EVALUATION OF ANTIULCEROGENIC EFFECT OF ALCOHOLIC EXTRACT OF
MAYTENUS EMARGINATA (WILLD.) DING HOU LEAVES

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ABSTRACT
Maytenus emarginata (Willd.) Dind Hou belongs to family Celastraceae, is an evergreen tree that tolerates various types of stresses of the desert, locally known as “Kankero”. Maytenus emarginata has been used for fever, asthma, rheumatism and gastrointestinal disorders worldwide. The effect of alcoholic extract of leaves of Maytenus emarginata was investigated in rats to evaluate the anti-ulcer activity by using aspirin induced gastric ulcer pyloric ligation model. The parameters taken to assess anti-ulcer activity were volume of gastric secretion, pH, free acidity, total acidity and ulcer index. In present study the orally administered alcoholic extract significantly (P < 0.05) increases pH and decreases the volume of gastric acid secretion, free acidity, total acidity and ulcer index in respect to control at a dose of 125 and 187 mg/kg body weight. This study lend support to the traditional use of Maytenus emarginata as antulcerogenic.

KEY WORDS: Maytenus emarginata (Willd.) Ding Hou, anti-ulcer, free acidity, pyloric ligation

INTRODUCTION
Peptic ulcer is the most common gastrointestinal disorder in clinical practice. Considering the several side effects (arrhythmia’s, impotence, and haematopoietic changes) of modern medicine, herbal drugs possessing fewer side effects should be looked as a better alternative for the treatment of peptic ulcer. There is evidence concerning the participation of reactive oxygen species in the etiology and pathophysiology of human diseases, such as neurodegenerative disorders, inflammation, viral infections, autoimmune pathologies and digestive system disorders such as gastrointestinal inflammation and gastric ulcer. The study assumes significance in the context that prolonged use of synthetic anti-ulcer drugs leads to adverse drug reactions and a search for new anti-ulcer agents that retain therapeutic efficacy and are devoid of adverse drug reaction is warranted. A study of the efficacy of an alcoholic extract of Maytenus emarginata in gastric ulcer with aspirin induced pylorus ligation model was undertaken in a rat model.

Maytenus emarginata (Willd.) Hou belongs to family Celastraceae, is an evergreen tree that tolerates various types of stresses of the desert, locally known as “Kankero”, “Baikal” in Hindi, and “Thorny staff tree” in English. Traditionally species of Maytenus has been used for fever, asthma, rheumatism and gastrointestinal disorders worldwide. Recently some biomolecules from Maytenus species has been reported to be active against HIV-Protease, Carcinoma and leukemia and MDR (Multi Drug Resistance). Roots are used in gastrointestinal troubles, especially dysentery, Stem tender shoots of the plant help for treating mouth ulcer. The bark is ground to a paste and applied with mustard oil to kill lice in the hair. Pulverized leaves of Maytenus emarginata are given with milk to children as a vermifuge. A decoction of the leafy twigs is used as a mouthwash to relieve toothache. Ash of leaves used to heal up sores and wound by giving cooling effect. The leaves are burnt and mixed with ghee to form an ointment used to heal sores. The tender leaves are chewed raw in the treatment of jaundice. Fruits are used in medicines to purify blood. In the present study Maytenus emarginata (Willd.) Ding Hou plant has been selected to investigate the anti-ulcer activity.

MATERIALS AND METHODS

Plant material
Collection of leaves of Maytenus emarginata was done personally from the rocky area of kaylana, Jodhpur (Raj.) in the month of June-September 2010. Taxonomical identification and authentication of the plant (Herbarium voucher no. BSI/AZ/RC/A.19014/SE-1/Estt.) was done at Botanical Survey of India, Arid Zone Regional Center, Jodhpur, Rajasthan, India.

Leaves of plant material were air dried at room temperature under shade and the dried parts were then ground to coarse powder with the help of suitable grinder. This powder was passed through a 40-mesh sieve to get a uniform particle size for further study. The powdered plant material was then kept in airtight polythene bags and stored in cool and dark place to avoid deterioration by elevated temperature, light and moisture.

