**Reviewer’s Comments**

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***IN VIVO*** ANTI-DIARRHOEAL **ACTIVITY OF METHANOL EXTRACT OF BOMBAX BUONOPOZENSE**

**ABSTRACT**

Diarrhoeal disease is responsible for mortality and morbidity, especially among children in developing countries causing a major healthcare problem.

The purpose of the present study was to evaluate scientifically the anti-diarrhoeal effects of methanolic extract of whole plant of *Bombax buonopozense* was studied against castor oil-induced-diarrhoea model and small intestine transit model in rats.

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 extract 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 results obtained establish the efficacy and substantiate the folklore claim as an anti- diarrheal agent.

**Keywords:**

Antidiarrhoeal activity, *Bombax buonopozense,* Castor oil induced diarrhoea, Small intestinal transit, charcoal.

**INTRODUCTION**

Diarrhoea can be defined as an alteration in the normal bowel movement, characterized by at least three loose or liquid bowel movements each day and adult daily stools exceeds 300 g and contains 60 – 95 % water. It can result in dehydration due to fluid loss and can lead to death1. The common reason for causing diarrhoea is gastrointestinal infection by various types of bacteria, virus, and parasites2.

According to WHO, after pneumonia it is second most common cause of infant mortality in developing countries, and it is responsible for the death of about 2 - 4 million children every year, especially in Africa and it is still a big public health challenge in developing countries3.

Due to unhygienic livelihood condition specially in rural areas, peoples are very prone to diarrhea and several other diseases. This infection can be spread out through food, drinking water, and unhygienic environment. Furthermore rural populations live very far away from health centers, so there is lack of availability of proper medical facilities4.

There are many antibiotics available that are used as anti-diarrhoeal drug, but these drugs sometimes show some adverse effects and microorganisms are tends to develop resistance towards them5.

In these conditions, medicinal plants appear as an alternative and available health care solution in the management of diarrhoea. The roles of plants in maintaining human health are well documented. Many plant species have been screened for substances with therapeutic activity. For the treatment of diarrhoea, medicinal plants are a potential source of antidiarrhoeal drugs

*Bombax buonopozense* P. Beauv.(Bombacacea*)* is a tropical tree that grows up to 40 meters in height with large buttress roots that can spread six meters. It is native primarily in West Africa where it is found in rainforests of Sierra Leone in the northwest, east Gabon6.

It has common vernacular names in different languages such as Vabga (Dagbani) and Kurya (Hausa) and different parts are used for different purposes.

The bark of younger trees is covered with spine and shedding the spines with age to some degree and large deep pink-to-red flowers emerge while the tree is leafless. The leaves are compound and have 5 to 9 leaflets and 15 to 25 secondary veins. The individual leaﬂets have entire margins and are large. The undersides of the leaﬂet may be glabrous or puberclous. The conspicuous flowers emerge while the tree is leafless and are either solitary or arranged in small axillary cymes7.

The petals are deep pink or red in colour and are 5.5 to 9.5cm in length by 2.7 to 3.7 cm in width. The fruits are oblong and fairly large being 8 to 18cm in length by 3.5 to 6cm in diameter. More so, they contain many seeds that are 5 to 6mm in length, all of which have a woolly indument, which is cotton-like fiber covering. The buds are conical.

 According to many parts of this plant is utilized for medicinal and traditional purposes. However, there is limited scientific evidence supporting the potential use of *B.buonopozense* as an antidiarrhoeal agent. Present study therefore investigated the scientifc basis for the efficacy of its antidiarrheal properties8.

**MATERIALS AND METHODS-**

 **Plant collection**

**Animals used**

Albino wistar rats (150-230g) of either sex were selected for the study. The animals were maintained in a well-ventilated room with 12:12 hour light/dark cycle in polypropylene cages.

**Determination of antidiarrheal activity**

**Castor oil-induced diarrhoea**

**Table 1: Effect of methanolic extract of *Bombax buonopozense* on castor oil induced diarrhea in mice**

|  |  |  |  |
| --- | --- | --- | --- |
| **Group**  | **Treatment**  | **Number of wet feaces in 4h** | **% Inhibition** |
|  |  |  |  |
| **I**  | Castor oil + Saline(2 mL /kg p.o) | 13.3 ± 0.43  | -- |
| **II**  | Castor oil + Loperamide(5 mg/kg i.p) | 9.20 ± 0.37 | 66.43 |
| **III**  | Castor oil + Extract(200 mg/kg i.p) | 13.00 ± 0.89 | 52.62 |
| **IV**  | Castor oil + Extract(400 mg/kg i.p) | 10.20± 0.58  | 56.31 |

Animals were fasted for 24 h but allowed free access to water. One hour after administration, all animals were given 0.5 ml of castor oil orally and individually placed in cages in which the floor was lined with transparent paper and changed every hour.

