Diuretic effect of Trigonella foenum graecum seed extracts
R Rohini, N Nayeem, A Das

Abstract
Purpose: Trigonella foenum-Graceum Linn commonly known as fenugreek has a long history of traditional use in ayurveda and Chinese medicine and has been used for numerous indications, including labor induction, aiding digestion and to improve metabolism and diuresis. The present study was to evaluate the diuretic activity as traditionally claimed, from successive petroleum ether, benzene, ethanol and aqueous extract of fenugreek seed. Method: The diuretic activity of the successive extract of fenugreek seeds was investigated in wistar rat, according to Lipschitz method. Phytochemical analyses of the extracts were carried out. The Na+, K+ ion concentrations were estimated by flame photometer, and Cl⁻ ion concentration was estimated by titration against silver nitrate. Results: The diuretic response and electrolyte excretion potency from petroleum ether, and benzene extract were remarkable in comparison with the control animals. The extract at 150 and 350 mg/kg body weight showed a dose dependent increase in volume of urine, the naliuretic activity seen by increase in Na⁺/K⁺ ions ratio with respect to control. Conclusion: The study indicates that aqueous and benzene extract as an effective diuretic and naluretic; thus, the work supports the traditional claim about the fenugreek seeds being used as diuretic.

INTRODUCTION
Nature has provided a complete storehouse of remedies to cure all ailments of mankind in which plant kingdom plays a major role. Fenugreek has a long history in Chinese medicines and in ayurveda; the seeds have been used for numerous indications, including labor induction, aiding digestion and to improve metabolism. The seed extract has shown antinoceptic, antidiabetic, antioxidant, hypoglycaemic, antiulcer and gastroprotectivity and anti-hyperlipimedic activity. Previous phytochemical study have revealed the presence alkaloids trigonellie and choline, the leaves contain diosgenin, a saponin and furostanol glycoside.

MATERIALS AND METHOD
Collection: Seeds of Trigonella foenum-graceum was purchased from Bangalore (India) market and air dried and powdered with grinder.

Preparation of the extracts: the powered seed material was extracted with polar solvents like petroleum ether (40-60°C) (PE), benzene (BE), chloroform (CE), ethanol (EE) by using soxhlet method and aqueous extract (AE) was obtained from maceration process using distilled water. The extracts were concentrated under vacuum at 60°C to obtain a constant weight of dry extract. Phytochemical analysis was carried for respective solvent extracts.

Evaluation of diuretic activity: The method of Lipschitz was employed for the assessment of diuretic activity. The male Wister rats weighing 160-200 g were used to study the diuretic activity. The animals were housed in metabolic cage under standard environmental conditions. Food and water were provided ad libitum. Animals were deprived of food and water for 18 hours prior to the experiment and were divided in seven groups of six rats in each. The first group of animals, serving as control, received normal saline (25 ml/kg, p.o.); the second group-received furosemide (10 mg/kg i.p.) in normal saline other groups received doses of extract 150 mg/kg and 350 mg/kg (i.p). Immediately after administration, the animals were placed in metabolic cages (2 per cage), specially designed to separate urine and faeces, kept at 22-25°C. The volume of urine collected was measured at the end of 5 h. During this period, no food and water was made available to animals. The parameters taken were total urine volume, concentration of Na⁺, K⁺ and Cl⁻ in the urine. Na⁺ and K⁺ concentrations were determined by flame photometer and Cl⁻ concentration was estimated by titration with standard silver nitrate solution using potassium chromate solution as indicator.

Statistically analysis: The data was analyzed statistically using ANOVA followed by Dunnett's Multiple Comparison
test. All results are expressed as mean ± standard error.

