



## Anti-ulcer activity of *Mimosa pudica* on Experimental animal models

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### ABSTRACT

*Mimosa pudica* is a species belonging to the Mimosaceae family. This plant is popularly known as Touch-me-not and is widely grown as weed in tropical crops particularly when fields are hand cultivated. This species has many uses in folk medicine for example the leaf preparations were used as traditional herbal medicine for the treatment of ulcers, fever and burning sensation. The main objective of present work is to investigate the anti ulcer activity of the leaves of *Mimosa pudica*. The effect of aqueous and methanolic extracts of *Mimosa pudica* was investigated in rats by using two models, i. e. pylorus ligation and indomethacin induced ulcer models. The PH, total acidity and ulcer index were determined. The results indicated that the alcoholic extract significantly ( $P < 0.001$ ) decreases the PH, total acidity and ulcer index with respect to control at dose 400mg/kg p. o.

**Keywords:** *Mimosa pudica*; anti-ulcer; pylorus ligation; Indomethacin.

### INTRODUCTION

*Mimosa pudica* (Mimosaceae) known as chue Mue, is a stout straggling prostrate shrubby plant with the compound leaves sensitive to touch. It has spinous stipules and globose pinkish flower heads and grows as weed in almost all parts of the country. Leaves and stem of the plant have been reported to contain an alkaloid mimosine. Leaves also contain mucilage and the root contains tannins (Ghani A, 1998).

Leaves and stem of the plant have been reported to contain an alkaloid mimosine, leaves also contain mucilage and root contains tannins. Leaves and roots are used in treatment of piles and fistula. Paste of leaves is applied to hydrocele. Cotton impregnated with juice of leaves is used for dressing sinus. In Ayurvedic and Unani system of medicine, this plant has been used in diseases arising from corrupted blood and bile, billious fever, piles, jaundice, leprosy, ulcers, small pox (Kirthikar).

*Mimosa pudica* is used for its hepatoprotective (Gauri Karwani, 2011), hypolipidemic (rekha, 2010), Anti fertility (Maysumi Ganguly, 2007), Anti hepato toxic (Nazeema T. H, 2009) Anti convulsant (Ngo Bum, 2004), Anti depressant (Molina, 1999), Wound healing (Dnyaneshwar, 2009) properties.

This plant is found to possess polyphenolic constituents like flavonoids, Quercetin, Naringin, Saponins, glycosides, tannins, gums and mucilages. Hence in the

present study, the *Mimosa pudica* plant has been selected to investigate the anti-ulcer study.

### MATERIALS AND METHODS

The fresh leaves of *Mimosa pudica* were collected from GKVK, Agricultural University, Bangalore and were authenticated by the taxonomist Dr. Rajanna, GKVK, Bangalore. The leaves of *Mimosa pudica* were shade dried and coarsely powdered. The coarse powder 63gms was subjected to aqueous and alcoholic extraction by Soxhlet apparatus and extracts were concentrated using rotary evaporator under reduced pressure. Then extract was stored in a refrigerator at 4°C until use for biological testing and phytochemical screening.

#### Preliminary phytochemical screening

The aqueous and alcoholic extracts of *Mimosa pudica* was screened for the presence of various phytoconstituents (Harborne 1998).

#### Animals

Healthy wistar rats of either sex and weighing 180-200g were acclimatized to the laboratory at a temperature (25±1°C), relative humidity (15±15%), 12hr light-dark cycles, kept in standard polypropylene cages of maximum 5 animals each and given standard diet (Kamadhenu Enterprises, Bangalore) and water ad-libitum in accordance with the instructions given by Institutional Animal Ethical Committee, CPCSEA (Dept. of Health and Human services).

#### Acute oral toxicity

Aqueous and alcoholic extracts of *Mimosa pudica* was studied for the acute oral toxicity according to the guidelines set by OECD (Organization for Economic cooperation and development) guidelines No. 423

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The acute toxicity of aqueous and alcoholic extracts of leaves of *Mimosa pudica* were determined by using female albino mice (18 -22g) under standard husbandry conditions.

The two doses of 1000mg/kg (p. o) and 2000mg/kg (p. o) of the test samples were given to two groups with 5 animals in each group. The mortality and general behavior of treated groups were monitored for 14 days. The extract was devoid of any toxicity in rats when the dose up to 2000mg/kg was given orally. Hence for further studies 200-400mg/kg doses of extract were selected.

