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Research Article

EVALUATION OF ANTIDIARRHOEAL ACTIVITY OF MEBARID: AN AYURVEDIC FORMULATION

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ABSTRACT

Antidiarrhoeal effect of Mebarid, an Ayurvedic formulation was subjected to pharmacological evaluation. Mebarid, at a dose of 2.5 to 10 ml/kg showed antidiarrhoeal activity in castor oil and magnesium sulphate induced diarrhoea. It has also produced antimotility and antisecretory activity in castor oil induced intestinal transit and intraluminal fluid accumulation in mice. Phytochemical analysis showed the presence of carbohydrates, steroids, triterpenoids, alkaloids, flavonoids and tannins as major constituents.

These results suggest that Mebarid possesses antidiarrhoeal effect may be due to its antimotility and antisecretory effect. Antimotility and antisecretory effect of Mebarid may be due to the presence of different phytochemicals.

Keywords: Mebarid, Diarrhoea, Intestinal transit, Intestinal fluid accumulation.

INTRODUCTION

Diarrhoea is a frequent medical problem.[1] Intestinal infection is the most common cause of diarrhoea worldwide and is responsible for the deaths of 3–4 million individuals each year, mostly in preschool-age children.[2] The major cause of diarrhoea among children in developing countries is malnutrition. In some developing countries, children may suffer from repeated attacks of acute diarrhoea, which contribute to the infection–malnutrition cycle and consequent impairment of growth and development. Acute diarrhoea in children leads to significant morbidity and mortality, even in the wealthy industrialized countries. Chronic diarrhoea is also a major problem in some other clinical situations.[3, 4]

In order to combat the problems of diarrhoea globally, the World Health Organization in Diarrhoeal Disease Control Programme has given a special emphasis on the use of traditional medicines in the control and management of diarrhea, as medicinal herbs constitute an indispensable component of the traditional medicine practiced worldwide due to the economical viability, accessibility and ancestral experience.[5, 6, 7, 8]

As Mebarid is widely used Ayurvedic antidiarrhoeal pediatric syrup, present study was conducted to investigate the antidiarrhoeal, antimotility and antisecretory effect of Mebarid in mice.

MATERIALS AND METHODS

Drugs

i) Mebarid – SG Phyto Pharma (P) Ltd., ii) Castor oil (refined pure) – Paras Chemical Industries, iii) Loperamide hydrochloride – Cipla Pharmaceuticals Ltd., iv) Chlorpromazine hydrochloride – Rhone Poulene (India) Ltd., v) Activated charcoal – E. Merck, vi) Magnesium sulphate – Merck, vii) Atropine sulphate – Sigma chemicals Ltd.

Composition of Mebarid

Each 10 ml of Mebarid contains i) Ajmoda (100 mg), ii) Bael (100 mg), iii) Lodhara (100 mg), iv) Dadim (100 mg), v) Badishep (100 mg), vi) Daruhalad (100 mg), vii) Jaiphal (50 mg), viii) Sunth (50 mg), ix) Ativish (50 mg), x) Kuda (50 mg), xi) Sugar (q.s.).

Animals

"Swiss albino mice" of either sex, weighing; 20 – 25 gm obtained from VIPER, Pune, were used for the experiments. They were kept in standard environmental condition, fed standard food and water ad libitum. All experiments were performed after an overnight fast. The study was approved by Institutional Animal Ethical Committee of Government College of Pharmacy, Aurangabad, Maharashtra, India (GCPA/IAEC/2011/235, 11/03/2011).

Experimental procedure for antidiarrhoeal activity

Acute toxicity

Initially the Mebarid was studied for acute oral toxicity as per revised OECD guidelines number 423. Mebarid was devoid of any toxicity up to 20 ml/kg in albino mice by oral route. Hence for further studies 2.5 to 10 ml/kg doses of these formulations were used.

