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ANTI-EMETIC AND ANTI-INFLAMMAOTRY ACTIVITY OF FRUIT PEEL OF *LUFFA CYLINDRICA* (L.) ROEM

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ABSTRACT

Ethanol extract of Luffa cylindrica (L.) Roem. Fruit peel was evaluated for anti-emetic and anti-inflammatory effect using chick emesis model and carrageenan induced rat paw edema. The antiemetic effect was observed at a dose of 150 mg /kg body weight whereas anti-inflammatory effect was observed at doses of 500, 750 and 1000 mg /kg body weight orally. Chlorpromazine 150mg/kg and indomethacin 10mg/kg orally were used as standard anti-emetic and anti-inflammatory drugs. The anti-emetic effect was determined by calculating the mean decrease in number of retching in contrast with those of control after 10 minutes of copper sulfate (50mg/kg orally) administration. The degree of paw edema of all the groups was measured using a plethysmometer at 5th hour of carrageenan (1% w/v) administration. The extract exhibited statistically significant anti-emetic (P< 0.001) and anti-inflammatory (P< 0.05) effects.

Keywords: *Luffa cylindrica*, antiemetic activity, anti-inflammatory activity, fruit peel, chick emesis model, carrageenan induced rat paw edema

INTRODUCTION

Luffa cylindrica (L.) Roem., (family Cucurbitaceae) is an annual climbing or trailing herb which is cultivated in Pakistan at Jehlum, Jammu and Kashmir, Loralai and Karachi (Nazimuddin & Naqvi, 1984). It has long been used as medicinal herb to treat asthma, intestinal worms, sinusitis (Chakravarty 1990; Schultes 1990), edema, pharyngitis and rhinitis (Khare 2007). Leaves are used in amenorrhea, decayed teeth, parasitic affections, skin diseases (Porterfield 1955), chronic bronchitis (Khare 2007) pain, inflammation, carbuncles, abscesses, (Perry 1980). Stem is used in respiratory complaints (Porterfield 1955) fruits in hemorrhage from bowels or bladder, hernia, hemorrhoids, jaundice, menorrhagia, scarlet fever (Porterfield 1955), bronchitis, haematuria, leprosy, spleenopathy and syphilis (Prajapati *et al.,* 2003). Flowers are effective in migraine (Khare 2007).

Phytochemically, leaves contain flavonoids (Schilling & Heiser, 1981), saponins (Liang *et al.,* 1993 & 1996) and triterpenes (Nauking Institute of Materia Medica 1980) where as in fruits ascorbic acid, anthocyanins, flavonoids (Bor *et al.,* 2006), triterpenoid saponins (Partap *et al.,* 2012) are present. The flowers are rich in flavonoids (Schilling & Heiser, 1981), carotenoids, flavonoids and oleanolic acid were found in the peel (Kao et al., 2012) where as polypeptides are reported in Seeds (Abirami *et al.,* 2011).

Pharmacologically, anti-tussive, anti-asthmatic, cardiac stimulant, hepatoprotective, hypolipidemic properties (Partap *et al.,* 2012) analgesic (Velmurugan *et al.,* 2011), anti-inflammatory (Muthumani *et al.,* 2010; Abirami *et al.,* 2011; Khan *et al.,* 2013) and anti-emetic activities (Khan *et al.,* 2013) are reported.

MATERIALS AND METHODS

Plant material and crude extract preparation

The fruits were collected from Malir, Karachi in June 2012 and compared with already deposited voucher specimen (G.H.No.85993) in the herbarium of Department of Botany, University of Karachi. The peel of fruits was soaked in ethanol for a week then was filtered and concentrated to dryness in vacuum at 40°C by rotary evaporator.

Animals

Twenty one chicks in three different groups (N=7 for each group) were used for the study. Young male chicks, 4 days of age, weighing from 32-52 g were taken from local market. After 24 hrs fasting, the anti-emetic activity was evaluated. The animals were maintained under standard laboratory conditions (temperature 25±2°C) and fed with standard pellet diet and fresh water *ad libitum*. The first group (Group I) represented control group which received 0.9% normal saline solution, orally at a dose of 10 ml/kg b.w., and the second group (Group II) received standard anti-emetic drug chlorpromazine (150mg/kg orally). Group III-received ethanol extract of fruit peel (150mg/kg orally).

