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AN EVALUATION ON PHARMACOLOGICAL ACTIVITY OF *HEVEA BRASILIENSIS*

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ABSTRACT
The biological investigation is carried out on *Hevea brasiliensis*, a plant belonging to the family Euphorbiaceae. The roots of *Hevea brasiliensis* was collected from the planted forest of Chittagong region of Bangladesh. It was extracted with methanol. The extract was used for the observation of analgesic and antidiarrheal activity. Extract of *Hevea brasiliensis* has showed significant (P value<0.001) analgesic activity in mice producing an inhibition of 21.62% and 36.53% from extract(250 mg/kg) and extract (500 mg/kg) respectively, where Diclofenac sodium (Positive control) had an inhibition of 45.02%.In antidiarrheal test, percentage of distance travelled by charcoal for control, loperamide, extract (250mg/kg) and extract(500mg/kg) were 89.32%, 40.19%, 62.61% and 56.98% respectively, that indicate reduction in gastrointestinal motility.

KEYWORDS Analgesic, Antidiarrhea, gastrointestinal motility, Positive control, biological investigation.

1. INTRODUCTION

Heveabrasiliensis, better known as the rubber tree, is the primary source of natural rubber. It is widely used for the production of rubber. It is cultivated in the tropics and subtropics of Southeast Asia and Western Africa.[1] Rubber tree is a quick-growing tree, rarely exceeding 25 m in height in plantations, but wild trees of over 40 m have been recorded. As a latex producing crop, the bark is regularly tapped.[2] Latex, the source of hevea or para rubber, is obtained by tapping the trunks of the trees. [3] It is cultivated for rubber, food, apiculture, fibre, timber, lipid,fuel etc.It also cultivated for soap, insect repellent in Brazil.[4] Proteins present in natural rubber latex may cause allergic reactions. [5] In this study, analgesic and antidiarrheal properties of the roots of *Hevea brasiliensis* are explained. Analgesic activity was performed by the model of acetic acid induced writhing in mice and antidiarrheal actinity test was done according to the methods of Qnais *et al.*,and Meite *et al.*, with slight modifications.

2. MATERIALS AND METHODS

2.1. Plant collection and identification

The roots of *Hevea brasiliensis* was collected from Fatikchari, Chittagong. It was taxonomically identified by the experts of Bangladesh National Herbarium, Mirpur, Dhaka, Bangladesh and also deposited there for further study.

2.2. Preparation of *Hevea brasiliensis* extract

After collection, the roots were cut into pieces, dried under sun for three weeks and finally ground to coarse powder by a suitable grinder. By cold extraction method, dried and powdered roots (500 g) was soaked in distilled methanol at room temperature for two weeks. The filtrate was concentrated by evaporation. Dried extract was stored at 4°C in air tight container and was diluted with methanol prior to any pharmacological screening.

2.3. Chemicals and Drugs

DICLOFENAC SODIUM and LOPERAMIDE were obtained from Square Pharmaceuticals Ltd, Bangladesh.

2.4. Experimental Animals

Young Swiss-albino mice aged 4-5 weeks, average weight 20-25 gm were used for the experiment. The mice were purchased from Jahangirnagar University, Dhaka. They were kept at standard environmental condition for adaptation before tests for one week. They were supplied with the standard rodent pellet diet, water *ad libitum* and fasted 18 hours prior to their use. Experiments were carried out according to animal ethics guidelines.[6] Animals were marked for proper identification.

2.5. Acetic Acid Induced Writhing Test

Analgesic activity of the methanol roots extract of *Hevea brasiliensis* was tested using the method of Koster et al., 1959.[7] Experimental animals were randomly selected and splitted into four groups, i.e. control, positive control and the two doses of the extract, consisting of 6 mice in each group. Each group received particular treatment. Test groups received the methanol roots extract at the doses of 250 and 500 mg/kg in oral route. Standard analgesic drug DICLOFENAC SODIUM (25 mg/kg, p.o.) was administered orally to the positive control group. Control group orally received 1% tween-80 in saline at the dose of 10 ml/kg. After 40 min, each animal was given an intraperitoneal (i.p.) injection of 0.1% acetic acid at the dose of 10 ml/kg to induce the characteristic writhing. 5 minutes after the administration of acetic acid, number of writhing were counted for each mouse for ten minutes. The percent writhing inhibition was calculated and compared with control to assess analgesic activity.