#### Evaluation of anti ulcer activity

Two animal models (Indomethacin and pylorus ligation) were employed to evaluate the anti ulcer activity of *Mimosa pudica* leaf extract.

#### Experimental design and treatment schedule

##### Indomethacin induced ulcer model

Healthy female wistar albino rats of weighing between 18-22gms were taken for the studies. The animals were divided in to seven groups (each contain 6 animals) as follows

Group I (vehicle): Distilled water (10ml/kg)

Group II (control): Indomethacin (25mg/kg b. w)

Group III (Standard): Omeprazole (10mg/kg)

Group IV: Aqueous extract (200mg/kg)

Group V: Aqueous extract (400mg/kg)

Group VI: Alcoholic extract (200mg/kg)

Group VII: Alcoholic extract (400mg/kg)

The animals in all the groups were fasted for 24h before the test substance administration but had free access to water. The test drugs were administered orally in 0.1% Tween 80 solution 10 min prior to oral indomethacin in a dose of 25 mg/kg (Hakan, 2009). Six hours later, the rats were sacrificed by cervical dislocation and their stomachs were removed. Stomach was cut along the greater curvature and observed for ulcers and its content drained into a graduated centrifuge tube and centrifuged at 3000 rpm for 10 min. The volume of the gastric content was measured. The pH of the solution was noted using pH strips (Ashoka, 2009).

##### Pylorus ligation induced ulcer

Forty two rats of either sex were divided in to seven groups each consists of six animals. Group I served as the vehicle control (10 ml/kg orally); Group II served as the pylorus ligation control (4 h); Group III served as standard control (Omeprazole – 10 mg/kg); Group IV and V – ALMP (at the dose levels of 200 and 400 mg/kg b. w); and Group VI and VII – AQMP (at the dose levels of 200 and 400 mg/kg b. w); were administered orally.

The animals were fasted for 24 h before the test substance administration but had free access to water.

After 1 hr administration of test doses, under light ether anesthesia, abdomen was opened by a small midline incision below the xiphoid process. Pylorus ligation was done without causing any damage to the blood supply of the stomach. The stomach was placed inside carefully and the abdominal wall was closed by interrupted sutures. Animals were allowed to recover and stabilize in individual cage and were deprived of water during post-operative period. Four hours later, the animals were sacrificed, the abdomen was opened and another ligature placed around the esophagus close to the diaphragm. The stomach was removed, and its content drained into a graduated centrifuge tube and centrifuged at 3000 rpm for 10 min and stomach was cut along the greater curvature and observed for ulcers. Volume was noted. Pipetted 1 ml of supernatant liquid and diluted it to 10 ml with distilled water. The volume of the gastric content was measured. Acidity was determined by titration with 0.1 N NaOH using phenolphthalein indicator. The pH of the solution was noted using pH strips (Kulkarni SK. 1987).

#### Scoring of ulcer

0-Normal coloured stomach, 0.5-red coloration, 1.0-spot ulcers, 1.5-hemorrhagic streaks, 2.0-ulcers more than 3 mm and less than 5 mm, 3.0-ulcers more than 5 mm. Mean ulcer scores for each experimental group were calculated and expressed as the ulcer index (UI).

#### Calculation of ulcer index

$$U_1 = U_N + U_S + U_P + 10^{-1}$$

$U_1$  = Ulcer index

$U_N$  = Average number of ulcer per animal

$U_S$  = Average of severity score

$U_P$  = Percentage of animal with ulcer

#### Determination of acidity

$$\text{Acidity} = \frac{\text{volume of NaOH} \times \text{Normality of NaOH} \times 100 \text{ mEq/litre}}{0.1}$$

#### Determination of percentage protection

$$\% \text{ protection} = \frac{\text{Control mean ulcer index} - \text{Test mean ulcer index}}{\text{Control mean ulcer index}}$$

#### Statistical analysis

The results are expressed as mean  $\pm$  SEM. Statistical difference was tested by using one-way analysis of variance (ANOVA) followed by Dunnetts test. A difference in the mean P value < 0.05 was considered as significance.