For anti-inflammatory activity, male Whistar Albino rats (weighing 150-200 g), were procured from animal house of Aga Khan University and Hospital, Karachi. The animals were grouped with six animals per cage. Group I received 1% tween 80 in water, orally at a dose of 10 ml/kg b.w., as control. Group II received indomethacin (10mg/kg orally) as standard anti-inflammatory drug. Whereas group III, IV and V received ethanol extract of fruit peel in a dose of 500, 750 and 1000mg/kg b.w., orally respectively. Permission and approval for animal studies were obtained from Board of Advanced Studies and Research, University of Karachi [BASR.Res.No.5 (4)-2007].

Drugs and Chemicals

Copper sulfate (Scharlau Chemie S.A. Barcelona, Spain), chlorpromazine (ICN, USA), Dimethyl sulfoxide (DMSO) and ethanol (Merck, Darmstadt, Germany) and indomethacin (Sigma-Aldrich Corporation) were used in the experiment.

Anti-emetic activity

The anti-emetic activity was determined by following the protocols of Akita *et al.,* 1998. Each chick was set aside in a large beaker for 10 minutes to stabilize. Chlorpromazine and the extract were dissolved in 0.9 % saline containing 5 % DMSO and 1 % tween 80 and administered abdominally at a dose of 150 mg/kg b.w., to the test animal. After 10 minutes copper sulfate was administered orally at 50 mg/kg b.w., to each chick, then the number of retching was observed during the next 10 minutes.

The percent inhibition was calculated by the following formula:

Inhibition (%) = [(A-B)/A] x 100

Where A = Frequency of retching in control group

B = Frequency of retching in test groups

Anti-inflammatory activity

The anti-inflammatory activity of the extracts was determined according to the method of Vogel and Vogel, 1997. All the suspensions were administered 30 min before the induction of edema by administering 0.1 ml of 1% w/v carrageenan in saline. The degree of paw edema

of all the groups was measured using a plethysmometer at 5th hour of carrageenan administration to each group.

% Inhibition was calculated using the formula: % Inhibition (treated) = $\frac{V_5 - V_o}{V_o} \times 100$

Where V_5 and V_o represent Right hind paw volume at 5th hour after and before sub-plantar injection of carrageenan, respectively.

Statistical Analysis

All data were expressed as the mean ± S. E. M. The data was analyzed by using unpaired Student's *t*-test and P<0.001 (for antiemetic activity) P<0.05 (for anti-inflammatory activity) were considered statistically significant.

RESULTS

Antiemetic Activity

The results of anti-emetic effect of fruit peel of *Luffa cylindrica* are shown in table 1. The ethanol extract of peel showed 69.34 % and standard drug chlorpromazine 34.87 % inhibition of retches.

Table 1. The anti-emetic effect of fruit peel of *Luffa cylindrica.*

Treatments	*Number of Retches (Mean ± SEM)*	*%Inhibition of retches*
Control (Normal saline solution)	71.28 ± 3.54	-
CZ 150mg/kg p.o.	46.42 ± 4.25*	34.87
PE 150mg/kg p.o.	21.85 ± 0.93*	69.34

CZ = Chlorpromazine; PE = Ethanol extract of fruit peel ; N=7; Dose=150 mg/kg orally ; *P* < 0.001 is significantly different from control value using unpaired student's *t*-test.

Anti-inflammatory activity

The data of anti-inflammatory activity is presented in table 2. The anti-inflammatory activity on carageenan induced rat paw edema was compared to that of control on the basis of percent inhibition of paw edema volume.

Table 2. The anti-inflammatory effect of fruit peel of *Luffa cylindrica.*

Group	*Dose (mg/kg) orally*	*Mean paw volume ± S. E. M. At 5th hr in ml*	*Percent inhibition of edema*
Control	-----	0.97 ± 0.07	-----
IN	10	0.32 ± 0.01*	67.01
PE	500	0.33 ± 0.06*	65.97
	750	0.23 ± 0.03*	76.28
	1000	0.24 ± 0.02*	75.25

IN = Indomethacin; PE = Ethanolic extract of peel; N = 6.*P<0.05 is statistically significant values as compare to control using unpaired student's *t*-test.

