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# ANTIDIABETIC ACTIVITY OF DELONIX REGIA LEAVES IN NORMAL AND ALLOXAN-INDUCED DIABETIC MICE

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***ABSTRACT***
*The present study was performed to investigate the local Unani ethnomedical claim scientifically by screening the acute hypoglycemic effect of Delonix regia leaves in normal and alloxan-induced diabetic mice, by monitoring the blood glucose level with glucometer. Methanolic extract of Delonix regia leaves has shown statistically significant hypoglycemic activity in normal and alloxan-induced diabetic mice. It also has significantly improved the glucose tolerance in normal mice.*

***Keywords****: Delonix regia,alloxan,tolerance,glucometer,methanolic.*

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## INTRODUCTION

Diabetes mellitus (DM) is a major public health problem in the developed as well as developing countries. India has been projected by W.H.O. as the country with fastest growing population of DM. A local Unani ethnomed ical claim was found regarding the hypoglycemic effect of D. regia leaves. D. regia, syn. Poinciana regia, is an ornamental tree planted in avenues, gardens and roadside in all the warmer and damper parts of India. Hamdard Pharmacopoeia reported D. regia as one of the medicinal plant used in Karachi (Pakistan). Further, there is no scientific study on the hypoglycaemic properties of this plant. Hence in this preliminary study, an attempt has been made to investigate the local Unani ethnomedical claim scientifically by screening the methanolic and aqueous extracts of D. regia leaves in terms of control of blood sugar level and improvement in glucose tolerance.

D.regia has been reported to have broad-spectrum antibacterial and antifungal activities in flower, anti-inflammatory and analgesic activities in flower

and bark5 and it also has been reported to have antioxidant activity.

**OBJECTIVE:**

To investigate the local Unani ethnomedical claim scientifically by screening the acute hypoglycemic effect of Delonix regia leaves in normal and alloxan-induced diabetic mice, by monitoring the blood glucose level with glucometer.

**MATERIALS AND METHODS:**

**Plant material:**

The leaves of D. regia were collected from the local area of Yavatmal district, Maharashtra during Feburary 2014.

**Extraction:**

The fresh leaves (i.e. leaflets) were washed with water, dried under shade and crushed into coarse powder. The powder was loaded into soxhlet extractor in 8 batches of 75 gms each and was subjected to complete extraction with methanol (methanolic extract, ME). Completion of extraction was directly related to the extent that chlorophyll is removed into the solvent and when the tissue debris, on repeated extraction, is completely free of green colour.[7] Aqueous extract (AE) was prepared in the form of decoction, by boiling 400 gms of coarse powder in 500 ml of water. After extraction, the solvent was distilled off and extract was concentrated on heating mantle to a dry residue.

**Pharmacological study:**

**Acute Toxicity Study:**

During preliminary toxicity study, no adverse effect or mortality was observed in albino mice with oral administration of ME and AE up to a high dose of 5 gm/kg B.W. observed for 24 hrs. Hence a high dose of 500 mg/kg B.W. was selected as a test dose.

**Animals:**

Wistar albino mice of both sexes, weighing between 20-30 gm, were used. They were housed in polypropylene cages and were fed on standard laboratory diet and water adlibitum. Animals were fasted overnight before commencing the experiment, but had free access to water. All the drugs (standard and test) as well as vehicle were administered per-orally using infant feeding tube.

**Standard drug:**

Metformin tablet manufactured by USV Limited was used as a standard drug. The dose was selected on the trial and error basis (75 mg/kg B.W.)

**Determination of blood glucose level:**

Blood glucose level was monitored by using Hypoguard Advance Blood Glucose Meter, imported and marketed by Nicholas Piramal Ltd.

**Hypoglycemic screening in normal mice:**

The normal fasted mice were divided into four groups of five each. One group received vehicle only (0.5% CMC) and served as control group. Other group received standard drug Metformin (75 mg/kg) and served as standard group. Remaining two groups received ME and AE at a dose of 500 mg/kg B.W. Blood Glucose level was monitored just prior to and after 2 hrs of drug administration.

**Induction of diabetes:**

Mice were fasted for 18 hrs and experimental diabetes was then induced by three i.p. administration of alloxan monohydrate (150 mg/kg) at intervals of 48 hrs.[8] Seven days after the last administration, the animals were fasted for 18 hrs and blood glucose levels were determined. Animals with fasting blood glucose levels between 200-300 mg/dl were used for the study.

