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TO EVALUATE ANTIULCER ACTIVITY OF MOMORDICA CHARANTIA FRUITS IN ALBINO RAT

Nazim Ali*[1], Yogendra Pal[2] and Shashi Bhooshan Tiwari[1]

[1]Department of Pharmacy, MJP Rohilkhand University, Bareilly 243006 (U.P) India.

[2]JDSR Institute of Pharmacy, Harpara, Shahjahanpur (U.P) India.

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***Corresponding Author**
Nazim Ali
Department of Pharmacy,
MJP Rohilkhand University,
Bareilly 243006 (U.P) India.

ABSTRACT

Objective: The aim of the present study is to evaluate the antiulcer activity of ethanolic extract of *Momordica charantia* in pylorus ligation gastric ulcer in rats. **Method:** Gastric ulcer was induced by pylorus ligation method. In pylorus ligation model, Rats of either sex were divided into four groups, group 1 received vehicle (distilled water 1ml/kg), group 2 (standard) received Ranitidine (10mg/kg) and group 3,4 were treated with ethanolic extract of 150mg/kg and 300mg/kg 10 days pretreatment. Rats were deprived of food, but not water, for 24 hours prior to the experiment. On 10th days, 1 hour after the respective treatments, pylorus ligation performed under diethyl ether anesthesia. Four hours later, animal were sacrificed by cervical dislocation and stomach was open along the curvature and ulcer was graded. Percentage of ulcer protection, ulcer index, free acidity, total acidity, and pH were observed and calculated. **Result:** The extract of *Momordica charantia* in pylorus ligation model. It decreased the ulcer index (0.80), and there was a decreases in total acid and free acid ($p<0.001$), and increases the pH value ($p<0.001$), increases the percentage of ulcer protection (65.66). **Conclusion:** Ethanolic extract of *Momordica charantia* was clearly shows a protective effect against total acid, free acid and ulcer index and also increase pH, and percentage of protection against ulcer in pylorus ligation model.

KEYWORD: *Momordica charantia*, pylorus ligation, Ranitidine, peptic ulcer, prostaglandin.

1. INTRODUCTION

Ulcers sore means open and painful wounds and the peptic ulcers are erosion of lining of stomach (the lining is a wrinkly bag holds acid to help digest food) or the duodenum.[1] Still the etiology of peptic ulcer is not clearly known, but it has been well established that peptic ulcer occurrence take place due to an imbalance between aggressive factor (like acid, pepsin, bile, and *H. pylori* infection), and defensive factors (like gastric mucosa, bicarbonate secretion, prostaglandins, nitric oxide and innate resistance of the mucosal cell). In gastric ulcer, acid secretion may be normal or low, while in duodenal ulcer, volume of acid secretion is high in half of patients and may be normal in rest.[2] Mucosal cell death results from increase in H^+ in its immediate environment (decrease pH).

Among various causes of gastric ulceration lesions caused by stress, alcohol consumption, Helicobacter pylori infection, and use of NSAIDs etc.[3]

The plant *Momordica charantia Linn. Cucurbitaceae* is known variously as bitter gourd, balsam pear, bitter melon, bitter cucumber, and African cucumber.[4,5] *Momordica charantia* primarily consist of glycosides, proteins, sterols, alkaloids, fatty acids flavonoids, tannins, saponins, fixed oil, triterpenoids gums and mucilage volatiles constituents.[6,7] Fruits of *Momordica charantia* used in asthma, burning sensation, colic constipation, cough, diabetes, malaria, gout, helminthiases, inflammation, leprosy, skin disease, ulcer and wound,. It has also been shown to have hypoglycemic properties in animal as well as human studies. Karela leaves used to treat piles completely. *Momordica charantia* is used as a blood purifier due to its bitter tonic properties. Karela can heal boils and other blood related problems that show up on the skin. Juice of karela is also beneficial in treating and preventing liver damage.[8]

Number of drug including proton pump inhibitor, prostaglandin analogue, histamine receptor antagonists, and cytoprotective agents, are available for the treatment of peptic ulcer, but most of these drugs produce several adverse reactions, including toxicities, and may even alter biochemical mechanism of the body upon chronic usage. Hence the present work was undertaken to investigate the antiulcer activity of alcoholic extract of *Momordica charantia* fruits in rats.

