Effect of ethyl acetate extracts of *Acanthospermum hispidum* on STZ-induced diabetic animals

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Abstract : The aim of present study was to evaluate the effects of *Acanthospermum hispidum* DC plant aqueous and ethyl acetate extracts (AHEAEs) on blood glucose, Plasma insulin and lipid profile in streptozotocin (STZ) - induced diabetic rats. The STZ-induced diabetic rats were treated by plant aqueous extract of *Acanthospermum hispidum* aqueous and ethyl acetate extracts at doses (300 and 450 mg/Kg bw) and glibenclamide (0.6 mg/Kg bw) for 21 days by gavage. Blood glucose levels and body weights of rats were measured and total lipid levels were determined in normal and STZ-induced diabetic rats after administration of the AHEAE for 21 days. STZ-induced diabetic rats showed a significant increases in the levels of blood glucose, triglycerides (TG), total cholesterol (TC), low density lipoprotein LDL-cholesterol (LDL-C) while body weight and high density lipoprotein HDL-cholesterolan (HDL-C) were significantly decreased compared to normal rats. Daily administration of (AHAEx) did not possess the hypoglycemic and hypolipidaemic activity in STZ- diabetic rats during 3-week treatment period. Results indicate the usage of AHEAE is more effective in traditional medicine for the treatment of diabetes may need more investigation.

IndexTerms – Ethyl Acetate, STZ, Diabetes, *Acanthospermum hispidum*.

I. INTRODUCTION

Since olden days, plants are used to treat many ailments. India has about 45,000 plant species and several thousands have been claimed to possess medicinal properties. Medicinal plants used to treat hypoglycemic or hyperglycemic conditions are of considerable interest for ethno-botanical community as they are recognized to contain valuable medicinal properties in different parts of the plant and number of plants have shown varying degree of hypoglycemic and antihyperglycemic activity. The active principles of many plant species with desired properties are isolated to cure ailments, such as diabetes type-I and type-II respectively. Though, medical heritage is centuries old, million people in rural area still depend on traditional medicine to congregate their healthcare needs. The major mode of controlling diabetes can be achieved by diet, exercise and insulin replacement therapy. The use of hypoglycemic drugs, like insulin, biguanides, sulfonylureas and α-glucosidase inhibitors are accompanied by unpleasant side effects such as severe hypoglycemia, lactic acidosis, peripheral edema and abdominal discomfort.

II. RESEARCH METHODOLOGY

2.1 Plant material

Whole plant of *Acanthospermum hispidum* were collected Sri Venkateswara Agricultural University and surrounding areas of Tirupati and identified & voucher specimen was deposited in the herbarium of the plant in Department of Botany, Sri Venkateswara university, Tirupati.

2.2 Preparation of plant extracts

About 500g of the air dried powder of the plant material *Acanthospermum hispidum* L was extracted successively with the following solvent in soxhlet extractor, and identified as fractions 1-3 as shown below; n-hexane-Fraction-1, Ethylacetate-Fraction-2, and ethanol fraction-3. Every time before extracting with the next solvent, the plant material was dried in hot air oven below 50°C. The extracts were in rotary vacuum evaporator. Extracts were stored in air tight container in refrigerator at below 10°C.

2.3 Chemicals

Streptozotocin was obtained from Himedia laboratory Limited , Mumbai, India. All other chemical were analyticalgrade laboratory reagents and were used as such without further testing

2.4 Animals

Adult male albino rats of the Wistar strain weighing approximately 200 to 230g were procured from department of zoology, Sri Venkateswara University, Tirupati. Andhra Pradesh, India. They were acclimatized to animal house conditions, and fed with standard rat feed supplied by Hindustan Lever Ltd., Bangalore, India. All the animal experiments were conducted according to the ethical norms approved by the Ministry of Social Justice and Empowerment, Government of India and the guidelines of the Institutional Animal Ethics Committee S.V.University.

