Evaluation of Diuretic Activity of Aqueous Extract of Ripe Fruit Pulp of *Tamarindus indica L.* in Rats

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ABSTRACT

Introduction: Diuresis is an important pharmacological property which is useful in many clinical conditions. There is a need of better diuretics with lesser adverse effects in comparison to currently available diuretics. The study aimed to evaluate the diuretic activity of aqueous extract of fruit pulp of *Tamarindus indica* L. in rats.**Methods:** The study was undertaken with aqueous extract of fruit pulp of *Tamarindus indica* in three doses: 300 mg/kg, 600 mg/kg and1200 mg/kg for its diuretic activity in comparison with standard (furosemide) and vehicle control (normal saline) in Wistar rats. Urine volume and electrolytes were measured after 24 hours of drug administration. **Results:** Aqueous extract of fruit pulp of *Tamarindus indica* at the dose of 1200 mg/kg exhibited significant diuretic activity ($p<0.05$) without significant natriuretic effect. Magnesium excretion was also significantly increased in comparison to control group. **Conclusion:** Aqueous extract of fruit pulp of *Tamarindus indica* has significant diuretic activity in Wistar rats.

Keywords: aqueous extract; diuretic activity; metabolic cage; t*amarindus indica*

INTRODUCTION

Diuresis is an important pharmacological property, useful in many clinical conditions like hypertension, congestive cardiac failure, renal failure, nephrotic syndrome and many other situations where there is fluid overload or electrolyte imbalance.[1] Although numerous diuretics belonging to different classes are available for clinical use, all of them have adverse effects, some of which may prove to be very serious and sometimes even fatal.[1, 2] In addition to this, there is possibility of emergence of resistance to many of the diuretics after prolonged use.[2] For these reasons, there is considerable interest in search for an ideal diuretic which is free of the adverse effects seen with existing diuretics and remains efficacious without development of resistance.

The recent resurgence of plant remedies results from several factors like their effectiveness and lesser side effects compared

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to modern medicines in most of the cases. This is evident by the increase in number of reports supporting the claim of efficacy of medicinal plants. [3,4] *Tamarindus indica* L. is an evergreen tree which is cultivated worldwide and also partly naturalized in the tropics and subtropics. [4] In traditional medicine, different parts of the plants viz. pulp of fruits, seeds, leaves, flowers and bark have been used for various indications.[5-9] Ethno-botanical uses of leaves and ripe fruit pulp of *Tamarindus indica* as diuretic have been reported.[10,11] There is lack of systematic preclinical or clinical evaluation of diuretic activity of fruit pulp. Hence this study was carried out to obtain preclinical evidence for diuretic efficacy of the ripe fruit pulp of *Tamarindus indica* L.

METHODS

The experiment was conducted as per CPCSEA guidelines after obtaining approval from the Institutional Animal Ethics Committee (IAEC).

Materials

- Aqueous extract of ripe fruit pulp of *Tamarindus indica* Linn.(AEFTI)
- Animals: Inbred albino rats of Wistar strain, of either sex, weighing 150-200 g.
- Drugs: Furosemide, (Lasix, 10 mg/mL, Aventis Pharma Limited, India), Normal saline
- Metabolic cages (Nalgene, USA, Model 650-0100. Figure 1)

Procedure

Fresh ripe fruits of *Tamarindus indica* were procured from local market and verified by a botanist. A sample of fruits of *T. indica* was deposited in the department for future reference. Ripe dried pods of *T.amarindus indica* were peeled, deseeded, and air-dried in shade for ten days. Dried fruit pulp is a gummy substance easily miscible in water. Half a kilogram of the dried pulp was tied in a thin muslin cloth and soaked in 1 liter of distilled water for 60 minutes and then boiled for a duration of two hours. The residue tied in the cloth was discarded after lightly squeezing out the liquid part. The extract was concentrated over low flame for about one hour to a thick consistency and then kept in hot air oven for 48 hours at 50°C. The yield was 24% of the pulp. It was then kept in a desiccator for daily use. Fresh solutions of the drug were prepared before experiments . Required quantity, according to the dose and body weight of animals, was dissolved in normal saline for oral administration.

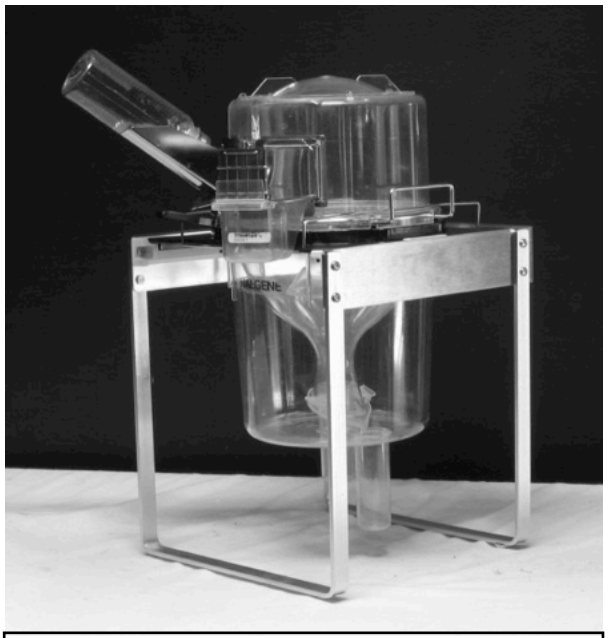

Figure 1: Metabolic cage (Nalgene, USA. Model 650-0100)

Acute toxicity study of the extract was not done as it was reported previously with no

Table 1: Effect of aqueous extract of fruit pulp of *Tamarindus indica* (AEFTI) on 24 hour urine volume, electrolytes and oxalate level in urine (Mean±SEM)(n=6)

