Vidyodaya J. of Sci, (2005) Vol. 12, pp 57-63

Antidiarrhoeal activity of decoction of *Scoparia dulcis in rats*

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Received on : 10/18/04
Accepted on : 11/01/04

Abstract

The aim of this study was to investigate the antidiarrhoeal potential of a decoction (D) made from *Scoparia dulcis* Linn (Family: Scrophulariaceae), as indicated by traditional practitioners in Sri Lanka, using the caster oil-induced diarrhoeal rat model. Different doses of D (500,1500,3000 mg/kg) were orally administrated and antidiarrhoeal activity monitored over a 6 h period. The D induced a significant ($P<0.05$) and dose- dependent antidiarrhoeal effect from 1h post treatment. This antidiarrhoeal action was mediated via an inhibition in intestinal transit due to impairment of intestinal peristalsis, rather than to an increment in intestinal fluid absorption and/or reduction in secretion. The decoction may have a potential in symptomatic relief of non-specific acute diarrhoeas.

Key words: *Scoparia dulcis*, antidiarrhoeal activity, peristalsis, intestinal transit, traditional medicine

1. Introduction

In Sri Lanka, about 35% of the population is primarily dependent on Ayurvedic and traditional systems of health care, and there is a long history of traditional knowledge associated with plant use (Mahindapala, 2001). Diarrhoea is one of the commonest gastrointestinal disorders treated by traditional practitioners, especially in the rural areas. One Sri Lankan traditional practitioner claims that decoction made from *Scoparia dulcis* Linn (Family Scrophulariaceae, Sinhala: Wal Kottamalli) is therapeutically effective against acute and non-specific forms of diarrhoea. It is and erect perennial herb with a stem up to 60 cm high and with ternately whorled tapering leaves, which are sub acute at apex. The flowers are small, white in colour and arranged in inflorescences of 2-5 and carry white erect hairs at throat and a corolla tube with inserted filaments. In Sri Lanka, this plant is

commonly found in paddy fields and along roadside in dry and low land areas (Dassanayaka, 1981). Tannins, phenolic compounds (Ediriweera and Ratnasooriya, 2002) and a tetracyclic diterpenoid scopadulciol (Hayashi and Hayashi, 1996) are present in this plant. Tannin containing plants are used in treatments of diarrhoea in many other countries (Henirich et al., 1992). Further, experimentally phenoic compounds (Ogata et al., 1993) and tannins (Galvez et al., 1991) are shown to have antidiarrhoeal activity. Therefore, a possibility exists that this plant may possess marked antidiarrhoeal potentials as is claimed. But this has not been scientifically examined so far.

The aim of this study was to evaluate the antidiarrhoeal activity of a decoction (D) of *S. dulcis* using caster-oil induced rat diarrhoea model.

2. Materials and methods

2.1 Plant Material

Fresh plants were collected from several paddy fields in Kalutara, Sri Lanka in May 2001 and authenticated by Dr. (Mrs.) I. Senvirathna, Department of Botany, University of Colombo, Sri Lanka. A voucher specimen (SN-32) has been deposited in the museum of the Department of Zoology, University of Colombo, Sri Lanka.

2.2 Preparation of the decocotion

D was prepared according to the instructions given by the traditional practitioner. Briefly, the plants were cut into small pieces and 120 g of these were added to 1920 ml distilled water (DW) and boiled slowly until the volume was reduced to 240ml. This was filtered and the filtrate was evaporated at 30°C until final volume became 24 ml (yield: 2% w/w). The D was freeze-dried and stored at 4°C. Appropriate weights of the freeze-dried D were dissolved in DW to obtain desired concentrations (500,1500,3000 mg/kg) in 2ml aliquots to be given orally. The highest dose selected was ten times the recommended human dose.

2.3 Animals

Male Wistar rats (250-300g) reared and kept under standardised animal have conditions were used. The animals had free access to water and pelted food (Master Feed Ltd, Colombo, Sri Lanka) at all times.