Extract Preparation
A weighed quantity (500 g) of the powder was then subjected to continuous hot extraction in Soxhlet apparatus with 1 liter ethanol after defatting with petroleum ether and the residual marc was collected. The extract was filtered through a cotton plug, followed by Whatman filter paper (no.1). The extract was evaporated under reduced pressure using a rota evaporator at a low temperature (40-60°C) until all the solvent had been removed to give an extract sample with a yield of 2.9% w/w in relation to the dried starting material.
Preliminary Phytochemical Analysis
Alcoholic extract of *Maytenus emarginata* was then subjected to preliminary phytochemical analysis to assess the presence of various phytoconstituents, it revealed the presence of alkaloids, steroids, flavonoids and tannins. Preliminary Thin layer chromatography studies also confirmed these constituents.

Animals
Wistar albino rats weighing 150-200g of either sex maintained under standard husbandry conditions (temp 23±2°C, relative humidity 55 ±10% and 12 hours light dark cycle) were used for the screening. Animals were fed with standard laboratory food and water during the study period. The experimental protocol has been approved by Institutional Animal Ethics Committee (Reg. No. 541/02/c/CPCSEA), Lachoo Memorial College of Sci. and Tech., Jodhpur, India.

Aspirin Induced Pylorus Ligation Model
Either sex of albino rats weighing 150-200 gm were selected for modified pyloric ligation ulcer model. Rats were divided into five groups, each group consisting of six animals. Animals were fasted for 24 h before experiment. First group received normal saline 2 ml/kg i.p. (negative control), the second group received Ranitidine 100 mg/kg body wt. by oral route (positive control) and the third, fourth and fifth groups received alcoholic extract *Maytenus emarginata* (62.5, 125.0, 187.0 mg/kg body wt. respectively) by oral route. All groups were given aspirin orally (200 mg/kg b.w.) 3 hour prior to this treatment. Treatment was continued for 3 days and pylorus was ligated on fourth day under ether anaesthesia. The abdomen was closed and the animals were left to recover. Animals were sacrificed 4 h later with over dose of ether and the stomach was opened to collect the gastric contents. The total volume of gastric content was measured.

Table 1: Effect of alcoholic extract of *Maytenus emarginata* (Willd.) Ding Hou against pylorus ligated aspirin induced gastric ulcer in rats

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg)</th>
<th>pH</th>
<th>Vol. of gastric juice (ml)</th>
<th>Free acidity (mEq/L)</th>
<th>Total acidity (mEq/L)</th>
<th>Ulcer index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Normal saline)</td>
<td>2 ml/kg b. w.</td>
<td>3.9 ±0.341</td>
<td>2.26 ±0.298</td>
<td>27.03 ±0.882</td>
<td>70.06 ±2.481</td>
<td>6.16 ±2.460</td>
</tr>
<tr>
<td>Standard (Ranitidine)</td>
<td>100 mg/kg b. w.</td>
<td>5.51 ±0.367</td>
<td>2.03 ±0.188</td>
<td>10.33 ±0.442</td>
<td>22.26 ±1.241</td>
<td>2.583 ±0.533</td>
</tr>
<tr>
<td>Alcoholic extract</td>
<td>62.5 mg/kg b. w.</td>
<td>5.0 ±0.645</td>
<td>2.56 ±0.567</td>
<td>28.03 ±0.626</td>
<td>33.31 ±0.470</td>
<td>6.16 ±2.460</td>
</tr>
<tr>
<td></td>
<td>125 mg/kg b. w.</td>
<td>4.16 ±0.812</td>
<td>2.08 ±0.273</td>
<td>14.0 ±1.205</td>
<td>22.93 ±1.903</td>
<td>1.91 ±0.786</td>
</tr>
<tr>
<td></td>
<td>187 mg/kg b. w.</td>
<td>4.83 ±0.745</td>
<td>1.81 ±0.389</td>
<td>24.16 ±1.610</td>
<td>37.48 ±1.374</td>
<td>0.75 ±0.629</td>
</tr>
</tbody>
</table>

All values are expressed as mean ± S.E.M. (n = 6). Statistical comparison was performed by using ANOVA coupled with Dunnet’ t test. P<0.05 were consider statistically significant as compared with control group.

