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Antidiarrheal Evaluation of *Ficus racemosa* Linn. Leaf Extract

Subhash C. Mandal, Pulok K. Mukherjee, Kakali Saha, M. Pal, and B. P. Saha[1]

Department of Pharmaceutical Technology, Faculty of Engineering and Technology, Jadavpur University, Calcutta-700 032, India

Abstract – A study was undertaken to evaluate the effect of petroleum ether extract of leaves of *Ficus racemosa* Linn. for its antidiarrheal potential against several experimental models of diarrhea in rats. *Ficus racemosa* leaves extract (FRLE) treated animals showed significant inhibitory activity against castor oil induced diarrhea and inhibited significantly PGE_2 induced enteropooling in rats. It also showed significant reduction in gastrointestinal motility following charcoal meal in rats. The results obtained establish the efficacy of FRLE as an antidiarrheal agent.

Keywords – *Ficus racemosa*, Leaf Extract, FRLE, Antidiarrheal.

Introduction

To combat the problems on diarrhea which is leading cause of mortality in developing countries, the World Health Organization (WHO) has constituted Diarrheal Diseases Control Program (DDCP) which includes studies on traditional medical practices, together with the evaluation of health education and prevention approaches (WHO 1964, DDCP 1979, Lutterodt 1989, Syder and Marson 1989).

Ficus racemosa Linn Syn. *Ficus glomerata* Roxb. (Family Moraceae commonly known as "Jagya-dumur" (Bengali), "Gular" (Hindi) and "Udumbara" (Sanskrit) is a well known moderate sized to large spreading evergreen tree with ovate, ovate-lanceolate leaves (Anonymous 1952, Kirtikar and Basu 1975). The leaves are used in dysentery, billious affection and in dysmenorrhea, barks and fruits are also used in dysentery, diarrhea and in diabetes (Kirtikar and Basu 1975, Nadkarni *et al.*, 1976) In view of the above information and folklore use of the barks and fruits of this plant as an antidiarrheal, the present study was undertaken to evaluate the antidiarrheal activity of petroleum ether extract of *Ficus racemosa* leaves in several experimental models of diarrhea in rats.

Experimental

Plant material – The leaves of *Ficus racemosa* were collected from Hetyasole, Bankura district of West Bengal, India, during the month of July and August. Taxonomical identification of the plant (Reference No. (CNH/7-3(20) Tech.II/95/239) was performed by Botanical Survey of India, Shibpur, Howrah. The specimen sample has been kept in our laboratory for future references. The leaves were shade dried, pulverized by a mechanical grinder, passed through 40 mesh sieve and stored in a well closed container for further use.

Preparation of extract – The powdered

[1]Author for correspondence.

leaves were extracted with petroleum ether (B.P. 60-80 ℃) in a soxhlet extractor. On evaporation of petroleum ether from the petroleum ether extract *in vacuo*, a greenish colored residue was obtained (yield 6.43% (w/w) with respect to the dry starting material) and was stored in a desicator. On preliminary screening the extract showed the presence of steroidal compound (Trease and Evans 1985) which was confirmed by TLC with the solvent system, Hexane : Ethylacetate (1:1) over sililca gel G (Stahl 1969). Further separation of the specific compound is under process. For pharmacological experiments, weighed amount of the dried extract was suspended in a 2% (w/v) aqueous Tween 80 solution.

Animals used – Wistar rats weighing between 180-200 g either sex were used. The animals were housed in standard metal cages and provided with food and water *ad libitum*.

Castor oil induced diarrhea in rats – The method followed here was the method of Awouters *et al.*, 1978, with modification. The original method has included only male wistar rats (220-250 g) and they were starved overnight before treatment with the selected drug in the next morning. In the present study rats of either sex (180-200 g) were fasted for 18 hours. Animals were housed in six perforated steel cages containing six in each. None of the animals died even at an oral dose of 3 g/kg of FRLE. The doses of FRLE used were selected on a trial basis and was administered orally (100, 200, 400 and 600 mg/kg) by gavage as suspension to four groups of animals. The fifth group received diphenoxylate (5 mg/kg) orally as suspension as a standard drug for comparison. Sixth group which served as control, received 2% (w/v) aqueous Tween 80 solution only.

One hour after treatment each animal received 1 ml of castor oil orally by gavage and then observed for defecation. Upto 4th hour after the castor oil challenge the presence of characteristic diarrheal. droppings were noted in transparent plastic dishes placed beneath the individual rat cages.

Gastrointestinal motility tests – Rats were tested for 18 h and placed 6 cages containing six in each. Each animal was administered orally with 1 ml of charcoal meal (3% deactivated charcoal in 10% aqueous Tween 80). Immediately after that, the first four groups of animals were administered orally with the extract (FRLE) suspension (100, 200, 400 and 600 mg/kg). The fifth group received atropine (0.1 mg/kg, i.p.), the standard drug for comparison. The sixth group was treated with aqueous Tween 80 solution as control. Thirty minutes later, each animal was killed and the intestinal distance moved by the charcoal meal from the pylorus was cut and measured and expressed as a percentage of the distance from the pylorus to the caecum.