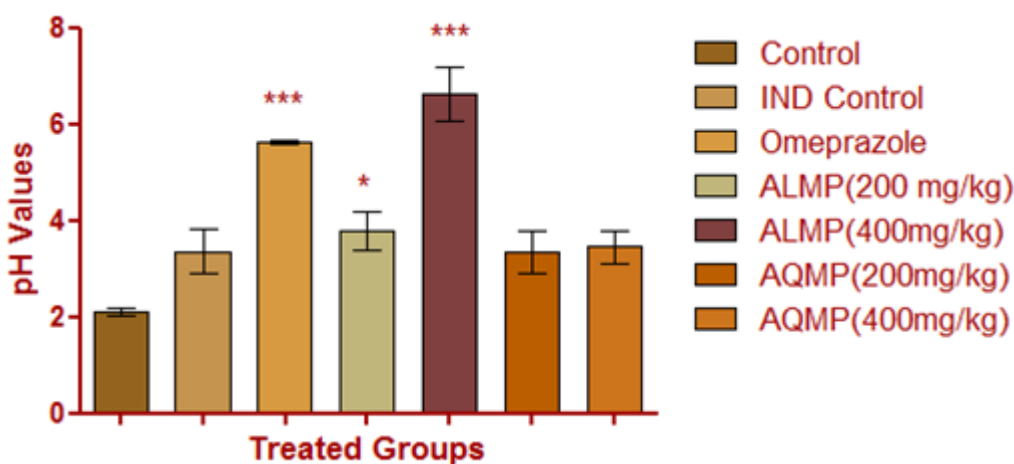
## RESULT AND DISCUSSION

### Preliminary phytochemical screening

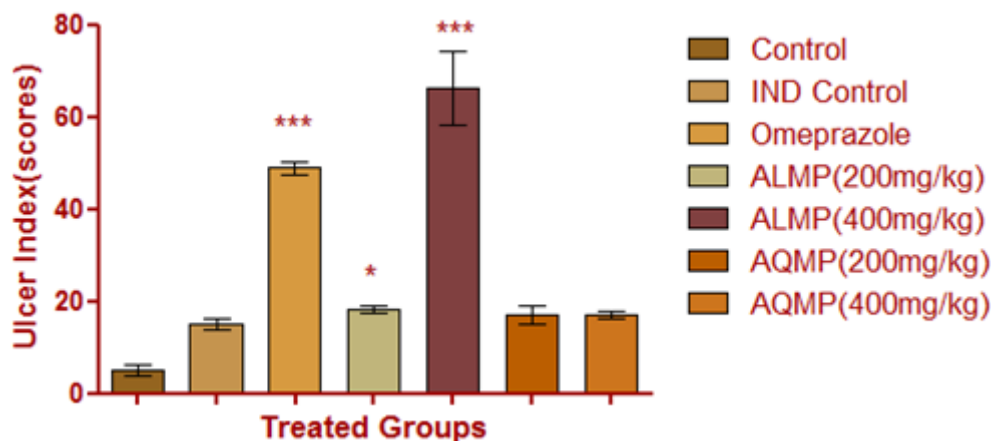
The percentage yield of AQMP and ALMP was 14.61 %w/w and 11.20% w/w respectively. These extracts

**Table 1: Anti ulcer activity of ALMP and AQMP in Pylorus ligation induced ulcer model**

Groups	TREATMENT	Mean pH	SEM ±	MEAN TOTAL ACIDITY	SEM ±	MEAN ULCER INDEX
I	Vehicle control (10ml/kg)	2.667	0.43	0	0	0.825
II	Pylorus ligation control (4h)	3.683	0.78	37.02	8.581	3.69
III	Omeprazole 10mg/kg	9.367***	0.426	50.35***	6.364	2.912***
IV	ALMP (200mg/kg)	7.69**	0.798	91.86**	12.01	2.628***
V	ALMP (400mg/kg)	6.63***	0.704	66.25***	8.056	3.600***
VI	AQMP (200mg/kg)	6.88	1.125	13.90	0.47	2.733
VII	AQMP (400mg/kg)	4.418	0.867	22.51	2.871	4.463

**Figure 1: pH values of treated groups in indomethacin induced ulcer model using albino wistar rats**

Values are expressed as mean  $\pm$ S.E.M. n=6. Significant values were compared with \* $p < 0.05$ , \*\* $p < 0.01$ \*\*\* and  $p < 0.001$  control versus treated groups using one way ANOVA followed by Dunnett's test.

