Antiulcer Activity Of Methanolic Extract Of Physalis minima Leaves

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Abstract: The anti-ulcer activity of methanolic extract of Physalis minima leaves was investigated on ethanol induced ulcer models and pylorus ligation in wistar rats. In both models the common parameter determined was ulcer-index. methanolic extract of dosage 100, 200mg/kg.p.o produced significant inhibition of gastric lesions induced by pylorus ligation and ethanol induced ulcers. The extract 100mg/kg and 200mg/kg showed significant (p<0.01) reduction in gastric volume, free acidity and ulcer index as compared to control. The present study indicates that Physalis minima leaves extract have potential anti ulcer activity in both models. This results may further suggests that methanolic extract was found to posses anti-ulcerogenic as well as ulcer healing property, which might be anti-secretory activity.

Key words: Physalis minima, antiulcer, methanol, pylorus ligation.

INTRODUCTION

Peptic ulcer disease is a serious gastrointestinal disorder that requires a well targeted therapeutic strategy. A number of drugs including proton pump inhibitors and H₂ receptor antagonists are available for the treatment of peptic ulcer, but clinical evaluation of these drugs has shown incidence of relapses, side effects and drug interactions(1). This has been the rationale for the development of new antiulcer drugs and the search for novel molecules has been extended to herbal drugs that offer better protection and decreased relapse. Drugs of plant origin are gaining popularity and are being investigated for a number of disorders including peptic ulcer. Indian Medicinal plants and their derivatives have been an invaluable source of therapeutic agents to treat various disorders including peptic ulcer. An indigenous drug possessing fewer side effects is the major thrust area of the present day research, aiming for a management of peptic ulcer. Gastric hyperacidity and ulcer are very common cause of human sufferings today (2). Although prolonged anxiety, emotional stress, hemorrhagic surgical shock, burns and trauma are known to cause severe gastric irritation, the mechanism is still poorly understood. The present work attempts to evaluate the antiulcer potential of Physalis minima leaves.

MATERIALS AND METHODS

Plant material

Physalis minima purchased from nursery of Government Siddha Medical College, Arumbakkam, Chennai, India. Its botanical identification was done by Dr. Jayaraman, Director, Plant Anatomy Research Centre, Tambaram, Chennai. The voucher specimen (281/TN/2010) was deposited at the herbarium, Department of Botany, Presidency College, Chennai, India.
Preparation of extract
The fresh leaves were shade dried, powdered and extracted (200 g) successively with 600 ml of methanol (60–80°C) in a soxhlet extractor for 18–20 hr. The extract was concentrated to dryness under reduced pressure and controlled temperature (40–50°C) to form a dark brown solid, weighing 500mg (25%w/w). Ethanol extract of Physalis minima was tested for its phytochemical screening.

Animals
Albino Wistar rats of either sex, weighing 150–200 g, were used in the study. They were kept in standard laboratory conditions under natural light and dark cycle, and are housed at ambient temperature (22±1°C), relative humidity (55±5%). Animals had access to standard pellet diet and water given ad libitum. IAEC/137/2010.

EXPERIMENTAL PROCEDURES
Ethanol-induced ulcer model
Swiss albino mice of either sex were divided into five groups, each group consists of six animals. All groups of animals received following treatments for 5 days. Group-1 (Normal) and 2(Control) received vehicle 10 ml/kg, groups 3 and 4 (Test) were given 100 and 200 mg/kg, respectively. On the 5th day, 1hr after final dose of treatment, the gastric ulcers were induced in rats by administering 96% ethanol (5ml/kg). After 1hr animals were sacrificed by cervical dislocation and stomach was incised along the greater curvature and examined for ulcers index. Percentage ulcer inhibition was calculated for each group on comparison with vehicle control group.

Pylorus ligation model
Wister albino rats of either sex were divided into five groups, each group consists of six animals. All groups of animals received following treatments for 5 days. Groups-1 (Normal) and 2 (Control) received vehicle 10 ml/kg, groups 3 and 4 (Test) were given 100 and 200 mg/kg, respectively. All the doses calculated with respective to body weights of animals and administered orally. On 5th day pylorus part was ligated following 36hr fasting. After the pre-treatment period of 1hr animals were anaesthetized using pentobarbitone (35 mg/kg, i.p.), the abdomen was opened and pylorus ligation was done without causing any damage to its blood supply. The stomach was replaced carefully and the abdomen wall was closed in two layers with interrupted sutures. After 4hr of pylorus ligation, stomachs were dissected out and cut open along the greater curvature and examined for ulcers index. The gastric juice was titrated against 0.01N sodium hydroxide using Topfer’s reagent as indicator to find out the free acidity and total acidity.

Calculation of ulcer index and Percentage ulcer inhibition
Ulcer index has been calculated by adding the total number of ulcers per stomach and the total severity of ulcers per stomach. A score for the ulcer was made as follows:

- 0: normal colored stomach.
- 0.5: red coloration.
- 1: spot ulcers.
- 1.5: haemorrhagic streak.
- 2: ulcers.
- 3: perforation.