PGE_2-induced enteropooling – In this method, rats of the same stock as above were deprived of food and water for 18 h and were placed in 6 perforated cages with 6 animals per cage. The first four groups of rats were treated with FRLE (100, 200, 400 and 600 mg/kg p.o.) The fifth and the sixth groups were treated with 2%(w/v) aqueous Tween 80 solution. The sixth group was then treated with 1 ml of 5% (v/v) ethanol in normal saline (i.p.), which served as control. Immediately afterwords, PGE_2 (Astra-IDL Limited, India) was administered orally to each rat (except the control group) (100 μg/kg) in 5% (v/v) ethanol in normal saline. Fifth group served as a PGE_2 control group. After 30 minutes each rat was killed and the whole length of the intestine from the pylorus to the caecum dissected out and its contents were collected in a test tube and the volume was measured.

Statistical analysis was performed by student's 't' test and in all the cases results are expressed as mean ± SE.

Results

Inhibition of castor oil-induced diarr-

Table 1. Effect of *F. racemosa* leaves extract on castor oil-induced diarrhea in rats (Mean±SE)

Oral Pre-treatment at 1-hour	Mean defecations/ group	Mean No. of wet faeces/ group
Tween 80	5.00	5.00
Solution	±0.38	±0.38
Diphenoxylate	1.84**	0.0**
(5 mg/kg)	±0.45	
FRLE	2.79*	1.83**
(100 mg/kg)	±0.41	±0.27
FRLE	2.04**	1.17**
(200 mg/kg)	±0.32	±0.61
FRLE	1.83**	0.93**
(400 mg/kg)	±0.51	±0.19
FRLE	1.32**	0.53**
(600 mg/kg)	±0.25	±0.75

p-values were calculated Vs. control (Tween 80 solution group), **p<0.001, *p<0.01. FRLE=Petroleum ether extract of *F. racemosa* leaves.

Table 2. Inhibition of gastro-intestinal motility by *F. racemosa* leaves extract

Treatment after Charcoal meal	Movement of Charcoal meal as %	p-value
Tween 80 Solution	84.36±3.72	
Atropine (0.1 mg/kg)	45.71±4.47	<0.001
FRLE (100 mg/kg)	73.48±3.16	<0.05
FRLE (200 mg/kg)	65.16±4.25	<0.01
FRLE (400 mg/kg)	54.72±4.51	<0.001
FRLE (600 mg/kg)	43.36±4.15	<0.001

p-values calculated with respect to Tween 80 solution control group (n=6). FRLE=Petroleum ether extract of *F. racemosa* leaves.

hea – The extract (FRLE) like the standard antidiarrheal agent, diphenoxylate, inhibited significantly the frequency of defecation when compared to untreated rats (Table 1.) Both substances also reduced greatly the wetness of faecal droppings.

Effects on gastro-intestinal motility – The extract decreased propulsion of the charcoal meal through the gastrointestinal tract when compared with the control group. Atropine reduced the motility of the intestine significantly (Table 2).

Anti-enteropooling activity – PGE_2 induced significant increase in the fluid volume of rat intestine when compared with control animals receiving only ethanol in normal saline and control vehicle. FRLE significantly inhibited PGE_2-induced enteropooling (Table 3).

Table 3. Anti-enteropooling effect of *F. racemosa* leaves extract

Treatment	Volume of intestinal fluid (ml)	p-value
Ethanol in saline	0.79±0.17	-
PGE_2 in ethanol (100 μg/kg)	2.67±0.11	<0.001*
FRLE (100 mg/kg)	2.11±0.19	<0.05†
FRLE (200 mg/kg)	1.93±0.11	<0.001†
FRLE (400 mg/kg)	1.65±0.09	<0.001†
FRLE (600 mg/kg)	1.25±0.12	<0.001†

Significant: *with respect to ethanol in saline treatment control group. †with respect to PGE_2 treatment (n=6). FRLE=Petroleum ether extract of *F. racemosa* leaves.

Discussion

There has been a statistically significant reduction in the incident and severity of diarrhea produced in experimental animal models. FRLE (100, 200, 400 and 600 mg/kg) like the standard antidiarrheal agent, diphenoxylate, inhibited significantly the frequency of defecation, wetness of faecal droppings when compared with untreated control rats (i.e. rats receiving nither FRLE nor diphenoxylate but castor oil only). The antimuscarine drug atropine and FRLE (in graded doses) decreased intestinal propulsive movement in charcoal meal treated animal models, the former being more potent that then later. The mechanism of this inhibition of motility may be due to the nonspecific spasmolytic activity of FRLE as reported earlier (Chopra *et al.*, 1958, Kirtikar and Basu 1975, Nadkarni *et al.*, 1976). Similarly FRLE inhibited significantly the PGE_2-induced enteropooling. Steroids are useful in treating the diarrhea and also may enhance intestinal absorption of Na^+ and water (Goodman and Gilman, 1996).

The above observations suggest that FRLE in graded doses reduced diarrhea by inhibi-

ting intestinal peristalsis gastrointestinal motility and PGE_2-induced enteropooling. These inhibitory effects of FRLE support the use of the leaves in folk medicine which justify its use as a nonspecific antidiarrheal agent. Hence, FRLE on preliminary studies can be claimed as a potential antidiarrheal agent, the underlying mechanism appears to be spasmolytic and anti-enteropooling property by which the leaves and or its extract produced relief in diarrhea.

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