**Figure 2: Ulcer index of treated groups in indomethacin induced ulcer model using albino wistar rats**

Values are expressed as mean  $\pm$ S.E.M. n=6. Significant values were compared with \* $p < 0.05$ , \*\* $p < 0.01$ \*\*\* and  $p < 0.001$  control versus treated groups using one way ANOVA followed by Dunnett's test.

contained alkaloids, carbohydrates, glycosides, saponins, phytosterols, phenolic compounds, tannins, flavonoids, proteins and amino acids.

#### Acute toxicity study

The acute oral toxicity test showed the normal behavior of the treated rats. No toxic effects were observed at a higher dose of 5 g/kg body weight. Hence there were no lethal effects, which indicated that it may have

a reasonable safety margin with regards to acute toxicity.

#### DISCUSSION

Gastric hyperacidity and gastro duodenal ulcer is a very common global problem today. It is now believed that the peptic ulcers results from an imbalance between defensive (cytoprotective) and offensive factors (gastric acid). Major aggressive factors are acid, pepsin, helicobacter pylori and bile salts. Defensive factors

mainly involve mucous bicarbonate secretion and prostaglandins.

Flavonoids are phenolic compounds found in many green plants. A wide variety of pharmacodynamics effects such as anti inflammatory, antimicrobial, antiallergic, hepatoprotective, antithrombotic and antineoplastic activities have been attributed to flavonoids. Flavonoids are known to inhibit several enzymes e. g. alkaline phosphatase, cAMP phosphodiesterase, lipases, hydrolases, lysosomal H<sup>+</sup>-ATPase and Na<sup>+</sup>/K<sup>+</sup>-ATPase. Since flavonoids antagonize aggressive factors which play a crucial role in the pathogenesis of gastric lesions and also gastric augment defensive factors to protect the gastric mucosa from injury. Flavonoids decrease histamine secretion from mast cells by inhibition of histidine decarboxylase and stimulate prostaglandin biosynthesis, it was postulated that these mechanisms of action may be responsible for the anti-ulcer activity of flavonoids. All flavonoids blocked acid formation in parietal cells in response to histamine. H<sup>+</sup>, K<sup>+</sup>ATPase the gastric proton pumps are also inhibited by flavonoids. These findings indicate that the site of action for the inhibition of acid formation by parietal cells is the H<sup>+</sup>/K<sup>+</sup>-ATPase, the gastric proton pump are also inhibited by all flavonoids. These findings indicate that the site of action for inhibition of acid formation by parietal cells is the H<sup>+</sup>/K<sup>+</sup>-ATPase. Flavone and flavonone were effective to stimulate PGE<sub>2</sub> production in gastric mucosal cells. Exogenous prostaglandins particularly E series protects gastro intestinal mucosa from the damage induced by a wide range of irritants and there is evidence that endogenous prostaglandins are important in maintaining gastro duodenal integrity (Bafna P A).

The result of our study on *Mimosa pudica* revealed that, the ALMP contains alkaloids, carbohydrates, glycosides, saponins, flavonoids, tannins, and phenolic compounds. AQMP contained carbohydrates, glycosides, saponins, flavonoids, tannins and phenolic compounds.

The concentration of flavonoids in *Mimosa pudica*, an important medication for the treatment of this disease. This is because flavonoids possess anti oxidant properties, antihistaminic and anticholinergic activity which plays a major role in repairing the gastric damage.

The anti-ulcer effect observed in the present study may be due to a possible relationship between the protection of mucosal injury, inhibition of acid secretion and the antioxidant nature of *Mimosa pudica*. Aqueous extracts of *Mimosa pudica* have not showed any activity against gastric ulcers in both the models. The alcoholic extracts of *Mimosa pudica* presents anti secretory and cytoprotective mechanism. This study indicates that *Mimosa pudica* extract has a potential anti ulcer activity. However further study is required to isolate the active molecule responsible for the activity.

## CONFLICT OF INTEREST

We declare that we have no conflict of Interest.

## ACKNOWLEDGEMENTS

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