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PHARMACOLOGICAL SCREENING OF ETHANOLIC FLOWER EXTRACT OF *COUROUPITA GUIANENSIS* FOR ITS ANTI- ULCER ACTIVITY

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ABSTRACT

The antiulcer Potential of ethanolic flower extract of *Couroupita guianensis* (EFCG) was investigated in indomethacin plus pylorus ligation induced gastric ulcer model in experimental rats. Ethanolic flower extract of *Couroupita guianensis* at a dose of (200 and 400 mg/kg) produced significant inhibition of the gastric lesions induced by indomethacin plus pylorus ligation induced gastric ulcer. The extract (200mg/kg and 400mg/kg) showed significant ($p<0.01$) reduction in gastric volume, free acidity and ulcer index as compared to control. The present study indicates that EFCG have potential anti-ulcer activity in indomethacin plus pylorus ligation induced gastric ulcer model. These results may further suggest that the extract was found to possess anti-ulcerogenic activity which might be due to the presence of phenolic compounds.

KEYWORDS: *Couroupita guianensis* (EFCG), anti-ulcerogenic.

INTRODUCTION

Peptic ulcers disease refers to a group of disorders characterized by circumscribed lesions of the mucosa of the upper gastrointestinal tract (especially of the stomach and duodenum). The lesions occur in regions exposed to gastric juices. When the stomach's natural protections form acid stop working ulcers will occur. Duodenal ulcers almost always develop in the duodenal bulb (the first few centimeters of the duodenum. A few, however, arise between the bulb and the ampulla. Gastric ulcers form most commonly in the antrum or at the antral-fundal junction. Nearly 80% of peptic ulcers are duodenal the others are gastric ulcers. Most duodenal ulcers appear in people between ages 20 and 50 years, while gastric ulcer usually occurs between ages 45 and 55 years. Duodenal ulcer is twice as common in men as in men as in women and gastric ulcers affect men and women equally. Approximately 10 to 20% of gastric ulcer patients also have a concurrent duodenal ulcer.

India is one of the country rich in medicinal plants which were used by ancestors. Traditionally, plants were used as medicine in traditional way such as Ayurveda, Naturopathy, Siddha and Unani. After knowing the used of plants in medicine, synthetic drugs were now started replacing by herbal products.

Since prehistoric times plants have been used by humans in many countries for the treatment of many ailments and in some other useful purposes. It was continued for the past 5000 years. Useful plants and its uses have been described in books like Materia medica, Wealth of India, Indian Medicinal Plants etc. Now a days many countries are having interest in using Indian medicinal plants as it can cure many diseases and other purposes.

Couroupita guianensis known by a variety of common names including cannonball tree, is a deciduous tree in the family *Lecythidaceae*. It is native to the rain forests of central and South America. It has large interesting fruits, beautiful, fragrant flowers. Chemical constituents include Alkaloids, Glycosides, Flavonoid glycosides, Saponin, Triterpinoids, Tannins, Anthroquinine glycosides, Isatin, Couroupitine, Couroupitone. Flowers yield an aliphatic hydrocarbon, stigmasierol, phenolic and flavonoids.

MATERIALS AND METHODS

Collection and Extraction of Plant

The Plant *Couroupita guianensis* collected from Malabar in Kerala and it was authentified by Botanical Survey of India, Coimbatore, India. It was shade dried. The shade dried leaves was grinded in mixer grinder to obtain particle size coarser size. The grinded product was extracted with ethanol using Soxhlet apparatus. Evaporation of solvent was done to obtain semisolid product which was used for the studies.

Animals

The study conducted on Wistar albino rats of 150-200g and maintained under standard (room temperature 24 C - 27C and humidity 60-65%). The food in the form of dry pellets (M/s Hindustan Lever Foods, Bangalore) and water were available ad *libitum* rats of either sex were selected into groups of 6 animals each. All the experimental procedures and protocols used in this study were reviewed by the Industrial Animal Ethics Committee (688/2/C-CPCSEA) and were in accordance with the guidelines of the IAE.

Anti- ulcer activity

Albino rats weighing 150-200g were selected and divided into five groups of six animals each. Animals were abstained for 24 hours before the study, but had the access to water.

