**Department of Pharmacology**

GRANT MEDICAL COLLEGE, MUMBAI - 400 008, INDIA.

# Effects of ayurvedic drugs used in the management of diarrhea and dehydration on gastrointestinal motility, Castor oil-induced diarrhea & PGE2-induced enteropooling: An experimental study in rats

## INTRODUCTION

Diarrheal diseases are amongst the leading cause of morbidity and mortality especially in rural parts of the developing countries. The World Health Organization (1) has constituted a diarrhoeal disease control programme (CDD) which includes studies of traditional medicinal practices, together with the evaluation of health education and prevention approaches. Due to limited access to modern medicine as well as due to ethnic and cultural influences, large parts of the population are using traditional botanical remedies for treating diarrheas. Ayurveda, the traditional Indian system of medicines describes a number of botanical drugs in the management of diarrhoeal diseases. In ayurveda many of these drugs are recommended for diverse uses and not only for treating diarrhea (2, 3) and hence cannot be categorised as "classical pharmacotherapeutic anti-diarrheals". In 1976 the World Health Organization launched a global program for prevention and treatment of acute diarrhoea with emphasis on ORT. Oral rehydration therapy rather than conventional 'anti-diarrheals' are being advocated. In Ayurveda as well, the use of herbal extracts to treat diarrhea is recommended in addition to oral rehydration by replacing fluid and salts (4). Over 3,000 years ago Sushruta, the father of ayurveda, recommended 'tepid water with rock salt and molasses' for treatment of diarrhea and dehydration (5, 6).

In the present study we have investigated the effects of the combination of extracts of botanical drugs from an ayurvedic medicine Lomotral oral powder on i)gastrointestinal motility, ii)Castor oil-induced diarrhea and iii)PGE2-induced enteropooling. Lomotral comprises of oral rehydration salts from ayurveda & siddha, namely, Samudra Lavana (Sodium Chloride), Jambira Lavana (Citric Acid), Bida Lavana (Potassium salt) and sugar coupled with extracts of botanical drugs used in ayurveda for treating diarrhoea & dysentery, namely Kutaja (Holarrhena antidysenterica), Vidanga (Embelia ribes) and Daruhalad (Berberis aristata).

MATERIALS AND METHODS

Plant material:

The test drug (TD) was provided by the manufacturers, M/s Pharmaveda in 60 mesh powdered blend. Each kilogram of the blend comprised of the following extracts: 714.28 gms of Holarrhena antidysenterica extract, 142.86 gms Embelia ribes extract and 142.86 gms Berberis aristata extract.

Castor oil-induced diarrhoea in rats:

Diarrhea was induced with castor oil modifying the reported method (7). Our laboratory bred Wistar albino rats, Haffkine strain, of either sex (180-200g) were fasted for 18 hours. Animals were divided into five groups and housed in perforated steel cages containing six animals and were provided with food and water ad libitum. Group I was control group and received only vehicle (1% carboxy methyl cellulose) orally by gastric gavage, Group II was standard reference group and received diphenoxylate (5 mg/kg) orally as suspension in 1% carboxy

methyl cellulose, while Groups II, IV & V received the test drug at dose of 175, 350 & 700 mg/kg respectively by gavage as suspension in 1% carboxy methyl cellulose. The doses of the test drug was selected on the basis of prior pilot work.One hour after treatment each animal received 1 ml of castor oil orally by gavage and then observed for defecation. Up to 4th hour after the castor oil challenge the presence of characteristic diarrhoeal droppings were noted in the transparent plastic dishes placed beneath the individual rat cages.

Gastrointestinal motility tests:

Groupings similar to the castor oil induced diarrhea study were made. Rats were fasted for 18 h and placed in five cages containing six in each. Each animal was administered orally with 1 ml of charcoal meal (5% deactivated charcoal in 10% aqueous tragacanth suspension). Immediately after that, Group I (control group) received vehicle (aqueous tragacanth suspension) orally by gastric gavage, Group II (standard reference group) received atropine (0.1 mg/kg, i.p.), while Groups III, IV & V received the test drug at dose of 175, 350 & 700 mg/kg respectively by gavage as suspension. Thirty minutes later, each animal was killed and the intestinal distance moved by the charcoal meal from the pylorus was measured and expressed as a percentage of the distance from the pylorus to the caecum.

PGE2-induced enteropooling:

In this study, groupings similar to the above studies were made. The animals were deprived of food and water for 18 h and were placed in 5 perforated cages with 6 animals per cage. The Groups I was treated with 1 ml of 5% v/v ethanol in normal saline (i.p.) and then it was

treated with 1% aqueous carboxy methyl cellulose suspension (vehicle control. group). Group II were treated with a solution of PGE2 at a dose of 100 micrograms/kg was made in 5% v/v ethanol in normal saline. Groups III, IV & V received the test drugs at doses of 175, 350 & 700 mg/kg respectively by gavage followed immediately by PGE2 at a dose of 100 micrograms/kg made in 5% v/v ethanol in normal saline. 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 Students't' test.