Castor oil induced diarrhea

The animals were divided in to control, positive and test groups containing six in each group. Each mouse was kept for observation under a glass funnel, the floor of which was lined with blotting paper and observed for 4 h. Diarrhea was induced by administering 0.2 ml. of castor oil orally to mice.[9, 10] The control group received only distilled water (10 ml/kg, p.o.); the positive control group received loperamide (2 mg/kg, p.o.); test group received Mebarid at doses of 2.5, 5, 10 ml/kg, p.o., body weight 30 min before the administration of castor oil. During an observation period of 4 h, the parameters observed were: onset of diarrhoea, total weight of stool output, total weight of wet stools, total number of stool output, and number of wet stools. [11]

Magnesium sulphate induced diarrhea

A similar protocol as for castor oil induced diarrhoea was followed.[12] Magnesium sulphate was given in the dose of 2 g/kg, p.o., to the animals 30 min after pre-treatment with distilled water (10 ml/kg, p.o.,) to the control group, loperamide (2 mg/kg, p.o.) to the positive control group, Mebarid at doses of 2.5, 5, 10 ml/kg, p.o., to test group.

Gastrointestinal motility by charcoal meal

The animals were divided in to control, positive and test groups of six mice each. Each animal was given orally 0.2 ml of charcoal meal (3% charcoal in 5 % gum acacia). The test groups received the Mebarid at doses of 2.5, 5, 10 ml/kg, p.o., body weight immediately after charcoal meal administration. The positive control group received atropine sulfate (5 mg/kg, i.p.), while the control group received distilled water (10 ml/kg, p.o.). After 30 min., the animals were sacrificed and the movement of charcoal from pylorus to caecum was measured. The peristaltic index, which is the distance travelled by charcoal meal to the total length of small intestine expressed in terms of percentage.[13]

Small intestinal secretions

Effect of Mebarid on intestinal secretion was indirectly studied by entero-pooling assay. The mice were divided into different groups and treated with Mebarid (2.5, 5, 10 ml/kg, p.o.), distilled water (10 ml/kg, p.o.) and standard chlorpromazine (30 mg/kg, i.p.) before the oral administration of castor oil 0.2 ml per mouse. These mice were sacrificed 30 min later and entire small intestine from each animal was

weighed and their group average was calculated. The difference in the weight of intestine in control and castor oil treated group was considered as the castor oil induced accumulation of intestinal fluid.[14]

Preliminary phytochemical analysis

Chemical tests were carried out on Mebarid using standard procedures, to identify its major groups of chemical constituents.[15, 16]

Statistics

The results of all experiments were reported as mean ± S.E.M. Statistical analysis was carried out using Student's '*t*'-test. A level of significance of $P < 0.05$ was regarded as statistically significant.

RESULTS

Effect of Mebarid on castor oil induced diarrhoea

In the course of observation for 4 h. after castor oil administration, all the mice in control group produced copious diarrhoea. Pretreatment of mice with the different doses of Mebarid caused a significant dose dependent decrease in the frequency of purging (reduction of number of wet stools and total no of stools) and, weight of wet stools. Mebarid showed 56.09 %, 75.81 %, 90.90 % inhibition of diarrhoea at doses of 2.5 ml/kg, 5 ml/kg and 10 ml/kg while loperamide at dose of 2 mg/kg showed 92.45 % inhibition of diarrhoea as shown in Table 1.

Table 1: Effect of Mebarid on castor oil induced diarrhoea in mice.

Group	Dose (/kg)	Onset of diarrhoea (min)	Total weight of stools (g)	Weight of wet stools (g)	Total number of stools	Number of wet stools	% Inhibition
Control		53 ± 2.11	0.372 ± 0.010	0.35 ± 0.010	13.33 ± 0.33	11.00 ± 0.36	
Mebarid	2.5 ml	83 ± 2.09	0.177 ± 0.006	0.16 ± 0.006	6.16 ± 0.30	4.83 ± 0.30	56.09
Mebarid	5 ml	110 ± 4.47	0.102 ± 0.008	0.091 ± 0.006	3.16 ± 0.30	2.66 ± 0.211	75.81
Mebarid	10 ml	170 ± 4.58	0.040 ± 0.002	0.035 ± 0.003	1.16 ± 0.16	1.00 ± 0.25	90.90
Loperamide	2 mg	223 ± 5.16	0.036 ± 0.002	0.030 ± 0.003	1.0 0± 0.25	0.83 ± 0.16	92.45

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's '*t*' test.

