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Research Article

Antiulcerogenic effect on the ethanol extract of the Fruits of *Garcinia mangostana* on Experimental Gastric Ulcer in Rats

Pankaj Nainwal*[1], Deepak Nanda[1], Kapil Kalra[1], Sachchidanand Mani Tripathi[2]

[1] *Dev Bhoomi Institute of Pharmacy and Research, Dehradun, Uttarakhand, INDIA*
[2] *Sri Krishana Chaithnya College of Pharmacy; Madanapalle; Chittore dist. A.P, INDIA*

ABSTRACT:
In the ethanol-induced ulcer protocol, *Garcinia mangostana* crude extract (250 and 500 mg/kg) significantly reduced the lesion index, the total lesion area and the percentage of lesion, in comparison with control group. In indomethacin-induced ulcer only the treatments with 500 mg/kg of *Garcinia mangostana* extract and 100 mg/kg of cimetidine reduced significantly in comparison with control group ($p < 0.05$). Regarding the stress-induced ulcer protocol, it was observed a significant reduction ($p < 0.05$) in lesion index, total lesion area and in the percentage of lesion in animals treated with *Garcinia mangostana* crude extract (250 and 500 mg/kg) and cimetidine (100 mg/kg), In the gastric secretion determination model, using ligated pylorus, the treatment with *Garcinia mangostana* crude extract (250 and 500 mg/kg) and cimetidine (100 mg/kg), respectively, reduced the volume of gastric juice, total acidity and raised gastric pH appreciably.
Keywords: lesion index, indomethacine, lesion area, cimetidine

INTRODUCTION

The Purple Mangosteen (*Garcinia mangostana*), colloquially known simply as "the mangosteen", is a tropical evergreen tree. The tree grows from 7 to 25 m (20–80 ft) tall. The rind (exocarp) of the edible fruit is deep reddish purple when ripe. Botanically an aril, the fragrant edible flesh can be described as sweet and tangy, citrusy with peach flavor and texture. The Purple Mangosteen belongs to the same genus as the other — less widely known —mangosteens, such as the Button Mangosteen (*G. prainiana*) or the Lemondrop Mangosteen (*G. madruno*).

MATERIALS AND METHODS

Preparation of Garcinia mangostana extract

Garcinia mangostana samples were collected from Nilgiri hills, Southern India in June 2009. Samples were air-dried at 40 °C for 48 h. The dried friuts of Garcinia *mangostana* was kept in a freezer overnight, powdered (100 g) and submitted to ethanolic extraction (70% ethanol) at room temperature by maceration. After filtration, the extract was concentrated under reduced pressure, yielding 5.2 g of crude *Garcinia mangostana* extract.

Animals

Healthy Female Wister albino rats weighing 150-200g for acute toxicity study and for antiulcer studies, obtained from the animal house of Govt. veterinary college, Pantnagar, (Protocol no. 236/2007) were used in the Present study. All animals were housed in standard propylene cages and fed with standard chow diet (Hindustan Liver, Kolkata) and water *ad libitum* for 2 days before the experiment, maintained with light/dark cycle of 12/12 hours in the temperature 25±2°C. Rats were starved for overnight & was allowed fresh water before administration of the plant extract.

Pharmacological assays

Ethanol-induced ulcer[2]

The experiment was performed according to the method of Morimoto et al. (1991). After 12 h of fasting, rats were randomly divided into four groups of six animals each. To the first group it was given 1 mL of vehicle (1% Tween-80 aqueous solution), and the second group was treated with omeprazole (30 mg/kg). The remaining two groups received 250 and 500 mg/kg of *Garcinia mangostana* crude extract, respectively. All treatments were administered orally. One hour after treatment, all rats received 1 mL of 99.5% ethanol to induce gastric ulcer. After 1 h the animals were sacrificed by cervical dislocation, the stomachs were removed and opened along the greater curvature. Stomachs were gently rinsed with water to remove gastric contents and blood clots before scanning[3]. The obtained images were analyzed by software "EARP" for measuring each lesion point. The ulcers were classified as level I, ulcer area <1 mm^2; level II, ulcer area 1–3 mm^2; level III, ulcer area >3 mm^2. The following parameters were determined: (i) Ulcerative Lesion Index (ULI) as 1 × (number of ulcers level I) + 2× (number of ulcers level II) + 3 × (number of ulcers level III); (ii) curative ratio, which was determined as follow**:**

