

Research Article

Anti Diabetic Activity of the Ethanolic Extract of root of *Mangifera indica* on Alloxan Induced Diabetes Rats

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ABSTRACT

Root powder of *Mangifera indica* was subjected to hot extraction (soxhlet) with ethanol. After preliminary phytochemical investigation ethanolic extract of the roots of *Mangifera indica* was evaluated for anti diabetic activity against alloxan-induced diabetes in albino rats. The ethanolic extracts showed significant activity as compare to standard glibenclamide.

Keywords: *Mangifera indica*, alloxan-induced, anti diabetic activity, glibenclamide.

INTRODUCTION

Mangifera indica is generally called as Mango tree family (Anacardiaceae). The plant is perennial, diffuse, prostrate, stems 60-120 cm long, slender, more. It is distributed throughout South India, up to 900m in the hills and also in Gujarat. Literature review reveals that root and leaves contain proteins, tannins and flavonoids and also showed significant inhibitory activity against some fungal pathogens causing major diseases in crop plants and stored food grains. Traditionally plant is use in cases of biliousness, rheumatism, excessive heat, intestinal poison, fever, diarrhea, asthma, heart diseases, worms and piles. Here the experiment of root has been evaluated for anti diabetic activity against alloxan induced model using standard drug glibenclamide.

MATERIALS AND METHODS

The Plant *Mangifera indica* (Linn) collected from different regions of Nugivedu, vijayawada, after proper identification by an expert taxonomist Dr.A.Srinivas Rao, Department of botany, VRS&YRN Degree college, Chirala. After due to authentication the roots were dried in shade and powdered to obtain coarse powder.

PREPARATION OF EXTRACT

The coarse powder material (300 g) was successively extracted with ethanol by using soxhlet apparatus. The extract was finally dried

and kept in a desiccator and used for further study.

ANIMALS

Wistar albino rats (150-200g) were selected for either sex, for studies and they were kept in a standard polypropylene cage at room temperature of 27±2 °C, relative humidity 60-70% and well ventilated. They were fed a standard rat pellet and water ad libitum. Animals were deprived of food initially for 16 hrs but had free access to water. Animal ethical committee of the institute approved animal experiment. The acute toxicity study of ethanolic extract was carried out according to OECD guidelines. The LD50 of the ethanolic root extract as per OECD guidelines - 420 is greater than 2000mg/kg (LD50 >2000mg/kg).

ANTIDIABETIC ACTIVITY

Hyperglycemia / diabetes was induced by single Intraperitoneal injection of freshly prepared aqueous solution of alloxan monohydrate 150 mg/kg, to overnight fasted rats. After 48 hrs of alloxan injection, the animals which did not developed hyperglycemia i.e glucose level >200mg/dl, were rejected/replaced with new animals. Immediately after confirmation of diabetes, rats were classified into five groups of six rats each. Standard drug used for treatment, Glibenclamide, 5 mg/kg, ethanolic test extract were prepared, 200mg/kg and 400mg/kg in 2%

Carboxy Methyl Cellulose (CMC) and were given orally. Taking six rats in each five groups did evaluations of antidiabetic effect. Treatment was continued for 21 consecutive days. Before the treatment (0day) and at the end of 7th, 14th and 21st day, blood samples were collected from the tip of the tail and on 21st day blood is collected from retro orbital plexus of each rat under mild ether anesthesia in 1 ml Eppendorf tubes containing 50 μ l of anticoagulant (heparin). Serum separated by centrifugation of blood at 4000rpm for 10mins was subjected for estimating glucose by Glucose oxidase method using semi auto- analyzer. It was done with 1 ml of blood withdrawn on 21st day, from all five groups rats (normal, diabetic control, extracts of 200 and 400mg/kg and standard treated) and stored in a refrigerator until analyzed. And the serum was subjected for the estimation of triglyceride (TGL), HDL, LDL, VLDL and total cholesterol level.

EXPERIMENTAL GROUPS

Rats were divided in different groups as follows.
 Group I: as normal control where rats received normal saline daily.
 Group II: as diabetic control where diabetic rats received normal saline.
 Group III: diabetic rats received glibenclamide 5mg/kg, an oral hypoglycemic agent.
 Group IV: diabetic rats received ethanolic root extract of *Mangifera indica* 200 mg/kg.

Group V: diabetic rats received ethanolic root extract of *Mangifera indica* 400 mg/kg.

Experimental procedure

Blood glucose estimation

Fasting blood glucose levels were determined in all experimental rats initially to determine the diabetic status and thereafter every week during the 21 day study period. Blood was obtained by snipping tail of rat with the help of sharp razor and blood glucose levels were determined using glucometer (Ultra Touch Two, Johnson and Johnson). Each time the tail of the rat was sterilized with spirit.

