EFFECT OF ADIANTUM CAUDATUM IN STREPTOZOTOCIN INDUCED DIABETES MELLITUS IN RATS

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ABSTRACT

The alleged anti-diabetic potential of ethanolic extract of Adiantum caudatum (Convolvulaceae) whole plant, on fasting blood sugar levels and serum biochemical analysis in Streptozotocin induced diabetic rats were investigated. Ethanolic Extract of Adiantum caudatum produced a significant anti-diabetic activity at dose levels 1/10 of their lethal doses (2000 mg/kg b.w). The activity was found comparable with standard drug Glibenclamide. The present investigation of the plant established some pharmacological evidence to support the folklore claim.

KEY WORDS: Adiantum caudatum, Streptozotocin (STZ), Antidiabetic effect

INTRODUCTION

Diabetes mellitus (DM) is a chronic disease caused by inherited and/or acquired deficiency in production of insulin by the pancreas or by ineffectiveness of the insulin produced. Such a deficiency results in increased concentration of glucose in the blood (hyperglycemia), which in turn leads to metabolic disorders and also damage many of the body’s systems, in particular the blood vessels and nerves1. It is also estimated that there are 30-33 million diabetic patients in India now and every fourth diabetic in the world today is an Indian. Indians are genetically more susceptible to diabetes and the WHO predicts the number of diabetic persons in India would go up to 74 million by 20252. Apart from currently available therapeutic
options, many herbal medicines have been recommended for treatment of diabetes. Furthermore after the recommendation made by WHO on diabetes mellitus, investigation on hypoglycemic agent from medicinal plants has become more important.

The plant *Adiantum caudatum* is distinctive in appearance, with dark, often black stipes and rachises, and bright green, often delicately cut leaf tissue. The sori are borne sub marginally, and are covered by reflexed flaps of leaf tissue which resemble indusial. It generally prefers humus rich, moist, well-drained sites, ranging from bottomland soils to vertical rock walls.

The present study aimed at investigating the effect of ethanolic extract of *Adiantum caudatum* on blood glucose levels and other biochemical parameters in normoglycemic and Streptozotocin induced diabetic rats.

**MATERIALS AND METHODS**

**Collection of plant material:** Fresh plant of *Adiantum caudatum* were collected in the month of June from local areas of Chhaygaon, Guwahati, Assam and authenticated by Botanist Dr. G.C Sarma Dept. of Botany, Gauhati University. The voucher specimen (Acc. no.15001) was deposited in dept of Pharmacognosy, GIPS, Azara, Guwahati.

**Preparation of Extract:** The collected whole plants were shade dried at room temperature. The dried plants were subjected to size reduction to a coarse powder by using dry grinder and passed through a sieve. This powder was packed into a Soxhlet apparatus and extracted successively with pet. Ether (40—60), Chloroform and ethanol at a temp 30---50°C. All the extracts were further concentrated to semisolid mass using Rota flash evaporator and were stored in desicator. The suspension of ethanolic extract was prepared by using CMC in distilled water for the experiment.

**Qualitative Chemical test:** All the extracts were tested to know the different constituents present in them by the standard procedures. Terpenoids, phenyl propanoids, steroids, flavonoids have been found in the plant *A. caudatum*. Triterpenoids and flavonoids are the dominant constituents within the plant *Adiantum caudatum*.

**Animals:** All the experiments on animals were conducted according to protocols that were approved by the Institutional Animal Ethics Committee/IAEC clear/2011. Anti-diabetic activity was carried out using healthy albino wistar rats of either ex
weighing between 200----300 g. Animals were housed in polypropylene cages and allowed free access to tap water and pellet diet and libitum throughout the study. After randomization into various groups and before initiation of experiment, the rats were acclimatized for a period of 7 days under standard environmental conditions of temperature, relative humidity and dark/ light cycle.

**Acute toxicity study:** Albino mice of either sex weighing between 20---30 g were used. The animals were fasted overnight. Acute toxicity was performed according to OECD guidelines; method followed is according to number 420\(^6\). Effective dose for the extract was found to be 200 mg/ kg body weight.

**Standard drug used:** Glibenclamide tablet (Daonil tablet, Aventis Pharma) was used as standard drug. It was purchased from new multicare, Chhaygaon. The tablets were suspended in distilled water using CMC as suspending agent and used for study.

**Streptozotocin induced diabetes:** The albino rats weighing 200---300 g of either sex were allowed to fast for 24 h prior to experimentation and rendered diabetic by a single dose of IP injection of STZ. 50 mg/ kg body weight. After one hr of STZ induction the animals were given feed and libitum and 5% dextrose solution for a day to avoid early hypoglycemic phase. The hypoglycemic activity on these animals was carried out after one week of STZ injection when the stabilization of diabetes was ensured\(^7\).The animals with sugar level more than 240mg/dl. were selected for the study\(^8\).

**Design:** All the animals were divided into four groups.

Group I: Healthy normal animals received only the vehicle (CMC).

