

Diuretic activity of extracts of *Mimusops elengi* Linn. bark

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In the present study, petroleum ether, chloroform, and alcoholic extracts of *Mimusops elengi* Linn. bark (200 mg/kg body weight, p. o.) were tested for diuretic activity. The animals were grouped into five of six animals each. The first group received only 0.9% sodium chloride solution (25 ml/kg body weight) and the second group received the standard drug furosemide (20 mg/kg body weight) in 0.9% sodium chloride solution. Rest of the three groups received each of extracts viz. petroleum ether, chloroform, and alcohol of *M. elengi* bark in a dose of 200 mg/kg body weight suspended in 0.9% sodium chloride solution (p. o). After oral administration, urine was collected and volume was recorded at 5 hours. The highest diuretic activity was presented by the alcoholic extract. Diuretic activity was not observed in chloroform and petroleum ether extracts. We observed a potent diuretic and electrolyte excretion activity in alcoholic extract of *M. elengi* bark.

Key words: Diuretic activity, furosemide, *Mimusops elengi*

INTRODUCTION

Diuretics play an important role in situations of fluid overload, like acute and chronic renal failure, hypercalciuria, cirrhosis of liver, and also as an antihypertensive agent. A number of diuretics like mannitol, thiazides, furosemide, and ethacrinic acid are used in practice.^[1] Drug-induced diuresis is beneficial in many life-threatening disease conditions such as congestive heart failure, nephritic syndrome, cirrhosis, renal failure, hypertension, and pregnancy toxemia.^[2] In traditional medicine, *Mimusops elengi* bark used as a diuretic, dental disease, burning sensation, uterine disorders, ulcer, cardiac diseases, fever, astringent, and aphrodisiac.^[3,4] There are reports available regarding the activity of *M. elengi* bark showing calcium-channel blocking,^[5] antimicrobial^[6] antibacterial activity,^[7] anthelmintic activity,^[8] and hypotension activities.

Since *M. elengi* is used as a diuretic in folklore medicine, the present work has been designed for systematic investigation of the diuretic activity of this plant in albino rats.

MATERIALS AND METHODS

Animals

Male Albino rats (Wistar strain) weighing 150-200 gm, procured from animal house of K.L.E.S's College of Pharmacy, Hubli, were used for the study. Animals were kept for 1 week to acclimatize to laboratory conditions

before starting the experiment; they were given free access to water and standard rat feed except during experimentation.

Preparation of the Extract

The stem barks of *M. elengi* Linn. (Family: Sapotaceae) were collected from mature trees grown locally. The bark of the plant was authenticated by Head of Botany Department, H.S Kothambri Science Institute, Hubli.

The bark of *M. elengi* were shade dried at room temperature and were subjected to size reduction to get coarse powder of desired particle size. The powdered material was subjected to successive extraction in a Soxhlet apparatus using solvents petroleum ether (60-80°C), chloroform, and alcohol. Appearance of colorless solvent in the siphon tube was taken as the end point of extraction. The extracts were concentrated to $\frac{3}{4}$ of its original volume by distillation. The concentrated extracts were taken in a china dish and evaporated on a thermostat controlled water bath until it forms a thick paste. The yield was 12.8% w/w, 3.5% w/w, and 18.2% w/w for petroleum ether, chloroform, and alcohol extract, respectively. Qualitative chemical tests were conducted for above extracts of *M. elengi* to identify the various phytoconstituents.^[9,10] The results of preliminary phytochemical investigation are shown in Table 1.

Diuretic Activity

The method of Lipschitz *et al.*^[11] was employed for the assessment of diuretic activity. The animals were

grouped into five of six animals each and they were fasted and deprived of food and water for 18 hours prior to experiment. The first group received only 0.9% sodium chloride solution 25 ml/kg, p.o. The second group served as the standard group, received the standard drug furosemide 20 mg/kg, p.o. Rest of the three groups received each of extracts viz. petroleum ether, chloroform, and alcohol of *M. elengi* bark in a dose of 200 mg/kg, p.o. suspended in 0.9% sodium chloride solution. All the animals received priming dose of 0.9% sodium chloride solution (25 ml/kg, p.o.).

