To Investigate The Antidiabetic Activity Of Althaea Officinalis And Thespesia Populnea Extract In Alloxan-Induced Diabetic Rats

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Abstract: Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia, which can lead to various microvascular and macrovascular complications. The present study investigated the antidiabetic potential of a combined extract of Althaea officinalis and Thespesia populnea (AOTPE) in Alloxan-induced diabetic rats. The extraction yield of the AOTPE was found to be 25.6% w/w, and phytochemical screening revealed the presence of various bioactive compounds such as flavonoids, alkaloids, glycosides, phenolic acids, sterols, and tannins. The AOTPE was evaluated for its effects on body weight, oral glucose tolerance, blood glucose levels, and glycosylated hemoglobin (HbA1c) in Alloxan-induced diabetic rats. The results showed that the AOTPE at doses of 200 mg/kg and 400 mg/kg exhibited a protective effect on body weight, improved glucose tolerance, reduced blood glucose levels, and lowered HbA1c levels compared to the positive control group. These findings suggest that the AOTPE possesses potent antidiabetic properties and could be a promising therapeutic option for the management of diabetes and its associated complications.

Index Terms - Diabetes mellitus, Althaea officinalis, Thespesia populnea, Antidiabetic activity, Alloxan-induced diabetes, Phytochemicals, Glucose tolerance, Blood glucose levels.

I. INTRODUCTION

Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia, which can lead to various microvascular and macrovascular complications. The disease is caused by either the body's inability to produce sufficient insulin (type 1 diabetes) or the body's inability to effectively utilize the insulin produced (type 2 diabetes). Diabetes has become a global epidemic, with the World Health Organization (WHO) estimating that the number of people with diabetes will rise from the current 150-220 million to 300 million by 2025[1]. The pathophysiology of diabetes involves multiple factors, including impaired insulin secretion, insulin resistance, and dysregulation of glucose metabolism. In type 1 diabetes, the destruction of pancreatic beta cells leads to insulin deficiency, while in type 2 diabetes, insulin resistance and impaired insulin secretion contribute to the development of hyperglycemia[2,3].

Diabetes can lead to various microvascular and macrovascular complications, including diabetic retinopathy, nephropathy, neuropathy, and an increased risk of cardiovascular diseases[4,5]. Effective management of diabetes and its complications is crucial to prevent or delay the onset of these debilitating conditions. Conventional treatment for diabetes involves the use of synthetic drugs, such as metformin, sulfonylureas, and insulin. However, these drugs can often have adverse side effects and may not be suitable for all patients[6,7]. Therefore, there is a growing interest in exploring alternative and complementary therapies, including the use of herbal medicines, to manage diabetes and its associated complications.
Althaea officinalis, commonly known as marshmallow, is a perennial herb that has been traditionally used in the treatment of various ailments, including diabetes\(^8\). Thespesia populnea, also known as the portia tree, is another medicinal plant that has been reported to possess antidiabetic properties\(^9\). The synergistic effects of these two plants may offer a promising approach to the management of diabetes.

The present study aimed to investigate the antidiabetic potential of a combined extract of Althaea officinalis and Thespesia populnea (AOTPE) in an Alloxan-induced diabetic rat model. The study evaluated the effects of the AOTPE on body weight, oral glucose tolerance, blood glucose levels, and glycosylated hemoglobin (HbA1c) to assess its therapeutic potential in the management of diabetes.

II. MATERIALS AND METHODS

Experimental animals:
Healthy Wistar albino rats, aged 6-8 weeks and weighing 150 ± 10 g, were obtained from the Vidyabharati College of Pharmacy, Amravati, India (CPCSEA Registration no. 1504/PO/RE/S/11/CPCSEA). The animals were housed in a temperature-controlled (22 ± 3°C) environment with a 12-hour light/dark cycle and had free access to standard rodent chow and water. The experimental protocol was approved by the Institutional Animal Ethical Committee (IAEC) and conducted in accordance with the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

Chemicals and reagents:
Alloxan monohydrate (CAS number: 2244-11-3) was purchased from Loba Chemie Pvt. Ltd., Mumbai, India. Glibenclamide, used as the standard antidiabetic drug, was obtained from Abmole BioScience.

Preparation of plant extract:
The aerial parts of Althaea officinalis and Thespesia populnea were collected from the Nashik region of Maharashtra, India. The plant materials were authenticated by a botanist, Prof. Kajal Apale, from Vidyabharti Mahavidyalaya, Amravati, Maharashtra, India. The dried and powdered plant materials were mixed in equal proportions (1:1) and subjected to Soxhlet extraction using ethanol as the solvent. The resulting Althaea officinalis and Thespesia populnea Extract (AOTPE) was dried and used for further experiments.

Phytochemical screening:
The AOTPE was screened for the presence of various phytochemicals, including carbohydrates, flavonoids, alkaloids, glycosides, phenolic acids, triterpenoids, sterols, fatty acids, tannins, proteins, and amino acids, using standard qualitative methods.