Group I- Vehicle control (0.1% CMC)
Group II- Ulcer control, Indomethacin (20mg/kg)
Group III- Ulcer control + Ranitidine (50mg/kg)
Group IV- Ulcer control + EFCG (200mg/kg)
Group V- Ulcer control + EFCG (400mg/kg)

Gastric lesions were induced to all groups of rats by oral administration of indomethacin 20 mg/kg suspension in 0.1% CMC for five consecutive days followed by pylorus ligation under pentobarbitone 45 mg/kg i.p anaesthesia on 6th day. The standard drug ranitidine 50 mg/kg was administered immediately after oral administration for five consecutive days. In the same manner, test compounds (EFCG dose 200 mg/kg and 400 mg/kg p.o) were administered after oral administration for five consecutive days. The rats were sacrificed after four hours of pylorus ligation and stomach was removed after clamping the esophagus. Gastric contents were collected and then assessed for the ulcer index (UI), free acidity and total acidity, gastric pH, and gastric volume.

Determination of Ulcer index

Ulcer index was resolute according to the score method. The lesions of each stomach were used for the calculation of ulcer index and the ulcer protection percentage was calculated from the following formula.

Table 1: Ulcer Score and Descriptive Observation.

Ulcer Score	Descriptive Observation
1	Less than 1mm (Pin point)
2	1-2 mm
3	Greater than 2 mm and above

The ulcer score was divided by a factor of 10 to get the ulcer index. % Ulcer protection was calculated using Ulcer index values.

$$\frac{\text{Ulcer Index in Control} - \text{Ulcer index in Test}}{\text{Ulcer Index in Control}} \times 100$$

Determination of Gastric Volume

The gastric juice was collected and poured into the measuring cylinder.

Determination of gastric pH

The gastric content was collected earlier and used for the determination of pH by using a digital pH meter.

Determination of Free Acidity and Total Acidity

1 ml of gastric juice was pipette out into a 100 ml conical flask. 3 drops of Topfer's reagent was added and titrated with 0.01N sodium hydroxide (which was previously standardized with 0.01 N of oxalic acid) until all traces of the red colour disappears and the colour of the solution was yellowish orange. The volume of alkali added was noted. This volume corresponds to free acidity. Then 2 drops of phenolphthalein solution was added and titration was continued until a definite red tinge reappears. Again the total volume of alkali added was noted. This volume corresponds to total acidity.

Acidity was calculated by using the formula,

$$Acidity = \frac{Volume\ of\ NaOH \times Acutal\ Normality\ of\ NaOH \times 100}{0.1}\ \mu eq/ml/100g$$

Statistical Analysis

The results were expressed as the mean ± SEM. Statistical differences were evaluated using a one-way analysis of variance (ANOVA) followed by Dunnett's test. Results were considered to be statistically significant at $p<0.01$.

RESULTS

The animals treated with EFCG 200 mg/kg and 400 mg/kg showed significant as well as dose dependent inhibition ($p<0.01$) of ulcer index when compared with control group. Also animals treated with EFCG 200 mg/kg and 400 mg/kg significantly ($p<0.01$) decreased gastric volume, gastric pH, total and free acidity when compared with control group.

Table 2: Effect of ethanolic flower extract of *Couroupita guianensis* on Ulcer index and Percentage ulcer protection in indomethacin plus pylorus ligation induced gastric ulcer.rats.

Drug Treatment	Ulcer Index	Percentage Ulcer Protection
Group I Normal control	---	---
Group II Ulcer control + Indomethacin (20mg/kg)	11.92±0.24	---
Group III Ranitidine (50mg/kg)	3.36±0.32**	72
Group IV EFCG (200mg/kg)	4.28±0.28**	64
Group V EFCG (400mg/kg)	3.82±0.16**	68

*Values are represented as mean ± SEM (n=6), **p<0.01 when compared with Control group.*

Table 3: Effect of ethanolic flower extract of *Couroupita guianensis* on pH, volume, free acidity and Total acidity of gastric secretions in indomethacin plus pylorus ligation induced gastric ulcer rats.