After oral administration, each animal were placed in an individual metabolic cages specially designed to separate feces and urine at room temperature. The observed parameters were total urine volume for 5 hours, Na⁺, K⁺, and Cl⁻ excreted in urine.

The concentration of the electrolytes in urine is expressed in terms of mmol/l and the urine volume is expressed in ml/100 g/5 hours. Na⁺ and K⁺ concentrations were measured by Flame photometer and Cl⁻ concentration was estimated by titration with silver nitrate solution (N/50) using three drops of 5% potassium chromate as an indicator.^[12]

Statistical Analysis

All values are expressed as mean ± SEM. The values obtained for the above parameters in case of the extracts were compared with standard drug and control group by using one-way Analysis of Variance (ANOVA) followed by Dunnett's test. $P < 0.05$ is considered as significant.

Table 1: Preliminary phytochemical investigation of *Mimusops elengi* bark

| Chemical constituents | Petroleum ether extract | Chloroform extract | Alcoholic extract |
|-----------------------|-------------------------|--------------------|-------------------|
| Flavonoids | – | + | + |
| Glycosides | + | – | + |
| Sterols | + | + | – |
| Saponins | – | – | + |
| Phenolic compounds | + | – | + |
| Alkaloids | + | + | + |

+: Present; -: Absent

Table 2: Summary of the parameters of diuretic activity of different extracts of the *Mimusops elengi* bark

| Group | Urine volume (ml/100 g/5 hours) | Urine pH | Na ⁺ (mmol/l) | K ⁺ (mmol/l) | Cl ⁻ (mmol/l) | Na ⁺ /K ⁺ | T/C (Diuretic index) |
|--------------------|---------------------------------|---------------------------|-----------------------------|----------------------------|----------------------------|---------------------------------|----------------------|
| Control | 4.56 ± 0.35 | 7.00 ± 0.85 | 111.80 ± 3.42 | 70.26 ± 1.08 | 58.38 ± 5.30 | 1.59 | – |
| Standard | 8.51 ± 0.75*** | 6.71 ± 0.06 ^{ns} | 173.00 ± 8.33*** | 82.17 ± 2.75** | 92.62 ± 5.36** | 2.10 | 1.87 |
| Pet ether extract | 5.79 ± 0.20 ^{ns} | 6.73 ± 0.06 ^{ns} | 120.62 ± 1.88 ^{ns} | 72.33 ± 2.99 ^{ns} | 56.62 ± 4.75 ^{ns} | 1.67 | 1.27 |
| Chloroform extract | 6.12 ± 0.38 ^{ns} | 6.73 ± 0.46 ^{ns} | 132.20 ± 3.69 ^{ns} | 74.83 ± 1.85 ^{ns} | 55.86 ± 6.82 ^{ns} | 1.77 | 1.34 |
| Alcoholic extract | 7.43 ± 0.25*** | 6.66 ± 0.09 ^{ns} | 159.30 ± 8.26*** | 78.94 ± 1.28* | 86.30 ± 6.23** | 2.02 | 1.63 |

Data are presented as mean ± SEM; n=6, ns=non-significant, * $P < 0.05$ compared to control, ** $P < 0.01$ compared to control, *** $P < 0.001$ compared to control (ANOVA followed by Dunnett's test). 'T' stands for urine collected for extracts, 'C' stands for urine collected for control and 'S' stands for urine collected for standard drug

RESULTS

In the present study, petroleum ether, chloroform, and alcoholic extracts of *M. elengi* bark were subjected to preliminary chemical tests. Table 1 shows the presence of various chemical constituents. All extracts were subjected to pharmacological screening to evaluate acute toxicity studies and diuretic activity.

