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# EFFECT OF EXTRACTS OF *MURRAYA KOENIGII* LEAVES ON THE LEVELS OF BLOOD GLUCOSE AND PLASMA INSULIN IN ALLOXAN-INDUCED DIABETIC RATS

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**Abstract :** The effect of daily oral administration of aqueous extract (600 mg/kg b.wt.) and methanol extract (200 mg/kg b.wt.) of *Murraya koenigii* Spreng leaves for a period of eight weeks was studied on blood glucose and plasma insulin level in alloxan-induced diabetic rats. Blood glucose levels of diabetic rats treated with aqueous and methanol extracts of *Murraya koenigii* Spreng showed significant reduction ($P<0.05$) as compared to diabetic control groups. Plasma insulin showed significantly high on $43^{rd}$ and $58^{th}$ days of treatment in aqueous and methanol extracts of *Murraya koenigii* treated groups. This suggests that the hypoglycemic effect may be mediated through stimulating insulin synthesis and/or secretion from the beta cells of pancreatic islets of Langerhans.

**Key words :** *murraya koenigii* alloxan diabetes glucose insulin hypoglycemia aqueous extract methanol extract

## INTRODUCTION

Many herbal agents have been described for the treatment of diabetic mellitus in ancient literature (1, 2). They have been shown to have hypoglycemic action in both animals and humans (3). Similarly, fresh leaves as well as aqueous and methanol extract of *Murraya koenigii* have also found to be hypoglycemic in nature (4, 5, 6). However, the actual cause of hypoglycemic due to *Murraya koenigii* treatment has not been established.

The aim of the present study was to examine the effect of aqueous and methanol extracts of *Murraya Koenigii* leaves on the levels of blood glucose and plasma insulin in alloxan-induced diabetic rats.

## METHODS

### Plant material

*Murraya koenigii* leaves were obtained from the plants grown in University of Agricultural Science campus, Bangalore.

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The leaves were cleaned dried and finely powdered. Each gram of dry powdered leaf was equal to five grams of fresh leaves.

The aqueous and methanol extract from the powdered leaf were obtained as per the procedure standardized (5) and were given orally for a period of eight weeks.

**Animals**

Adult male and female rats of Sprague-Dawley strain weighing between 200-300g were obtained from the Laboratory Animal House, University of Agricultural Science, Bangalore. They were kept in polypropylene cages and allowed to get acclimatized to a standard laboratory diet (Amrut Laboratory Animal feed, Nav Maharashtra Chakan Oil Mills Ltd.) and constant room temperature at 22 ° C–24 ° C with 12 hour day and night cycle. Feed and drinking water were provided *ad libitum*.

**Induction of diabetes**

Diabetes was induced by intraperitoneal injection of alloxan monohydrate (5% w/v) in physiological saline at a dose of 200 mg/ kg body weight (5, 6). Alloxan monohydrate was purchased from the IOBA Chemie, Bombay. The diabetic state was confirmed 48h after alloxan injection by weight loss, glucosuria (7) and hyperglycemia (8). Rats with a fasting blood glucose level higher than 200 mg/dl were selected for the study.

**Experimental design and procedures**

The rats were randomly divided into four groups (diabetic T0, diabetic T1, diabetic T2 and diabetic T3) of six animals each of either sex. Diabetic T0-group (comprising T0-male and T0-female rats) was kept as diabetic control. Diabetic T1 (comprising T1-male and T1-female rats) and Diabetic T2-groups (comprising T2-male and T2-female rats) were given orally 600 mg/kg b.wt. of aqueous extract and 200 mg/kg b.wt. of methanol extract (used DMSO as vehicle as methanol extract was completely miscible in it) of *Murraya koenigii* respectively. Diabetic T3-group (comprising T3-male and T3-female rats) served as DMSO control, which was given orally. Retro orbital venous blood samples were collected in the fasting state at specific intervals (Day 0, 7, 14, 21, 28, 43 and 58) and were tested for presence of glucose and insulin concentration.