Effect of Mebarid on magnesium sulphate induced diarrhoea

All the mice in control group produced diarrhoea after magnesium sulphate administration during the observation period of 4 h. Pretreatment of mice with the different doses of Mebarid caused a significant dose dependent decrease in the frequency of purging (reduction of number of wet stools and total no of stools) and, weight of wet stools. Mebarid showed 59.19 %, 75.49 %, 91.91 % inhibition of diarrhoea at doses of 2.5 ml/kg, 5 ml/kg and 10 ml/kg while loperamide at dose of 2 mg/kg showed 91.11 % inhibition of diarrhoea (Table 2).

Effect of Mebarid on small intestinal transit

The results revealed that Mebarid inhibited the gastrointestinal transit of charcoal in mice by 20.48 %, 31.02 % and 45.16 % at doses of 2.5 ml/kg, 5 ml/kg and 10 ml/kg respectively while atropine hydrochloride at dose of 5 mg/kg showed 55.94 % inhibition of gastrointestinal transit as shown in Table 3.

Effect of Mebarid on small intestinal secretion

Mebarid reduced the castor oil induced intraluminal accumulation of fluid by 50.69 %, 67.72 % and 80.19 % at doses of 2.5 ml/kg, 5 ml/kg and 10 ml/kg respectively while chlorpromazine hydrochloride at dose of 30 mg/kg showed 89.50 % inhibition of castor oil induced intraluminal accumulation of fluid (Table 4).

Phytochemical screening of Mebarid

The phytochemical analysis of the Mebarid showed the presence of carbohydrates, steroids, triterpenoids, alkaloids, flavonoids and tannins as major constituents.

Table 2: Effect of Mebarid on magnesium sulphate induced diarrhoea in mice.

X	Dose (/kg)	Onset of diarrhoea (min)	Total weight of stools (g)	Weight of wet stools (g)	Total number of stools	Number of wet stools	% Inhibition
Control		41 ± 2.06	0.32 ± 0.01	0.291 ± 0.009	11.50 ± 0.42	8.16 ± 0.30	
Mebarid	2.5 ml	86 ± 3.31	0.135 ± 0.006	0.123 ± 0.005	4.66 ± 0.33	3.33 ± 0.49	59.19
Mebarid	5 ml	113 ± 4.41	0.083 ± 0.005	0.067 ± 0.004	2.83 ± 0.30	2.00 ± 0.30	75.49
Mebarid	10 ml	201 ± 5.20	0.027 ± 0.002	0.024 ± 0.002	0.83 ± 0.16	0.66 ± 0.21	91.91
Loperamide	2 mg	207 ± 6.58	0.030 ± 0.004	0.027 ± 0.006	0.83 ± 0.16	0.66 ± 0.21	91.11

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's '*t*' test.

Table 3: Effect of Mebarid on castor oil induced intestinal transit in mice.

Group	Dose (/kg)	Percent intestinal transit	% Inhibition
Normal		73.30 ± 1.60	
Control		81.33 ± 2.13	
Mebarid	2.5 ml	58.28 ± 1.73	20.48
Mebarid	5 ml	50.55 ± 1.84	31.02
Mebarid	10 ml	40.19 ± 1.48	45.16
Atropine sulphate	5 mg	32.29 ± 1.02	55.94

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's '*t*' test.

Table 4: Effect of Mebarid on castor oil induced intraluminal fluid accumulation in mice.