RESULTS

Effect on castor oil-induced diarrhea:

The test drug (botanical extracts from Lomotral) in a dose related manner significantly inhibited the frequency of defecation and wetness of faecal droppings similar to the reference standard diphenoxylate when compared to vehicle control group (Table 1).

Effects on gastro-intestinal motility:

The test drug marginally but significantly ($p<0.05$) decreased transit of the charcoal meal through the gastrointestinal tract at the highest dose studied (700 mg/kg) when compared with the control group. While at the lower doses there was no significant difference and the results were comparable to the vehicle control group. The standard reference atropine however significantly ($p<0.001$) reduced the gastrointestinal motility (Table 2).

Anti-enteropooling activity:

PGE2 induced significant increase in the fluid volume of rat intestine when compared with control animals receiving only ethanol in normal saline and vehicle. The test drugs from Lomotral however significantly inhibited PGE2-induced enteropooling (Table 3).

REMARKS

There has been a statistically significant reduction in the incidence and severity of diarrhea produced in experimental animal model. Test Drug (botanical extracts from Lomotral) (175, 350 and 700 mg/kg) like the standard anti-diarrhoeal agent, diphenoxylate, inhibited significantly the frequency of defecation, wetness of faecal droppings when compared with untreated vehicle control rats. Botanical extracts from Lomotral at the highest dose used and atropine decreased intestinal transit in charcoal meal treated animal models, atropine being more potent than the Test Drugs. Botanical extracts from Lomotral similarly significantly inhibited the PGE2-induced enteropooling.

Embelia ribes, H. antidysenterica and Berberis aristata have been used over centuries in traditional Indian medicine, amongst other uses, as antihelmintics and in the treatment of diarrhoea and dysentery (8, 9, 10, 11). While the mechanism of anti-diarrheal action of H. antidysenterica is not as yet known, Berberine the active marker from Berberis has been shown to be effective in prevention and treatment of animal models of diarrhea (12, 13, 14). Berberine has a varied pharmacology including anti-microbial (11),

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anti-motility (15) and anti-secretory (16, 17 Guandalini et al., 1987) activities, each of which may contribute to an anti-diarrhoeal effect. Though how each or all of these mechanisms may contribute to the therapeutic usefulness of berberine in the treatment of diarrhoea is not yet firmly established

While castor oil induces diarrhea by increasing peristaltic activity and altering the permeability of the intestinal mucosa to water and electrolytes, PGE2 induces a significant increase in the fluid volume of the rat intestine as compared with control animals receiving only ethanol in normal saline. The above observations suggest that botanical extracts from Lomotral in graded doses reduced diarrhea by mildly inhibiting intestinal peristalsis, minimally affecting gastrointestinal motility and strongly inhibiting PGE2-induced enteropooling. However it is probable that the anti-secretory effects may have also contributed to the anti-diarrheal effects.
Collectively, the present findings suggest that the pharmacological actions produced by botanical extracts from Lomotral are probably due to a synergic action of its constituents

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Table 1
Effect of Test Drugs (botanical extracts from Lomotral) on castor oil-induced diarrhea in rats

| Oral pretreatment at 4 hours | Mean No. of wet faeces/ group |
|---|---|
| Carboxy methyl cellulose suspension (5 ml/ kg) | 24.00 ± 1.63 |
| Diphenoxylate (5 mg/ kg) | 13.26 ± 1.40** |
| Test Drug (175 mg// kg) | 20.08 ± 1.98* |
| Test Drug (350 mg// kg) | 16.83 ± 1.40** |
| Test Drug (700 mg/ kg) | 14.33 ± 1.51** |

Results are (mean ± S.E.), (n = 6), Statistical significance test with control was done by students 't' test, * $p < 0.01$; ** $p < 0.001$

Table 2
Inhibition of gastro-intestinal motility by Test Drugs
(botanical extracts from Lomotral)

| Treatment after charcoal meal | Movement of charcoal meal as % of intestinal length | P-value |
|---|---|---|
| Saline (5 ml/ kg) | 87.40 ± 2.76 | |
| Atropine (0.1 mg/ kg) | 45.09 ± 2.24 | < 0.001 |
| Test Drug (175 mg/ kg) | 85.31 ± 3.12 | |
| Test Drug (350 mg/ kg) | 83.52 ± 4.42 | |
| Test Drug (700 mg/ kg) | 78.43 ± 4.66 | < 0.05 |

Results are (mean ± S.E.), P-value calculated with respect to saline control group (n = 6)

Table 3

Anti-enteropooling effect of Test Drug (botanical extracts from Lomotral)

| Treatment | Volume of intestinal fluid in ml | P-value |
|---|---|---|
| Ethanol in saline (1 ml) | 0.78 ± 0.21 | |
| PGE2 in ethanol (100 microgram/ kg) | 2.97 ± 0.17 | < 0.001* |
| Test Drug (175 mg/ kg) | 2.23 ± 0.34 | < 0.05** |
| Test Drug (350 mg/ kg) | 1.79 ± 0.18 | < 0.01** |
| Test Drug (700 mg/ kg) | 1.28 ± 0.37 | < 0.05** |

Results are mean ± S.E.M.

* With respect to ethanol in saline treatment.

** With respect to PGE2 treatment (n = 6).

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