2.5 Induction of experimental diabetes

The animals were fasted overnight and diabetes was induced by a single intraperonal injection of freshly prepared solution of Streptozotocin (55 mg/kg body weight) in 0.1M cold citrate buffer pH 4.5. The animals were allowed to drink 5% glucose solution overnight to overcome drug-induced hypoglycaemia. The control rats were injected with citrate buffer alone. After a week’s delay for the development of diabetes, the rats with moderate diabetes, i.e. with glycosuria and hyperglycaemia (blood glucose range above 250 mg/dl) were considered as diabetic and used for the drug treatment. The bark ethyl acetate extract was administered orally througha gavages at a concentration of 300 and ,450mg/kg body weight/rat/day for 3 weeks
2.6 Experimental Groups

- Group 1: Normal treated animals
- Group 2: Diabetic treated animal
- Group 3: Diabetic animals treated with plant extract 300 mg/kg body weight
- Group 4: Diabetic animals treated with plant extract 450 mg/kg body weight

III. RESULTS

Table 1: Effect of alcoholic extract of *Acanthospermum hispidum* on fasting blood glucose of normal and diabetic rats.

Changes in plasma glucose, plasma insulin of normal, diabetic, diabetic treated rats with aqueous extract 300 mg/kg body weight and ethyl acetate extract 450 mg/kg body weight. (Means±S.D).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Plasma glucose</th>
<th></th>
<th></th>
<th>Plasma insulin</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before treatment</td>
<td>After treatment</td>
<td>Before treatment</td>
<td>After treatment</td>
</tr>
<tr>
<td>Group 1</td>
<td>79.62±2.86</td>
<td>24.12±2.85</td>
<td>76.68±4.13</td>
<td>23.42±2.62</td>
</tr>
<tr>
<td>Group 2</td>
<td>282.34±5.26</td>
<td>9.25±2.12</td>
<td>276.45±3.46</td>
<td>9.12±1.26</td>
</tr>
<tr>
<td>Group 3</td>
<td>276.62±6.12</td>
<td>8.56±1.13</td>
<td>145.26±5.26</td>
<td>14.11±2.09</td>
</tr>
<tr>
<td>Group 4</td>
<td>276.23±5.62</td>
<td>8.52±1.02</td>
<td>126.12±3.48</td>
<td>13.16±1.15</td>
</tr>
</tbody>
</table>

F-Value: 1142.70 152.31 1156.84 78.135

Significance: 0 0 0 0

T-test: Values are expressed as mean ± SEM (n=6). ***P<0.0001 compared with diabetic control (one way ANOVA followed by Duncan post-hoc tests)

Fig 9: Changes in plasma glucose, plasma insulin of normal, diabetic, diabetic treated rats with aqueous extract 300 mg/kg body weight and ethyl acetate extract 450 mg/kg body weight. (Means±S.D)
Table 2: Levels of serum cholesterol, triglycerides, HDL, LDL, and VLDL cholesterol in normal, diabetic, diabetic treated rats (Mean±SD)

<table>
<thead>
<tr>
<th>Groups</th>
<th>Mean ±SD</th>
<th>Cholesterol (mg/dl)</th>
<th>Triglycerides (mg/dl)</th>
<th>HDL-Cholesterol (mg/dl)</th>
<th>LDL-Cholesterol (mg/dl)</th>
<th>VLDL-Cholesterol (mg/dl)</th>
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</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>Mean±SD</td>
<td>39.99±3.49a</td>
<td>8.96±1.35a</td>
<td>47.29±3.55d</td>
<td>18.54±1.52a</td>
<td>1.82±0.26a</td>
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<td>Group 2</td>
<td>Mean±SD</td>
<td>96.42±4.82c</td>
<td>46.63±3.48c</td>
<td>15.22±1.26a</td>
<td>74.85±3.86d</td>
<td>8.96±1.06d</td>
</tr>
<tr>
<td>Group 3</td>
<td>Mean±SD</td>
<td>62.15±2.99b</td>
<td>25.42±2.72c</td>
<td>29.42±1.48b</td>
<td>41.56±2.25c</td>
<td>6.15±0.52c</td>
</tr>
<tr>
<td>Group 4</td>
<td>Mean±SD</td>
<td>45.26±2.55a</td>
<td>15.54±1.56b</td>
<td>41.23±2.44c</td>
<td>24.75±1.63b</td>
<td>2.11±0.23b</td>
</tr>
<tr>
<td>F-Value</td>
<td></td>
<td>152.42</td>
<td>258.12</td>
<td>139.048</td>
<td>565.216</td>
<td>136.026</td>
</tr>
<tr>
<td>Significance</td>
<td></td>
<td>0.000</td>
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</tbody>
</table>

