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## Analgesic Activity Evaluation of Methanol Extract of *Luffa cylindrica* Fruits

Jakia Sultana, Md. Mainul Hasan Sarker, Ashif Rahman, Mirza Nipa Monalisa, Moytry Marzan, Md. Faisal, Mohammed Rahmatullah

Faculty of Life Sciences, University of Development Alternative, Dhanmondi, Dhaka-1209, Bangladesh

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### ABSTRACT

The analgesic effect of methanol extract of *Luffa cylindrica* fruits was evaluated in Swiss albino mice in intraperitoneally administered acetic acid induced pain model. At doses of 50, 100, 200, and 400 mg extract per kg body weight, the extract significantly and dose-dependently reduced the number of abdominal constrictions induced by acetic acid by, respectively, 30.0, 36.7, 40.0, and 46.7%. By comparison, a standard analgesic drug, aspirin, when administered at doses of 200 and 400 mg/kg reduced the number of abdominal constrictions, respectively, by 40.0 and 56.7%. Thus the extract at doses of 200 and 400 mg demonstrated comparable or higher activity than aspirin at the dose of 200 mg/kg. The results suggest that the extract has significant analgesic potential and could be used for further experiments to isolate and identify the responsible bioactive constituent(s).

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## INTRODUCTION

*Luffa cylindrica* (L.) M.J. Roem. (Cucurbitaceae) is a vinous plant grown in Bangladesh for its edible fruits, which are cooked and consumed as vegetable. In English, the plant is known as “ridged luffa” or “sponged gourd”, and in Bengali as “dhundol”.

A number of polyphenols have been reported from the fruit pulp of the plant. These polyphenols include p-coumaric acid, 1-O-feruloyl-b-D-glucose, 1-O-p-coumaroyl-b-D-glucose, 1-O-caffeoyl-b-D-glucose, 1-O-[4-hydroxybenzoyl]-glucose, diosmetin-7-O-b-D-glucuronide methyl ester, apigenin-7-O-b-D-glucuronide methyl ester, and luteolin-7-O-b-D-glucuronide methyl ester (Du *et al.*, 2006; Azeez *et al.*, 2013).

The Mullu kuruma tribe of Wayanad district, Kerala, India use leaf paste of the plant externally for treatment of hemorrhoids and leprosy (Silja *et al.*, 2008). The local and tribal people of North Gujarat, India use unripe fruits for treatment of anorexia, indigestion, calculi, fatigue, and hemorrhoids (Patel *et al.*, 2013). Leaves are used to treat snake bites by the Malayali tribals of Kolli Hills in Eastern Ghats, Tamil Nadu, India (Vaidyanathan *et al.*, 2013). The Tharu tribes of Dudhwa National Park, India use fruits for treatment of hemorrhoids (Kumar and Bharati, 2014).

We had been conducting extensive ethnomedicinal surveys among the folk and tribal medicinal practitioners of Bangladesh followed by evaluation of the medicinal plants obtained for their antidiabetic, analgesic and cytotoxic activities (Rahmatullah *et al.*, 2009a-c; Anwar *et al.*, 2010; Jahan *et al.*, 2010; Rahman *et al.*, 2010; Rahmatullah *et al.*, 2010a-h; Shoha *et al.*, 2010; Ali *et al.*, 2011; Barman *et al.*, 2011; Hossan *et al.*, 2011; Jahan *et al.*, 2011; Rahman *et al.*, 2011; Rahmatullah *et al.*, 2011a,b; Sutradhar *et al.*, 2011; Ahmed *et al.*, 2012; Arefin *et al.*, 2012; Haque *et al.*, 2012; Sathi *et al.*, 2012; Rahmatullah *et al.*, 2012a-d; Haque *et al.*, 2013). The objective of the present study was to evaluate the analgesic potential of methanol extract of *Luffa cylindrica* fruits.

## MATERIALS AND METHODS

Fruits of *Luffa cylindrica* were collected from Kolatia in Dhaka district, Bangladesh during September 2013. The fruits were taxonomically identified at the Bangladesh National Herbarium at Dhaka (Accession Number 39,207). The sliced and air-dried fruits were grounded into a fine powder and 150g of the powder was

**Corresponding Author:** Professor Dr. Mohammed Rahmatullah, Pro-Vice Chancellor and Dean, Faculty of Life Sciences, University of Development Alternative, House No. 78, Road No. 11A (new), Dhanmondi, Dhaka-1205, Bangladesh  
 Tele: +88-01715032621 Fax: +88-02-815739 E-mail: rahamatm@hotmail.com

extracted with 750 ml methanol for 48 hours. The extract was evaporated to dryness at 40°C. The final weight of the extract was 11.3g.

#### Chemicals:

Glacial acetic acid was obtained from Sigma Chemicals, USA; aspirin was obtained from Square Pharmaceuticals Ltd., Bangladesh.

