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Evaluation of Anti-Diarrhoeal Activity of Methanolic Extract of *Polycarpaea Aurea Wight & Arn* in Albino Wistar Rats

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ABSTRACT

The purpose of the present study was to evaluate scientifically the anti-diarrhoeal effects of methanolic extract of whole plant of *Polycarpaea aurea Wight & Arn* (MEPA) was studied against castor oil-induced-diarrhoea model and small intestine transit model in rats. Antidiarrhoeal activity of methanolic extract of whole plant of *Polycarpaea aurea Wight & Arn* was investigated in this study using castor oil induced diarrhoea, Small intestinal transit models in rats.the number of droppings and the distance traveled by charcoal in intestine were measured. Standard drug Loperamide (2.5 mg/kg, p.o) was shown significant reductions in fecal output and frequency of droppings whereas MEPA at the doses of 200 and 400 mg/kg p.o significantly ($P<0.001$) reduced the castor-oil induced frequency and consistency of diarrhoea. The gastrointestinal transit rate was expressed as the percentage of the longest distance travelled by the charcoal divided by the total length of the small intestine. MEPA at the doses of 200 and 400 mg/kg significantly inhibited ($P<0.001$) the castor oil induced charcoal meal transit. The MEPA showed marked reduction in the number of diarrhoea stools as well as a modest reduction in intestinal transit. The results obtained establish the efficacy and substantiate the folklore claim as an anti- diarrheal agent. Further studies are needed to completely understand the mechanism of anti-diarrhoeal action of *Polycarpaea aurea Wight & Arn.*

Keywords: Antidiarrhoeal Activity, *Polycarpaea aurea Wight & Arn*, Traditional medicine, Castor oil induced diarrhoea, Small intestinal transit, charcoal.

INTRODUCTION

Polycarpaea aurea Wight & Arn is a herb of annual or perennial, small shrubs with taproots slender to stout, stems erect, branched, terete, leaves opposite, sometimes appearing whorled belonging to the family Caryophyllaceae. It is widespread but mainly of temperate or warm-temperate occurrence in the North hemisphere, with principal centers of distribution in the Mediterranean region and West Asia to West China and the Himalayas, fewer species in Africa, South of the Sahara, America, India and Oceania; 30 genera and 390 species. The whole plant is used as a diabetic and diaphoretic. The native practitioners in and around Chittoor District, India, have claimed that the whole plant is being traditionally used in diarrhea (Madhava Chetty K, 2008). Hence the present work is carried out to evaluate the effect of methanolic extract of whole plant of *Polycarpaea aurea Wight & Arn* in an adult albino wistar rats with anti-diarrhoeal activity by castor oil-induced-diarrhoea and small intestine transit model.

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MATERIALS AND METHODS

Plant collection

The Plant material of *Polycarpaea aurea Wight & Arn* used for investigation was collected from S.V. University at Tirupathi, Chittor (Dist.), Andhra Pradesh, India. The plant was authenticated by Dr K. Madhava Chetty, Assistant Professor, Department of botany, S.V. University, Tirupathi. The voucher specimen of the plant was deposited at the college for further reference.

Preparation of extract

The whole plant of *Polycarpaea aurea Wight & Arn* was collected, washed, cleaned, dried and pulverized in a grinder- mixer to obtain a coarse powder and then passed through 40 mesh sieves. Weighed quantity of powdered drug was extracted successively with methonol

using Soxhlet apparatus. The extraction was carried out until the extract becomes colorless. . The extract was prepared. The solvent was evaparator from extract by distillation under reduced pressure. The dried extract thus obtained was kept in desiccator and was used for further experiment. Percentage yield of Methonolic Extract of *Polycarpaea aurea Wight & Arn* was found to be 15.8% w/w.

Preliminary phytochemical screening

The freshly prepared crude methanolic extract of *Polycarpaea aurea Wight & Arn* was qualitatively tested for the presence of phytochemical constituents by standard methods (Harborne JP, 1973).

Acute toxicity studies

The acute toxicity studies of was determined as per the OECD guideline no. 423 (Acute toxic class method) (OECD, 2002). It was observed that the test extract was not lethal to the rats even at 2000mg/kg dose. Hence, $1/10^{th}$ (200mg/kg) and $1/5^{th}$ (400mg/kg) of this dose were selected for further study.

