mg/dl)	Urea (mg/dl)	Creatinine (mg/dl)
Group-I	88.05 $\pm$ 5.17	23.55 $\pm$ 2.90	0.83 $\pm$ 0.07
Group II	317.52 $\pm$ 16.47*	45.54 $\pm$ 20.15*	3.32 $\pm$ 0.36*
Group III	105.91 $\pm$ 8.89*	30.94 $\pm$ 1.12*	2.53 $\pm$ 0.31*
Group IV	173.07 $\pm$ 18.34*	28.21 $\pm$ 1.94*	2.70 $\pm$ 0.18*
Group V	168.90 $\pm$ 12.50*	23.07 $\pm$ 1.92*	1.72 $\pm$ 0.22*

Values are expressed as mean  $\pm$  SD of six samples

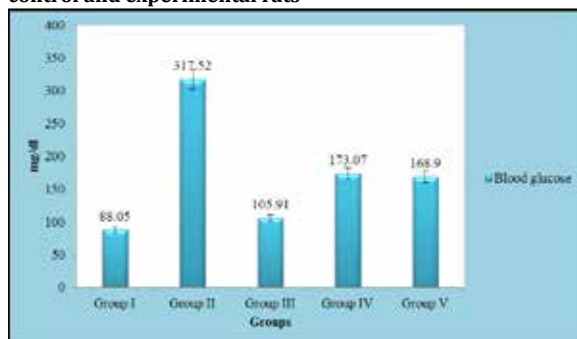
Group comparison : Group I Vs III, IV and V

Statistical significance : \*Significant ( $p < 0.05$ )

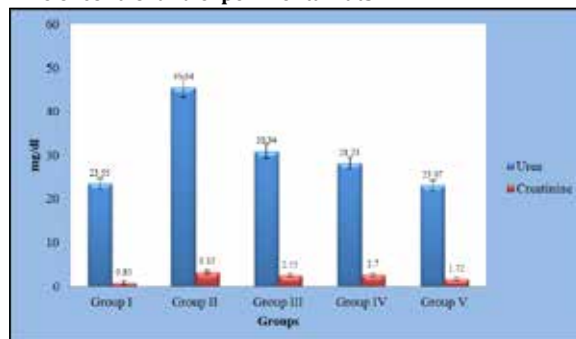
Decrease in blood glucose might be due to improving the glycemic control mechanisms and insulin secretions from remnant pancreatic cells in diabetic animals.

Many studies have shown significant increase in the rate of kidney cell damage in diabetes disorders. changes in the structure of kidney in diabetes. These changes lead to hypertrophy and changes in physiological function and structure of kidney, as well as increase in serum Urea and Creatinine levels<sup>13</sup>.

**Figure-1 Effect of *Ixora macrothyrsa* on blood glucose of control and experimental rats**



**Figure-2 Effect of *Ixora macrothyrsa* on Urea and Creatinine of control and experimental rats**



### Lipid profile

Table-2 shows the significant decrease in Total cholesterol, Triglyceride, LDL and VLDL levels in Group-III, IV and V when compared to Group II. Meanwhile there was a reduction in HDL level in diabetic rats and increased significantly in all the treated groups such as Group III, IV and V.

**Table-1 Effect of *Ixora macrothyrsa* on Total cholesterol, Triglyceride, LDL, HDL and VLDL levels of control and experimental rats**

Groups	Total cholesterol (mg/dl)	Triglyceride (mg/dl)	LDL (mg/dl)	HDL (mg/dl)	VLDL (mg/dl)
Group-I	154.83 $\pm$ 5.12	66.81 $\pm$ 5.48	93.70 $\pm$ 6.03	47.71 $\pm$ 4.06	13.36 $\pm$ 1.10
Group-II	235.64 $\pm$ 5.36*	182.98 $\pm$ 12.85*	181.59 $\pm$ 9.21*	17.45 $\pm$ 2.55*	36.39 $\pm$ 2.57*
Group-III	160.72 $\pm$ 4.33*	87.58 $\pm$ 5.38*	98.29 $\pm$ 6.46*	44.91 $\pm$ 2.70*	17.51 $\pm$ 1.07*
Group-IV	191.87 $\pm$ 6.82*	145.36 $\pm$ 4.06*	117.98 $\pm$ 2.88*	32.79 $\pm$ 3.24*	29.31 $\pm$ 0.80*
Group-V	180.00 $\pm$ 7.84*	136.15 $\pm$ 5.84*	105.20 $\pm$ 3.09*	41.96 $\pm$ 1.59*	27.23 $\pm$ 1.17*

Values are expressed as mean  $\pm$  SD of six samples

Group comparison : Group I Vs III, IV and V

Statistical significance : \*Significant ( $p < 0.05$ )

Derangement of glucose and fat metabolism results in the development of hyperlipidaemia. Significant lowering of total cholesterol and rise in HDL is a very desirable biochemical state for the prevention of atherosclerosis and ischemic conditions<sup>14</sup>.

### Conclusion

The present work implies that the Hydroethanolic leaf extract of IM exerts antidiabetic effect by lowering blood glucose, Urea and Creatinine levels and in maintaining lipid profile on Streptozotocin induced diabetic rats. However, further pharmacological and biochemical investigations are underway to find out the mechanism of action of active components involved in IM.

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