Introduction

An ulcer is a sore or lesion that forms in the lining of the stomach or duodenum where acid and pepsin are present. Ulcers in the stomach are called gastric or stomach ulcers. Those in the duodenum are called duodenal ulcers. In general, ulcers in the stomach and duodenum are referred to as peptic ulcers. Ulcers occur in the esophagus or in the first portion of the duodenum, the duodenal bulb. Ulcers can also be caused or worsened by drugs such as ethanol, aspirin, indomethacin, ibuprofen, and other NSAIDs. The phytochemical profile of this plant chrozophora plicata reveals the presence of sterols, alcohols, hydrocarbons, flavonoids, lignans, coumarins, tannins, phenanthrenes, quinones, phenolic acids, alkaloids, cyanogenic glucosides and glucosinolates. It is evident from the literature that Chrozophora plicata leaves possess flavonoids. The results obtained indicate that Chrozophora plicata possesses ulcer protective principles and flavonoids may be responsible for gastroprotective activity.

Key words: Chrozophora plicata, Flavonoids, gastroprotective activity

Materials and Methods

For this study, the leaves of chrozophora plicata were collected from the surrounding gardens of the gajwel (mandal), medak dist, Andhra Pradesh, India. After the fresh leaves were authenticated by botanist, leaf specimens have been deposited at the museum of the college. Fresh mature leaves were shade dried at room temperature, coarse powdered and extracted with chloroform by soxhlet’s extraction method. Thereafter, the extracts were concentrated using electric water bath to obtain semisolid crude extract. The percentage yields of the leaf extract were found to be 9.8% and 10.7% respectively. The extract was stored in airtight container in refrigerator below

Abstract

Safi is an herbal medicine and is said to be used as a blood purifier. The study was designed to investigate the anti-ulcer effect of chloroform extract of Chrozophora plicata leaves (600mg/kg) an essential herb in unani medicine Safi against ethanol induced gastric ulcer model in albino rats. In the present study, anti-ulcer activity was assessed by using ethanol (1 ml/200 g of 99.80% p.o) induced Ulcer models in albino rats. The gastroprotective activity was determined by comparing gastric volume, acidity, ulcer score and ulcer index in control, test extract and standard (ranitidine 10mg/kg) treated rats. An ulcer index of 1.66 ± 0.42 has been observed in chrozophora plicata treated rats, whereas standard ranitidine (10mg/kg) treated rats exhibited an ulcer index of 1.41 ± 0.30. Both extract treated and standard ranitidine treated rats has shown ulcer indices which are less than the ulcer index in control rats i.e 5.66 ± 0.80. The percentage protection of ulcers was found to be 70.67 with chloroform extract of chrozophora plicata and with standard ranitidine a percentage protection of 75.04 has been obtained. It is evident from literature that Chrozophora plicata leaves possess flavonoids. The results obtained indicate that Chrozophora plicata possesses ulcer protective principles and flavonoids may be responsible for gastroprotective activity.

Key words: Chrozophora plicata, Flavonoids, gastroprotective activity
Appropriate concentration of stock solution of extract were prepared using distilled water and used for the following studies.

**Preliminary phytochemical screening**

Preliminary phytochemical tests were performed for the chloroform extract of Chrozophora plicata to detect the presence of phytochemicals by following the standard methods described in the practical pharmacognosy of kokate and khandelwal. The results have been tabulated in table 1.

**Experimental animals**

Albino rats (180-225g) were used in the experiments. They were procured from sainath agencies, musheerabad. After randomization into various groups and before initiation of experiment, the rats were acclimatized for a period of 10 days. Animals were housed in polypropylene cages and maintained under standard environmental conditions such as temperature (26 ± 2°C), relative humidity (45-55%) and 12hr dark/light cycle. The animals were fed with rodent pellet diet (Golden Mohur Lipton India Ltd.) and water ad libitum. The study protocol was approved from the institutional animal ethics committee (IAEC) before commencement of experiment (1230/a/08/CPCSEA).

**Determination of acute toxicity**

The chrozophora plicata chloroform extract was studied for acute toxicity study at a dose of 5 mg/kg, 50mg/kg, 300 mg/kg, and 2000 mg/kg P.O in albino mice for each dose 3 mice are used (up and down procedure, OECD guidelines No. 425). The extract was found safe to all the animals 12 mice. The mice are subjected to a dose of 5000 mg/kg. Even at 5000mg/kg no mortality is seen, but few symptoms of CNS depression such as sedation and drowsiness is seen in all the mice at 5000mg/kg. Hence a dose of 3000mg/kg is selected as safer dose and 1/5th of 3000mg/kg i.e. 600mg/kg is selected for our study.

**Effect of chloroform extract of Chrozophora Plicata leaves on ethanol induced ulcers in albino rats**

The experiment was performed on albino rats (150 – 200gms) of either sex procured from Sainath agencies, Musheerabad. The animals were housed in colony cages at an ambient temperature of 26±2˚C and, relative humidity (45 - 55%), with a 12h/12h light dark cycle and free access to food and water ad libitum. Food was restricted during experiments. Ranitidine (10 mg/kg) and chloroform extract of Chrozophora plicata (600mg/kg) are prepared in 2% acacia suspension. Weigh and mark the animals. Albino rats of either sex weighing between 150-200 g were selected and divided into 3 groups of 6 animals each.