**Figure 1**

Table 1: diuretic activity of successive fenugreek seed extracts

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Volume of urine (ml)</th>
<th>Sodium (mEq/l)</th>
<th>Potassium (mEq/l)</th>
<th>Chloride (mEq/l)</th>
<th>Na/K ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>2.5 ± 0.01</td>
<td>39.2 ± 3.154</td>
<td>392.0 ± 11.5</td>
<td>193.0 ± 9.36</td>
<td>0.256</td>
</tr>
<tr>
<td>Fenugreek</td>
<td>11.5 ± 8.17**</td>
<td>789.0 ± 3.09</td>
<td>1213.7 ± 11.4**</td>
<td>1534.12.2**</td>
<td>0.582</td>
</tr>
<tr>
<td>PE 100mg/kg</td>
<td>1.38 ± 0.56**</td>
<td>192 ± 3.93</td>
<td>826.162.8</td>
<td>131.264.3**</td>
<td>0.367</td>
</tr>
<tr>
<td>BE 100mg/kg</td>
<td>8.38 ± 3.12**</td>
<td>316.8 ± 3.16**</td>
<td>875.8 ± 4.3**</td>
<td>1226.14.4**</td>
<td>0.309</td>
</tr>
<tr>
<td>BB 100mg/kg</td>
<td>1.5 ± 0.60*</td>
<td>119.6 ± 4.15*</td>
<td>356 ± 1.66</td>
<td>137.7 ± 2.6*</td>
<td>0.295</td>
</tr>
<tr>
<td>BB 200mg/kg</td>
<td>3.5 ± 0.18**</td>
<td>316 ± 0.54**</td>
<td>875 ± 1.75**</td>
<td>115.2 ± 0.3**</td>
<td>0.417</td>
</tr>
<tr>
<td>CE 50mg/kg</td>
<td>1.38 ± 0.26**</td>
<td>119.6 ± 14.1*</td>
<td>368 ± 4.11</td>
<td>131.1 ± 0.1*</td>
<td>0.355</td>
</tr>
<tr>
<td>CE 150mg/kg</td>
<td>3.58 ± 0.37**</td>
<td>184 ± 1.60**</td>
<td>516 ± 1.84**</td>
<td>130.3 ± 0.6*</td>
<td>0.582</td>
</tr>
<tr>
<td>ES 100mg/kg</td>
<td>1.5 ± 0.51*</td>
<td>39.4 ± 3.94</td>
<td>594.3 ± 4.9</td>
<td>153 ± 4.2**</td>
<td>0.351</td>
</tr>
<tr>
<td>ES 150mg/kg</td>
<td>3.5 ± 0.34**</td>
<td>205.0 ± 4.2**</td>
<td>682.1 ± 5.12**</td>
<td>93.7 ± 7.7*</td>
<td>0.403</td>
</tr>
<tr>
<td>AE 150mg/kg</td>
<td>1.42 ± 0.20*</td>
<td>189 ± 0.19</td>
<td>287 ± 3.09</td>
<td>96.3 ± 7.2</td>
<td>0.296</td>
</tr>
<tr>
<td>AE 150mg/kg</td>
<td>4.08 ± 0.93**</td>
<td>668.6 ± 0.46**</td>
<td>856.5 ± 3.84**</td>
<td>97.9 ± 8.7*</td>
<td>0.701</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.E. (n = 6). * P<0.05 and ** P<0.01

Photochemical analysis: of successive extract obtained revealed the presence of terpenoids in petroleum ether extract, flavonoids in benzene, alkaloids in chloroform extract, glycosides from ethanol extract, proteins, carbohydrates, saponin glycoside from aqueous extract. Table 2 presents the yields of successive extract of fenugreek seeds.

**RESULTS AND DISCUSSION**

Significant increase in the volume of urine was seen at 150 mg/kg (p<0.05) and 350 mg/kg (p<0.01) and dose dependent increase in the excretion of electrolyte was exhibited from petroleum ether, benzene chloroform, ethanol and aqueous extracts of fenugreek seeds respectively. Frueosmide treated rats showed a significant increase in volume of urine and marked increase in excretion of sodium, potassium and chloride (p< 0.01) compared to control.

Diuretics are developed for treatment of sodium and water retention in edematous disorders and clinically, they remain the most potent drugs available to relieve symptoms and eliminate edema in the congested patient with heart failure. In the non-congested patient, however, diuretics continue to be used on a purely clinical basis potential to influence on mortality. Diuretics have important role in treatment of hypertension they are useful in reducing the syndrome of volume overload, including orthopnea and paroxysmal nocturnal dyspnoea. They decrease plasma volume and subsequently venous pressure to the heart. Thereby decrease cardiac workload oxygen demand and plasma volume, thus decrease blood pressure. In the present study, it was observed that the successive extracts of fenugreek seeds, showed a dose dependent increase urine output along with increase in excretion of Na+, K+, and Cl ions when compared with the control. The increase in the ratio of concentration of excreted sodium and potassium ion from the aqueous extract, compared to control, is a very essential quality of a good diuretic with less hyperkalaemic side effect.
CONCLUSION

The study supports the diuretic effect of fenugreek seed as traditionally claimed. The active phytochemicals terpenoids, alkaloid, flavonoid, steroidal glycoside are known to be responsible for diuretic activity\(^\text{13-14}\). These active constituent present in the fenugreek seed extracts may be responsible for diuretic and natriuretic activity which is prominently exhibited from benzene and aqueous extract.

ACKNOWLEDGEMENT

The authors are thankful to Prof Suresh Nagpal, Chairman, Krupanidhi Trust of Education and Prof Sunil D, Head Department of Pharmacology for their encouragement and support.

References

Author Information

R.M. Rohini, M.Pharm
Asst Professor, Department of Pharmaceutical Chemistry, Krupanidhi College of Pharmacy

Naira Nayeem, M. Pharm.
Asst Professor, Department of Pharmaceutical Chemistry, Krupanidhi College of Pharmacy

Amit Kumar Das, M Pharm PhD
Professor, Department of Pharmaceutical Chemistry, Krupanidhi College of Pharmacy