T-test: Values are expressed as mean ± SEM (n=6). ***P<0.0001 compared with diabetic control (one way ANOVA followed by Duncan post-hoc tests)

Fig 2: Levels of serum cholesterol, triglycerides, HDL, LDL, and VLDL cholesterol in normal, diabetic, diabetic treated rats (Mean±SD)

IV. DISCUSSION

Diabetes mellitus, the most common endocrine disease, is not a single disease but a group of disorders. In fact, hyperglycemia, polyphagia, polydipsia and reduction. Table 1 illustrates the levels of blood glucose, plasma insulin, and body weight in normal and experimental animals. The levels of blood glucose was significantly increased Where as the levels of plasma insulin were significantly decreased in diabetic rats when compared with normal rats. Body weights were also significantly reduced in diabetic rats when compared to normal rats while it was significantly recovered in the AHEt treated animals. The recovery effect with plant extract (450 mg/kg) was significant .

Tables 2 show the levels of plasma and tissues glycoproteins in normal and experimental animals. There was a significant increase in the level of plasma glycoproteins in diabetic rats. In liver and kidney of diabetic rats, the levels of serum cholesterol, triglycerides, HDL, LDL, and VLDL cholesterol were significantly raised, but the level of sialic acid was significantly decreased. Oral administration of AHEts significantly restored the changes in total cholestrols of diabetic rats. The recovery effect with plant extract (450 mg/kg) was more significant but normal rats treated with AHEt did not show significant changes.

In diabetes, the levels of plasma lipids are usually raised and such an elevation represents a risk factor for coronary heart disease5. (Grundy, 1999). Also, an increase in levels of plasma cholesterol, phospholipids, free fatty acids and triglycerides was observed in alloxan diabetic rats. The abnormally high concentration of plasma lipids in diabetes is mainly due to the increase in the mobilization of free fatty acids from the peripheral deposits, since insulin inhibits the hormone sensitive lipase6 (Dhandapani et al., 2002). In the present study, there was a decrease in levels of triglyceride and cholesterol in diabetic alloxan rats treated with cinnamon7. Blevins et al. (2007) reported that oral administration of cinnamon (20 mg/Kg body weight) significantly decreased serum total cholesterol, triglyceride levels and at the same time markedly increased plasma insulin8. Amin and Abd ElTwab
(2009) proposed that cinnamon extract may improve the postprandial overproduction of intestinal apoB48-containing lipoproteins by ameliorating intestinal insulin resistance and may be beneficial in the control of lipid metabolism. However, treatment with cinnamon essential oil significantly decreased and improved the diabetic status including protection of DNA against oxidative damage and hypocholesterolemic effect⁹. (Thresa, Christieand, Andrea, 2004).

These results were in agreement with the study proposed by Anderson.¹⁰ which revealed that the polyphenols, polymers and anthocynins, found in cinnamon functions as antioxidants, potentiate insulin action and may be beneficial in the control of glucose intolerance and diabetes.¹¹ Aderson also reported that cinnamon extract and polyphenols improved the lipid profile of people with type 2 diabetes. On the other hand, our results showed a significant decrease in TG and TC levels in cinnamon-treated rats. Similar results were obtained by Qin et al¹² who reported that TG and TC were decreased by administration of cinnamon extract in rats treated with streptozotocin for 2 weeks, in our study A. hispidum plant extracts were significant activity of diabetic treated animals.

V. CONCLUSION

Our results concluded that the extract of Acanthospermum hispidum for antidiabetic activity have shown appreciable results in ameliorate the blood glucose level and related other complications. Our findings also may open the door for a new, alternative, leading drug for treating diabetic patients in future.

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