From preliminary toxicity studies, it was observed that animals were found to be safe upto a maximum dose of 2000 mg/kg b. w. However, there were few changes in the behavioral response like alertness, touch response, and restlessness

All three extracts viz. petroleum ether, chloroform, and alcoholic extract of *M. elengi* bark were screened for diuretic activity, and the extracts were administered orally at the dose of 200 mg/kg b.w.

Urine volume (ml), urine pH, concentration of Na⁺, K⁺, and Cl⁻ electrolytes (mmol/l) in the urine were recorded. The ratio of the concentration of Na⁺/K⁺ and diuretic index at the end of 5 hours were calculated to assess the diuretic potential of the extracts of *M. elengi* [Table 2].

In the control group, the volume of urine for 5 hours was found to be 4.56 ± 0.35 and in standard group it was found to be 8.51 ± 0.75. In the petroleum ether, chloroform, and alcoholic extract groups, the volume of urine for 5 hours was found to be 6.70 ± 0.20, 6.12 ± 0.38, and 7.43 ± 0.25, respectively.

In the control group, the urine pH for 5 hours was found to be 7.00 ± 0.85. In standard group it was found to be 6.71 ± 0.06. In the petroleum ether, chloroform, and alcoholic extracts groups the urine pH for 5 hours was found to be 6.73 ± 0.06, 6.73 ± 0.46 and 6.66 ± 0.09, respectively.

In the control group, the excretion of sodium for 5 hours was found to be 111.8 ± 3.42 mmol/l and in the standard group it was found to be 173.0 ± 8.33 mmol/l. In the petroleum ether, chloroform, and alcoholic extracts groups, the excretion of sodium for 5 hours was found to be 120.62 ± 1.88, 132.20 ± 3.69, and 159.30 ± 8.26 mmol/l, respectively.

In the control group, the excretion of potassium for 5 hours was found to be 70.26 ± 1.08 mmol/l, and in the standard group, it was found to be 82.17 ± 2.75 mmol/l. In the petroleum ether, chloroform, and alcoholic extracts groups the excretion of potassium for 5 hours was found to be 72.33 ± 2.99 , 74.83 ± 1.85 , and 78.94 ± 1.28 mmol/l, respectively.

In the control group, the excretion of chloride for 5 hours was found to be 58.38 ± 5.30 mmol/l, and in the standard group, it was found to be 92.62 ± 5.36 mmol/l. In the petroleum ether, chloroform, and alcoholic extracts groups the excretion of chloride for 5 hours was found to be 56.62 ± 4.75 , 55.86 ± 6.82 , and 86.30 ± 6.23 mmol/l, respectively

The ratio of the concentration of sodium ions to that of the potassium ions in the control group was found to be 1.59, and in the standard group, it was found to be 2.10. In the petroleum ether, chloroform, and alcoholic extracts groups it was found to be 1.67, 1.77, and 2.02, respectively.

DISCUSSION

An attempt has been made in the present study to evaluate the diuretic activity of petroleum ether, chloroform, and alcohol extracts of *M. elengi* bark.

The results of the present study revealed that the alcohol extract of *M. elengi* present a potent diuretic activity. The diuretic potency was comparable to that of the standard drug furosemide. Here, the alcohol extract of *M. elengi* increases the Na^+ and K^+ excretion, which may be acting like a loop diuretic. A number of compounds have been identified like alkaloids, tannin, saponins, taraxerone, taraxerol, ursolic acid, betulinic acid, α -spinosterol, β -sitosterol, quercitol,^[13] lupeol,^[14] isoretronecyl tigalate,^[15] and mixture of triterpenoid saponins,^[16,17] steroidal saponin, β -sitosterol in the bark of *M. elengi* and may be these constituents are responsible for the diuretic, natriuretic, and kaliuretic activities of *M. elengi*.

The preliminary phytochemical analysis revealed that flavonoids, phenolic compounds, and alkaloids are present in alcoholic extract of *M. elengi*. These natural products might be acting synergistically or individually to produce diuresis. It is also possible that the alcohol extract might manifest cumulative effect of several active principles in the extract.^[18] In conclusion, the present studies support traditional use of *M. elengi* for its diuretic effect.

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