2. MATERIAL AND METHODS

2.1 Collection of drugs and procurement of chemicals

The fruits of *Momordica charantia* were collected from the local market, Bareilly. The drugs and chemicals used in the study were Ranitidine (Cadila pharmaceutical), sodium hydroxide, phenolphtheline, tofer's reagent, diethyl ether, etc. All the analytical grade chemicals were used in this study.

Rats: 150-200gm.

2.2 Identification and authentication

The plant material was identified and authenticated by the department of plant science, M.J.P. Rohilkhand University Bareilly, (U.P). A voucher specimen has been preserved at our laboratory for future reference.

2.3 Preparation of extract

The dried fine powder of the *Momordica charantia* powder was weighed on balance 30gm. And taken into the cloth material and placed in the soxhlet apparatus. 300ml of ethyl alcohol was taken as solvent into the soxhlet flask. Through the inlet and outlet of the condenser cold tap water must flow. For proper extraction the soxhlet apparatus kept running for 24 hours. The *Momordica charantia* extract laden solvent falling from the soxhlet basket is dark in color and it become clearer, that indicate the extraction process is finished. At the end of the extraction the solvent is then evaporated, total yields was 5gm, percentage yields was 16.6% as mg per gm dried powder.[9]

2.4 Experimental animals

Albino rats of either sex were employed in this study. The rats were obtained from the Indian Veterinary Research Institute at Izatnagar, Bareilly in Uttar Pradesh State and were used after acclimatization for 10 days for this study. The animals were housed in polypropylene cages under standard husbandry conditions (12 hrs light/dark cycle, 25±2^{o}C) were used for the experiment. Rats were provided water and pellet diet *ad libitum.* The animal house and breeding facility is registered with Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), government of India. All the experimental protocols were approved by the Institutional Animal Ethics Committee (IAEC). All efforts were made to reduce animal suffering and to use only the number of animals necessary to produce reliable scientific data.

2.5 Administration of drug

Ranitidine: 10mg/kg i.p[10]

2.6 Experimental design

The animals used for the experiment were divided into 4 groups for model, 6 rats in each group.

2.7 Method to induced ulcer

2.7.1 Pylorus-ligation induced ulcer[11]

Rats of either sex were divided into four groups with six rats in each group. Group 1 received vehicle (distilled water, 1ml/kg, p.o) group-2 (standard) received Ranitidine (10mg/kg) and Group-3, 4 were treated with ethanolic extracts 150mg/kg and 300mg/kg 10 days pre-treatment. Rats were deprived of food, but not water, for 24 hours prior to the experiment. On 10th day, 1hour after the respective treatments, pylorus ligature was performed under diethyl-ether anaesthesia. Four hours later, animals were sacrificed by cervical dislocation and their stomachs were dissected out and cut open along the greater curvature, inspected internally for ulcer index, free acidity, total acidity, pH, percentage of ulcer protection. The gastric juice collected into centrifuge tubes was centrifuged at 1000 rpm for 10 min and volume was noted. The stomach were washed under running tap water and then focused under microscope to note the ulcer in the glandular portion. The number of ulcers per stomach was scored microscopically with help of (10x) hand lens and the scoring is done as per standard procedure.

Pylorus ligation induced model.

2.8 Parameters: Used for anti-ulcer activity

2.8.1 pH

Gastric content collected from pylorus ligated rats was centrifuged and the volume of gastric juice as well as pH of gastric juice was noted. The gastric juice was subjected to biochemical estimations as follows

2.8.2 Determination of free and total acidity

1 ml of gastric juice was pipette into a 100 ml conical flask, 2 or 3 drops of Topfer's reagent was added and titrated with 0.01N Sodium hydroxide until all traces of red color disappears and the color of the solution turns to yellowish orange. The volume of alkali added was noted. This volume corresponds to free acidity. Then 2 or 3 drops of phenolphthalein solution was added and titration was continued until a definite red tinge appears. Repeatedly the final total volume of alkali added was noted now this volume corresponds to total acidity. Acidity was calculated by using the formula,

$$\text{Acidity} = \frac{\text{volume of NaOH} \times \text{Normality of NaOH} \times 100}{0.1} \text{meq/lt/100g}$$