**Biochemical estimations**

Blood glucose was estimated by Ortho-toluidine method (8). Plasma was obtained and stored at –20°C prior to determination of insulin level. Plasma insulin was determined by radioimmunoassay (9) using a commercially available RIA kit (Board of Radiation and Isotope Technology, BARC, Mumbai, India).

**Statistical analysis**

The quantitative measurements were made on six animals in each group and the values were expressed as mean ± SE. Data obtained were subjected to oneway ANOVA using Bonferroni test with level of significance as $P<0.05$.

## RESULTS

The concentrations of glucose and insulin in both sexes between diabetic T0-group (control) and diabetic T3-group (DMSO control) revealed no significant (P>0.05) difference. On the other hand, significant changes were observed in the experimental groups i.e. in diabetic T1-group and diabetic T2-group.

The injection of alloxan monohydrate resulted in elevation of the blood glucose level. The aqueous and methanol extracts of *Murraya koenigii* leaves exhibited progressive, significant hypoglycemic effect from very first week in diabetic T1 and diabetic T2-groups respectively. This decrease was significant (P<0.05) and continuous up to the end of 8 weeks. (Table I). The insulin concentration was low in alloxan diabetic T0 and diabetic T3-groups. The extracts of *Murraya koenigii* caused significant rise (P<0.05) in the plasma insulin concentration on days 43 and 58 in diabetic T1 and diabetic T2-groups (Table II).

TABLE I: Effect of *Murraya koenigi*i Spreng (Curry leaf) on blood glucose concentration (mg/dl) in diabetic male and female rats.

| *Sex* | *Group* | *Days* | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | *0* | *7* | *14* | *21* | *28* | *43* | *58* |
| **Male** | | | | | | | | |
| | Diabetic T0 | 261.78±7.49 | 270.00±9.66 | 281.78±10.14 | 293.33±11.45 | 296.78±9.89 | 300.00±10.95 | 308.30±11.95 |
| | Diabetic T1 | 265.00±6.71 | 256.78±5.58* | 256.78±5.58* | 218.33±6.54[2]* | 198.33±7.92* | 180.00±7.60* | 153.30±7.61* |
| | Diabetic T2 | 250.00±10.33 | 236.78±8.43* | 213.33±9.89* | 196.78±10.22* | 184.00±10.30* | 168.00±6.63* | 135.00±6.45* |
| | Diabetic T3 | 255.00±8.47 | 263.33±8.03 | 268.33±7.03 | 275.00±8.47 | 276.78±5.58 | 283.33±7.15 | 290.00±7.30 |
| **Female** | | | | | | | | |
| | Diabetic T0 | 260.00±9.66 | 266.67±8.03 | 275.00±6.19 | 288.33±7.92 | 291.67±7.03 | 297.60±4.79 | 305.00±6.45 |
| | Diabetic T1 | 245.00±4.28 | 233.33±4.22* | 218.33±6.01* | 203.33±6.15* | 180.00±5.15* | 168.00±5.58* | 143.30±4.22* |
| | Diabetic T2 | 243.30±10.85 | 226.67±12.02* | 206.67±11.16* | 195.00±9.16* | 174.00±9.92* | 162.00±8.41* | 135.00±8.06* |
| | Diabetic T3 | 246.67±9.55 | 253.33±9.19 | 261.67±9.10 | 270.00±7.30 | 272.00±8.60 | 266.67±8.82 | 280.00±5.77 |