Serum lipid profile estimation

At the end of 21 days, the blood samples were collected from retro orbital plexus of rats under anesthesia using a glass capillary tube, serum separated for determination of parameters like total cholesterol, HDL- cholesterol and triglycerides using commercially available kits (Span diagnostics), VLDL cholesterol and LDL- cholesterol were calculated using the Friedewald's formula.

$$\text{VLDL} = \text{Triglycerides} / 5$$

$$\text{LDL} = \text{Total cholesterol} - (\text{HDL-CH} + \text{VLDL-CH})$$

Statistical analysis

All results are expressed as the mean \pm SEM. The results were analysed for statistical significance by one way ANOVA followed by Dunnett's Multiple Test for comparison.

Table 1: Effect of ethanolic extract of root of *Mangifera indica* on Blood glucose levels

| S.no | TREATMENT | TC(mg/dL) | TG(mg/dL) | HDL(mg/dL) | LDL(mg/dL) | VLDL(mg/dl) |
|------|----------------------|--------------------|--------------------|------------------|-------------------|--------------------|
| 1. | Vehicle control | 81.48 \pm 0.37 | 73.22 \pm 0.31** | 23.73 \pm 0.36 | 39.72 \pm 0.81 | 14.15 \pm 0.25 |
| 2. | Diabetic control | 222.53 \pm 0.94* | 114.46 \pm 0.60 | 20.91 \pm 0.33 | 132.58 \pm 0.93 | 22.52 \pm 0.52** |
| 3. | Glibenclamide 5mg/kg | 84.85 \pm 0.90 | 81.52 \pm 0.55 | 32.40 \pm 0.37 | 34.55 \pm 0.56 | 17.00 \pm 0.06 |
| 4. | 200mg/kg EEMI | 132.9 \pm 0.48** | 95.02 \pm 0.05** | 40.22 \pm 0.37 | 68.48 \pm 0.51 | 20.04 \pm 0.03 |
| 5. | 400mg/kg EEMI | 108.8 \pm 0.41 | 78.03 \pm 0.05 | 34.02 \pm 0.02 | 45.89 \pm 0.41 | 17.98 \pm 0.06 |

Values are expressed as Mean \pm SE, n = 6 by Dunnett's *t* test; *P < 0.01 Vs Control **P > 0.001 Vs Control

Table 2: Effect of ethanolic extract of root of *Mangifer indica* on lipid levels

| S.no | Groups | BODY WEIGHTS (gm) | |
|------|----------------------|---------------------|----------------------|
| | | 0 th day | 21 st Day |
| 1. | Vehicle control | 148.83 ± 2.04 | 164.11 ± 0.67 |
| 2. | Diabetic control | 156.34 ± 2.72 | 157.53 ± 1.06 |
| 3. | Glibenclamide 5mg/kg | 157.08 ± 0.44 | 161.22 ± 0.90 |
| 4. | 200mg/kg EEMI | 158.69 ± 0.21 | 158.71 ± 1.04 |
| 5. | 400mg/kg EEMI | 151.13 ± 0.65 | 153.66 ± 1.02 |

Values are expressed as Mean ± SE, n = 6 by Dunnett's *t* test;
*P < 0.01 Vs Control **P > 0.001 Vs Control

Table 3: Effect of ethanolic extract of root of *Mangifer indica* on alloxan induced diabetic rats on body weight

| S.no | Groups | Blood glucose levels(mg/dl) | | | |
|------|----------------------|-----------------------------|---------------------|----------------------|----------------------|
| | | 0 th day | 7 th day | 14 th day | 21 st day |
| 1. | Vehicle control | 90.05±0.096 | 90.3±0.20 | 90.23±0.24 | 89.81±0.63 |
| 2. | Diabetic control | 262.53±0.821 | 261.35±0.95* | 260.35±0.39 | 256.87±0.44 |
| 3. | Glibenclamide 5mg/kg | 259.83±1.32 | 120.5±1.87 | 119.05±0.28** | 108.82±0.37 |
| 4. | 200mg/kg EEMI | 260.33±1.366 | 186.88±0.47 | 173.16±3.32 | 169.97±1.09* |
| 5. | 400mg/kg EEMI | 261±2.6430 | 130.66±1.21 | 121.9±0.60 | 105.54±0.64 |

Values are expressed as Mean ± SE, n = 6 by Dunnett's *t* test; *P < 0.01 Vs Control **P > 0.001 Vs Control

Blood glucose levels

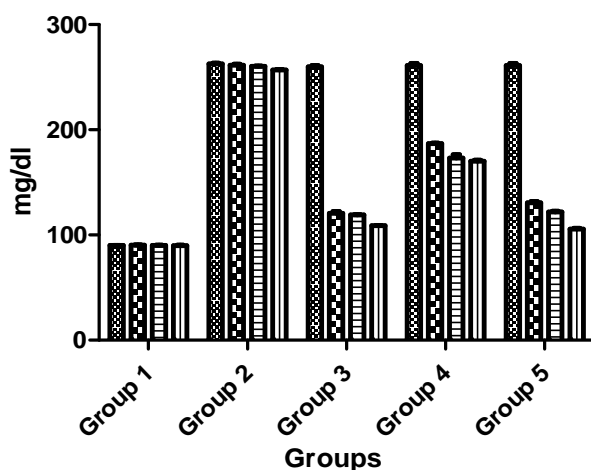


Fig. 1: Effect of EEMI on over all blood glucose levels on alloxan induced diabetic rats. Values are expressed as Mean ± SD of 6 animals in each group.