2.8.3 Determination of ulcer index

Scoring of ulcer

0	Normal coloured stomach
0.5	Red colouration
1.0	Spot ulcer
1.5	Haemorrhagic
2.0	Ulcer ≥ 3 but ≤ 5
3.0	Ulcer > 5

2.8.4 Calculation of ulcer index

$U1 = UN + US + UP \times 10^{-1}$

U1	Ulcer index
UN	Average of number of ulcer per animal
US	Average of severity score
UP	Percentage of animal with ulcer

2.8.5 Determination of ulcer area

$$\% \text{ protection } = \frac{\text{control mean ulcer index} - \text{test mean ulcer index}}{\text{Control mean ulcer indexdx}} \times 100$$

4.9 Experimental groups

In the present study following groups of animals will be designed and each group contains six rats.

Group-I: Control (pylorus-ligation) given only vehicle.

Group-II: Pylorus ligation + standard drug (Ranitidine: 10mg/kg), i.p.

Group-III: Pylorus ligation + plant extract (150mg/kg), p.o.

Group-IV: Pylorus ligation + plant extract (300mg/kg), p.o.

4.10 Treatment schedule

The dosing of schedule of fruit extract of *Momordica charantia* and ranitidine administered in the different groups of animal for 10 days continuously after 10 days ant-ulcer activity in rats are examined/ observe by pylorus ligation method.

5. OBSERVATION AND RESULT

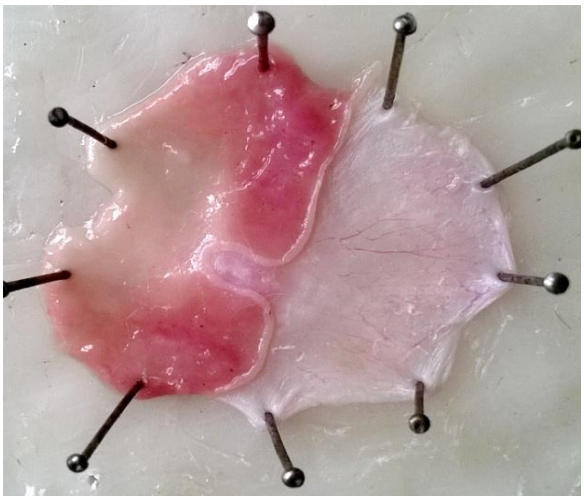
In pylorus ligation model, the fruit extract of *Momordica charantia* 150mg/kg and 300mg/kg treated animals has shown significant reduction in ulcer index, free acid, total acid, ($p<0.0001$) also it increases the pH ($p<0.0001$) the percentage of ulcer protection at low dose 150mg/kg (50.21%) and at high dose 300mg/kg (65.66%), the standard drug treatment with ranitidine (10mg/kg. i.p.) also showed significant reduction in ulcer index, free acid, total acid it also increases in pH and percentage of protection (74.24%), when compared with control group.

High dose of *Momordica charantia* fruit extract (300mg/kg) more active than low dose (150mg/kg) but less active than standard drug ranitidine (10mg/kg).

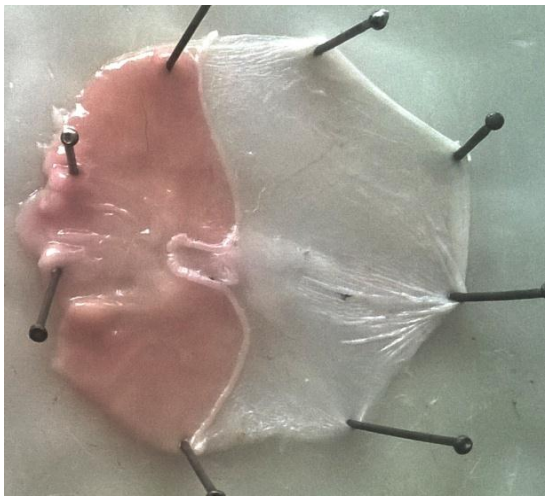
Table: 5. Pylorus ligation induced gastric ulcer in rats.

