



Investigation of Antidiarrhoeal and Antimotility Activities of Methanolic Extract of *Musa Sapientum* Flowers and Fruit Peels

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ABSTRACT

The objective of the present study was investigation of antidiarrhoeal and antimotility activities of methanolic extract of *musa sapientum* (family- Musaceae) flowers and fruit peels. The phytochemical analysis of both extracts of *Musa sapientum* revealed the presence of alkaloids, carbohydrates, glycosides, saponins, proteins, coumarins, flavonoids, triterpenoids, tannins and phenolic compounds. The extracts were evaluated for castor oil- induced diarrhoea and intestinal motility test in rats. Both the methanolic extracts were significantly ($p < 0.05$) and dose dependently reduced frequency of stooling in castor oil-induced diarrhoea and anti-motility action in intestinal motility test. These findings suggest that the methanolic extract of the *musa sapientum* flowers and fruit peels may contain some biologically active ingredients that are useful for the treatment of diarrhoea.

Key words: *Musa sapientum*, Antidiarrhoeal, castor oil, antimotility, phytochemical

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INTRODUCTION

Diarrhoea is characterized by increased frequency of bowel movement, wet stool and abdominal pain. Diarrhea is an important health problem worldwide, especially in developing countries¹. It accounts more than 5-8 millions of death each year in infants and children under five years^{2, 3}. Despite massive technological advancement in modern medicine, many people in the developing countries still depend on the healing practices of use of medicinal plants for their daily health care needs. To fight this problem, the world health organization (WHO) has started a diarrhoea disease control program to study traditional medicine practices and other related aspects, together with the evaluation of health education and prevention approaches. Many public and private research institutions trying to control this disease, but the mortality rate of diarrhoea is still high in developing countries⁴⁻⁶.

Musa sapientum (banana) belongs to the Musaceae family. It is a large, perennial, monocotyledonous herb 2–9 m in height that arises from large, subterranean rhizomes, from which the leaves emerge. The entire above-ground portion of the plant is not a true woody trunk, as in other trees, but a “false trunk” or “false stem” that consists of leaves and their fused petiole bases, referred to as a pseudo-stem. The pseudo-stem supports a canopy consisting of 6–20 (or more) leaves. In the center of the leaves, a growing point forms from the top of the rhizome, grows up and emerges as an overhanging inflorescence with a succession of reddish brown bracts. The bracts unfold from the base to the tip and fall off. Within the lower 1-12 bracts arise 14-18 female flowers in double rows which develop into parthenocarpic fruits. The next few bracts contains bisexual flowers that are rich in nectar but do not develop any further. In the upper bracts only male flowers are formed⁷⁻¹⁰. About 300 varieties of banana have spread throughout the tropical and subtropical countries and are widely used for its nutritional values all over the world. The fruits as well as the other parts of the plant are used to treat different diseases in human in traditional medicine. The fruit of *Musa sapientum* is traditionally used in diarrhoea (unripe), dysentery, intestinal lesions in ulcerative colitis, diabetes (unripe), uremia, nephritis, gout, hypertension, cardiac disease. *Musa sapientum* is also used in the treatment of excess menstruation. Banana leaves (ashes) are used in eczema, as cool dressings for blister and burns. Flowers are used in dysentery and menorrhagia. Stem juice of fruited plant is used for treating diarrhoea, dysentery, cholera, otalgia, haemoptysis and flower is used in dysentery, diabetes and menorrhagia. The root is used as anthelmintic, blood disorders, venereal diseases. The plant is also used in inflammation, pain and snakebite¹¹⁻¹⁷.

All these above mentioned traditional uses indicate that there must be some antidiarrhoeal property lying with the plant. In the present investigation the methanol extracts of *Musa sapientum* flowers and fruit peels were subjected for antidiarrhoeal activities.

MATERIALS AND METHOD

Plant material

The flowers and fruit peels of *Musa sapientum* were collected from local areas of Berhampur, Odisha, India, in the month of august 2017 and were identified by Dr. S. K. Dash, former professor, Department of Biosciences, College of Pharmaceutical Sciences, Mohuda, Dist-Ganjam, Odisha. The plant materials were cleaned with deionized water and were air-dried under shade, coarsely powdered, and kept in airtight container.

Animals

Wistar albino rats weighing 150-200 g of either sex were used for this study. They were brought from animal house of Royal College of Pharmacy and Health Sciences (RCPHS), Berhampur, Odisha. Animals were maintained under controlled room temperature ($24 \pm 2^\circ\text{C}$) and a 12:12 hour light: dark cycle. The animals were fed with standard laboratory food diet made in-house recommended by National institute of nutrition, Hyderabad and pure drinking water ad libitum. The animals were acclimatized to laboratory hygienic conditions in the departmental laboratory for 7 days before commencing the experiment. The ethical clearance was granted for the study by Institutional animal ethics committee of RCPHS, Berhampur, Odisha (bearing registration number 1018/C/06/CPSEA and date of registration 19th Dec 2006) prior to the beginning of the experimental works.

