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# ETHNO MEDICAL VALUE OF *HOUTTUYNIA CORDATA* THUNB METHANOL EXTRACT IN EXPERIMENTALLY INDUCED DIARRHOEA

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**ABSTRACT:** *Houttuynia cordata* Thunb. (*H. cordata*) is used as a remedy for diarrhoea and dysentery in Asia, but has not been investigated for its antidiarrhoeal properties. Antidiarrhoeal activity of 80% "methanol extract of *H. cordata*" (HCM) was investigated in this study using castor oil induced diarrhoea, castor oil induced intraluminal fluid accumulation as well as charcoal transit in wister albino rat. The extract significantly increased the latent period of diarrhoea in all the models. Preliminary phytochemical screening revealed the presence of flavonoids, saponins, and tannins. It is suggested that the extract may contain biologically active components that may be useful against diarrhoea, thereby justifying its use in ethnomedical practice as an antidiarrhoeal agent. The HCM produced a significant ($P < 0.05$) dose dependent decrease castor oil induced diarrhoeal faeces with values of 54.35% and 65.22% respectively at doses of 200 and 400 mg/kg vs. 93.48 for reference dose loperamide. The extract also significantly decreased the gastrointestinal motility 33.22% and 51.53% as well as fluid accumulation 26.53% and 43.54 at both doses 200 and 400mg/kg respectively. The results suggest that *H. cordata* showed antidiarrhoeal activity and justify its use in traditional medicine.

**INTRODUCTION:** *Houttuynia cordata* Thunb. (*H. cordata*) Family Saururuacea, locally known as "Moshundari" is a traditional Indian medicinal plant and edible vegetable in Assam, India. Similar reports have been made from regions of the Eastern-Asia viz. Chinese, Japan & Himalaya and Vietnam of using the tender young shoots and leaves of *H. cordata*, either raw or cooked, as vegetable and leaves for flavoring salads or as a salad crop Japanese use *H. cordata* leaves, as a beverage under the name "dokudami cha"; literally known as "*H. cordatta* tea"[1].

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It composes of major active ingredients such as flavonoids, volatile oils and alkaloids. The major identified flavonoids in *H.cordata* are rutin, quercetin, quercitrin, isoquercitrin and hyperin. Flavonoids provide a wide range of pharmacological activities including antiviral, anti-microbial, antioxidant, anti-inflammatory, antileukemic, anticancer, and immunomodulatory effects[2, 3]. In order to substantiate the claims made by local people, the current study was undertaken to evaluate the methanol extract of *H. cordata* (HCM) on different experimental models of antidirrhoeal activity.

W.H.O. estimates for 1998, about 7.1 million deaths were caused by diarrhoea. The incidence of diarrhoeal diseases still remains high despite the

efforts of many governments and international organisations to curb it. More than 5-8 Million infants and children below 5 years old die every year of Diarrhoea especially in developing countries. A nationwide study conducted has estimated that Diarrhoea kills more than 1 million children per year in India alone [4-7]. There is therefore an urgent need for the intensification of research into medicinal plants claimed to be effective in the management of diarrhoea.

## MATERIALS AND METHODS

**Collection and authentication of plant material:** The entire plants of *H. cordata* were collected from District Dibrugarh Assam (India) in the month of Feb 2010 and were authenticated by Dr Tariq Hussain Scientist, Taxonomy division at National Botanical Research Institute (NBRI), Lucknow, Uttar Pradesh (India). A voucher specimen (NBRI/CIF/145/2010) has been deposited in the institute for further reference.

**Preparation of the extract:** Entire plants of *H. cordata* were washed with distilled water to remove dirt and soil and shade dried in a ventilated place at room temperature. The dried plant materials were reduced to coarse powder by mechanical grinder, extracted with 80% methanol as solvent in soxhlet extractor for 18 h. The extract was filtered and concentrated under reduce pressure using rotavapor (Buchi, USA), then freeze-dried (Freezone® 4.5, Labconco, USA) and stored in deep freezer for further use. Solutions of the extracts were prepared freshly for each study.