Group (n=6)	Dose (mg/kg)	Gastric pH	Free Acidity	Total Acidity	Ulcer Index	% Protec-tion of Ulcer
Control (normal saline)	(normal saline)	3.41±0.14	31.16±0.45	37.50±0.50	2.33±0.12	
Standard (Ranitidine)	10mg/kg	6.65±0.21	21.50±0.37	26.83±0.42	0.60±0.06	74.24%
Plant extract (low dose)	150mg/kg	5.10±0.18	28.50±0.43	33.60±0.47	1.16±0.08	50.21%
Plant extract (high dose)	300mg/kg	6.10±0.20	25.10±0.40	29.30±0.44	0.80±0.07	65.66%

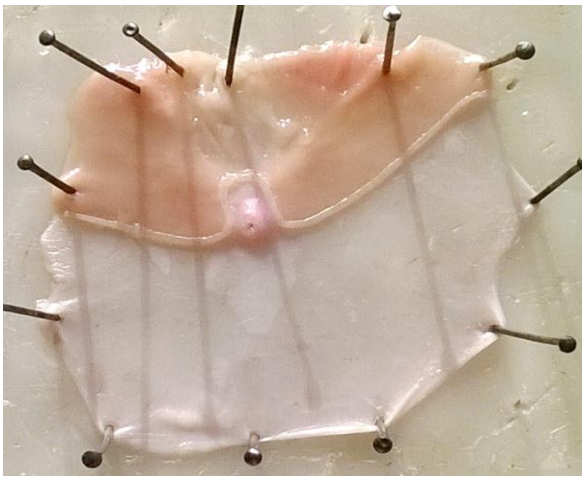
(Effect of *Momordica charantia* fruit on gastric pH, free and total acidity, ulcer index and percentage protection of ulcer of in pylorus ligation (Mean ± SEM).