**Hypoglycemic screening in alloxan-induced diabetic mice:**

The diabetic mice were divided into four groups of six each. Group one received vehicle only (0.5 % CMC) and served as control group. Group two received standard drug Metformin (75 mg/kg) and served as standard group. Remaining two groups received ME and AE at a dose of 500 mg/kg B.W. Study for the acute hypoglycemic activity involved determination of blood glucose levels at 0, 1, 2, 3, 5 and 24 hrs after administration of single dose.

**Oral glucose tolerance test:**

The normal fasted mice were divided into 3 groups of five each. Group One received vehicle only (0.5% CMC) and served as control group.

Other group two received standard drug Metformin (75 mg/kg) and served as standard group. Group three received ME at a dose of 500 mg/kg B.W. All the mice were loaded with Glucose solution 2.5 gm/kg B.W. per orally after 0.5 hr of drug administration. Blood Glucose level was monitored just prior to drug administration and 30, 90 and 120 min. after glucose loading.

The project was undertaken with prior approval from Institutional Animal Ethics Committe IBSS College of Pharmacy, Malkapur (Regd.no.-723/07/abc/ CPCSEA). Care was taken to ensure that the animals were treated in the most humane and ethically acceptable manner.

**STATISTICAL ANALYSIS:**

Results are expressed as Mean±SD and analyzed statistically by One Way ANOVA followed by Dunnett's multiple comparison test using Graph Pad In Stat software. All the

The experimental results were compared with control group.

** $p < 0.01$, very significant; *$p < 0.05$, significant; and $^{ns}p>0.05$, Non significant.

**RESULTS:**

**Table no. 1- Hypoglycemic screening in normal mice.**

| Groups | 0 hour | 2 hours |
|---|---|---|
| Control | 107±11.640 | 98.4±11.104 |
| Standard | 102±7.949 | 64±9.471 * |
| ME | 108±8.556 | 70±4.359 * |
| Aqueous extract | 102±6.325 | 103±4.278 |

**Table no. 2- Hypoglycemic screening in diabetic mice.**

| Groups | 0 hr | 1 hr | 2 hr | 3 hr | 5 hr | 24 hr |
|---|---|---|---|---|---|---|
| Control | 225.67±19.7 | 227.33±16.8 | 220.33±13.2 | 224.5±11.9 | 213±14.7 | 233.5±23.4 |
| Standard | 271.33±22.5 | 240±19.05 | 192.8±19.7** | 167±17.32* | 152±21.03* | 274±26.7 |
| ME | 242.5±30.1 | 221.33±28.2 | 181.3±27.2** | 178.6±26.2* | 165.6±24.8* | 249.83±25.2 |
| Aqueous Extract | 230.5±26.86 | 231.17±23.1 | 232.17±27.5 | 230.83±27.3 | 222.5±28.71 | 239.33±26.1 |

**Table no. 3- Oral glucose tolerance test in normal mice.**

| Groups | 0 min | 30 min | 90 min | 120 min |
|---|---|---|---|---|
| Control | 93.8±16.42 | 399.8 ±15.16 | 200.2 ±30.22 | 161.2 ±34.99 |
| Standard | 105.4 ±19.06 | 185.6 ±30.43* | 127.4 ±31.64* | 91 ±17.39 * |
| ME | 107 ±10.51 | 209.4 ±22.38* | 115 ±21.78 * | 111.6 ±17.8** |

**DISCUSSION AND CONCLUSION:**

ME of D. regia leaves significantly reduced the fasting blood glucose level in normal mice after 2 hrs as compared to control group. It also showed significant reduction in fasting blood glucose level in diabetic mice after 2, 3 and 5 hrs interval as compared to control group. Maximum reduction in fasting blood glucose level (%) was seen after 5 hrs of administration of dose with a significance level of $p<0.01$. Metformin showed maximum reduction (%) after 5 hrs with a significance level of $p<0.01$. After 24 hrs, the mean fasting blood glucose levels of all the groups were almost equal to that at 0 hr.

ME of D. regia leaves and Metformin significantly depressed the peak of blood glucose level at 90 min. after glucose loading with a significance level of $p<0.01$.

AE (500 mg/kg B.W.) neither had shown any hypoglycemic activity nor improved glucose tolerance.

Results clearly indicate that the ME of D. regia leaves have significant hypoglycemic activity. These hypoglycemic effects were produced after oral administration of extract to normal and diabetic mice. The exact mechanism of action needs further studies. Acute toxicity studies (no death even with 10 times the effective dose), indicates high margin of safety. However, the above are the preliminary indications and it is worth undertaking further studies on possible usefulness of D. regia leaves in DM.

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