Animals used

Albino wistar rats (150-230g) of either sex were obtained from the animal house in C.L. Baid Metha College of Pharmacy, Chennai. The animals were maintained in a well-ventilated room with 12:12 hour light/dark cycle in polypropylene cages. The animals were fed with standard pellet feed (Hindustan Lever Limited., Bangalore) and water was given *ad libitum*. Ethical committee clearance was obtained from IAEC (Institutional Animal Ethics Committee) of CPCSEA (Ref No. SVCP/IAEC / 16-0049 dated 10/02/2011).

Castor oil-induced diarrhoea

Animals were fasted for 24 h but allowed free access to water. Rats were divided into four groups of six animals each, diarrhoea was induced by administering 2 ml of castor oil orally to rats (Nwafor PA *et al*., 2005, Robert A *et al*., 1976). Group I treated as control (2 ml/kg, p.o. saline), group II received loperamide (2.5 mg/kg p.o) served as standard and group III and IV received MEPA (200 and 400 mg/kg, p.o) 1 hr before castor oil administration. Then observed for consistency of faecal matter and frequency of defaecation for 4 hrs.

Small intestinal transit

Rats were fasted for 18 hr divided into five groups of six animals each, Group I received 2ml normal saline orally, group II received atropine (2.5 mg/kg, i.p.), group III and IV received MEPA 200 and 400 mg/kg p.o respectively, 1 hr before administration of castor oil. One ml of marker (10% charcoal suspension in 5% gum acacia) was administered orally 1 hr after castor oil treatment (Izzo AA, 1996). The rats were sacrificed after 1h and the distance traveled by charcoal meal from the pylorus was measured and expressed as percentage of the total length of the intestine from the pylorus to caecum**.**

RESULTS

Acute toxicity studies

Acute toxicity study in which the animals treated with the MEPA at the higher dose of 2000mg/kg did not manifest any significant abnormal signs, behavior changes, body weight changes or macroscopic findings at any time of observation. There was no mortality in the above mention dose at the end of the 14 days of observation.

Phytochemical Screening

The results of preliminary phytochemical screening of the methonolic extract of Polycarpaea *aurea Wight & Arn* revealed that presence of alkaloids, carbohydrates, tannins, phenols, gums and mucilage, aminoacids and absence of saponins and steroids.

Castor oil-induced diarrhoea

After 30 min administration of castor oil the diarrhoea was clinically apparent in all the animals of control group, for the next 4 h. This was markedly reduced by loperamide (2.5 mg/kg p.o) (75%). A similar marked reduction in the number of defecations over four hours was achieved with *P. aurea* at the doses of 200 or 400 mg/kg p.o. MEPA 200 and 400 significantly inhibited the defecation (25% and 50%) MEPA 200 and 400 mg/kg, p.o. dose of extract delayed the onset of diarrhoea. (Table.1 and Figure.1).

Small intestinal transit

The percent intestinal transit was increased with control (96%), but it was reduced in both doses of extract, and much more markedly by atropine (79%). MEPA 200 mg/kg, p.o dose of extract produced 81% intestinal transit induced by castor oil respectively. Whereas, MEPA 400 mg/kg, p.o dose produced 86% of castor oil induced charcoal meal transit (Table.2 and Figure.2).

DISSCUSSION

Diarrhoea results from an imbalance between the absorptive and secretory mechanisms in the intestinal tract, accompanied by excess loss of fluid in the faeces. At doses of 200 and 400 mg/kg, the methanol extracts of *Polycarpeae aurea Wight & Arn.* showed significant anti-diarrhoeal activity against castor oil-induced diarrhoea as compared with the control group. It significantly ($P<0.01$) reduced the frequency of diarrhoea and consistency of defecations. The MEPA also showed a dose related decrease in castor oil-induced diarrhoea.