Preparation of extract

Methanol extract of *Musa sapientum* flowers (MEMF) and Methanol extract of *Musa sapientum* fruit peels (MEMFP) were prepared by soxhlet apparatus by successive extraction with petroleum ether (60–80°C), chloroform and methanol. Petroleum ether and chloroform were used in initial steps of extraction for defatting the plant materials. The methanol extracts were collected and dried using rotary vacuum evaporator followed by lyophilization and stored in desiccator until further use^{18, 19}. The type and extractive yield of different extracts of *Musa sapientum* were observed and results of such observation are tabulated in table no 1.

Table 1 Types and percentage yield of MEMF and MEMFP

Sl. no.	Extracts	Colour of the extracts	Physical appearance of the extracts	%Yield (w/w)
1	MEMF	Light brown	Dried powder	17.08
2	MEMFP	Brown	Dried powder	15.64

Phytochemical screening

Qualitative analysis of MEMF and MEMFP were carried out for presence of various phytoconstituents based on standard protocols²⁰⁻²³ and results of such observation are tabulated in table no 2.

Table 2 Preliminary phytochemical investigation MEMF and MEMFP

phytoconstituents	MEMF	MEMFP
Alkaloids	+	+
Carbohydrates	+	+
Glycosides	+	+
Cardiac glycosides	-	-
Saponin glycosides	+	+
Proteins and Amino acids	+	+
Tannins and phenolic compounds	+	+
Triterpenoids	+	+
Flavonoids	+	+
Coumarins	+	-
Steroids	-	-
Fats and oils	-	-

Antidiarrheal Study

Castor oil-induced diarrhea

After acclimation in the departmental laboratory for 7 days, the castor oil induced diarrhoea test was performed in wistar albino rats. Thirty six number of rats were randomly divided into six groups of six rats each (n=6) and administered different drugs as per the following schedule. Group 1 was received only distilled water 5 ml/kg and served as control group animals; group 2, treated with 3 mg/kg of loperamide and served as standard group; group 3 and group 4 animals were treated with 200 and 400 mg/kg of MEMF respectively; group 5 and group 6 were treated with 200 and 400 mg/kg of MEMFP respectively. The rats were fasted for 18h (free access to water) and were housed separately in metabolic cages. The animals administered the above drugs to respective groups. Thirty minutes after drug treatment the animals were administered with 1 ml of castor oil orally. The numbers of both wet and dry diarrhoeal droppings were counted

every hour for a period of 4 hours. At the end of 4th hour, the cumulative wet and dry weight of stool mass was weighed. Percent reduction in stool mass was calculated²⁴⁻²⁶.

Intestinal motility test

Intestinal motility test was performed in wistar albino rats. Thirty six number of rats were randomly divided into six groups of six rats each (n=6) and administered different drugs as per the following schedule. Group 1 was received only distilled water 5 ml/kg and served as control group animals; group 2, treated with 5 mg/kg of atropine sulfate and served as standard group; group 3 and group 4 animals were treated with 200 and 400 mg/kg of MEMF respectively; group 5 and group 6 were treated with 200 and 400 mg/kg of MEMFP respectively. The rats were fasted for 18h (free access to water) and administered the above drugs to respective groups. Thirty minutes after drug treatment, 2ml of charcoal meal (consisting of activated charcoal 12 g, tragacanth 2 g, water 130 ml) was administered orally to all the rats²⁶. Thirty minutes later, the rats were sacrificed and the abdomen was opened. The small intestine was dissected out from the pyloric sphincter to the ileocecal junction and the distance covered by the charcoal meal in the small intestine was measured. Calculate the percentage of the distance traveled and statistically compare between the different groups²⁶⁻²⁸.

Statistical Analysis

The values are expressed as mean \pm Standard Error Mean (SEM). The results were analyzed for statistical significance using one-way ANOVA (and nonparametric), followed by Bonferroni's Multiple Comparison Test (Graph pad prism 5.04 version). $P < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Phytochemical screening of the extracts

The percentage yield (w/w) of MEMF was 17.08% and MEMFP was 15.64% (table 1). Preliminary phytochemical analysis of MEMF and MEMFP revealed the presence of alkaloids, carbohydrates, glycosides, saponins, proteins, coumarins, flavonoids, triterpenoids, tannins and phenolic compounds (table 2).

Notably the tannins, flavonoids and alkaloids have been reported to possess anti diarrhoeal activity. Tannins may precipitate proteins of enterocytes, reduce peristaltic movement and intestinal secretion. Flavonoids possesses anti diarrhoeal activity as it inhibits intestinal motility and hydroelectrolytic secretions. Flavonoids also have antioxidant properties for which it inhibits several enzymes including those involved in the arachidonic acid metabolism²⁹⁻³¹.