$\%C = 100 - (IU_{treated} \times 100 / IU_{control})$; (iii) total area of lesion; (iv) percentage of lesion area in relation to the

***Corresponding author: Pankaj Nainwal**
Sr. Lecturer, Dev Bhoomi Institute of Pharmacy and Research
Dehradun, Uttarakhand-248001
Email: pankajherbs@gmail.com

total stomach area.

Nonsteroidal anti-inflammatory Garcinia mangostana Linn (NSAID)-induced ulcer[3]

The experiment was performed according to the method of Nwafor et al. (2000). After 12 h of fasting, rats were randomly divided into four groups of six animals each. To the first group it was given 1 mL of vehicle (1% Tween-80 aqueous solution), and the second group was treated with cimetidine (100 mg/kg). The remaining two groups received 250 and 500 mg/kg of *Garcinia mangostana* crude extract, respectively. All treatments were administered orally. One hour after treatment, all rats received indomethacin (100 mg/kg) to induce gastric ulcer. Four hours later the animals were sacrificed by cervical dislocation, the stomachs were removed, and opened along the greater curvature. Stomachs were gently rinsed with water to remove gastric contents and blood before scanning. The obtained images were analyzed using the parameters previously described.

Stress-induced ulcer[4]

The method described by Basile et al. (1990) was employed in this assay. Groups of six animals were treated as previously described, and 30 min later, each animal was placed in a tube and immersed vertically until the water reached the neck region in a tank with current water at 25 °C for 17 h. After this period, the rats were sacrificed by cervical dislocation. The stomachs were removed, opened along the greater curvature, followed by gently washing with water to remove gastric contents and blood clots before scanning[5]. The obtained images were analyzed using the parameters described above.

Determination of gastric secretion[6]

The assay was performed by using the method of Shay et al. (1945) with a few modifications. The animals were divided into groups ($n = 6$), according to the employed treatment, as previously described. After 24 h of fasting, the animals were anesthetized with ketamine hydrochloride (10 mg/kg, i.p.), the abdomen was incised and the pylorus ligated. Immediately after pylorus ligature, *Garcinia mangostana* extract was administered at the doses of 250 and 500 mg/kg, respectively. Cimetidine (100 mg/kg) was used as positive control, and 1 mL of vehicle (1% Tween- 80 aqueous solution) was administered as negative control. All samples were administered intraduodenally. Four hours later, the animals were sacrificed by cervical dislocation, and the abdomen was opened to place another ligature at the oesophageal end. The stomachs were removed and the gastric content was collected and centrifuged at 3000 rpm ($8000 \times g$, 25 °C, 10 min). The amount of gastric-juice acid (mL) and the pH values were determined. Total acid secretion was determined in the supernatant volume by titration to pH 7 using a 0.01 mol^{-1} NaOH solution, and phenolphthalein as indicator.

RESULTS

In the ethanol-induced ulcer protocol, it was observed that the treatment with *Garcinia mangostana* crude extract (250 and 500 mg/kg) and omeprazole (30 mg/kg) significantly reduced the lesion index, the total lesion area and the percentage of lesion, in comparison with control group ($p < 0.05$). On the other hand the indomethacin-induced ulcer protocol, only the treatments with 500 mg/kg of *Garcinia mangostana* extract and 100 mg/kg of cimetidine reduced significantly all the evaluated parameters in comparison with control group ($p < 0.05$). Regarding the stress-induced ulcer protocol, it was observed a significant reduction ($p < 0.05$) in lesion index, total lesion area and in the percentage of lesion in animals treated with *Garcinia mangostana* crude extract (250 and 500 mg/kg) and cimetidine (100 mg/kg), respectively (Table 1).In the gastric secretion determination model, using ligated pylorus, the treatment with *Garcinia mangostana* crude extract (250 and 500 mg/kg) and cimetidine (100 mg/kg), respectively, reduced the volume of gastric juice, total acidity and raised gastric pH appreciably ($p < 0.05$) in comparison with control group (Table 2).