2.4 Evaluation of antidiarrhoeal activity

Sixty rats were randomly assigned into 5 equal groups (N=12/groups) and oally treated in the following manner. Group 1 with 1ml DW, 2 with 40 mg/kg of reference antidiarrhoeal drug loperamide (Heel et al., 1978) (Janssen Pharmaceuitca, Seoul, South Korea) and groups 3-5 with 500,1500,3000 mg/kg of

D respectively. The doses of D selected was comparable to what we have used previously to investigate antidiuretic (Ediriweera and Ratnasooriya, 2002) analgesic and antihyperalgesic (Rantnasooriya et al, 2003) potential of *S. dulcis.* One hour after dosing, each rat was challenged with 1 ml of castor oil orally to induce diarrhoea and was placed individually in wire-bottomed cages over a clean paper (Ogata et al., 1993). Faeces excreted were then observed at hourly intervals over the next five hours and number of rats in each treatment group producing watery and muddy faececs was recorded.

2.5 Charcoal meal test

Twelve rats were randomly divided in to equal groups (N=6/group).They were starved for 24 h and orally treated with either 1 ml DW or 1500 mg/kg of D. One hour later, 0.5 ml of 10% charcoal suspension in DW was orally administrated to each rat. 20 min later, the rats were killed with an overdose of ether. Their small intestines (pylorus to caecum) were removed, blotted free of blood, unfolded and placed on a white paper. The total length of the small intestine and the distance to which the charcoal plug had moved was determined. The results are expressed as the % distance traveled (Green, 1959).

2.6 Tied-off rat intestine test

This test was performed as described by Ogata et al., 1993. Briefly, 12 rats were randomly assigned in to 2 equal groups (N = 6 / group) and were anaesthetized with ketamine hydrochloride (Neon Laboratories Limited, Mumbai, India) (14 mg/kg, i. m.). In each rat a silicone rubber tube (2 mm outer diameter and 20 mm length) was inserted and tied at the oral end of jejunum and at the anal end of the ileum through an abdominal incision. The entire small intestine was rinsed cautiously with 20 ml saline and then with 10 ml air (to remove the fluid) using a syringe. Subsequently, either 3 ml of D (n=6) (1500 mg/kg) or 3 ml of saline (N=6) was injected into the intestinal loop between the two tubes, using a 27-guage needle and the abdomen was closed. One hour later, the abdomen was opened and the volume of the injected fluid remaining in the intestinal loop was measured to determine the net amount of fluid absorbed.

2.7 Organ bath test

Six rats were sacrificed with an overdose of ether and their small intestine excised, cut into small segments (approximately 20 mm), luminal contents carefully flushed and placed in 50 ml organ baths containing Standard Tyrode solution (Siddiqi, 1982). The organ baths were maintained at 37 ± 1^{0}C and continuously bubbled with air. The spontaneous longitudinal phasic contractions of these segments were recorded under a resting tension of 1g using an isometric transducer (Star Medicals,

Tokyo, Japan) and a pen recorder (Rickadenki Kogyo Company Ltd. Tokyo, Japan). The contractile responses to the D, added in cumulative doses (20, 50, 100 or 200 mg/ L) were recorded. Contact time for each drug concentration was 5-10 min, the pH of the Tyrode solution in the bath was determined using a pH meter (Toa Electronics, Tokyo, Japan), before and after the addition of the D. The results are expressed as % inhibition of the pretreatrtment spontaneous contractions. Complete inhibition was regarded as 100%.

2.8 Statistical analysis

The results are given either as raw data or mean ± SEM. Dose-response relations were determined using Spearman correlations. Statistical comparisons were made using LSD test and Mann-Whitney U test as appropriate. The significance level was set at $P<0.05$.

