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Protective effects of *Moringa oleifera* on experimentally induced gastric ulcers in rats

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ABSTRACT

To study the possible antiulcer effects of water extracts of *Moringa oleifera* in two animal models of ulcers. The water extract of leaves was prepared and tested for antiulcer activity at the dose level of 200mg and 400 mg/kg PO in pyloric ligation and ibuprofen induced gastric ulcer models. The effect of the extract was compared famotidine(3.6 mg/kg PO). The antiulcer effects of the drugs were assessed on the parameters such as number, size and index of ulcers and the volume, acidity and pH of gastric juice. The extract of *Moringa oleifera* and famotidine significantly ($p<0.001$) reduced ulcer index in the model employed. Both Moringa oleifera extract and famotidine reduced significantly ($p<0.001$) the free and total acidity of gastric juice. *Moringa oleifera* possesses gastric ulcer protective principles.

Key words: *Moringa oleifera,* gastric ulcer, famotidine, ibuprofen, antiulcer

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INTRODUCTION

Man has been using herbs and plant products for combating diseases since time immemorial. The traditional system of medicine is so engrained in our culture that, even now 75% of the Indian population depend on this indigenous system for relief of symptoms. With such a huge section of an ever-increasing population relying on herbal remedies, it is imperative that the plant products, which have been in use from ages, be scientifically supported for their efficacy [1]. Ayurveda is an original holistic system of medicine whose principles of therapeutics are applicable universally. A continued search for ayurvedic medicinal plants during the last several centuries has given rise to a long list of plants which are of great use in the treatment of diseases, and for promoting health. It can be stated that every disease has a cure in a plant growing in nature. Drugs used in medicine today are either obtained from nature or are of synthetic origin. Ayurvedic medicinal plants, as a basis for new drugs, have great promise and it is gratifying to note that the World Health Organization (WHO) have shown an abiding interest in plant derived medicines described in the folklore of various countries [2].

Peptic ulcer is an epidemic disease. It is the disease of the 20th century. Its incidence has been estimated to be 10% of the general population [3]. This disease affects males and females equally in West, whereas in India, men are affected 18 times more commonly than women. In a large country like India, differences are bound to exist between regions. South India, where rice is the staple food, has a higher incidence of peptic ulcer than the north. Peptic ulcer disease is best reviewed as an imbalance between mucosal defense factors (bicarbonate, prostaglandins, mucin, NO, other peptides and growth factors) and injurious factors (acid, Pepsin). *H.pylori and* exogenous agents such as non-steroidal anti-inflammatory drugs (NSAIDS) interact in complex ways to cause an ulcer. Upto 60% of peptic ulcers are associated with *H.pylori* infection of the stomach. Last two decades have witnessed introduction of a number of new drugs for the treatment of peptic ulcers. None of these drugs are free from toxicities. Efforts have also been made to find suitable alternative remedies from plant and animal origins for the treatment of peptic ulcers. *Moringa oleifera*, commonly known as drumstick tree, is the most widely cultivated variety of the genus *Moringa*. It is of the family Moringaceae. *Moringa* is native to the southern foothills of the Himalayas, and possibly Africa and the Middle East. It is widely cultivated in Africa, Central and South America, India, SriLanka, Mexico, Malaysia and the Philippines. The tree has its origin from the South Indian state of Tamilnadu and Kerala. It is an exceptionally nutritious vegetable tree with a variety of potential uses. Its leaves are highly nutritious and are full of medicinal properties [4]. It finds its use as an anti-pyretic, anti-spasmodic, anti-hypertensive, anti-anemic etc and is also used for various bacterial, fungal and viral infections [4]. In a preliminary investigation *M.oleifera* is proved to be an anti ulcerogenic [5].

Recent studies on *Moringa oleifera* indicate it to possess antioxidant property [6]. A number of studies have implicated the role of oxidative stress in the pathophysiology of peptic ulcer [7, 8]. Drugs possessing antioxidant propertyhave been observed tom mitigate. Thus *M.oleifera* could be expected to play a promising role in the treatment of peptic ulcers [8, 9]. In

view of these the present study has been undertaken to evaluate in detail the anti-ulcer effect of the water extract of *M.oleifera* in two different peptic ulcer models.

MATERIALS AND METHODS

Anti-ulcer Study

Plant material

The plant materials were collected locally from Udupi district, Karnataka. The plants were authenticated by Department of Botany, MGM College, Udupi, Karnataka.

Preparation of Extract

The leaves were separated, shade-dried and powdered in a grinder. Leaves of the plants were crushed in to moderately coarse powder and immersed in distilled water in a flask and allowed to stand for seven days. The solid residue obtained by straining was pressed and filtered. The filtrate was concentrated on a water bath to get a viscous paste (10). It was finally dried in a desiccators and the yield was calculated according to the body weight of rat.

Animals

Studies were conducted in Wistar rats of 180-200 g weight. The animals were locally bred and maintained under standard laboratory conditions. They had free access to water and standard food; *ad libitum.* However, rats were fasted for 24 h and water was withdrawn 1h before administration of drugs. The experimental protocol was approved by the Institutional Animal Ethics Committee.

Drug treatment

The extracts and drugs were suspended in 1% sodium carboxy methyl cellulose to administer them orally in the volume of 2.5 ml/kg. Eight groups of animals each containing 6 rats were used. Four groups (Group I- Control, Group II- Famotidine 3.6 mg/kg, Group III- Test 1, Group IV- Test 2) of animals, each was used for pylorus ligation and ibuprofen induced ulcer models. *M. oleifera* was administered in the dose of 200mg/kg (Test 1) and 400mg/kg (Test 2) [5].

