Evaluation of the anti diarrheal activity of the plant extracts of *Ficus* species

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**OBJECTIVE:** The Khandesh region of Jalgaon district, India has a dense forest with plenty of medicinal plants which have been used as folklore medicines by the local people for many years. They use different parts of *Ficus* species to treat and cure diarrhea. Depending on the traditional use of some plants as anti diarrheal by local people of that region, the authors have selected three plants (specific parts) to evaluate their anti diarrheal activities in different animal models.

**METHODS:** Wistar albino rats weighing 180 to 200 g of either sex were used in this study. There were eight groups for each individual study with 10 animals in each group. The anti diarrheal profile of the ethanolic extracts of the bark of *Ficus bengalensis* and the leaves of *Ficus racemosa* and *Ficus carica* from the region of Khandesh in Jalgaon district of Maharashtra, India were evaluated by different experimental models, namely, castor oil-induced diarrhea, gastrointestinal motility test, prostaglandin E<sub>2</sub> (PGE<sub>2</sub>)-induced enteropooling in Wistar albino rats.

**RESULTS:** The extracts of *F. bengalensis* (bark), *F. racemosa* (leaves) and *F. carica* (leaves) showed significant inhibitory activities against castor oil-induced diarrhea and PGE<sub>2</sub>-induced enteropooling in rats. The ethanolic extracts at 400 and 600 mg/kg significantly inhibited diarrhea. There was a significant dose-dependent decrease in diarrhea produced by all the three models in rats as compared to that of the standard drug group (*P* < 0.01). Based on the results in experimental rat models, the ethanolic extract of *Ficus* species demonstrated significant reductions in faecal output and frequency of droppings when compared to the castor oil-treated rats (*P* < 0.01). All plant extracts also significantly retarded the propulsion of charcoal meal and significantly inhibited PGE<sub>2</sub>-induced enteropooling.

**CONCLUSION:** All these plant materials can be claimed as potential anti diarrheal agents. The
underlying mechanism appears to be spasmylytic and an anti-enteropooling property by which the different plant extracts produced relief in diarrhea. Tannins and flavonoids present in the plant extracts may be responsible for the antidiarrheal activity.

**KEYWORDS:** antidiarrheals; *Ficus*; plant bark; plant leaves; plant extracts; rats

In recent years, there has been a global trend towards the use of natural phytochemicals presenting in natural products such as fruits, vegetables, and their extracts. From the vast array of the materia medica of the indigenous system, so many plants have been reported to have the activity against diarrhea and act as very useful remedies for the alleviation of human sufferings[8,12]. Recently, it has come to our notice that the people of the Khandesh region of Jalgaon district, India use different parts of various plants to treat and cure diarrhea. The Khandesh region of Jalgaon district has a dense forest with plenty of medicinal plants which have been used as folklore medicines by the local people for many years. Depending on the traditional uses of some plants as antidiarrheal by local people of that region, the authors selected three plants (specific parts) to evaluate their antidiarrheal activities in different animal models.

The tree *Ficus bengalensis* contains tannins, carbohydrates[3] and flavonoids[13]. The barks were reported to have antidiabetic[5], anti-inflammatory[6,7], analgesic antipyretic[8] and antibacterial[9] activities. The leaves contain 9.6% crude protein, 26.84% crude fibres, 2.53% calcium oxide, and 0.4% phosphorus. It yields latex containing caoutchouc (2.4%), resin, albumin, cerin, sugar, and malic acid[10,11].

The root sap of *Ficus racemosa* is used for treating diabetes. Both the root and fruit are credited with hypoglycemic activity. The root juice is reportedly useful for treating dysentery. The stem bark is used to treat menorrhagia, leucorrhoea, gonorrhoea, urinary diseases, haemorrhage and skin diseases. Both the fruits and bark are used extensively in Ayurvedic and Unani medicines[5]. *F. racemosa* was documented to possess anti-inflammatory[13], antipyretic[14] and antiuretic[15] activities. The bark and leaves of *F. racemosa* are reported to possess antidiabetic activity[16,17].