**Preliminary phytochemical screening:** The methanol extract obtained was tested for the presence of various chemical constituents such as saponins, flavonoids, glycosides, alkaloids, tannins and reducing sugar by Trease and Evans [8, 9].

**Animals:** Wister albino rats of either sex weighing 150-200 g were kept at departmental animal house at a temperature $(25 \pm 2)^0$C and 12 h light/dark cycle respectively for one week before and during the experiments and fed with standard diet and water ad libitum. Animal studies were conducted according to the Institute Animal Ethics Committee.

All the experiments were performed in the morning according to the current guidelines for the care of laboratory animals and the ethical guidelines for the investigation of experimental pain in conscious animals

**Drugs and chemicals:** Atropine sulphate and Loperamide (Ranbaxy (I) Ltd, castor oil (Galaxo) all other chemicals were of analytical grade.

**Acute toxicity:** Acute toxicity study was performed according to OECD guidelines No. 423 (OECD, 2000). Swiss albino mice of either sex were used for acute toxicity of *H. cordata* methanol extract (HCM). It is nontoxic at 2000 mg/kg.

**Statistical analysis:** Results obtained were expressed as mean ±S.E.M. The data were analysed using the Student's t-test where appropriate. Results were considered significant when $P < 0.05$.

**Experiment Design:**

1. **Castor oil-induced diarrhoea:** This was determined according to the method of Amresh *et al* [10] modified by Adeyemi *et al* [11] were used to assess the antidiarrhoeal activity of the plant extract. Wister albino rats of either sex (150-200 g) were fasted for 24 h before starting the experiment. The animals were randomly housed in individual cages and divided into four groups (n = 5). The first group received 1% CMC (10 ml/kg p.o.) served as the control and group II was received loperamide (3mg/kg) acting as the standard. The last two groups received different doses (200 and 400mg/kg p.o.) of the plant extract. One hour after the treatment, each animal received castor oil (10ml/kg, p.o.) through a feeding needle. At 4th hour after dosing the castor oil, the individual mouse cages were inspected for the presence of unformed water fecal pellets; their absence was recorded as a positive result, indicating protection from diarrhoea at that time.

2. **Assessment of small intestine transit:** Animals were divided into four groups of five rats each and each animal was given orally 1 ml of charcoal meal (5% activated charcoal suspended in 1% CMC) 60 min after an oral dose of drugs or vehicle. Group I was

administered with 1% CMC (10 ml /kg), group II received atropine sulfate (0.1 mg/kg, i.p.) as standard drug and animals in groups III and IV received extracted drug (200 and 400 mg/ kg p.o.). After 30 min, animals were killed by cervical dislocation and the intestine was removed without stretching and placed lengthwise on moist filter paper. The length of the intestine (pyloric sphincter to caecum) and the distance travelled by the charcoal as a percentage of that length were evaluated for each animal, and group means were compared and expressed as percentage inhibition [10, 11].

**RESULT AND DISCUSSION:** The phytochemical analyses of the HCM (80% methanol extract) were revealed the presence of various chemical constituents such as flavonoids, tannins, alkaloids saponins particularly steroidal saponin (**Table 1**).

**TABLE 1: QULITATIVE ANALYSIS OF METHANOLIC EXTRACT OF *H. CORDATA***

| Test | HCM |
|---|---|
| **Alkaloids** | + |
| **Steroids/Terpenes** | ++ |
| **Flavonoids** | +++ |
| **Saponins** | + |
| **Tannins** | + |
| **Reducing sugars** | ++ |
| **Amino acid** | +++ |

The HCM showed dose-dependent inhibition of castor oil induced diarrhoea in Wister albino rats. This effect was significant at 400mg/kg dose of extract at 4 h in comparison to control group; however, this activity was less as compare to loperamide (Table 2). The castor oil induced intraluminal accumulation of fluid was inhibited by extract in a dose-dependent manner, shown in **Table 2**.

