Antihyperglycemic/ Hypoglycemic Effect of Celery Seeds (Ajwain/ Ajmod) in Streptozotocin induced Diabetic Rats

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Abstract

Background: To investigate the effect of Apium graveolens L. (Celery seeds) on blood glucose and insulin concentrations in streptozotocin (STZ) induced diabetic rats.

Methods: Forty Sprague Dawley rats weighing from 180 to 250 grams were randomly divided into four experimental groups A, B, C, and D, each containing ten (10) rats. Diabetes was induced by intraperitoneal administration of STZ (60mg/kg) in groups B, C, and D, while group A served as normal control. The experimental animals became diabetic within 48 to 72 hours after administration of STZ. Group B rats were taken as diabetic control. Alcoholic extract of Celery seeds, the test drug was administered (400mg/kg) orally to experimental group C, while group D received standard drug glibenclamide, induced diabetic rats for six weeks. Normal control group A & diabetic control B received only normal saline solution orally. Blood samples were collected from experimental groups after 43 days, 24 hours of the last dose. ANOVA and Tukey HSD (Honestly Significant Difference) test applied to all groups.

Results: The results show Celery seeds extract treatment caused a statistically significant decrease in the elevated serum glucose levels and increase in the serum insulin concentrations in test group C as compared to group B.

Conclusion: Findings of present study provide evidence for traditional use of apium graveolens in the control of diabetes.

Key Words: Streptozotocin, Apium graveolens

Introduction

Diabetes Mellitus is a clinical disorder characterized by increased blood glucose due to deficiency or decreased secretion of insulin. About 180 million of the world population have diabetes mellitus. In 1995, 4.3 million people had DM and in 2025 it will increase to 14.5 million. This data suggests that Pakistan becomes the fourth country among the top 10 countries of the world.
Group B: In rats of this group diabetes mellitus was induced by giving an intraperitoneal injection of STZ, 60mg/kg, after an overnight fasting. No drug or extract was given to this group. These rats were fed on Purina rat chow and normal saline (1.5ml) orally for six weeks of experimental study.

Group-C: In these diabetic rats celery seeds extract was given 400mg/kg as a single dose orally for six weeks of experimental study.

Group-D: In these diabetic rats glibenclamide (600µg/kg) body weight was given orally for six weeks.

For the preparation of ethanol extract of celery seeds, freshly collected fruits were shade-dried and coarsely powdered in a grinder. Powdered dried seeds 500g were soaked for 12hrs in 2.0 liters of ethyl alcohol. Then this suspension was filtered and the residue was again soaked in equal amount of ethanol for 48hrs and again filtered. The two filtrates obtained were evaporated and dried by distillation under reduced pressure at 40 to 50°C in rotary evaporator. The ethanol extract so obtained (42g) was suspended in distilled water in the required amount at the time of dosing.

Initial blood sample for glucose estimation was collected from all groups A, B, C, and D, from the tail veins of rats. Blood sample from group B, C, and D was collected after 72 hours of STZ-injection and diabetes mellitus confirmed by Ames One Touch Glucometer. Animals with blood glucose more than 200mg/dl were selected for this research study.

After 24hours of the last dose of celery seeds extract and after overnight fasting (12 to 14 hours) the animals were anesthetized under chloroform vapors. Blood samples (5ml) were collected by cardiac puncture from all groups. Blood was collected in vacutainers and then centrifuged for ten minutes at a speed of 5000 rev/min. Plasma was separated and stored at -20°C and used for the measurement plasma glucose and plasma insulin. The significance between two means was calculated by the ANOVA and Tukey HSD. The difference was regarded statistically significant if the ‘p’ value was < 0.05.

Results

At the end of experimental study, body weights of rats in group A (normal control) were increased as compared to their initial body weights. Significant body weight loss was observed in the STZ induced diabetic rats in group B (STZ induced Diabetic control group), when their final body weights were compared with their initial body weights. The final body weights of group B (STZ induced Diabetic control group) were significantly less than group A. The body weights of group C (STZ induced Diabetes + Celery seeds extract) were less than group A, but it was still significantly greater than the STZ induced diabetic rats in group B. Similarly the body weights of group D (STZ induced Diabetes+ Glibenclamide) were also increased significantly (Table 1).

In group C rats at the end of experimental study mean blood glucose level ± SEM was 139.65 ± 0.257 mg/dl in Group C rats compared with groups Group-B (Diabetic group). Mean blood glucose values were statistically significant p< 0.001, when fasting blood glucose levels of Group C (STZ induced Diabetes + Celery seeds extract) was compared with groups Group-B (Diabetic group) and other groups (Table 2).