Antidiarrhoeal Study

Castor oil-induced diarrhea

As expected, 30 min after administered the castor oil, diarrhea was found in all the animals of the control group. The treatment of loperamide had significantly ($P < 0.05$) decreased the total number of diarrhoeal faeces and cumulative fecal mass in the standard group of rats. Loperamide (3 mg/kg) treated animals had shown 73.94% inhibition of diarrhoeal faeces and 86.44% inhibition of total weight of faeces as compared to control group animals. Treatment with both the extracts of MEMF and MEMFP of 200 and 400 mg/kg showed significant antidiarrhoeal effects in castor oil induced rats. Treatment with MEMF of 200 and 400 mg/kg caused 44.17% and 62.78% inhibition of diarrhoeal faeces; 45.79% and 73.36% inhibition of total weight of faeces respectively. The MEMFP of 200 and 400 mg/kg caused inhibition of diarrhoeal faeces 39.49% and 58.46%; inhibition of total weight of faeces 40.18% and 61.21% respectively (Table 3).

Table 3 Effect of MEMF and MEMFP in castor oil induced diarrhoea

Groups	Treated Drugs and Doses	Total number of Diarrhoeal faeces	% inhibition of Diarrhoeal faeces	Cumulative fecal mass (gm)	% inhibition total weight of faeces
Group 1 Control	Distilled water- 0.5 ml/ 100 g	8.33±1.15	-	2.14±0.39	-
Group 2 Standard	Loperamide- 3 mg/kg	2.17±0.90*	73.94	0.29±0.15*	86.44
Group 3 Test-I	MEMF- 200 mg/kg	4.65±2.05*	44.17	1.16±0.67*	45.79
Group 4 Test-II	MEMF- 400 mg/kg	3.1±1.62*#	62.78	0.57±0.53*#	73.36
Group 5 Test-III	MEMFP- 200 mg/kg	5.04±2.12*	39.49	1.28±0.44*	40.18
Group 6 Test-IV	MEMFP- 400 mg/kg	3.46±1.43*#	58.46	0.83±0.58*#	61.21

The results were expressed as mean \pm SEM, $n=6$.

* $P < 0.05$; compared standard and test groups vs control group.

'#' indicates there is no significant difference between standard and test drug at $P < 0.05$ significant level.

The antidiarrhoeal effects of 400 mg/kg of both the extract were comparable with reference standard group treated with loperamide. The MEMF showed more significant antidiarrhoeal effect than the MEMFP in castor oil administered rats.

Intestinal motility test

In intestinal motility test all the rats of different groups were sacrificed, thirty minutes after administering 2ml of charcoal meal and distance covered by the charcoal meal in the small intestine was measured. In the control group animals 75.7% transit of charcoal meal was found. Atropine pretreated group produced a marked decrease in the propulsive movement and the intestinal length travelled by charcoal meal was 40.32%. The percentage transit of charcoal meal in the animals pretreated with MEMF 200 and 400 mg/kg were 58.93% and 47.52% respectively. The percentage transit of charcoal meal in the animals pretreated with MEMFP 200 and 400 mg/kg were 61.34% and 51.64 % respectively (Table 4). The MEMF and MEMFP treated animals had significantly ($P < 0.05$) decreased the intestinal transit of charcoal meal. The anti-motility effects of 400 mg/kg of both the extract were comparable with standard drug atropine. The MEMF showed more anti-motility effect than the MEMFP in intestinal motility test.

Table 4 Effect of MEMF and MEMFP in intestinal motility test

Groups	Treated Drugs and Doses	% Transit of charcoal meal	% Inhibition
Group 1 Control	Distilled water- 0.5 ml/ 100 g	75.7 ± 6.41	-
Group 2 Standard	Atropine- 3 mg/kg	40.32 ± 4.78*	46.73
Group 3 Test-I	MEMF- 200 mg/kg	58.93 ± 7.08*	22.15
Group 4 Test-II	MEMF- 400 mg/kg	47.52 ± 4.16*#	37.22
Group 5 Test-III	MEMFP- 200 mg/kg	61.34 ± 5.18*	18.96
Group 6 Test-IV	MEMFP- 400 mg/kg	51.64 ± 4.25*#	31.78

The results were expressed as mean ± SEM, n=6.

* $P < 0.05$; compared standard and test groups vs control group.

indicates there is no significant difference between standard and test drug at $P < 0.05$ significant level.

CONCLUSION

The results of the present work provide evidence that methanol extract of *Musa sapientum* flowers and fruit peels have antidiarrhoeal and antimotility activity in a dose dependent manner. The phytochemical study showed that the extract contains tannins, flavonoids and alkaloids may be responsible for management of diarrhoea. This study also scientifically justifies the traditional claim of usefulness of this plant against diarrhoea. Further investigation is necessary for isolation, identification and characterization of different active compounds from the extract and for elucidating their mode of action, responsible for these properties on different biological systems.

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