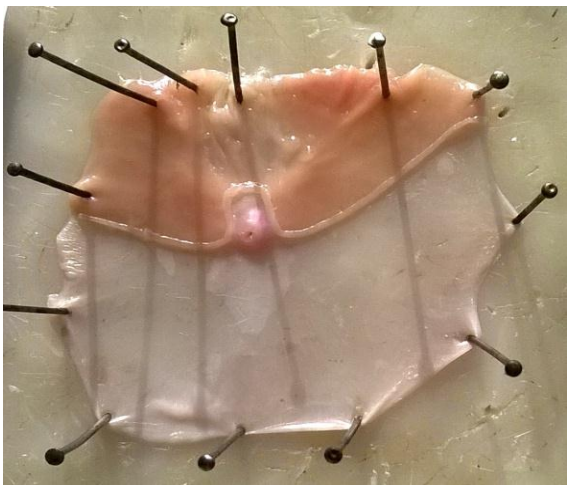
Control group I



Plant extract150mg/kg III

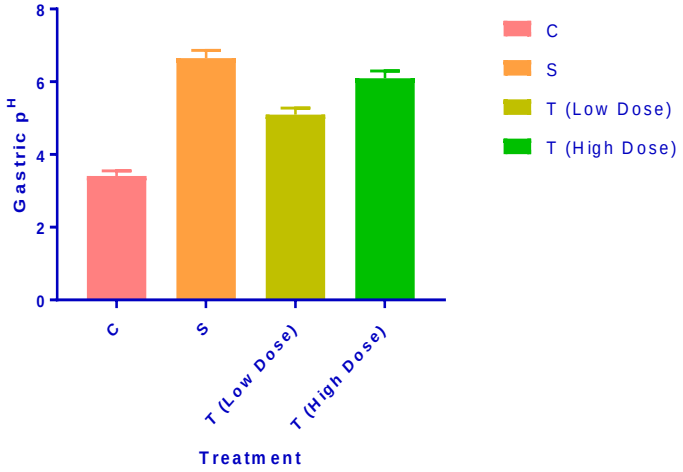


Ranitidine treated II

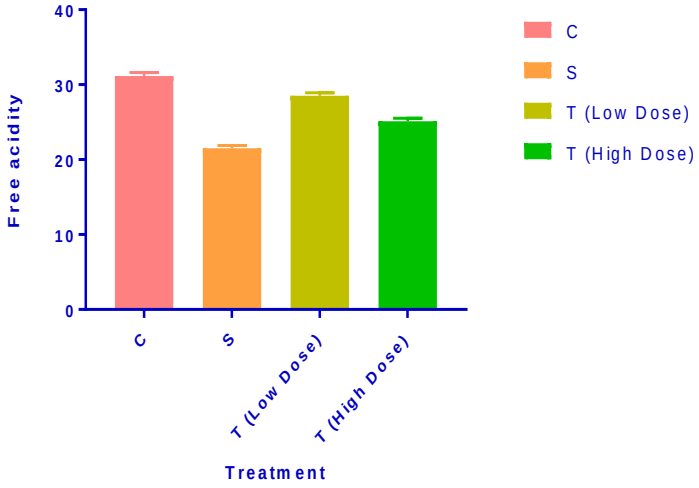


Plant extract 300mg/kg I

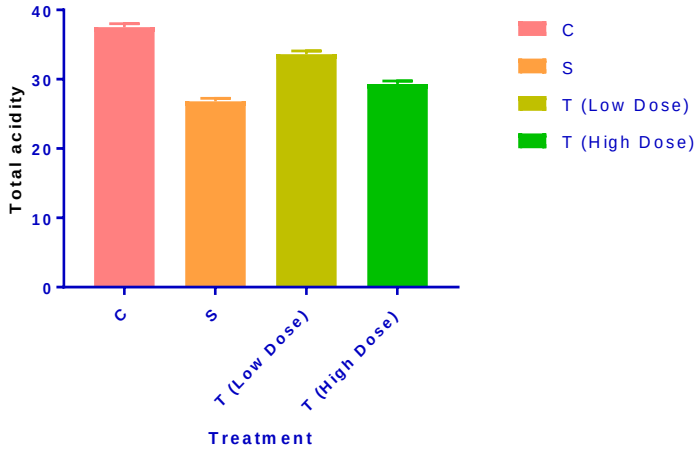
Fig. 5: Pylorus ligation induced model.



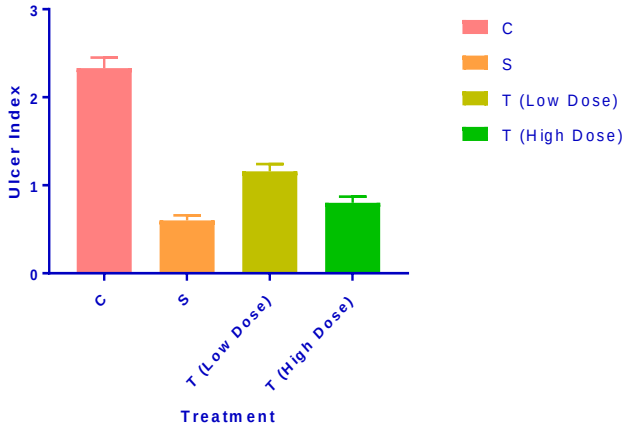
Effect of *momordica charantia linn.* On gastric pH.



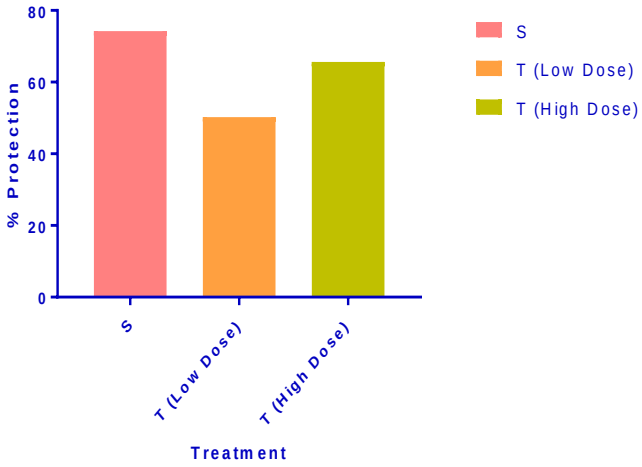
Effect of *momordica charantia Linn.* On free acidity.