3. Results

3.1 Castor oil test

The results obtained are depicted in Table 1. As shown, the mid- and high- doses of D and the reference drug, loperamide, induced marked and significant (LSD test, $P<0.05$) antidiarrhoeal effect during the entire study period, except at 2 h. This antidiarrhoeal effect had a rapid onset (within 1 h). The peak effect was also evident at 1 h. There was no significant difference between the antidiarrhoeal effect induced by mid-and high-doses and loperamide. Further, D induced antidiarrhoeal effect was dose-dependent ($r=0.88$; $P<0.05$). At the peak effect, EC_{50} was 499 mg/kg.

3.2 Charcal meal test

1500 mg/kg dose of D significantly (Mann-Whitney U test, $P<0.05$) and markedly impaired the distance the charcoal plug moved in the small intestine (control vs. treatment; 83.5±3.3%vs. 41.8±3.9%).

3.3 Tied off rat intestine test

In this test, a dose of 1500 mg/kg of D failed to significantly (Mann-Whitney U test, $P>0.05$) alter intesntional fluid absorption (control vs. treatment; 1.31± 0.15 vs. 2.20 ±0.30 ml).

3.4 Organ bath test

None of the concentrations of D significantly altered the frequency of spontaneous rhythmic contractions of isolated small intestine segments. However, the D inhibited the amplitude of those contractions (see Table 2) in a dose-related manner, ($r=0.99$;$P<0.05$) following a lag period of 1-2 min. The pH of the Tyrode solution remained unchanged at 7.4, after the addition of D.

4. Discussion

A marked antidiarrhoeal activity was evident in the D of *S. dulcis* with oral administration when tested against castor oil induced-diarrhoea model of rats. This is a novel finding which provides scientific evidence in favour of the claim that *S.dulcis* is therapeutically effective against acute non-specific diarrhoea. The antidiarrhoeal effect of the D was dose-dependent. Further, it had a rapid onset and a fairly long duration of action as loperamide, a clinically used potent antidiarrhoeal agent (Heel et al., 1978) This is indicative of fast absorption and slow metabolism and / or clearance.

The D induced antidiarrhoeal effect was neither due to increased fluid absorption nor decreased fluid secretion as evident in the tied off rat small intestine study. In the charcoal meal test, the D caused a profound impairmnet in intestinal transit possibly related to impairment in peristalsis: since in the isolated rat ileum the D caused a dose-related suppression of the amplitude of spontaneous rhythmic contraction, without altering the organ bath pH. Tannins and phenolic compounds are present in the D (Ediriweera and Ratnasooriya, 2002) and these are known to suppress intestinal motililty (Ogata et al., 1993; Galvez et al., 1991). Thus, the antidiarrhoeal effect of the D may be attributed to its anitmotility action. D is well tolerated in rats even after chronic oral treatment (Ediriweera and Ratnasooriya, 2002).

In conclusion, this study shows antidiarrhoeal activity of *S. dulcis.* The D may be used especially in rural settings, for short-term symptomatic relief of non-specific acute diarrhoeas.

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Table 1

Effect of decoction of *Scoparia dulcis* on caster oil induced diarrhoea in rats

Dose of decoction (mg/kg)	Number of rats used	Number of rats without diarrhoea at various times after castor oil administration				
		1h	2h	3h	4h	5h
0	12	2	4	3	10	5
500	12	4	2	7	3	4
1500	12	10	4	9	9	7
3000	12	10	4	8	10	9
Loperamide (40mg/kg)	12	12	5	8	6	7

Table 2

Inhibitory effect of decoction of *Scoparia dulcis* on spontaneous phasic longitudinal contractions of isolated small intestinal segments of rats (mean ± S E M, N = 7)

Concentration of decoction (mg/L)	% Inhibition	
	Amplitude	Frequency
20	25.86 ±1.47	1.34±0.76
50	21.42±3.97	-0.68±5.35
100	33.80±8.33	-0.49±2.14
200	38.80±0.86	-2.49±1.84

* Spearman Correlation, r = 0.99; < 0.05