#### Animals:

In the present study, Swiss albino mice (male), which weighed between 13-18g were used. The animals were obtained from International Centre for Diarrheal Disease Research, Bangladesh (ICDDR,B). All animals were kept under ambient temperature with 12h light followed by a 12h dark cycle. The animals were acclimatized for three days prior to actual experiments. The study was conducted following approval by the Institutional Animal Ethical Committee of University of Development Alternative, Dhaka, Bangladesh.

#### Analgesic activity:

Analgesic activity of the methanol extracts of fruits (MELC) was examined using previously described procedures (Shanmugasundaram and Venkataraman, 2005). Briefly, mice were divided into seven groups of six mice each. Group 1 served as control and was administered vehicle only. Groups 2 and 3 were orally administered the standard analgesic drug aspirin at a dose of 200 and 400 mg per kg body weight, respectively. Groups 4-7 were administered MELC at doses of 50, 100, 200 and 400 mg per kg body weight, respectively. Following a period of 60 minutes after oral administration of standard drug or extract, all mice were intraperitoneally injected with 1% acetic acid at a dose of 10 ml per kg body weight. A period of 15 minutes was given to each animal to ensure bio-availability of acetic acid, following which period, the number of abdominal constrictions (writhings) was counted for 10 min. The following formula was used for calculation of percent inhibition of the number of writhings in aspirin and MEAS administered animals compared to control mice, Percent inhibition =  $(1 - W_e/W_c) \times 100$  where  $W_e$  and  $W_c$  represents the number of writhings in aspirin or MELC administered mice (Groups 2-7), and control mice (Group 1), respectively.

#### Statistical analysis:

Experimental values are expressed as mean  $\pm$  SEM. Independent Sample t-test was carried out for statistical comparison. Statistical significance was considered to be indicated by a p value  $< 0.05$  in all cases.

## RESULTS AND DISCUSSION

At doses of 50, 100, 200, and 400 mg extract (MELC) per kg body weight, the extract significantly and dose-dependently reduced the number of abdominal constrictions induced by acetic acid by, respectively, 30.0, 36.7, 40.0, and 46.7%. By comparison, a standard analgesic drug, aspirin, when administered at doses of 200 and 400 mg/kg reduced the number of abdominal constrictions, respectively, by 40.0 and 56.7%. Thus the extract at doses of 200 and 400 mg demonstrated comparable or higher activity than aspirin at the dose of 200 mg/kg. The results are shown in Table 1.

**Table 1:** Analgesic effect of MELC in the acetic acid-induced gastric pain model mice.

Treatment	Dose (mg/kg body weight)	Mean number of writhings	% inhibition
Control (Group 1)	10 ml	6.0 $\pm$ 0.20	-
Aspirin (Group 2)	200 mg	3.6 $\pm$ 0.40	40.0*
Aspirin (Group 3)	400 mg	2.6 $\pm$ 0.51	56.7*
MELC (Group 4)	50 mg	4.2 $\pm$ 0.58	30.0*
MELC (Group 5)	100 mg	3.8 $\pm$ 0.37	36.7*
MELC (Group 6)	200 mg	3.6 $\pm$ 0.75	40.0*
MELC (Group 7)	400 mg	3.2 $\pm$ 0.49	46.7*

All administrations (aspirin and extract) were made orally. Values represented as mean  $\pm$  SEM, (n=5); \* $P < 0.05$ ; significant compared to control.

The analgesic activity of leaves, flowers and fruit peels of *Luffa cylindrica* has been previously noted (Khan *et al.*, 2013). It is also noteworthy that the fruits of the plant have been reported to contain a number of polyphenols, like apigenin and luteolin derivatives. Apigenin and luteolin are flavonoids; various flavonoids including apigenin and luteolin are known to have anti-inflammatory and analgesic properties (Sandhar *et al.*, 2011). Luteolin and luteolin-7-O-glucoside have been shown to inhibit lipopolysaccharide-induced inflammatory responses through modulation of NF-kappaB/AP-1/PI3K-Akt signaling cascades in RAW 264.7 cells (Park and Song, 2013). *Cardiospermum halicacabum* leaves are known to give analgesic effects; apigenin

and luteolin have been identified as constituents of the leaves (Rajasekaran *et al.*, 2013). Thus these compounds may be responsible for the observed analgesic effects of MELC.

Pain is caused by increased synthesis of prostaglandins like PGE<sub>2</sub>, which is mediated through cyclooxygenase and/or lipoxygenase activities. Thus the extract may be postulated to contain phytoconstituents with inhibitory effects on the synthesis of prostaglandins. Although the exact constituent was not identified in this preliminary study, the strong analgesic activity demonstrated by the extract merits further work towards isolation of the responsible bio-constituents of the fruits.

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