Several mechanisms have been supposed to be involved in the diarrhoeal effect of castor oil. These include Castor oil is decreases fluid absorption, increases secretion in the small intestine and colon, and affects smooth muscle contractility in the intestine. Castor oil produces diarrhoeal effect due to its active component of recinoleic acid, inhibition of intestinal Na^+, K^+-ATP ace activity to reduce normal fluid absorption, activation of adenylyl cyclase, stimulation of prostaglandin formation, platelet-activating factor and recently nitric oxide was contribute to the diarrhoeal effect of castor oil (Ammon HV *et al*., 1985)**.** Despite the fact that number of mechanisms has been involved for the diarrhoeal effect of castor oil, it has not been possible to define its correct mechanism of action**.** MEPA may act against to above any one of the mechanism (Nell G *et al*., 1984).

Table.1 Effect of methanolic extract of *Polycarpaea aurea Wight & Arn* on castor oil induced diarrhoea in rats

Groups	Treatment/ Dose (mg/kg)	Mean defecation in 4 hrs	% inhibition of defecation
1	Disease control (saline 2ml/kg)	4	---
2	Standard loperamide (2.5mg/kg)	1	75
3	MEPA (400mg/kg)	2	50
4	MEPA (200mg/kg)	3	25

Table.2 Effect of methanolic extract of *Polycarpaea aurea Wight & Arn* on small intestine transit in rats

Groups	Treatment / Dose (mg/kg)	Total length of intestine	Distance travelled by marker	% of intestinal transit
1	Disease control (saline 2ml/kg)	76	73	96
2	Standard atropine (2.5mg/kg i.p.)	78	62	79
3	MEPA (400mg/kg)	72	59	81
4	MEPA (200mg/kg)	76	66	86

Figure.1 Effect of methanolic extract of *Polycarpaea aurea Wight & Arn* on castor oil induced diarrhoea in rats

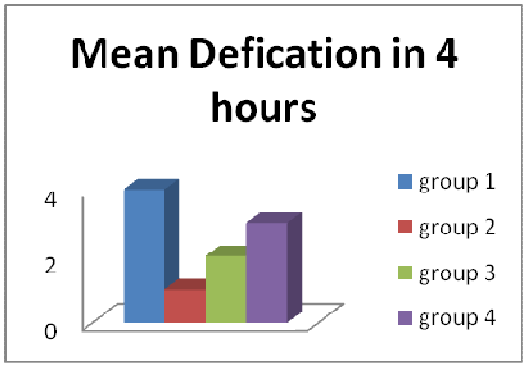
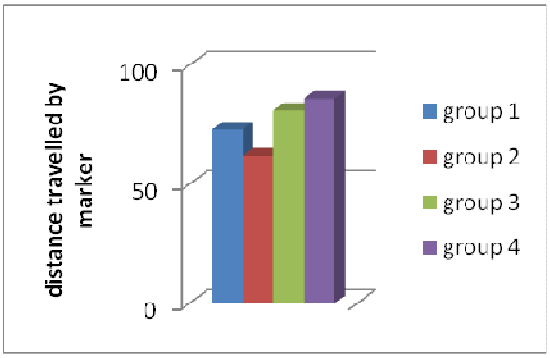


Figure.2 Effect of methanolic extract of *Polycarpaea aurea Wight & Arn* on small intestine transit in rats



The MEPA significantly reduced the castor oil induced intestinal transit as compared with control group**.** In this study, atropine decreased intestinal transit possibly due to its anti-cholinergic effect (Beyer T, 2001). In castor oil induced diarrhoea, the liberation of ricinoleic acid (Phillips RA *et al*., 1965) results in irritation and inflammation of the intestinal mucosa, leading to release of prostaglandins, which results in stimulation of secretion by prevents the reabsorption of NaCl and water (Greenbargena NJ *et al*., 1978). Probably MEPA increased the reabsorption of NaCl and water by decreasing intestinal motility as observed by the decrease in intestinal transit by charcoal meal.

CONCLUSION

The present study has shown that methanolic extract of *Polycarpeae aurea Wight & Arn.* is a potential therapeutic option in the effective management of diarrhoea, thus justifying its widespread use by the local population for these purposes. Concerted efforts are being made to fully investigate the mechanisms involved in the pharmacological activities of *Polycarpeae aurea Wight & Arn*. and phytochemical studies are also in progress to isolate and characterize the active constituents of *Polycarpeae aurea Wight & Arn*. The isolated compound may serve as useful prototypes of anti-diarrhoeal drugs of natural origin possessing the desired pharmacological activities while lacking certain untoward effects.

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