Drug Dose (mg/kg)	Urine volume (ml)	Sodium (mmol/L)	Potassium (mmol/L)	Chloride (mmol/L)	Magnesium (mg/dL)	Calcium (mg/dL)	Oxalate (mg/dL)
Control (0.9% saline)	8.80±0.53	61.30±1.48	54.22±3.02	127.77±6.66	3.72±2.06	7.85±1.42	44.40±4.61
Standard (Frusemide 25mg/kg)	16.57±0.38*	75.47±8.30	95.05±19.4	184.00±16.65	no data	no data	54.47±5.74
AEFTI 300 mg/kg	13.80±1.68	80.20±10.68	182±26.57*	154.97±28.81	40.60±10.24*	8.57±0.75	61.05±1.02
AEFTI 600 mg/kg	12.07±1.83	59.55±9.32	193.20±55.79*	222.30±44.88*	56.70±13.12*	6.50±0.28	76.97±0.80**,‡
AEFTI 1200 mg/kg	14.50±2.95*	80.72±11.65	139.42±34.09	207.32±32.39	60.50±16.74*	12.92±5.26	74.10±3.42**,‡

*p<0.05 vs control, ** p <0.001 vs control, ‡ p<0.05 vs standard.

evidence of acute toxicity up to 8 g/kg of the extract.[12]

The method for evaluation of diuretic activity described by Kau *et al*. with modifications was used.[13]

Inbred albino rats of Wistar strain of either sex weighing 150-200 g were used. They were housed in standard environmental conditions of temperature (25±2°C), relative humidity (50–55%) and approximately 12 hours natural light per day of in the institutional animal house. They were fed standard pellet diet (Hindustan Lever rat pellets) and water *ad libitum*.

Thirty animals were divided into five groups: control, standard drug and three treatment groups (n=6). Furosemide solution for injection (Lasix, 10 mg/mL, Aventis Pharma Limited, India) was used in a dose of 25 mg/kg orally as the standard drug. Three doses of aqueous extract of ripe fruit pulp of *Tamarindus indica* (AEFTI): 300 mg/kg, 600 mg/kg and 1200 mg/kg were tested in the treatment groups for diuretic activity. After overnight fasting, animals were orally administered normal saline, standard drug (furosemide) or different doses of test drug, all reconstituted to same volume. Urine output was collected for 24 hours housing each animal in a separate metabolic cage (Nalgene, USA, Model no 650-0100, Figure 1), kept in standard environmental conditions of institutional animal house. The animals had free access to water but not to food. The volume of urine was measured at 24 hours of

drug administration. Urine samples were taken separately for each animal and were quantitatively analyzed for sodium, potassium, chloride, calcium, magnesium and oxalate levels in the biochemistry laboratory. The procedure was repeated thrice after a wash out period of 2 weeks each and mean for each animal was taken for statistical evaluation.

Statistical evaluation was done by using ANOVA in SPSS version 10. Scheffe test was used for test of significance. P value < 0.05 was taken as significant and < 0.001 was taken as highly significant.

RESULTS

Results are summarized in Table 1. Aqueous extract of ripe fruit pulp of *Tamarindus indica* (AEFTI) revealed significant ($p<0.05$) increase in the volume of urine in the dose of 1200 mg/kg. There was no significant increase in the excretion of sodium, but excretion of potassium and chloride was significantly increased, although there was no linear relation with dose of the extract. There was significant increase in urinary magnesium level in all doses compared to control group. There was highly significant increase in urinary oxalate excretion ($p<0.001$) in the rats receiving all doses of AEFTI in comparison to control group and in the groups receiving 600 mg/kg and 1200 mg/kg doses in comparison to standard group.

DISCUSSION

The fruit pulp of *Tamarindus indica* has a strongly acid taste and contains chiefly tartaric acid (about 10%), acid potassium tartrate (about 8%), and invert sugar (from 25 to 40%).The total acidity varies from 11 to 16 %. [14] It contains around 400 compounds and elements with diverse biochemical properties and pharmacological activities including anti-inflammatory, analgesic, antipyretic, antidiabetic and diuretic properties.[6, 7]

Although the aqueous extract of fruit pulp of *Tamarindus indica* showed significant diuretic activity only in the highest dose (1200 mg/kg) used in the study, diuretic effect was seen in all the three doses used. Dose response relationship was not linear within the dose range which may be due to the small sample size and variability in the normal renal physiology of rats. Constituents of fruit pulp of *Tamarindus indica* already characterized to have diuretic activity are magnesium, ascorbic acid, calcium, potassium, and terpinen-4-ol which may be responsible for diuretic activity of the extract although other compounds not yet characterized may also be responsible.[15] Urine analysis also showed significant increase in magnesium levels in the treated rats with all three doses in comparison to the control group. Although this study shows significant increase in urinary oxalate level in rats treated with the extract, tamarind ingestion has been shown to decrease the oxalic acid level and reduced lithogenic property of urine mainly due to tartaric acid content in some preclinical and clinical studies.[16-19] Species difference in renal physiology may explain the apparent contradiction in oxalate excretion in human and rats with intake of fruit pulp of *Tamarindus indica*.

CONCLUSION

This study supports the traditional use of fruit pulp of *Tamarindus indica* as diuretic when significant sodium overload is not present,

although larger scale preclinical and clinical studies are required for confirmation of clinical efficacy and safety. This traditional herbal extract may be further analyzed for the diuretic constituents for development of diuretic agents with better efficacy and tolerability. It is likely that the plant products may prove to be better tolerated diuretics in comparison to the existing ones. Besides having diuretic property, preventive effect on urolithiasis as shown by previous studies could be an advantage.

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