The fig tree (*Ficus carica*) is one of the unique *Ficus* species widely spread in tropical and sub-tropical countries which has edible fruits with high commercial value. The fig has been traditionally used for its medicinal benefits as metabolic, cardiovascular, respiratory, antispasmodic and anti-inflammatory remedies[18,19]. The root is tonic, useful in leucoderma and ringworm. The fruit is sweet, antipyretic, tonic and purgative in inflammation, weakness, paralysis, thirst, diseases of liver and spleen, and pain in chest, and it could cure piles and stimulate the growth of hair. The milky juice is expectorant, diuretic, and dangerous for eyes. Fig latex is used as an anthelmintic remedy[20]. *F. carica* leaves have been reported to have hypoglycaemic[21], hepatoprotective[22], immunomodulatory[23], antipyretic[24] and anthelmintic[25] activities.

In view of the above mentioned references, the following plants of their specific parts were selected for evaluation of their antidiarrheal potential, namely, the bark of *F. bengalensis*, the leaves of *F. racemosa*, and the leaves of *F. carica*.

1 Materials and methods

1.1 Plant materials The bark of *F. bengalensis*, and the leaves of *F. racemosa* and *F. carica* were collected from the Pal forest in Khandesh region of Jalgaon, Maharashtra, India. The plants were identified by the Botanical Survey of India, Pune, Maharashtra, India and the voucher specimens of all the samples dated 09/09/2008 No. BSI/WC/Tech/2008/411, 09/11/2005 No. BSI/WC/Tech/2005/728 and 13/08/2008 No. BSI/WC/Tech/2008/355, respectively.

The plant materials were dried under shade, pulverized by a mechanical grinder, passed

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through a 40-mesh sieve and stored in separate tightly closed containers for further use. The coarse powder of the individual plant was extracted separately in a Soxhlet extraction apparatus by using ethanol as the solvent. After complete extraction, the solvent was removed by vacuum distillation. The semisolid mass (yield 6.5%, 7.20%, and 18.8% in weight ratio with respect to dry powdered material of F. bengalensis, F. racemosa and F. carica, respectively) of each plant extract was stored in a desiccator for further use. On preliminary phytochemical screening, the presence of alkaloids, steroids and tannins was confirmed in all the plant extracts under the tests. The weighed amount of the ethanol extracts of each individual plant material obtained was suspended in 2% (weight/volume) aqueous tragacanth suspension and administered to each individual group of animals at a dose of 400 mg/kg, per oral for evaluation of the anti-diarrheal activity.

1.2 Experimental animals Wistar albino rats weighing 180 to 200 g of either sex were maintained at the uniform laboratory condition in standard steel cages and provided with food and water ad libitum. The animals were maintained under the laboratory condition for an acclimatization period of 7 d before the experiments. There were eight groups for each individual study with 10 animals in each group.

1.3 Experimental methods

1.3.1 Castor oil-induced diarrhea in rats The method followed that of Awouters et al[26] with modifications. In this study, the rats of either sex weighing 180 to 200 g were fasted for 18 h. They were housed in eight steel cages with 10 in each. None of the animals died even at the dose of 3 g/kg of each extract. The doses for the different plant extracts used were selected on a trial basis and were administered orally at the doses of 400 and 600 mg/kg, respectively. The standard control group received diphenoxylate (5 mg/kg) orally in the form of a suspension as the standard drug for comparison. The control group received 2% aqueous tragacanth suspension only. After 1 h of treatment, each animal received 1 mL castor oil orally and then was observed for defecation. Up to 4 h after the castor oil challenge, the presence of characteristic diarrhea droppings was noted in the transparent plastic dishes placed beneath the individual rat cage[27].

1.3.2 Gastrointestinal motility test Rats were fasted for 18 h and placed in eight cages containing 10 in each. Each animal was administered orally with 1 mL of charcoal meal (3% deactivated charcoal in 10% aqueous tragacanth). Immediately after that, the first four groups of animals were orally administered with different extract suspensions (400 and 600 mg/kg), respectively. The standard control group received 0.1 mg/kg atropine intraperitoneally for comparison. The control group was treated with aqueous tragacanth suspension only. After 30 min, each animal was killed and the intestinal distance of the charcoal meal moving from the pylorus was measured and expressed as a percentage of the distance from the pylorus to the caecum[26].