% writhing Inhibition = Mean No. of Writhes in control- Mean No. of Writhes in test / Mean No. of Writhes in control x 100 .[8]

2.6. Intestinal motility test

Albino Swiss mice were preserved in standard conditions for 10 days before performing the experiment. They had free access to water and a normal commercial laboratory diet. On test day, the animals were divided into four groups of 5 mice each. They were weighed and deprived of food, with free access to water. Three hours after food deprivation, the animals in group A received 10ml/kg of normal saline, while those in group B received orally by gavage 5mg/kg of LOPERAMIDE as positive control. The methanolic roots extracts of *Hevea brasiliensis* (250 and 500 mg/kg, p.o.) were provided to the test groups. After 90min, 0.3 ml of an aqueous suspension of 5% charcoal in 10% water was administered to each animal orally by gavage. The animals are killed 45 min later to open the abdomen and remove the small intestine (from the pylorus to the caecum) to determine the length of intestine and distance travelled by charcoal meal as a fraction of the length of intestine.[9]

3. RESULTS AND DISCUSSION

3.1.1. Analgesic activity

The methanol roots extract of *Hevea brasiliensis* exhibited dose dependent inhibition of writhing. The doses of 250 and 500 mg/kg extract showed 21.62% and 36.53% inhibition of writhing significantly **(P < 0.001)**. Positive control DICLOFENAC SODIUM showed strong analgesic activity with 45.02% inhibition of writhing, compared to control. Activity of the extract was strongly comparable with DICLOFENAC SODIUM.

Table 1: **Effect of methanolic extract of roots of *Hevea brasiliensis* on acetic acid induced writhing mice.**

Sr. No.	Animal group	Treatment	Writhing count (Mean±SEM) (%writhing)	% Writhing Inhibition
1.	I (Control) n=5	1%tween-80 solution in water orally	12.95± 0.68 (100)	---
2.	II (Positive control) n=5	Diclofenac sodium (25mg/kg) orally	7.12 ± 0.57 a (54.98)	**45.02**
3.	III (Test group) n=5	Methanol extract (250mg/kg) orally	10.15 ± 0.93 b (78.38)	**21.62**
4.	IV (Test group) n=5	Methanol extract (500mg/kg) orally	8.22 ± 0.58 a (63.47)	**36.53**

n=number of mice, S.E.M = standard error of mean, aP<0.001; bP<0.01, Values are expressed as Mean±S.E.M

3.1.2.Intestinal motility test

The methanol roots extract of ***Hevea brasiliensis*** significantly (P < 0.001) and dose dependently decreased the intestinal transit charcoal meal as compared with control. Percentage of distance travelled by charcoal for control, LOPERAMIDE, extract(250mg/kg) and extract(500mg/kg) were 89.32%,40.19%,62.61% and 56.98% respectively. The results indicated a reduction in peristaltic activity and ultimately reduction in gastrointestinal motility.

Table 2: Effects of *Hevea brasiliensis* on charcoal meal-stimulated gastrointestinal transit in mice.

Sr. No.	Group	n	Mean intestinal length (cm) Mean ± SEM	Mean distance travelled by charcoal (cm) Mean ± SEM	Gastrointestinal Transit (%) (% of inhibition)
1.	Control	5	51.13 ± 1.34	45.67 ± 1.98	89.32
2.	Loperamide	5	56.67 ± 1.27	22.78 ± 1.86	40.19
3.	Extract(250mg/kg)	5	57.50 ± 1.50	36.00 ± 0.50	62.61
4.	Extract(500mg/kg)	5	59.75 ± 0.55	34.05 ± 0.45	56.98

3.2. DISCUSSION

Analgesic activity of the extract was evaluated by acetic acid-induced writhing method. This method was used to assess peripherally acting analgesic activity of the plant extract in which writhing results from the sensitization of pain receptors by prostaglandins release. The released prostaglandins, mainly prostacyclin (PGI_2) and prostaglandin-E have been reported to be responsible for pain sensation by exciting the Aδ-fibres. Activity in the Aδ-fibres cause a sensation of sharp well localized pain.[10, 11] Since the methanol roots extract of ***Hevea brasiliensis*** significantly inhibited the acetic acid-induced writhing in mice it suggests that the analgesic effect of the extract may be peripherally mediated.

Previous study shows that activated charcoal avidly absorbs drugs and chemicals on the surface of the charcoal particles thereby preventing absorption.[12] Thus, gastro-intestinal motility test with activated charcoal was carried out to find out the effect of methanol roots extract of ***Hevea brasiliensis*** on peristaltic movement. The results showed that, intestinal motility was decreased with increasing the dose of the plant extract. The methanol roots extract of ***Hevea brasiliensis*** significantly delayed gastrointestinal transit of charcoal meal, compared to control. So, the extract might have ability in greater extent to decrease gastrointestinal motility. The inhibition of peristaltic movement with methanol roots extract of ***Hevea brasiliensis*** may be due to the anti-histaminic and anticholinergic actions. From these models we can suggest that methanol roots extract of ***Hevea brasiliensis*** nonspecifically inhibit diarrhea by decreasing intestinal motility. LOPERAMIDE was used as positive control. The effects of this positive control on gastrointestinal motility were investigated.

CONCLUSION

It can be concluded that, methanol roots extract of ***Hevea brasiliensis*** comprise of analgesic and anti-diarrheal effects. Though, it requires further investigations to use in folk medicine.

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