Mean ulcer score for each animal was expressed as ulcer index. The percentage of ulcer inhibition was determined as follows:

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\text{Percentage of ulcer inhibition} = \left(\frac{\text{Control mean ulcer index} - \text{Test mean ulcer index}}{\text{Control mean ulcer index}}\right) \times 100
\]
Statistical Calculations

The data expressed are mean ± standard error of mean (SEM). All statistical comparisons between the groups are made by means of One Way Analysis of Variance (ANOVA) with post hoc Dunnett’s test or by Student’s t-test. The p value less than 0.01 is regarded as significant.

RESULTS AND DISCUSSION

Physalis minima leaf extract was found to non-toxic as it did not show any toxic symptoms and mortality up to the dose of 2000mg/kg. Effects of Physalis minima at dose of 100 and 200 mg/kg body weight, twice a day for 5 days prevented the acute gastric ulcers in a dose related manner. Administration of Physalis minima leaf extract 1hr before the induction of gastric lesions by ethanol showed significant activity (8) and inhibited the total ulcer index by percent in dose dependent manner. The oral administration of Physalis minima leaf extract at 100, 200mg/kg in pylorus ligature inhibited the total ulcer index by percent in dose dependent manner as compare to control. In pylorus ligation induced gastric ulcer the Physalis minima leaf extract showed significant reduction in gastric volume, free acidity, total acidity and ulcer score (9) (Table 1). Peptic ulcer is a common disease of modern life style due to changed food habits and ever increasing stress. The imbalance of aggressive (gastric juice, pepsin) and protective factors include mucosal blood flow, bicarbonate secretion, the secretion of mucosa integrality of cellular membrane, cell regeneration, prostaglandin and other hormones, are considered as the major mechanism (10). The allopathic drugs of ulcer inhibit the acid secretion, protect the mucosa and inhibit the Helicobacter pylori. We designed two different experimental models ethanol-induced gastric ulcer and Pylorus ligation induced gastric ulcer to investigate the effect and mechanism of Physalis minima leaf extract on gastric ulcer (11). Ulcer index parameter was used for the evaluation of anti-ulcer activity since ulcer formation is directly related to factors such as reduction in gastric volume, decrease in free and total acidity (12). Alcohol can cause the lesion of gastric mucosa, reinforcement of the aggressive factors while weakness of the protective factors, so the ulcer was formed. Studies suggest that the ethanol damage to the gastrointestinal mucosa starts with microvascular injury, namely disruption of the vascular permeability, edema formation and epithelial lifting (13). Ethanol induced gastric damage in mice possibly through leukotrienes production and also involvement of 5- lipooxygenase in the formation of ulcer lesion. Prostaglandins also play a role in ethanol-induced ulcer. So the protective effect of the Physalis minima leaf extract against the gastric damage might be due to protection against 5-lipooxygenase or leukotriene pathway. The cytoprotective action possibly stimulates the prostaglandin synthesis, which in turn is involved in cytoprotection of the gastric mucosa (14). The gastroprotective effect in ethanol model indicates that the Physalis minima leaf extract could enhance cytoprotective mechanism of the gastric mucosa. Pylorus ligation can lead to the accumulation of gastric juice in the stomach, damaging the balance of aggressive and create protective factors so ulcer is shaped (15). On Pylorus ligation-accumulated secretions and the related ulcers confirm gastric acid output to be the root cause of gastric ulcers. Physalis minima leaf extract attenuated the gastric volume free acidity, total acidity and ulcer index thus showing the anti-secretory mechanism (16). Our present study results clearly demonstrate that Physalis minima leaf extract is in possession of good preventive and therapeutic action on the gastric ulcers. It was a dose-dependent protection against gross damaging action of ethanol and Pylorus ligation on gastric mucosa of animals.

Table 1: Effect of alcoholic extract of Physalis minima leaves on gastric secretion, acidity and pH in plus pylorus ligated rats

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Volume of gastric secretion(ml/100g)</th>
<th>Free acidity (mequiv./l/100g)</th>
<th>Total acidity (mequiv./l/100 g)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group-1 (normal)</td>
<td>1.32±0.02</td>
<td>118±4.23</td>
<td>260±8.20</td>
<td>3.6±0.04</td>
</tr>
<tr>
<td>Group-2 (control)</td>
<td>2.43±0.06*</td>
<td>225±6.21*</td>
<td>504±10.50*</td>
<td>2.4±0.12*</td>
</tr>
<tr>
<td>Group-3 (Test extract 100mg/kg)</td>
<td>1.68±0.04**</td>
<td>142.5±5.12**</td>
<td>384±7.90**</td>
<td>2.98±0.07*</td>
</tr>
<tr>
<td>Group-4 (Test extract 200mg/kg)</td>
<td>1.46±0.03*</td>
<td>120.2±3.60*</td>
<td>346±7.35</td>
<td>3.21±0.06*</td>
</tr>
</tbody>
</table>

Values are expressed in mean ± SEM (n=6)
CONCLUSION

It was found that antiulcer activity exhibited was due to mucosal defensive factor hence it can be used for management of peptic ulcer. Present in the extract and experimentation on Physalis minima methanolic extract had the healing action of chronic ulcer as well as on the possible side effects. The investigation on mode of action may pave way for establishment of new anti-ulcer therapy regimen.

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