The results of present study revealed that the extract significantly inhibited the gastrointestinal transit of charcoal in rat. This activity was significant at 400mg/kg dose of extract as compared to control. Extract and loperamide significantly inhibited castor oil induced gastrointestinal transit of charcoal in rat. The activity of extract was dose dependent, and was significant at 200 and 400 mg/kg both doses (**Table 3**).

It is well know that the traditional uses of plants and their effects are due to the presence of secondary metabolites. These metabolites are may be alkaloids, glycosides, flavonoids, tannins, triterpenes etc. The medicinal value of plants are depend the presence of these metabolites qualitatively & quantitatively. So main aim of present study was provide scientific prove to the traditional claim.

**TABLE 2: EFFECT OF *H. CORDATA* METHANOL EXTRACT ON CASTOR OIL INDUCED DIARRHOEA IN RATS**

| Treatment | Dose (mg/kg) | Total no of faeces in 4 hr | Total no of wet faeces in 4 hr | Reduction (%) |
|---|---|---|---|---|
| **Control** (1%, 10ml/kg CMC) + **Castor oil** | -- | 10.4±0.50* | 9.2±0.37* | 0 |
| **Loperamide + Castor oil** | 3 | 1.6±0.24** | 0.6±0.24** | 93.48 |
| **HCM + Castor oil** | 200 | 5.8±0.20* | 4.2±0.20* | 54.35 |
| **HCM + Castor oil** | 400 | 5.6±0.24* | 3.2±0.37** | 65.22 |

Values are mean ± SEM for five rats; * = p < .0001 vs Control student's t-test; ** = p < .005 vs Control student's t-test

**TABLE 2: EFFECT OF H. CORDATA METHANOL EXTRACT ON SMALL INTESTINE TRANSIT**

| Treatment | Dose (mg/kg) | Total length of intestine | Distance traveled by charcoal | % Transit |
|---|---|---|---|---|
| **Control** (1%, 10ml/kg CMC) + **Charcoal** | -- | 83.82±0.61* | 71.5±0.54 | 84.53±4.32 |
| **Atropine sulphate + Charcoal** | 0.1 | 80.44±0.66* | 24.68±0.80* | 30.67±2.34 |
| **HCM + Charcoal** | 200 | 80.88±0.71* | 53.98±1.04* | 66.78±1.63 |
| **HCM + Charcoal** | 400 | 80.66±0.74* | 39.10±0.61* | 48.47±.56 |

Values are mean ± SEM for five rats; * = p < .0001 vs Control student's t-test

The study proven that the plant HCM has significant antidirrhoeal value. Number of factors, such as infective, immunological and nutritional has been involved in the perpetuation of the diarrhoeal syndrome [12]. Many plants conveniently available in India are used in traditional folklore medicine for the treatment of diarrhoea and dysentery.

Of the indigenous plants used, *Andrographis paniculata, Asparagus racemosus, Butea monosperma, Cassia auriculata*, and others are mentioned [13]. Several studies have shown that prior administration with some plant extracts had a protective effect on the intestinal tract [14-16]. In the present study, the newer plant have used by tribes and rural have not been studied so for, was evaluated for its anti-diarrhoeal potential against castor oil induced diarrhoea, in Wister albino rats.

80% Methanol is a strong polar solvent considered to extract most plant secondary constituents. Though several constituents were present in the extract, the compound responsible for the observed actions is unknown. Flavonoids possess a wide range of activities *in vitro* [17] including antidiarrhoeal activity [18-20] may have contributed to this activity, but further studies are required.

**CONCLUSION:** The 80% methanol extract of *H. cordata* showed antidiarrhoeal activity in primarily evaluation of diarrhoeic conditions in test animals. The obtained results thus give the experimental basis to understand the use of selected traditional medicine, as an antidiarrhoeal agent. However, further bioassay guided phytochemical and pharmacological studies are required to identify the active principle(s) and exact mechanism(s) of action.

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