LIPID PROFILE

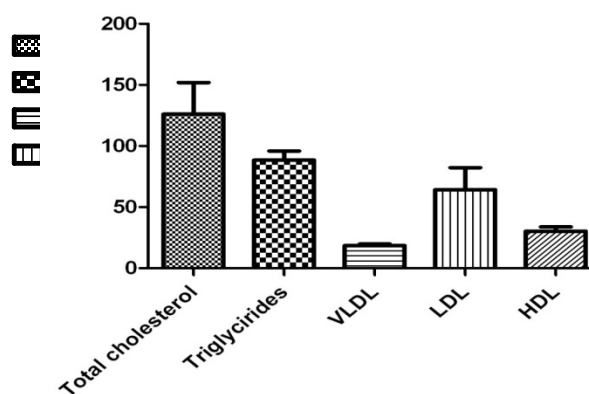


Fig. 2: Effect of EEMI on lipid profile in normal control and alloxan induced diabetic rats. Values are expressed as Mean ± SD of 6 animals in each group.

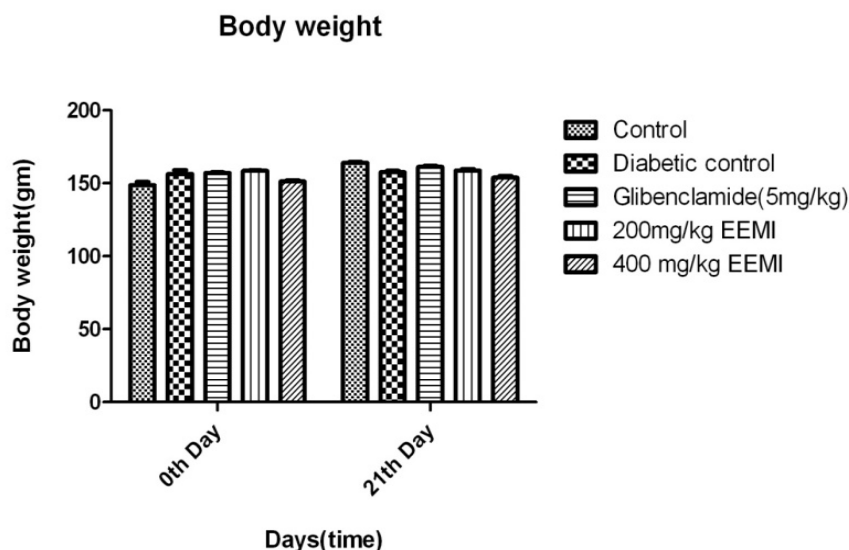


Fig. 3: Effect of EEMI on body weight in normal control and alloxan induced diabetic rats. Values are expressed as Mean \pm SD of 6 animals in each group

RESULTS AND DISCUSSION

Alloxan is widely used to induce diabetes in experimental animals. In alloxan diabetes rats the blood glucose levels were in the range of 260-265 mg/dl, which were considered as severe diabetes. In the standard drug Glibenclamide (5mg/kg) and ethanolic extract (200 mg/kg) and (400mg/kg) treated groups, (Table No.1) the peak values of blood sugar significantly decreased to 108 mg/dl, 165 mg/dl, and 105 mg/dl on the 21st day respectively. Thus, the ethanolic extract (400 mg/kg) was found to be almost significant as standard drug in lowering blood glucose level, whereas the ethanolic extract (400 mg/kg) treated group showed blood glucose level that is comparatively less to ethanolic extract (400 mg/kg) and standard drug.

The ethanolic extract has shown positive test for alkaloids, carbohydrates, glycosides, flavonoids and phenolic compounds, which may be active ingredient in a group or as an individual responsible for activity. The effects of plant extract on different biochemical parameters were tabulated in Table no.2 from which it is revealed that both ethanolic extract (200 mg/kg) and (400 mg/kg) has significantly ($p < 0.01$) reversed the diabetes-induced hyperlipidemia compared to standard drug. A significant percentage reduction of TGL (15.32%), HDL (52.16%), VLDL (15.32%), LDL (64.78%) and total cholesterol (57.46%) in ethanolic extract

(200mg/kg) treated was comparative to standard drug treated groups, TGL (27.38%), HDL (54.86%), VLDL (29.69%), LDL (75.08%) and total cholesterol (65.21%) and reached normal value compared to ethanolic extract (200mg/kg) were TGL (6.00%), HDL (44.58%), VLDL (6.00%), LDL (49.08%) and total cholesterol (57.46%) which was comparatively less.

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