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# Phytochemical Screening and Anti-Ulcer Activity of *Cleomo Gynandra Linn*

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## ABSTRACT

The anti-ulcer activity of leaves of benzene and pet ether extract of cleome gynandra inn was investigated in ethanol induced ulcer model in the male wistar albino rats. The parameters evaluated are ulcer index ,volume of gastric juice, gastric acidity, ph of gastric juice. Pet ether and benzene extract at doses of 150 mg/kg produced significant inhibition of gastric lesion induced by ethanol induced gastric ulcer. The extract shows significant reduction in gastric volume ,and ulcer index when compared to control. The present study indicates that leaves of benzene and pet ether extract of cleome gynandra inn have potential anti ulcer activity in ethanol induced ulcer model. Further the study also implies that dietary polyphenolic phytochemical especially the flavonoid, saponins accumulated in leaves may supply substantial anti-ulcer agents, which in turn may inhibit the development of several chronic diseases and there by provide health promoting effect.

**Keywords :** Cleome gynandra linn, Ethanol induced ulcer model, ulcer index

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# INTRODUCTION

Gastric ulcer, one of the most widespread, is believed to be due to an imbalance between aggressive and protective factors . The gastric mucosa is continuously exposed to potentially injurious agents such as acid, pepsin, bile acids, food ingredients, bacterial products (Helicobacter pylori) and drugs . These agents have been implicated in the pathogenesis of gastric ulcer, including enhanced gastric acid and pepsin secretion, inhibition of prostaglandin synthesis and cell proliferation growth, diminished gastric blood flow and gastric motility . Drug treatment of peptic ulcers is targeted at either counteracting aggressive factors (acid, pepsin, active oxidants, platelet aggravating factor "PAF", leukotrienes, endothelins, bile or exogenous factors including NSAIDs) or stimulating the mucosal defences (mucus, bicarbonate, normal blood flow, prostaglandins(PG), nitric oxide). The goals of treating peptic ulcer disease are to relieve pain, heal the ulcer and prevent ulcer recurrence. Currently there is no cost-effective treatment that meets all these goals. Hence, efforts are on to find a suitable treatment from natural product sources.[Raju.D-2009]

Cleome gynandra Linn. [Sync. pentaphylla Linn. Gynandropsis pentaphylla DC. G. gynandra (Linn.)Briq.] of Cleomaceae (Capparaceae)family is an erect glandular-pubescent annual herb, popularly used in the Ayurveda, Siddha, Folk and Tibetiansystems of medicine. It is known as Cat's whiskers and Spider flower in English; Cararvella, varvar, surjavarta andarkapushpika in Sanskrit; Arkahuli, karaila, hulul and churota in Hindi; and Velai keerai, neivayalla keeraiand katte kadugu in Tamil. This wildleafyvegetable is indigenous to the tropical and pan tropical regions and plays animportantrole in agricultural and nutritional systems of these regions [chewya JA-1997,Van den Heaver H-2007, the wealth of india-1956 and chatterjee A-1991]. Inmany cultures, the boiled leaves are regarded as medicinal meal for the treatment of various ailments. Bruised Leaves are reported to be rubefacient,vesicant,antiseptic,anti-inflammatory andAnalgesic and hence used to treat local pains, neuralgia, rheumatism and scorpion-sting. Oral administration of a decoction or an infusion of the boiled leaves or the leaf juice has been recorded to facilitate child birth, to relieve stomach pain, beneficial in constipation, thread-worm infection, conjunctivitis, oral ailments, convulsions and in certain bilious disorders [Van den Heever H-2007, the wealth of india-1956 and chatterjee A-1991,oliver-bever B-1983,kumar PS-1987,tabutiJRS-2003cano JH-2004, hebbar SS-2004 and kamatenesi-mugisha M-2007]. Earlier investigations on the leaves of the Egyptian taxon haveAfforded certain flavonoids, triterpenoid, saponins, sterols and fatty acids [rastogi RP-2007], atriterpene from the whole plant [das PC-1999], glucosinolates [songsak T-2002] and a number of anti-tick essential oil constituents