The results showed that the extract (500, 750 and 1000 mg/kg b.w.,) exhibited statistically significant (P< 0.05) inhibition of paw volume. Maximum percent inhibition of paw edema was found at 750 mg/kg b.w which is 76.28. The standard drug indomethacin showed 67.01 % inhibition at dose of 10mg/kg body weight.

DISCUSSION

Antiemetic Activity

The ethanol extract of fruit peel of *Luffa cylindrica* showed significant ($p < 0.001$) anti-emetic effect in young chicks. The protective effect of the extract against copper sulfate induced retching in young chicks is possibly by peripheral action as the oral copper sulfate induces emesis by peripheral action through excitation of visceral afferent nerve fibers of the GIT (Bowman and Rand, 1980). It has also been established that the peripheral 5-HT_3 (Fukui *et al.,* 1993), 5-HT_4 (Fukui *et al.,* 1994) or NK_1 (Ariumi *et al.,* 2000) receptors are involved in emesis. Therefore, it may be said that the ethanol extract of *Luffa cylindrica* fruit peel produced anti-emetic activity by receptor antagonism and has peripheral anti-emetic action.

Anti-emetic activity by using copper sulfate proposed 5-HT_3 (Fukui *et al.,*1993), 5-HT_4(Fukui *et al.,*1994) or NK_1 (Ariumi *et al.,* 2000) receptors antagonism. Therefore it may be said that the extract was able to effectively prevent its effect and has a peripheral anti-emetic action.

Anti-inflammatory activity

The anti-inflammatory effect of plant extracts and natural products are frequently assessed by carrageenan-induced rat paw edema (Panthong *et al.,* 2003). Edema development in carrageenan-induced paw edema model in rats is represented by two phases (Vinegar *et al.,* 1969). The first phase occurs within an hour of carrageenan injection and is partly due to the trauma of injection and also due to release of histamine and serotonin (Crunkhon and Meacock, 1971).Where as arachidonate metabolites (prostaglandins, leukotrienes) play a major role in the development of the second phase of reaction that is measured around 3-hours time (Vinegar *et al.,* 1969; Crunkhon and Meacock, 1971). The presence of prostaglandins in the inflammatory exudates from the injected foot can be demonstrated at 3-hour and period thereafter (Vinegar *et al.,* 1969). Non-steroidal anti-inflammatory agents inhibit cyclooxygenase (COX-2) enzymes involved in prostaglandin synthesis (Robinson, 1997; Kulkarni *et al.,* 2000). Based on these reports it is possible that the inhibitory effect of fruit peel of *Luffa cylindrica* (L.) Roem., on carrageenan-induced inflammation in rats could be due to inhibition of cyclooxygenase leading to inhibition of prostaglandin synthesis. Although the cyclooxygenase and lipoxygenase pathways are both involved in the inflammatory process, inhibitors of cyclooxygenase are more effective in inhibiting carrageenan-induced inflammation than lipoxygenase inhibitors (Flower *et al.,* 1980). In our experiment, rats pre-treated with *Luffa cylindrica* (L.) Roem., showed a significant edema inhibitory response at 5^{th} hour following carrageenan injection. This result suggests that *Luffa cylindrica* extracts may act by suppressing the later phase of the inflammatory process by the inhibition of cyclooxygenase.

Flavonoids possess anti-emetic (Kinoshita *et al.,* 1996) and anti-inflammatory (Rotelli *et al.,* 2003) activity. The ethanol extract of *Luffa cylindrica* peel contain the highest level of total flavonoids (Kao *et al.,* 2012). Therefore, it may be said that flavonoids may play important role in anti-emetic and anti-inflammatory effect of the extract besides other compounds. The present study is on preliminary level and results need to be verified in other experimental models and the compound(s) related activity is required to further specify the responsible anti-emetic and anti-inflammatory phytochemical.

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