Figure 1: Effect of alcoholic extract of *Maytenus emarginata* (Willd.) Ding Hou against pylorus ligated aspirin induced gastric ulcer in rats
(a) Stomach of control rat; (b) standard drug treated; (c) Effect of alcoholic extract (dose -62.5 mg/kg b.w.) on aspirin induced gastric ulcer in rat; (d) Effect of alcoholic extract (dose-125.0 mg/kg b.w.) on aspirin induced gastric ulcer in rat; (e) Effect of alcoholic extract (dose-187.0 mg/kg b.w.) on aspirin induced gastric ulcer in rat.


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The gastric contents were centrifuged at 5000 rpm for 5 min. One ml of the supernatent fluid was pipetted out and diluted to 10 ml with distilled water. The solution was titrated against 0.01N NaOH using Topfer’s reagent as indicator, to the endpoint when the solution turned to orange colour. The volume of NaOH needed was taken as corresponding to the free acidity. Titration was further continued till the solution regained pink colour\textsuperscript{14}. The volume of NaOH required was noted and was taken as corresponding to the total acidity (Table 1).

Acidity was expressed as:

\[ \text{Acidity} = \frac{\text{Volume of NaOH} \times \text{Normality of NaOH} \times 100 \text{ mEq/l}}{0.01} \]

**Ulcer Index (UI)**

The mucosa was flushed with saline and stomach was pinned on frog board. The lesion in glandular portion was examined using 10 X magnifying glass and length was measured using a divider and scale and gastric ulcer was scored [0 - Normal coloured stomach; 0.5 - Red colouration; 1.5 – Hemorrhagic streak; 2 – Ulcers; 3 – Perforations]. Ulcer index of each animal was calculated by adding the values and their mean values were determined\textsuperscript{16} (Table 1).

**Statistical analysis**

All data are represented as mean± SEM. Statistical analysis was performed using one-way ANOVA\textsuperscript{15} followed by Dunnet’s test \textsuperscript{16} and P<0.5 was considered significant when compared to control.

**RESULTS**

Preliminary phytochemical screening revealed the presence of alkaloids, steroids, flavonoids and tannins. In Pylorus ligation induced ulcer model alcoholic extract of the *Maytenus emarginata* (Willd.) Ding Hou in the doses of 125 and 187 mg/kg body wt. produced a reduction in the ulcer index, gastric volume, free acidity, total acidity and raised gastric pH significantly in comparison with control group (Table1). The control animals had ulcers and haemorrhagic streaks, whereas in animals administered with the extracts of *Maytenus emarginata* showed significant reduction in ulcer index (P < 0.05) (Figure 1).

**DISCUSSION**

The alcoholic extract of leaves of *Maytenus emarginata* (Willd.) Ding Hou has significantly protected the gastric mucosa against aspirin induced pylorus ligated model as shown by reduced values of ulcer index as compared to control group suggesting its potent cytoprotective effect. In aspirin induced pylorus ligation, the digestive effect of accumulated gastric juice, interference of gastric blood circulation and aspirin are responsible for induction of ulceration\textsuperscript{17}. It is generally accepted that gastric ulcers result from an imbalance between aggressor factors and the maintenance of the mucosal integrity through endogenous defence mechanisms. It is suggested that alcoholic extract of leaves of *Maytenus emarginata* suppress gastric damage induced by aggressive factors. The preliminary phytochemical studies revealed the presence of flavonoids and alkaloids in alcoholic extract of leaves of *Maytenus emarginata*. Various flavonoids and alkaloids have been reported for its anti-ulcerogenic activity with good level of gastric protection\textsuperscript{18, 19} so the possible mechanism of anti ulcer activity against gastric ulcer of alcoholic extract of leaves of *Maytenus emarginata* may be due to its flavonoid or alkaloid content.

**REFERENCES**


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