Rats were divided into four groups of six animals each, diarrhoea was induced by administering 2 ml of castor oil orally to rats. Group I treated as control (2 ml/kg, p.o. saline), group II received Loperamide (5 mg/kg p.o) served as standard and group III and IV received extract (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 hrs10.

Where, WFC = average number of wet feces in control group and WFT = average number of wet feces in test group

**Figure 1- Total number of diarrhoeal faeces in castor oil-induced diarrhoea in albino rats**

**Small intestinal transit**

**Table 2:** Effect of methanolic extract of *Bombax buonopozense* on small intestinal transit in mice

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Group**  | **Treatment**  | **Total length of intestine(cm)** | **Distance traveled bymarker (cm)** | **% Inhibition** |
| **I**  | Castor oil + Saline(2 mL/kg p.o) | 56.09 ± 0.85  | 39.83 ± 0.34  | **--** |
| **II**  | Castor oil + Loperamide(5 mg/kg i.p) | 54.16 ± 0.93 | 15.21 ± 0.63  | 74.27 |
| **III**  | Castor oil + Extract (200mg/kg i.p) | 48.20 ± 0.66 | 26.72 ± 0.63  | 42.58 |
| **IV**  | Castor oil + Extract (400mg/kg i.p) | 51.06 ± 0.83  | 23.81 ± 0.84  | 53.68 |

Albino rats of either sex (200-250g) were randomly divided into five groups of six rats each. Rats were fasted for 18 hr divided into four groups of six animals each, Group I received 2ml normal saline orally, group II received atropine (5 mg/kg, i.p.), group III and IV received extract
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. The rats were sacrificed after 1h and the distance traveled by charcoal meal from the pylorus to caecum was measured and expressed as mean distance traveled by charcoal meal caecum11**.**

**Figure 2- Percentage of distance travelled by charcoal meal (cm) in albino rats.**

**RESULTS**

**Castor oil-induced diarrhoea**

As shown in table 1, it was observed that administration of methanol extract of *Bombax buonopozense*, induced insignificant decrease in the total number of feces and number of diarrhoeal feces and percent inhibition of 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 *Bombax buonopozense* at the doses of 200 and 400 mg/kg p.o.

**Small intestinal transit**

The percent intestinal transit was increased with control, but it was reduced in both doses of extract, and much more significantly reduced by Loperamide (5 mg/kg i.p).

Methanol extract of *Bombax buonopozense* 200 mg/kg, p.o dose of extract produced 42.58 % intestinal transit induced by castor oil respectively, whereas, 400 mg/kg, p.o dose produced 53.68 % of castor oil induced charcoal meal transit (Table 2 and Figure 2).

 **Statistical analysis**

All analyses were carried out in triplicates. Data were presented as mean ± SEM. The significance of difference between the control and treated groups was determined using two way analysis of variance (ANOVA), followed by Student's *t*-test. *P* value of 0.05 or 0.01 was considered as significant.

**DISSCUSSION**Diarrhoea is due to imbalance between the absorptive and secretory mechanisms in the intestinal tract, associated with excess loss of fluid in the faeces. Castor oil produces diarrhoeal effect due to its active component of ricinoleic 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 oil12. Since the alcoholic extract successfully inhibits the castor oil induced diarrhoea, the action might be via anti-secretary mechanism.

Loperamide at present is one of the most efficacious and widely employed anti diarrheal agents and effectively antagonizes the action of castor oil due to its antimotility and antisecretary property13.

At doses of 200 and 400 mg/kg, the methanol extracts of *Bombax buonopozense* showed significant antidiarrhoeal 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. It also showed a dose related decrease in castor oil-induced diarrhoea.

In addition, the administration of the aqueous extract of *Bombax buonopozense* in rats caused a significant reduction in the progression of charcoal meal and in the intestinal transit time.

**CONCLUSION**

Plants play a vital role in the maintenance of human health**.** Results concludes that the leaves extract of *Bombax buonopozense* and its fractions contains bioactive natural substances with antidiarrheal properties thus justifying its widespread use by the local
population for these purposes..

This antidiarrheal activity probably results from the spasmolytic or may be due to a possible antisecretory effect of the plant extract on the intestinal smooth muscle. The isolated compound may serve as useful prototypes of antidiarrhoeal drugs of natural origin possessing the desired pharmacological activities while lacking certain untoward effects.

**CONFLICT OF INTEREST:**

The author has declared that there is no conflict of interest related to this paper.

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