All values are mean ± SE *P<0.05

TABLE II: Effect of *Murraya koenigi*i Spreng (Curry leaf) on Plasma insulin concentration (µU/ml) in diabetic male and female rats

| *Sex* | *Group* | *Days* | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | *0* | *7* | *14* | *21* | *28* | *43* | *58* |
| **Male** | | | | | | | | |
| | Diabetic T0 | 24.67±1.76 | 25.33±2.96 | 24.67±3.84 | 26.00±2.31 | 27.33±0.67 | 27.00±1.53 | 26.00±1.15 |
| | Diabetic T1 | 25.33±1.76 | 27.00±2.65 | 30.67±3.38 | 32.00±3.05 | 32.67±1.33 | 34.67±0.67* | 35.67±0.33* |
| | Diabetic T2 | 24.00±0.58 | 25.00±2.52 | 29.33±2.67 | 32.00±1.15 | 33.00±1.00 | 35.00±1.15* | 37.00±1.53* |
| | Diabetic T3 | 22.33±0.88 | 23.00±1.15 | 22.67±1.86 | 23.00±0.58 | 25.00±1.15 | 24.33±0.33 | 25.00±1.00 |
| **Female** | | | | | | | | |
| | Diabetic T0 | 22.67±1.86 | 22.33±2.33 | 24.67±1.86 | 25.33±3.71 | 23.33±2.19 | 25.67±2.60 | 25.33±1.45 |
| | Diabetic T1 | 25.00±0.58 | 26.33±0.88 | 26.67±1.76 | 28.67±2.40 | 28.33±0.88 | 31.00±0.58* | 33.67±0.33* |
| | Diabetic T2 | 24.67±0.67 | 26.67±2.33 | 29.67±2.60 | 32.33±1.95 | 33.00±2.89 | 34.67±0.88* | 36.00±1.15* |
| | Diabetic T3 | 22.33±0.88 | 21.67±1.20 | 22.33±0.67 | 24.00±1.53 | 25.00±1.86 | 24.67±1.86 | 25.00±0.58 |

All values are mean ± SE *P < 0.05

## DISCUSSION

Alloxan has been shown to destruct beta cells of pancreas producing hyperglycemia. In our experiments the diabetes was characterized by presence of sugar in urine and hyperglycemia.

In diabetic rats plasma glucose was significantly decreased ($P<0.05$) by treatment of aqueous and methanol extracts of *Murraya koenigii* leaves powder. Hypoglycemic effect of *Murraya koenigii* was also reported (4, 5, 6). The mode of action could be either due to increased glycogenesis or decreased glycogenolysis or gluconeogenesis (4) and/or due to insulin secretogogue effect of *Murraya koenigii,* which causes an increased glucose uptake and its utilization by cells (5).

*Murraya koenigii* leaves is found to have alanine, lysine, carbohydrates, iron, calcium, niacin and volatile oils (10). Aqueous and methanol extracts of *Murraya koenigii* treatment for eight weeks has caused significant increase ($P<0.05$) in the insulin concentration in diabetic T1 and T2-groups respectively. The increased insulin secretion in diabetic T1 and diabetic T2-groups could be due to presence of alanine, leucine (11), carbohydrate, niacin, iron and calcium (12, 13) in aqueous extract and the various constituents in the volatile oil (indole alkaloids such as mahanine and mahanimbine, sesquiterpene such as cadinene and monoterpene such as dipentene) (14) in methanol extract of *Murraya koenigii.* The increase in plasma insulin concentration could also be due to the longer lasting stimulant effect on β-cells of pancreatic islets or due to pancreatic β-cells regeneration of *Murraya koenigii* (6).

Plasma glucose concentration when compared between diabetic T1 and diabetic T2-groups revealed that aqueous and methanol extracts of *Murraya koenigii* are equally potent as hypoglycemic agents. The reason for this could be due to similar insulin response observed in both the groups. This revealed that both aqueous and methanol extracts of *Murraya koenigii* are equally potent as insulin secretogogues. Further studies are required for identifying individual effects of the constituents of *Murraya koenigii* leaves separately and/or in combination.

In conclusion, this study confirms that extracts of *Murraya koenigii* leaves have a modulatory role in the treatment of diabetes mellitus and can form a part of therapy in its management.

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