[Lwande W-1999] . Extracts of the leaves and certain isolated flavonoids have been reported to possess antibacterial, antifungal, anti-neoplastic and anti-arthritic properties and improved the levels of endogenous antioxidants and also modulated glucose metabolizing enzyme Activity [Ajaiyeoba EO-2000, pettit GR-2005, narendhirakannan RT-2007 and siyanesan D 2007]. Dietaryphytochemicals, especially the polyphenolic antioxidants such as the ubiquitous flavonoids [Middleton E-2000], polyunsaturated fatty acids, tocopherols,vitamin C and various inorganic micronutrients have been the subject of extensive research for their potential benefits to reduce the risk of degenerative diseases such as cardiovascular disease, several types of cancer, inflammation and neurological and other age-related disorders. Being diet derived, these compounds are generally regarded as safe chemicals based on their long history of use in the diet and have been demonstrated to possess strong antioxidant activities in vitro. Though the screening of antioxidant and radical scavenging activities of the taxon have been reported earlier [cook JA-1998 and Atwood SE-2005], but an investigation of the amounts of the potentially beneficial antioxidants available in the extracts of varying polarities of this leafy vegetable will also be of importance.

It is reported to contain of many biologically active phyto chemicals such as triterpenes, tannins, anthroquinones, flavonoids, saponins, steroids, resins, lectins, glycosides, sugars, phenolic compounds, and alkaloids in the extract of *C. gynandra* and these compounds might be responsible for the anti-arthritic properties observed in the present study. The possible mechanism of action of the *C. gynandra* extract may be through its stabilizing action on lysosomal membranes and there by preventing the spread of inflammation.Narendhirakannan R.T. 2007).By keeping these factors in this study we evaluated the plant for anti ulcer activity.

## MATERIALS AND METHODS

**Collection Of Plant Material***:*

a). *Cleome gynandra* Linn. Leaves were collected from the local gardens of houses and plant nursery and they are authenticated. The collected plant material was washed with water and dried at shadow room.

b) The dried plant material was powdered by using pulveriser.

**PREPARATION OF THE PLANT EXTRACT**

**Preparation of the *cleome gynandra* Linn leaves extract:**

50 gm of powdered *cleome gynandra* Linn was weighed accurately and is placed in the main chamber by either directly or by the use of thimble packing. The powder drug is placed onto a

flask containing the extraction solvent like benzene or pet.ether. The soxhlet is then equipped with a condenser. The solvent is heated to reflux the solvent vapour travels up distillation arm and floods into the chamber housing of solid. The condenser ensures that any solvent vapour cools, and drips back down into the chamber housing the solid material. When the soxhlet chamber is almost full the chamber is automatically emptied by side siphon arm, with the solvent running back to the distillation flask. The cycle may be allowed to repeat many times. After 7 days plant extracts will be collected.

**Phytochemical analysis**

Qualitative phytochemical evaluation was carried out to test the presence of alkaloids, flavones, sugar, phenol group, saponin, amino acid and essential oil in the extracts samples using modified method. Brindha *et al*. (1981).

**BIOLOGICALACTIVITY**

**Preparation of ranitidine solution:**

Ranitidine 150mg was taken and it was dissolved in 10ml of distilled water. It was administered at adose of 80mg/kg according to the body weights of the animals.

**Animal selection:**

A total of 25 male Wister rats were obtained from the animal facility Bharath College of pharmacy and used for study. All rats were certified good health at the time of receiving. Age of animals at the start of the treatment was approximately 8 to 10 weeks.

**Acclimatisation:**

Rats were allow to acclimatise to experimental room conditions for a period of 7 days prior to randomisations and treatment. During the acclimatisation period the rats are observed for the clinical signs**.**

**Environmental conditions**:

The rats were maintained in the separate polypropylene cages. In the experimental room 12 hrs of artificial lightening and 12 hrs of darkness cycle were maintained. The experimental room was cleaned and mopped with a disinfectant daily.

**Housing conditions:**

The rats were housed based on the group size per polycarbonate cage. Each cage was fixed with a poly propylene water bottle with stainless steel nozzle. Feed was provided *ad libitum* throughout the study. The bedding material was changed daily.

**Feeding conditions:**

Rats were provided with 150 gms of rat bed and sterilized water.rat feed and supplied water was

changed on alternative days. The amount of the feed consumed by the rats were calculated on successive days.