1.3.3 Prostaglandin E2-induced enteropooling Rats were deprived of food and water for 18 h and were placed in eight cages with 10 animals in each. The first six groups of rats were treated with different extracts at oral doses of 400 and 600 mg/kg, respectively. The normal control group was treated with 1 mL of 5% ethanol in normal saline intraperitoneally and then it was treated with aqueous tragacanth suspension as mentioned earlier. Immediately after the extract administration, the prostaglandin E2 (PGE2) (Astra-IDL, India) was orally administered to each rat (100 mg/kg) of the treatment groups, in 5% ethanol in normal saline. The model control group was treated with PGE2 as well as tragacanth suspension. After 30 min, each rat was killed and the whole length of the intestine from the pylorus to the caecum was dissected out and its contents were collected in a test tube and the volume was measured[26].

1.4 Statistical analysis In all the above experiments, the results were expressed as mean ± standard error of mean. Statistical significance tests were performed by one-way analysis of variance and Dunnett t-test by using GraphPad Prism 4.0 software. P<0.05 was considered significant.

2 Results

2.1 Inhibition of castor oil-induced diarrhea The extract of the individual Ficus species material tested, like the standard anti-diarrheal agent, diphenoxylate, significantly inhibited the frequency of defecation when compared with the untreated control rats (P<0.01). All the substances also reduced the wetness of the faecal droppings compared with the untreated control rats (P<0.01) (Table 1).

2.2 Gastrointestinal motility test The extracts of Ficus species plant materials decreased the propulsion of the charcoal meal passing through the gastrointestinal tract when compared with the control group (P<0.01). Atropine reduced the motility of the intestine to a greater extent in comparison with the control group (P<0.01) (Table 2).

2.3 Anti-enteropooling activity PGE2 induced a significant increase in the fluid volume of the rat intestine when compared with the control group (P<0.01). All the extracts of the individual Ficus species plant material significantly inhibited the PGE2-induced enteropooling in rats when compared with the PGE2 control group (P<0.01) (Table 3).
Table 1 Effects of some Ficus species extracts on castor oil-induced diarrhea in rats

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Mean defections in 4 h</th>
<th>Mean number of wet faeces in 4 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (aqueous tragacanth suspension, p.o.)</td>
<td>10</td>
<td>4.00±0.33</td>
<td>4.00±0.33</td>
</tr>
<tr>
<td>Diphenoxyline (5 mg/kg, p.o.)</td>
<td>10</td>
<td>1.37±0.39**</td>
<td>0.0**</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>2.31±0.26**</td>
<td>1.80±0.31**</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>1.54±0.18**</td>
<td>1.20±0.20**</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>2.72±0.25**</td>
<td>1.81±0.36**</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>1.82±0.17**</td>
<td>1.20±0.24**</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>3.72±0.51**</td>
<td>2.35±0.52**</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>2.48±0.35**</td>
<td>1.57±0.35**</td>
</tr>
</tbody>
</table>

** P<0.01, vs control group. p.o.: per oral.

Table 2 Inhibition of gastrointestinal motility by some Ficus species extracts

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Movement of charcoal meal as percentage of the intestinal length (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (aqueous tragacanth suspension, p.o.)</td>
<td>10</td>
<td>79.4±2.8</td>
</tr>
<tr>
<td>Atropine (0.1 mg/kg, i.p.)</td>
<td>10</td>
<td>34.2±1.9**</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>51.4±2.2**</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>42.3±1.7**</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>54.6±2.5**</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>43.2±1.8**</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract (400 mg/kg, p.o.)</td>
<td>10</td>
<td>65.2±2.2**</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract (600 mg/kg, p.o.)</td>
<td>10</td>
<td>40.2±1.6**</td>
</tr>
</tbody>
</table>