Experimental Group	Dose (/kg)	weight of small intestine mg	Castor oil induced intraluminal fluid (mg)	% Inhibition
Normal		1123 ± 25		
Control		1628 ± 23	505 ± 40	
Mebarid	2.5 ml	1372 ± 22	249 ± 17	50.69
Mebarid	5 ml	1286 ± 21	163 ± 12	67.72
Mebarid	10 ml	1223 ± 25	100 ± 11	80.19
Chlorpromazine	30 mg	1176 ± 24	53 ± 8	89.50

Values are mean ± standard error of mean. Each value represents average of six determinations. $P < 0.05$ vs. control, student's '*t*' test.

DISCUSSION

Castor oil induces diarrhoea by causing increased secretion of fluid and electrolytes into the lumen of the bowel by intestinal mucosa, resulting in fluid accumulation and a watery luminal content that flows rapidly through the small and large intestines[2]. This is brought about by the irritant effect of ricinoleic acid liberated by pancreatic lipases, which hydrolyse the oil derived from the seeds of *Ricinus communis*.[17, 18] As Mebarid effectively inhibited the castor oil induced diarrhoea, it can be assumed that the antidiarrhoeal action was exerted by antisecretory mechanism.

Magnesium sulphate produces the diarrhoea by osmotic properties, preventing reabsorption of water ions, leading to increase in the volume of the intestinal content. It promotes the liberation of cholecytokinin from the duodenal mucosa, which increases the secretion and motility of small intestine and thereby prevents the reabsorption of sodium chloride and water.[12, 19] Mebarid found to reduce the diarrhoeic condition in this model. Mebarid may have increased the absorption of water and electrolyte from the gastrointestinal tract, since it delayed the gastrointestinal transit in mice as compared to the control.

GI motility describes the contraction of the muscles that mix and propel contents in the gastrointestinal tract. Charcoal meal test in mice is a method used to study the effect of drugs on the motility of intestine.[20, 21] In present study Mebarid was found to be the inhibitor of intestinal motility.

Diarrhoea occurs when the bowels secrete more electrolytes and water than they absorb. Castor oil produces permeability changes in the intestinal mucosa membranes to water and electrolytes resulting in fluid and watery luminal content that flows rapidly through small and large intestines.[22, 23] Mebarid inhibited the castor oil induced intestinal fluid accumulation. Preliminary phytochemical analysis revealed the presence of carbohydrates, steroids, triterpenoids, alkaloids, flavonoids and tannins as major constituents.

CONCLUSION

Mebarid possesses antidiarrhoeal effect may be due to its antimotility and antiseretory effect. Antimotility and antisecretory effect of Mebarid may be due to the presence of different phytochemicals.