Experimental design:
The rats were divided into five groups, with six animals in each group:
- **Group I (Negative control):** Received saline solution.
- **Group II (Positive control):** Received Alloxan (100 mg/kg, i.p.).
- **Group III (Standard):** Received Alloxan (100 mg/kg, i.p.) + Glibenclamide (0.5 mg/kg, oral).
- **Group IV (Treatment 1):** Received Alloxan (100 mg/kg, i.p.) + AOTPE (200 mg/kg, oral).
- **Group V (Treatment 2):** Received Alloxan (100 mg/kg, i.p.) + AOTPE (400 mg/kg, oral).

Induction of diabetes:
Diabetes was induced in the rats by a single intraperitoneal (i.p.) injection of Alloxan monohydrate (100 mg/kg body weight) after an overnight fast. The development of hyperglycemia was confirmed by measuring the blood glucose levels using a glucometer after 72 hours. Rats with a blood glucose level above 250 mg/dL were considered diabetic and included in the study.

Evaluation of antidiabetic activity:
**Body weight:** The initial and final body weights of the rats were recorded.
**Oral glucose tolerance test (OGTT):** The rats were fasted for 12 hours, and their basal blood glucose levels were measured. Glucose (2 g/kg) was then administered orally, and blood glucose levels were measured at 30 and 90 minutes.
**Blood glucose levels:** Fasting blood glucose levels were measured on days 1, 7, 14, and 21 using a glucometer.
Glycosylated hemoglobin (HbA1c): HbA1c levels were measured at the beginning and end of the experiment using a commercial kit.

Statistical analysis:
The data was expressed as mean ± standard error of the mean (SEM). The statistical significance of the differences between groups was determined using one-way analysis of variance (ANOVA) followed by Dunnett's test. A p-value less than 0.05 was considered statistically significant.

III. RESULTS

Extraction yield and phytochemical screening:
The extraction yield of the AOTPE was found to be 25.6% w/w. The phytochemical screening revealed the presence of flavonoids, alkaloids, glycosides, phenolic acids, sterols, tannins, proteins, amino acids, phenols, and saponins in the extract.

Effect on body weight:
The positive control group (Alloxan-induced diabetic rats) showed a significant decrease in body weight compared to the negative control group (non-diabetic rats). However, the groups receiving the AOTPE at 200 mg/kg and 400 mg/kg doses exhibited a smaller decrease in body weight compared to the positive control group, indicating a protective effect of the AOTPE on body weight in the diabetic condition (Table 1, Figure No.1).

Table 1 Effect of AOTPE on body weight of rats

<table>
<thead>
<tr>
<th>Treatment/ Group</th>
<th>Body Weight of rats in Gram</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before Treatment</td>
</tr>
<tr>
<td>Group I Negative Control</td>
<td>155.7 ± 2.917</td>
</tr>
<tr>
<td>Group II Positive Control</td>
<td>150.8 ± 1.579</td>
</tr>
<tr>
<td>Group III Alloxan + Glibenclamide</td>
<td>151.7 ± 2.459</td>
</tr>
<tr>
<td>Group IV Alloxan + AOTPE 200mg/Kg</td>
<td>150.2 ± 2.509</td>
</tr>
<tr>
<td>Group V Alloxan + AOTPE 400mg/Kg</td>
<td>152.5 ± 2.717</td>
</tr>
</tbody>
</table>

Findings are demonstrated as mean ± SEM (n=6); statistically difference at **P=0.0001 and *P<0.0001 compared to positive control.
Oral glucose tolerance test (OGTT):
The positive control group displayed significantly higher blood glucose levels at 30 minutes and 90 minutes after the glucose load compared to the negative control group. In contrast, the groups receiving the AOTPE at 200 mg/kg and 400 mg/kg doses showed lower blood glucose levels compared to the positive control group, suggesting improved glucose tolerance. (Table 2, Figure No. 2).

Table. 2 Effect of AOTPE on oral glucose tolerance in diabetic Rat.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Blood Glucose Level (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 Min</td>
</tr>
<tr>
<td>Group I Negative Control</td>
<td>97.83±1.138</td>
</tr>
<tr>
<td>Group II Positive Control</td>
<td>244.8±1.195</td>
</tr>
<tr>
<td>Group III Alloxan + Glibenclamide</td>
<td>100.3±1.282</td>
</tr>
<tr>
<td>Group IV Alloxan + AOTPE 200mg/Kg</td>
<td>95.00±1.265</td>
</tr>
<tr>
<td>Group V Alloxan + AOTPE 400mg/Kg</td>
<td>99.00±1.366</td>
</tr>
</tbody>
</table>

Figure No.1 A graphical representation of Effect of AOTPE on body weight of rats.
Outcomes are demonstrated as mean ± SEM (n=6); statistically difference at *P<0.0001 compared to Positive control.

![Graphical representation of effect of AOTPE on oral glucose tolerance](image)

**Figure No.2** A graphical representation of effect of AOTPE on oral glucose tolerance

**Effect of AOTPE on blood glucose levels:**

The positive control group maintained high blood glucose levels throughout the 21-day study period. The groups receiving the AOTPE at 200 mg/kg and 400 mg/kg doses showed a significant reduction in blood glucose levels compared to the positive control group, especially from day 14 onwards (Table 3, Figure No. 3).