Table 1: Effects of *Garcinia mangostana* extract (*GME*) on ethanol, indomethacin and stress-induced gastric ulcers in rats

Method	Treatment	Dose (mg/kg)	Total area of lesion (mm^2)	% of lesion area	Ulcer index	Inhibition (%)
Ethanol induced ulcer	Control	—	223.24 ± 26.14	31.24 ± 4.06	48.46 ± 5.87	—
	Omeprazole	30	0.81 ± 0.81*	0.11 ± 0.23*	0.80 ± 1.02*	98.34
	GME	250	7.21 ± 0.87*	2.11 ± 0.63*	11.05 ± 1.12*	63.42
		500	3.72 ± 2.47*	0.58 ± 0.36*	5.62 ± 3.29*	88.42
Indomethancin induce ulcer	Control	—	32.06 ± 5.32	5.30 ± 0.52	43.12 ± 3.41	—
	Cimetidine	100	13.45 ± 4.33*	2.35 ± 0.78*	21.37 ± 5.81*	49.81
	GME	250	30.27 ± 5.35	3.85 ± 0.75	38.99 ± 6.81	5.85
		500	10.35 ± 5.64*	1.73 ± 0.98*	18.40 ± 6.29*	55.25
Stress Induce ulcer	Control	—	132.11±9.77	16.01±1.14	91.90±2.54	—
	Cimetidine	100	21.57±2.96*	2.46±0.30*	24.80±3.26*	73.16
	GME	250	70.05±9.18*	7.22±0.66*	52.38±4.37*	42.85
		500	34.54±8.75*	3.78±1.06*	27.65±2.74*	60.98

Results are mean ± S.E.M. for six rats. Statistical comparison was performed using ANOVA followed by Dunnet's test.
*$P < 0.05$ when compared to control group.

Table 2: Effects of *Garcinia mangostana* extract (*GME*) and cimetidine, administered intraduodenally, on the biochemical parameters of gastric juice obtained from pylorus-ligature in rats

Treatments	Dose (mg/kg)	Volume (mL)	pH	$[H^+]$(mEq/I/4h)
Control	—	2.87±0.50	2.05±0.27	96.69±10.67
Cimetidine	100	0.72±0.06*	6.14±1.07*	24.21±2.71*
GME	250	1.06±0.24*	2.43±0.16*	49.97±4.89*
	500	0.82±0.15*	3.09±0.36	49.65±5.84*

Results are mean ± S.E.M. for six rats. Statistical comparison was performed using ANOVA followed by Dunnet's test.* $p < 0.05$ when compared to control group.

DISCUSSION

Based on the obtained results it is suggested that *Garcinia mangostana* extract could prevent gastric lesions caused by ethanol, indomethacin, as well as by stress. These are the most commonly used protocols for the evaluation of anti-ulcer agents in rats[7,8]. The ability of the gastric mucosa to resist injury by endogenous secretions (acid, pepsin, and bile) and by ingested irritants (e.g. alcohol) can be attributed to a number of factors that have been generally referred as mucosal defense[9]. The formation of gastric mucosal lesions by necrotizing agents, such as ethanol, has been reported to involve the depression of these gastric defensive mechanisms[12]. On one hand, ethanol administration reduces mucus production, gastric mucosal blood flow, bicarbonate secretion, endogenous glutathione and prostaglandin (PG) levels, and on the other hand it increases the release of histamine, the influx of calcium ions, the generation of free radicals and the production of leukotrienes[10]. The results show that *Garcinia mangostana* extract probably display an antiulcerogenic effect related to cytoprotective activity, since it reduced significantly the ethanol- induced ulcer. Nonsteroidal anti-inflammatory *Garcinia mangostana Linn*s (NSAIDs) such as indomethacin have the ability to cause gastroduodenal ulceration, and this effect is related to the ability of these agents to suppress prostaglandin synthesis. Several studies have indicated that gastroduodenal protection by prostaglandins (PGs) is due to increasing mucosal resistance, as well as the decrease in aggressive factors, mainly acid and pepsin[1]. In this model, *Garcinia mangostana* crude extract was also significantly effective in reducing the mucosal damage. The increased synthesis of mucus and/or prostaglandins can be explained as the probable cytoprotective mechanism in this assay. Gastric stress ulceration is probably mediated by the release of histamine. It does not only enhance the gastric secretion, often called the aggressive factor, but also causes disturbances of gastric mucosal microcirculation, anormal motility, as well as reduces mucus production, which is called defensive factor. Moreover, stress-induced ulcer in animal models may be partially or entirely prevented by vagotomy, since the increased vagal activity has been suggested to be the main factor in stress induced ulceration[13].