Drug Treatment	pH of gastric juice	Volume of gastric juice (ml)	Free acidity (mEq/ l)	Total acidity (mEq/ l)
Group I Normal control	2.1±1.8	4.8±1.3	29.3±0.2	72.3±0.3
Group II Ulcer control + Indomethacin (20mg/kg)	2.3±0.1	7.1±0.6	48.6±2.8	66.8±2.6
Group III Ranitidine (50mg/kg)	4.6±0.3**	4.4±0.2**	21.3±1.6**	37.4±3.2**
Group IV EFCG (200mg/kg)	3.4±0.2**	5.2±0.4**	32.2±1.4**	44.2±2.4**
Group V EFCG (400mg/kg)	4.6±0.4**	4.8±0.3**	26.2±3.8**	40.2±1.8**

*Values are expressed as mean ± SEM (n=6), **p<0.01 Vs control*

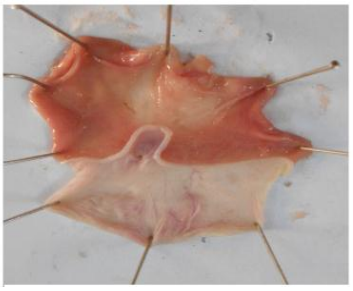

Figure 1: Control.

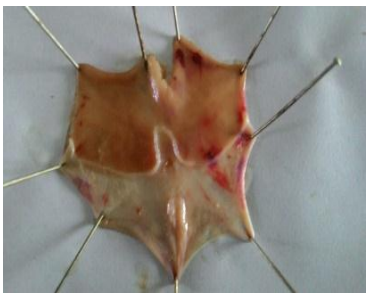

Figure 2: Ulcer control + Indomethacin.

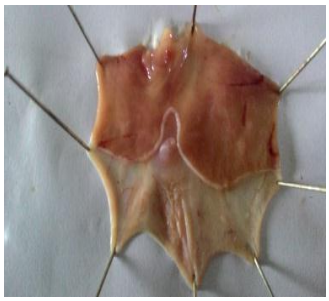

Figure 3: EFCG 200 mg/kg.

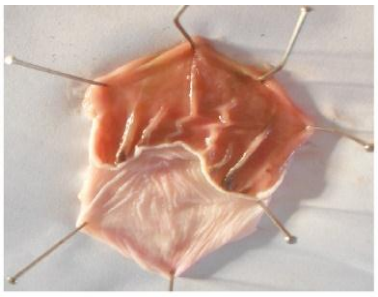

Figure 4: EFCG 400 mg/kg.

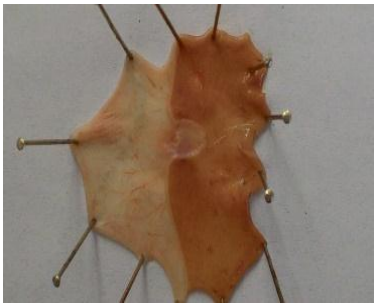

Figure 5: Ranitidine 50 mg/kg.

DISCUSSION

Couroupita guianensis is a medicinal plant widely used for the gastric problems. Literature survey shows the efficiency of agent for its anti-ulcer activity. The objective of the study is to screen the ethanolic flower extract of *Couroupita guianensis* for its anti-ulcer activity. Here, ethanolic extract is found to be safe on consumption and also it is helpful to yield polyphenols in higher amount. In Indomethacin plus pylorus ligation induced ulcer model, the ethanolic flower extract at dose of 200 and 400 mg/kg showed significant gastro protective activity 64% and 68% respectively as compared with control. Standard drug Ranitidine showed 72% of gastro protective activity in indomethacin plus pylorus ligation induced gastric ulcer. Both doses of ethanolic flower extract of *Couroupita guianensis* extract showed significant ($p<0.01$) reduction in ulcer index as compared to the control. The results of the present study indicate that the ethanolic extract significantly ($p<0.01$) reduces the total volume of gastric juice, free and total acidity of gastric secretion, pH of gastric juice and also has activity against indomethacin plus pylorus ligation induced gastric ulcers in rats. This effect is due to the presence of phenolic compound in EFCG.

CONCLUSION

The present study demonstrated that ethanolic flower extract of *Couroupita guianensis* possess significant dose dependent anti-ulcer activity in indomethacin plus pylorus ligation induced gastric ulcer model. The findings of the study could lead a compound for further isolation, characterization of active principle for its anti-ulcer activity.

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