![Table 1. Effect of Celery seeds extract on the body weight of normal and STZ-diabetic rats.](image)

<table>
<thead>
<tr>
<th>Groups</th>
<th>0th day</th>
<th>43rd day</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A (normal control)</td>
<td>208 ± 6.46</td>
<td>249.4 ± 3.99</td>
</tr>
<tr>
<td>Group B (STZ induced Diabetic control)</td>
<td>205 ± 6.83</td>
<td>176.9 ± 1.82</td>
</tr>
<tr>
<td>Group C (STZ induced Diabetes + celery seeds extract given)</td>
<td>200.5 ± 1.74</td>
<td>233.3 ± 6.46</td>
</tr>
<tr>
<td>Group D (STZ induced Diabetes + Glibenclamide given)</td>
<td>205.5 ± 6.16</td>
<td>230 ± 6.30</td>
</tr>
</tbody>
</table>

![Table 2. Comparison of all groups for plasma glucose (mg/dl).](image)

<table>
<thead>
<tr>
<th>Mean Plasma Glucose (mg/dl)</th>
<th>Group A (normal control)</th>
<th>Group B (STZ induced Diabetic control)</th>
<th>Group C (STZ induced Diabetes + celery seeds extract given)</th>
<th>Group D (STZ induced Diabetes + Glibenclamide given)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0th day</td>
<td>78.88±1.5</td>
<td>80.0±6.8</td>
<td>77.7±1.7</td>
<td>83.5±4.5</td>
</tr>
<tr>
<td>43rd day</td>
<td>78.88±1.5</td>
<td>254.7±1.8</td>
<td>139.6±1.6</td>
<td>102.8±1.8</td>
</tr>
</tbody>
</table>

There was statistically significant difference( p< 0.001) in mean fasting plasma insulin levels of Group C (STZ induced Diabetes + Celery seeds extract) compared with groups Group B (Diabetic group). Group C was also compared with groups (Group A, and D). The mean fasting plasma insulin levels of...
Group C (STZ induced Diabetes + Celery seeds extract) could not reach the level as that of the normal control group A, but it justifies the role of this plant extract in induction of insulin release from pancreatic islets (Table 3).

Table 3. Comparison of all Groups for plasma Insulin (µL/ml)

<table>
<thead>
<tr>
<th>Mean Plasma Insulin (µL/ml)</th>
<th>Group A (normal control)</th>
<th>Group B (STZ induced Diabetic control)</th>
<th>Group C (STZ induced Diabetes + celery seeds extract given)</th>
<th>Group D (STZ induced Diabetes + Glibenclamide given)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0th day</td>
<td>11.68±2.0</td>
<td>11.31±1.5</td>
<td>10.81±2.5</td>
<td>11.04±1.8</td>
</tr>
<tr>
<td>43rd day</td>
<td>11.53±2.0</td>
<td>4.422±2.0</td>
<td>8.913±4.0</td>
<td>10.93±3.0</td>
</tr>
</tbody>
</table>

Discussion

Diabetes is a chronic metabolic disorder affecting a major population worldwide. A sustained reduction in hyperglycemia will decrease the risk of developing microvascular complications. The conventional therapies for diabetes have many shortcomings like unwanted side effects and high rate of secondary failure. On the other hand, herbal extracts are expected to have similar efficacy with minimum side effects like that of conventional drugs. 20,21

Streptozotocin is a glucoseamine-nitrosourea compound used in medicinal research to produce animal models for diabetes mellitus. As with other alkylating agents in the nitrosourea class, it is toxic to cell by causing damage to the DNA, though other mechanisms may also contribute. Thus STZ injection results in diabetes mellitus due to the destruction of beta cells of islets of Langerhans. 20 Streptozotocin selectively destroys the pancreatic insulin secreting β-cells, leaving less active cells and resulting in a diabetic state. STZ-induced diabetes is characterized by severe loss in body weight, and this reduction is due to loss or degradation of structural proteins, as the structural proteins are known to contribute to body weight. This effect is evident by high level of glucose in animals. 20 Flavonoids are one of the most numerous and widespread groups of phenolic compounds in higher plants. Some of these, due to their phenolic structure, are known to be involved in the healing process of free radical mediated diseases including diabetes. Celery seeds possess several flavonoids such as apigenin, and luteolin, which are reported as the active principles. 7 Apigenin, isolated from celery seeds extract also possess an inhibitory effect on the aldose reductase enzymes.22 This enzyme is known to play a key role in the polyol pathway, by catalyzing the reduction of the glucose to sorbitol, which cannot diffuse out of cell membranes. In diabetics excessive intracellular accumulation of sorbitol result in chronic complications such as neuropathy, retinopathy, and cataract. A study was conducted in which antihyperglycaemic effects of herbal drugs Cardiospermum halicacabum L were investigated in STZ induced diabetic rats. 19 This plant has same active compounds apigenin and luteolin, 10 and has same effect on insulin release from pancreatic islets of rats as that of celery seeds. 7

Scientific studies have been conducted in which antihyperglycaemic effects of herbal drug Nigella sativa L was reported. Aegle marmelos L. investigated in STZ induced diabetic rats showed results are similar to the results of our current study. 22,23 In celery seeds treated diabetic rats, the significant elevation of plasma insulin may be due to the stimulation of insulin secretion from the existing β-cells of the pancreas. 20 The decrease in blood glucose level of diabetic rats treated with celery seeds extract might be due to elevated secretion of insulin, increasing repair or proliferation of β-cells, enhancing the effect of insulin and increasing the oxidative capability which in turn, increases the utilization of glucose by the tissues. 24 Extrapancreatic mechanisms such as enhanced glucose transport into the cells and improved formation of glycogen in the liver might be involved in celery seeds induced decrease in blood glucose concentration of STZ diabetic rats. 24 Several of the compounds found in celery seeds have been reported as possessing antidiabetic activity, like flavonoids, such as luteolin and apigenin, which not only have well documented antidiabetic activity but also have significant role in the prevention of diabetes mellitus complications. 25,26

References