<table>
<thead>
<tr>
<th>Group I</th>
<th>Control (1 ml/200 g of 99.80% alcohol p.o.)</th>
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</thead>
<tbody>
<tr>
<td>Group II</td>
<td>Standard (Ranitidine 10 mg/kg p.o.)</td>
</tr>
<tr>
<td>Group III</td>
<td>Test extract of chrozophora plicata 600mg/kg</td>
</tr>
</tbody>
</table>

The animals were fasted for 24 hrs with free access to water. Animals were given a dose of chrozophora plicata leaf extract 600mg/kg and standard drug ranitidine (10mg/kg) as mentioned above. Thirty minutes after the treatment 1ml/200 g of 99.80% alcohol was administered p.o. to each animal. Animals were sacrificed 1 hr. after alcohol administration, stomachs were isolated and cut open along the greater curvature and pinned on a soft board. Microscopic examination was carried out by observing in 10 x magnifications and the presence of lesion was scored. 0=Normal coloured stomach, 0.5 =Red colouration, 1.0 =Spot ulcer, 1.5 = haemorrhagic streaks, 2.0 =Ulcers ≥ 3 ≤ 5, 3.0=Ulcers > 5

Mean ulcer score for each animal is represented as Ulcer index. The ulcer index was measured. The results are compiled in Table No. 2.

The percentage of ulcer protection was determined as follows:

\[
\text{Control mean ulcer index} - \frac{\text{Test mean ulcer index}}{\text{Control mean ulcer index}} \times 100
\]

**Statistical Analysis**

The values are represented as mean ± S.E.M, and statistical significance between treated and control groups was analyzed using One way ANOVA, Followed by Dunnett’s test where P<0.001, P<0.01 and P<0.05 was considered statistically significant.

**Results and Discussion**

Results of the preliminary phytochemical investigation of chloroform extract of Chrozophora plicata leaves extract are shown in table no.1. The results obtained (Table II) by using alcohol induced ulcer model in rats indicates that the ulcer score in control animals are 1.5 to 2.0 which is an indication of ulceration, whereas the chrozophora plicata extract treated animals has shown less ulcer score i.e.0.5 to 1.0 and percentage protection from ulcer is 70.67% in comparison with control animals which is a clear indication of gastroprotective activity of chrozophora plicata leaf extract (600mg/kg). But the % protection of ulcer in test treated rats is less than that of the % protection of ulcer in ranitidine treated rats (75.04%). Safi is a Unani medicine which claims to be a blood-purifier. It is produced by
Hamdard Laboratories (Waqf) in Pakistan, as well as by Hamdard (Wakf) Laboratories in Ghaziabad, India and Bangladesh. Chrozophora plicata is one of the plant which is used as an ingredient in Safi. Chrozophora plicata leaves are available abundantly throughout the Andhra Pradesh. It is evident from the literature that chrozophora plicata contains flavonoids and flavonoids have antiulcer principles. Ethanol, acting as an acute pro-inflammatory agent, is known to produce erosions, ulcerative lesions and petechial bleeding in the mucosa of the stomach in humans. Ethanol-induced gastric ulcer model is widely used to evaluate the protective activity of new anti-ulcer agents. Furthermore, from this study, it could also be suggested that chrozophora plicata act mainly by stimulating the defensive factors in the gastric mucosa and also by increasing up regulation or increase in COX1 and COX2 which inturn would lead to increase in prostaglandin synthesis. Although the exact mechanism is not clear. However, it could be concluded that Chrozophora plicata (600mg/kg) anti-ulcer property probably by increasing defensive gastric mucosa.

Table I. Results of Phytochemical investigation of chloroform extract of chrozophora plicata

<table>
<thead>
<tr>
<th>Phytoconstituents</th>
<th>Chloroform extract of chrozophora plicata</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbohydrates</td>
<td>-</td>
</tr>
<tr>
<td>Steroids</td>
<td>-</td>
</tr>
<tr>
<td>Glycosides</td>
<td>-</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+++</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>++</td>
</tr>
<tr>
<td>Tannins</td>
<td>+++</td>
</tr>
</tbody>
</table>

- Absent  ++ present  +++ present with more clarity

Table II. Anti ulcer activity of chrozophora plicata chloroform leaf extract (600mg/kg) by using Ethanol induced ulcer model

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatment</th>
<th>Dose</th>
<th>Mean ulcer index ± SEM</th>
<th>% protection</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
<td>Alcohol 1ml/200 g</td>
<td>5.66 ± 0.80</td>
<td>--</td>
</tr>
<tr>
<td>II</td>
<td>Standard (Ranitidine)</td>
<td>10 mg/kg</td>
<td>1.41 ± 0.30***</td>
<td>75.04</td>
</tr>
<tr>
<td>III</td>
<td>Chloroform extract of chrozophora plicata leaves</td>
<td>600 mg/kg</td>
<td>1.66 ± 0.42***</td>
<td>70.67</td>
</tr>
</tbody>
</table>

P<0.001**, P<0.01** and P<0.05* was considered statistically significant
**Conclusion**

The results of the present study indicates that the chloroform extract of Chrozophora plicata leaves at 600mg/kg possessed significant anti-Ulcer effect and thus supports the use of Chrozophora plicata leaves in treatment of Ulcer. Ethanol rapidly penetrates the gastric mucosa, and causes membrane damage and erosions. Chrozophora plicata leaf extract had significantly reduced ulcer index to an extent of 1.66 ± 0.42 in comparison with the control group ulcer index i.e 5.66 ± 0.80.

**Acknowledgements**

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