In the current study, *Garcinia mangostana* crude extract inhibited the production of stress-induced ulcers. This finding indicates that *Garcinia mangostana* may enhance gastric mucosal defensive factors. Moreover, the pylorus ligature technique revealed that *Garcinia mangostana* (250 and 500 mg/kg) possess anti-secretory potency. This effect could suggest an anti-histaminic activity. The analysis of the chemical composition of *Garcinia mangostana* extract used in this study revealed the presence of prenylated cinnamic acid derivates and flavonoids. In this regard, numerous flavonoids have shown anti-secretory and cytoprotective properties in different experimental models of gastric ulcer. Besides, antioxidant activity was observed in extracts containing prenylated cinnamic acid derivates. Thus, this additional information contributes to explain the anti-ulcer activity observed for the extract of *Garcinia mangostana*. Overall, the obtained results demonstrate that *Garcinia mangostana* displays a good anti-ulcer activity, corroborating the folk use of *Garcinia mangostana* preparations, and contributing for its pharmacological validation.

REFERENCES

1. Kahre. CP. Indian Medicinal Plants, Springer-Verlag Berlin/Heidelberg, 2007, 275.
2. MorimotoI A, Toshiraru A, Sugimura F, Matsuo Y, Honda T. Recurrence during maintenance therapy with histamine H2 receptors antagonist in cases of gastric ulcers. Nihon Univ J Med 1986 , 28;69–74
3. Nwafor, VS, Castro, SL, Marcucci, MC. Propolis: Recent Advances in Chemistry and Plant Origin. Apidologie 2000,31;3–15
4. Basile AC, Sertié, JA, Panizza S, Oshiro,T T, Azzolini CA. Pharmacological assay of *Casearia silvestris* preventive anti-ulcer activity and toxicity of the leaf crude extract. Journal of Ethnopharmacology 1990,30;185–97
5. Belaiche J, et al. Observational survey of NSAID-related upper gastro- intestinal adverse events in Belgium. Acta Gastroenterologic Belgiam 2002, 65;65–73
6. Shay AE, Antônio MA, Kohn LK, Rehder VG, Foglio MA. Antiulcerogenic activity of a crude hydroalcoholic extract and coumarin isolated from *Mikania laevigata.* Bio Phytomed 2005,12; 72–7
7. Castaldo C, Capasso F. Garcinia: An old remedy used in modern medicine. Fitoterapia 2002, 73, 1–6
8. Castro SL. Garcinia: Biological and Pharmacological Activities Therapeutic uses of this bee product. Annual Review of Biomed Science 2001,3;49–83
9. Gonzalez FG, Di Stas LC. Anti-ulcerogenic and analgesic activities of the leaves of *Wilbrandia ebracteata* in mice. Phytomedicine 2002, 9;125–34
10. Glavin GB, Szabo S. Experimental gastric mucosal injury: laboratory models reveal mechanisms of pathogenesis and new therapeutic strategies. Exp Biol Journal 1992, 6;825–31
11. Havsteen B. The biochemistry and medical significance of the flavonoids. Pharmacol and Therap 2002, 96;67–202
12. Kinoshita M, Tsunehisa N, Tamaki H. Effect of a

combination of ecabet sodium and cimetidine on experimentally induced gastric-lesions and gastric-mucosal resistance to ulcerogenic agents in rats. Biol and Pharma Bull 1995,18; 223–226

13. Marcucci MC, Rodriguez J, Ferreres F, Bankova V. Chemical composition of Brazilian garcinia from Sa˜o Paulo. Zeists fur Nature 1998, 53;117–9.