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REFERENCES

1. Farthing MJG. Diarrhoea: a significant worldwide problem. International Journal of Antimicrobial Agents 2000; 14: 65-69.
2. Rouf ASS, Islam MS, Rahman MT. Evaluation of antidiarrhoeal activity *Rumex maritimus* root. Journal of Ethanopharmacology 2003; 84: 307-310.
3. Syder JD, Merson MH. The magnitude of the global problem of acute diarrhoeal disease: a review of active survelliance data. Bulletin of the World Health Organisation 1982; 60: 605-613.
4. Lutterodt GD. Inhibition of gastrointestinal release of acetyl choline by quercetin as possible mode of action of *Psidium gujava* leaf extracts in the treatment of acute diarrhoeal disease. Journal of Ethanopharmacology 1989; 25: 235-249.
5. Abdulkarim A, Sadiq Y, Garbiel OA, Abdulkadir UZ, Ezzeldin MA. Evaluation of five medicinal plants used in diarrhoea treatment in Nigeria. Journal of Ethanopharmacology 2005; 101: 27-30.
6. Gricilda S F, Molly T. Study of antidiarrhoeal activity of four medicinal plants in castor oil induced diarrhoea. Journal of Ethanopharmacology 2001; 76: 73-76.
7. Queen RSX, Arockiasamy P, Jeba SD, Ilavarasan R, Alex RV. Ant-Ulcer eavaluation of *Polygonum barbatum linn* leaf extract. International Journal of Pharmacy and Pharmaceutical Sciences 2011; 3: 81-84.
8. Vite MH, Nangude SL, Gorte SM. Anti-Inflammatory effect of ethanolic extract of *Embelia tsjeriam cottam*. International Journal of Pharmacy and Pharmaceutical Sciences 2011; 3: 101-102.
9. Mukhergi, PK, Scha K, Murugesan T, Mandal SC, Pal, M, Scha, BP. Screenig of antidiarrhoeal profiles of some plant extracts of specific region of West Bengal, India. Journal of Ethanopharmacology 1998; 60: 85-89.
10. Shoba, FG, Thomas M. Study of antidiarrhoeal activity of four medicinal plants in castor oil induced diarrhoea. Journal of Ethnopharmacology 2001; 76: 73-76.
11. Adeyemi, OO, Akindele AJ, Ogunleye EA. Evaluation of Antidiarrhoeal effect of *Sanseviera liberic*a Geerome & Labroy (Agavaceae). Journal of Ethanopharmacology 2009; 123: 459-463.
12. Afroz S, Alamgir M, Khan MTH, Jabbar S, Nahar N, Choudhari MSK. Antidiarrhoeal activity of the ethanol extract of *Paederia foetida* Linn (Rubiaceae). Journal of Ethanopharmacology 2006; 105: 125-130.
13. Adeyemi OO, Akindele AJ. Antidiarrhoeal activity of the ethyl acetate of *Baphia nitida* (Papilionaceae). Journal of Ethanopharmacology 2008; 116: 407-412.
14. Mujumdar AM, Misar AV, Upadhye AS. Antidiarrhoeal activity of ethanol extract of the bark of *Dalbergia lanceolaria*. Journal of Ethanopharmacology 2005; 102: 213-216.
15. Harborne JB. Phytochemical methods: A guide to modern techniques of plant analysis, 3rd ed. Delhi; Springer (India) Private Limited, 2005; 1-32.
16. Odebiyi OO, Sofowora EA. Phytochemical screening of Nigerian medicinal plants II. Lioydia 1978; 41: 234-236.
17. Uddin SJ, Shilpi, JA, Alam SMS, Alamgir M, Rahman MT, Sarker SD. Antidiarrhoeal activity of the methanol extract of the barks of *Xylocarpus Moluccensis* in castor oil and magnesium sulphate induced diarrhea models in mice. Journal of Ethanopharmacology 2005; 101: 139-143.
18. Uddin SJ, Mondal K, Shilpi, JA, Rahman MT, Sarker SD. Antidiarrhoeal activity of *Cyperus rotundus*. Fitoterapia 2006; 77: 134-136.
19. Parimala B, Boominath R, Mandal SC. Evaluation of anti-diarrhoeal activity of *Cleome Viscosa* L. extract in rats. Phytomedicine 2002; 9: 739-742.
20. Pillai NR. Antidiarrhoeal activity of Punica granatum in experimental animals. International Journal of Pharmacognosy 1992; 30: 161-168.
21. Rouf ASS, Islam MS, Rahman MT. Evaluation of antidiarrhoeal activity *Rumex maritimus* root. Journal of Ethanopharmacology 2003; 84: 307-310.
22. Mujumdar AM, Upadhye AS, Misar AV. Antidiarrhoeal activity of *Jatropa curcus* root extract in albino mice. Journal of Ethanopharmacology 2000; 70: 183-187.
23. Uchida M, Kato Y, Matsueda K, Shoda R, Muaoka A, Yamato S. Involvement of nitric oxide from nerves on diarrhoea induced by castor oil in rats. The Japnese Journal of Pharmacology 2000; 82: 168-170.