Effect of *momordica charantia linn.* On total acidity.



Effect of *momordica charantia linn.* On ulcer index.



Effect of *momordica charantia Linn.* On percentage protection.

6. DISCUSSION

In spite of tremendous development in the field of synthetic drugs during recent era, there are found to have various side effect, Whereas plant products having no side effect. Peptic ulcer disease is a chronic inflammatory disease characterized by ulceration in the upper gastrointestinal tract. The pathogenesis of ulcers is due to an imbalance between aggressive factors (acid, pepsin, *H. pylori* and NSAIDs) and local mucosal defensive factors (mucous, bicarbonate, blood flow and prostaglandin). The integrity of gastro duodenal mucosa is maintained through a hemostatic balance between these aggressive and defensive factor. The major cause of gastric ulcer is the chronic use of NSAIDs. Therapeutic and adverse effects of NSAIDs have been attributing to the ability of these drugs to inhibit the action of cyclooxygenase (COX). COX is responsible for the synthesis of the prostaglandin that normally inhibits acid secretion, as well as having a protective effect on gastric mucosa.

Infection of the stomach mucosa with *H. pylori* a gram negative spiral shaped bacterium is now generally considered as the major cause of gastro intestinal ulcers. Treatment includes H_2-receptor antagonist, proton pump inhibitors and cytoprotective antacid like aluminium hydroxide and magnesium hydroxide are used often to neutralize excess gastric acidity in the stomach. Due to problem associated with recurrence after treatment, there is a need to seek an alternative drug against gastrointestinal ulcer. The present investigation demonstrated the efficacy *Momordica charantia* of plant extract against gastric ulceration induced by experimental models viz. Pylorus ligation induced gastric ulceration.

In pylorus ligation model, the plant extract of *Momordica charantia* produces a decrease in the ulcer number, total gastric volume, total acid and free acid and increases the pH and percentage of ulcer protection at low dose 150mg/kg (50.21%) and at high dose 300mg/kg (65.66%) and in the standard drug (Ranitidine) treated animals the percentage of ulcer protection (74.24%) when compared with control group (0%). High dose of extract (300mg/kg) more active than low dose (150mg/kg) but less active than standard drug (Ranitidine).

The anti-ulcer property of *Momordica charantia* in the experimental models explained above is due to presence of flavonoids, terpenoids, tannins, and triterpenes. The triterpenes are known as anti-ulcer and their action has been mentioned to be due to activation cellular protein, reduction of mucosal prostaglandin metabolism, cytoprotective action and reduction of gastric vascular permeability and remaining compounds have been shown to scavenger free radicals. The result in the present study seems to provide support for the use of *Momordica charantia* as an anti-ulcer drug in folk medicine. Therefore, also in view of its large use in India further investigations are needed to know about its anti-ulcer activity in human beings.

7. CONCLUSION

The present study indicates that the plant *Momordica charantia* has potential anti-ulcer activity against pylorus ligation induced ulcer in experimental animals. This activity of plant probably due to the presence of compounds such as Flavonoids, triterpenes, tannins, and terpenoids. The mechanism of action triterpenes are as anti-ulcer agents due to activation of cellular proteins, reduction of mucosal prostaglandin metabolism cytoprotective action and reduction of gastric vascular permeability. The results in the present study seem to provide support for the use of *Momordica charantia* as an anti-ulcer drug in folk medicine. So the

plant *Momordica charantia* used for both Ayurvedic and modern drug development areas because of its phyto-medicinal uses but it needs further clinical trials before complete trust and usage.Fruit extract of *Momordica charantia* show lesser side effect is compare to the standard drug (Ranitidine).

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