Ulcer Induction

Pylorus-ligation method

One hour after the drug administration the pylorus was ligated under light ether anaesthesia as described by Shay et al [11]. Nineteen hours later the rats were killed and their stomachs were dissected out after ligating the cardiac end. Each stomach was cut open along the inner

curvature and the contents were collected. Then the mucosa was washed and the extent of ulceration was scored [12]. The gastric juice collected from each stomach was then centrifuged and its volume and pH were measured. Free and total acidity were estimated titrimetrically with 0.01 N NaOH using Toepfer's reagent and phenolphthalein as indicators. Acidity of the gastric juice was expressed as mEq/L/hr/100g body weight.

Ibuprofen-induced ulcers

Ibuprofen in the dose of 300mg/kg was administered orally at 15 h intervals to fasted rats to produce gastric ulcers [13]. The animals were killed 6 h after the second dose of ibuprofen and the ulcers were scored. The test drugs were administered 1 h before each dose of ibuprofen administration [10].

Statistical analysis

Results were expressed as Mean±SE. The data was analyzed by one way ANOVA followed by Bonferroni's post-hoc test.

RESULTS

The severity of gastric ulceration in both the models was assessed based on the means of ulcer index. Both the models produced moderate to severe ulcers in control group of animals; in that the maximum was by pylorus ligation method. Both famotidine and the extract of *M.oleifera* significantly ($p<0.001$) reduced the ulcer index as compared to control group in both ulcer models. (Table 1).

TABLE:1 Effect of water extract of *Moringa oleifera* on peptic ulcers in experimentally induced ulcers in rats.

Ulcer induction method	Ulcers	GROUPS			
		Control	Famotidine (3.6 mg/kg)	*M.oleifera* (200 mg/kg)	*M.oleifera* (400 mg/kg)
Pylorus ligation	Ulcer Index	0.5111±0.664	0.142±0.0401*	0.0841±0.0180*	0.1565±0.0179*
Ibuprofen induced	Ulcer Index	0.6541±0.06327	0.1125±0.01085*	0.1005±0.02225*	0.1173±0.01187*

Values are in Mean ± SEM; *p< 0.001 vs. control; n=6

The antiulcer effect of *M.oleifera* was comparable with that of the standard drugs in pylorus ligation and ibuprofen induced ulcer methods. Famotidine and *M.oleifera* extract significantly ($p<0.05$) reduced the free acidity and total acidity of gastric juice. (Table 2).

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TABLE:2 Effect of water extract of *Moringa oleifera* on gastric acid secretion in pyloric ligation method.

Drugs	Dose (mg/kg)	Volume of gastric juice (ml/100 g body weight)	pH of gastric juice	Free Acidity (mEq/L/hr/100g body weight)	Total Acidity (mEq/L/hr/100g body weight)
Control		9.8±0.5447	1.65±0.047	0.8618±0.009	1.7411±0.041
Famotidine	3.6	3.333±0.421*	2.31±0.050*	0.33±0.007*	0.6766±0.030*
M.oleifera					
Test 1	200	3.5±0.5627*	2.18±0.050*	0.4515±0.006*	0.8955±0.017*
Test 2	400	4.666±1.085*	2.28±0.041*	0.5095±0.005*	0.9776±0.036*

Values are in Mean ± SEM ; *p<0.001 vs control; n=6

DISCUSSION

The present study was undertaken to see if the water extract of *M.oleifera* could show antiulcer effect in two different models of peptic ulcers. The extract showed comparable antiulcer effects to the standard drug in pylorus ligation and ibuprofen induced ulcers. An increase in the acid secretion, a decrease in the gastric mucosal protection and an induction of oxidative stress in gastric mucosa are the important factors that are implicated in the pathogenesis of peptic ulcers. The causes of ulcer in the gastric mucosa after pylorus ligation are believed to be due to either stress induced increase in gastric hydrochloric acid secretion and/or stasis of acid. The volume of secretion is also an important factor in the production of ulcer due to exposure of unprotected lumen of the stomach to the accumulating acid. The results of our study showed that in pylorus ligated rats, there was a significant decrease in the mean ulcer index after treatment with the water extract of *M.oleifera* (Table 1). Moreover it resulted in a significant decrease in free acidity and total acidity. It is equally potent when compared to famotidine (Table 2).

In the ibuprofen induced ulcer model, ibuprofen, a NSAID produced spectrum of injury to the gastroduodenal mucosa ranging from hemorrhages and petechiae to erosions and ulcers. These changes could be induced due to denaturation of mucus glycoproteins in contact with high concentration of the drug. NSAIDs are known to cause gastric ulceration by inhibiting the synthesis of PGE_2 (a prostaglandin that protects the gastric mucosa). *M.oleifera* exhibited antiulcer actions against ibuprofen induced gastric ulcers. This, therefore, suggests that *M.oleifera* may have cytoprotective action on GI mucosa. In this study, the antiulcer activity of water extract of *M.oleifera* was compared with that of famotidine in the ibuprfen induced ulcer model. There is a significant decrease in the mean ulcer index in the *M.oleifera* extract treated group and equally potent as famotidine.

There are reports, which indicate that certain substances through free radical scavenging protect the gastric mucosa. Recent study on M.oleifera indicates it to possess antioxidant property [6]. We have confirmed this finding by doing the DPPH radical scavenging assay with the water extract of *M.oleifera.* Therefore, gastroprotective effect of *M.oleifera*

could be attributed to the modulation of various factors through improved gastric cytoprotection, partly due to antioxidant property. In brief, *Moringa oleifera* has got anti-ulcer activity. It being cheap, less toxic, widely used and easily available, might play as an adjunct to the existing drugs in the pharmacotherapy of peptic ulcer.

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