Group II: Untreated but diabetes induced animals served as a negative control.

Group III: Diabetes induced animals and treated with standard drug glibenclamide 10mg/ kg body weight/ day orally.

Group IV: Diabetic animals and treated with EEAC.

The extract was given orally at the dose of 200mg/kg body weight.

On 22\(^{nd}\) day of treatment, blood samples were collected by retro- orbital plexus puncture method under mild ether anaesthesia and serum was separated by centrifugation. The serum was analysed for blood glucose level, total cholesterol and total triglyceride level.

**Statistical Analysis:** The quantitative measurements were made on six animals
in each group & the values of biochemical estimations were expressed as mean ± SEM. The data obtained were subjected to one way ANOVA by multiple comparison tests.

RESULT

The anti diabetic effect of the extract on the fasting blood sugar levels of diabetic rats is shown in table 1.

Table 1: Effect of ethanolic extract of A. caudatum (whole plant) on blood glucose level & body weights of the rats

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>BLOOD GLUCOSE LEVEL (mg/dl)</th>
<th>BODY WEIGHT (gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before Treatment Mean ± SEM</td>
<td>After Treatment Mean ± SEM</td>
</tr>
<tr>
<td>Normal Control</td>
<td>84.83 ± 0.65</td>
<td>86.17 ± 0.64</td>
</tr>
<tr>
<td>Untreated</td>
<td>265.0 ± 1.39</td>
<td>271.3 ± 2.69b***</td>
</tr>
<tr>
<td>Standard</td>
<td>271.0 ± 1.52</td>
<td>204.2 ± 1.51b***</td>
</tr>
<tr>
<td>EEAC</td>
<td>260.0 ± 1.43</td>
<td>208.0 ± 1.71b***</td>
</tr>
</tbody>
</table>

Fig: Blood Glucose Level (mg/dl) and Body weight in different groups in before and after treatment.
Table 2: Effect of ethanolic extract of *A. caudatum* (whole plant) on Biochemical parameters

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>BIOCHEMICAL PARAMETERS (mg/dl)</th>
<th></th>
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</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total Cholesterol</td>
<td>Total Triglyceride</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mean ± SEM</td>
<td>Mean ± SEM</td>
<td></td>
</tr>
<tr>
<td>Normal Control</td>
<td>34.17 ± 0.23</td>
<td>75.94 ± 0.84</td>
<td></td>
</tr>
<tr>
<td>Untreated</td>
<td>52.53 ± 0.59&lt;sup&gt;a&lt;/sup&gt;***</td>
<td>143.6 ± 1.02&lt;sup&gt;a&lt;/sup&gt;***</td>
<td></td>
</tr>
<tr>
<td>Standard</td>
<td>45.22 ± 0.88&lt;sup&gt;b&lt;/sup&gt;***</td>
<td>82.55 ± 1.40&lt;sup&gt;b&lt;/sup&gt;***</td>
<td></td>
</tr>
<tr>
<td>EEAC</td>
<td>45.86 ± 0.92&lt;sup&gt;b&lt;/sup&gt;***</td>
<td>85.82 ± 0.94&lt;sup&gt;b&lt;/sup&gt;***</td>
<td></td>
</tr>
</tbody>
</table>

Fig: Total Cholesterol (mg/dl) & Total Triglyceride (mg/dl) in different groups
(N.B: Values represented in (Mean ± S.E.M, n=6), *p<0.05, **p<0.01, ***p<0.001;
<sup>a</sup>untreated vs. Normal control, <sup>b</sup>others group vs. untreated).

Administration of Streptozotocin (50mg/kg, I.P.) led to elevation of fasting blood glucose levels, which was maintained throughout the period of treatment. 21 days of daily treatment of EEAC led to a fall in blood sugar levels. Effect seems to reach to a maximum after 21 days of treatment. Vehicle controlled animals were found to be somewhat stable in their body weight but diabetic rats showed significant reduction in body weight during the entire experiment.

The ethanolic extract was effective in normalizing the elevated levels of blood sugar and blood lipid like cholesterol, triglycerides. Results are comparable with that of reference drug Glibenclamide. The phytochemical screening of the extract revealed the presence of flavonoids, alkaloids, carbohydrates and triterpenoids.
DISCUSSION

In light of the results, our study indicates that ethanolic extract of A. caudatum without significant change in body weight (200mg/kg body weight) exhibited significant anti diabetic activity in Streptozotocin induced hypoglycemic rat. The antidiabetic effect may be due to the presence of flavonoids, alkaloids, triterpenoids.

CONCLUSION

In conclusion, oral administration of ethanolic extract of A. caudatum lowers serum glucose, triglyceride & cholesterol in Streptozotocin administered rats. The ethanolic extract showed significant anti-diabetic effect in rats after oral administration. Thus the claim made by the traditional Indian systems of medicine regarding the use of this plant in the treatment of diabetes stands confirms. The results suggest the presence of biologically active principle flavonoids which may be worth further investigation, elucidation.

ACKNOWLEDGEMENT

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