** P<0.01, vs control group. p.o.: per oral; i.p.: intraperitoneally.

Table 3 Anti-enteropooling effects of some Ficus species extracts in rats

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Volume of intestinal fluid (mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (aqueous tragacanth suspension, p.o.)</td>
<td>10</td>
<td>0.78±0.11</td>
</tr>
<tr>
<td>PGE₂ control (PGE₂ 100 mg/kg, p.o.)</td>
<td>10</td>
<td>2.97±0.14**</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract plus PGE₂ (400 mg/kg, p.o.)</td>
<td>10</td>
<td>1.36±0.14△△</td>
</tr>
<tr>
<td><em>Ficus bengalensis</em> extract plus PGE₂ (600 mg/kg, p.o.)</td>
<td>10</td>
<td>1.17±0.10△△</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract plus PGE₂ (400 mg/kg, p.o.)</td>
<td>10</td>
<td>1.74±0.15△△</td>
</tr>
<tr>
<td><em>Ficus racemosa</em> extract plus PGE₂ (600 mg/kg, p.o.)</td>
<td>10</td>
<td>1.35±0.12△△</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract plus PGE₂ (400 mg/kg, p.o.)</td>
<td>10</td>
<td>2.16±0.13△△</td>
</tr>
<tr>
<td><em>Ficus carica</em> extract plus PGE₂ (600 mg/kg, p.o.)</td>
<td>10</td>
<td>1.55±0.10△△</td>
</tr>
</tbody>
</table>

** P<0.01, vs control group; △△ P<0.01, vs PGE₂ control group. p.o.: per oral; PGE₂: prostaglandin E₂.

3 Discussion

The above observations suggest that the ethanol extracts of the bark of *F. bengalensis*, and the leaves of *F. racemosa* and *F. carica* at the doses of 400 and 600 mg/kg reduced diarrhea by inhibiting gastrointestinal motility and PGE₂-induced enteropooling. The inhibitory effect of the extracts justifies their uses as nonspecific antidiarrheal agents in folk medicines. Hence, all these plant materials, from the preliminary studies, can be claimed as potential antidiarrheal agents. The underlying mechanism appears to be spasmolytic and the anti-enteropooling property by which the different plant extracts produced relief in diarrhea. Tannins present in many plants and can denature protein to form the protein tannate, which makes the intestinal mucosa more resistant and reduces secretion[10]. The tannins present in the plant extracts may be responsible for the antidiarrheal activity.

4 Acknowledgements

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5 Competing interests

The authors declare that they have no competing interests.

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榕属植物提取物止泻作用的研究

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目的：研究印度贾尔肯地区森林里的榕属植物多年来被当地人用来治疗腹泻。本研究根据当地人的用药传统选取 3 类榕属植物的特定部位来研究它们对不同大鼠腹泻模型的治疗及缓解作用。

方法：按性别随机地选取体重在180～200 g的Wistar大鼠240只，在每个独立实验中，大鼠分别被分为8组，每组10只。通过蓖麻油诱导大鼠腹泻、大鼠胃肠蠕动测试及前列腺素E2诱导大鼠小肠溢积的3个独立实验来评估榕属榕树树皮、榕果榕树叶以及无花果叶的乙醇提取物治疗腹泻的作用。

结果：分别将榕属榕树皮、榕果榕树叶和无花果叶的提取物具有显著抑制由蓖麻油诱发的大鼠腹泻及前列腺素E2诱导大鼠小肠溢积的作用。当给予大鼠400,600 mg/kg的乙醇提取物时，腹泻显著减少。与对照组比较，服用3种植物乙醇提取物的大鼠表现出显著的依赖性腹泻缓解现象（P<0.01）。与蓖麻油诱导大鼠腹泻实验中的对照组相比，榕属植物的乙醇提取物被证实具有显著的降低粪便排泄量及排泄次数的作用（P<0.01）。所有植物提取物具有显著减缓木糖供给大鼠小肠内的推进速度，并抑制前列腺素E2引起的旷场小肠溜积的作用。

结论：印度贾尔肯地区的榕属植物具有止泻作用。这些植物的作用机制主要通过解痉和抗小肠溢积来缓解腹泻，这可能与榕属植物的乙醇提取物内含有鞣酸及类黄酮有关。

关键词：止泻药；榕属；植物皮；植物叶；植物提取物；大鼠