**Table. 3 Estimation of Blood Sugar Level**

<table>
<thead>
<tr>
<th>Treatment/Group</th>
<th>Average Blood Glucose (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1st Day</td>
</tr>
<tr>
<td>Group I Negative Control</td>
<td>106.2±2.25</td>
</tr>
<tr>
<td>Group II Positive Control</td>
<td>288.2±2.16</td>
</tr>
<tr>
<td>Group III Alloxan + Glibenclamide</td>
<td>286.8±1.42</td>
</tr>
<tr>
<td>Group IV Alloxan + AOTPE 200mg/Kg</td>
<td>287.0±2.42</td>
</tr>
</tbody>
</table>
Findings are demonstrated as mean ± SEM (n=6); statistically difference at *P<0.0001 compared to Positive Control.

Effect on glycosylated hemoglobin (HbA1c):
The positive control group had significantly higher HbA1c levels compared to the negative control group. In contrast, the groups receiving the AOTPE at 200 mg/kg and 400 mg/kg doses exhibited lower HbA1c levels compared to the positive control group, indicating improved long-term glycemic control (Table 4, Figure No. 4).

Table. 4 Effect of AOTPE on HbA1c level in rat.

<table>
<thead>
<tr>
<th>Groups</th>
<th>HbA1c (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before Treatment</td>
</tr>
<tr>
<td>Negative Control Saline Treatment</td>
<td>5.633 ± 0.0210</td>
</tr>
<tr>
<td>Positive Control Alloxan (100mg/kg)</td>
<td>5.650 ± 0.0226</td>
</tr>
<tr>
<td>Standard Alloxan + Glibenclamide</td>
<td>5.700 ± 0.0365</td>
</tr>
<tr>
<td>Treatment I Alloxan + AOTPE (200mg/kg)</td>
<td>5.617 ± 0.0166</td>
</tr>
<tr>
<td>Treatment II</td>
<td>5.667 ± 0.0333</td>
</tr>
<tr>
<td>-------------</td>
<td>----------------</td>
</tr>
<tr>
<td>Alloxan + AOTPE (400mg/kg)</td>
<td></td>
</tr>
</tbody>
</table>

Findings are demonstrated as mean ± SEM (n=6); statistically difference at *P<0.0001 compared to Positive control.

**Figure No.4 A graphical representation of Effect of AOTPE on HbA1c level in rat.**

**IV. DISCUSSION**

The present study investigated the antidiabetic potential of a combined extract of Althaea officinalis and Thespesia populnea (AOTPE) in Alloxan-induced diabetic rats. The extraction yield of the AOTPE was found to be 25.6% w/w, suggesting an efficient extraction process in obtaining a reasonable amount of the active compounds from the plant materials.

The phytochemical screening of the AOTPE revealed the presence of various bioactive compounds, including flavonoids, alkaloids, glycosides, phenolic acids, sterols, and tannins. These phytochemicals have been reported to possess antioxidant, anti-inflammatory, and antidiabetic properties, which may contribute to the observed antidiabetic effects of the AOTPE\(^{[10,11]}\).

The study demonstrated the antidiabetic potential of the AOTPE in Alloxan-induced diabetic rats. The AOTPE at both 200 mg/kg and 400 mg/kg doses showed a protective effect on body weight, improved glucose tolerance, reduced blood glucose levels, and lowered HbA1c levels compared to the positive control group. The observed protective effect on body weight in the diabetic condition may be attributed to the ability of the AOTPE to regulate glucose metabolism and prevent the excessive loss of body weight, which is a characteristic feature of uncontrolled diabetes\(^{[12]}\). The improvement in glucose tolerance and the reduction in blood glucose levels suggest that the AOTPE possesses the ability to enhance insulin sensitivity and/or insulin secretion, thereby improving glycemic control\(^{[13]}\).
The observed reduction in HbA1c levels indicates that the AOTPE can effectively manage long-term glycemic control in the diabetic condition. HbA1c is a reliable marker of average blood glucose levels over the past 2-3 months and is closely associated with the development of diabetic complications\[^{14}\]. The lowering of HbA1c levels by the AOTPE suggests its potential in preventing or delaying the onset of diabetic complications. The antidiabetic effects of the AOTPE can be attributed to the synergistic actions of the various bioactive phytochemicals present in the extract. These compounds may exert their effects through multiple mechanisms, such as enhancing insulin secretion, improving insulin sensitivity, inhibiting carbohydrate-metabolizing enzymes, and exhibiting antioxidant and anti-inflammatory properties\[^{15,16}\].

**V. CONCLUSION**

In conclusion, the findings of this study suggest that the Althaea officinalis and Thespesia populnea Extract (AOTPE) possesses potent antidiabetic properties and could be a promising therapeutic option for the management of diabetes and its associated complications. Further research is warranted to elucidate the precise mechanisms of action and explore the clinical applications of this bi-herbal extract.

**REFERENCES**