**Grouping of animals:**

The animals were grouped into five groups. Each group contains six animals.

**Groping of animals as follow:**

**DOSE:150 mg/kg(R.T. Narendhirakannan 2007)**

**DOSING OF ANIMALS:**

The animals were dosed with the test and the standard drugs orally based on the body weights of the animals. The animals were dosed with the extracts for about 7 days. During dosing of animals, the body weights of animals and the food consumed by the animals were taken on successive days.

**METHOD:**

**Ethanol induced method:**

The healthy wistar rats were taken for the study. The rats were to be administered with the vehicle and extracts for about 7 days. After 7 days of dosing was completed they were to be fasted for 12hrs, then ulcer induced with ethanol, after 2hrs of the induction of ethanol the animals were sacrificed the stomachs of the rats were cut open along the greater curvature and parameters were checked.

**MONITORING OF PARAMETERS:**

**Determination of ulcer index, volume of gastric juice, ph of gastric secretions**

**Ulcer index:**

The dissected stomachs of the sacrificed rats were opened along the greater curvature and the ulcer index was calculated from the glandular portions of the stomach.

The ulcer index:

0 - normal coloured stomach

0.5 - red coloration

1.0 - spot ulcers

1.5 - haemorrhagic streaks

2 - Ulcers greater than or equal to 3, but less than 5

3 - Ulcer greater than 5

The ulcer index was calculated as

Ulcer index=10/x

Where x=total mucosal surface/total ulcerated surface

**Volume and pH of gastric juice:**

The contents of the dissected stomachs of the rats were taken in a graduated test tube and allowed to centrifuge at 2000rpm for 10 min.the supernatant fluid was measured for volume of gastric juice and expressed as ml/4hrs and Ph of the gastric juice was measured.

**Gastric acidity:**

The supernatant liquid of the gastric juice was taken in a conical flask and 2 drops of torpors reagent was added.0.01N NaoH was taken in a burette and allowed to triturate till the flask changed to yellow colour. Then 2 drops of phenolphthalein was added and titrated till orange colour was reached

## RESULTS AND DISCUSSION

**Phytochemical screening**

The phytochemical screening of both the leaf extract of benzene and pet ether were performed and it shows the presence of different amount of content

**Table 1 phytochemical screening of both the leaf extract of benzene and pet ether**

| S. No. | Phytochemical name | Solvent name | Plant parts | | | | |
|---|---|---|---|---|---|---|---|
| | | | Leaf | Root | Stem | Seed | Seed pod |
| 1 | Tannin | Ether | +++ | +++ | +++ | +++ | +++ |
| | | Benzene | +++ | +++ | +++ | +++ | +++ |
| 2 | Alkaloid | Ether | +++ | ++ | +++ | ++ | + |
| | | Benzene | +++ | ± | +++ | ++ | + |
| 3 | flavones | Ether | +++ | - | + | +++ | + |
| | | Benzene | +++ | - | ++ | +++ | + |
| 4 | sugar | Ether | +++ | +++ | ++ | +++ | +++ |
| | | Benzene | +++ | ++ | +++ | ++ | + |
| 5 | Phenolic group | Ether | +++ | +++ | +++ | +++ | +++ |
| | | Benzene | +++ | +++ | +++ | +++ | +++ |
| 6 | Essential oil | Ether | +++ | ++ | + | +++ | +++ |
| | | Benzene | ++ | +++ | ++ | +++ | +++ |
| 7 | Amino acids | Ether | +++ | +++ | +++ | +++ | +++ |
| | | Benzene | +++ | +++ | +++ | +++ | ++ |
| 8 | Saponin | Ether | +++ | ± | ++ | + | + |
| | | Benzene | +++ | + | +++ | ++ | + |

+ + + excellent; + + good; + moderately positive; ± doubtful

**Phytochemical screening:**

The results of preliminary phytochemical screening of the pet ether and benzene extract of cleome gynandra revealed that presence of tannins, flavanoids, essential oils, alkaloids.

**Ethanol Induced Gastric Ulcers**

In control animal ,oral administration of absolute ethanol produced characteristic lesions in the

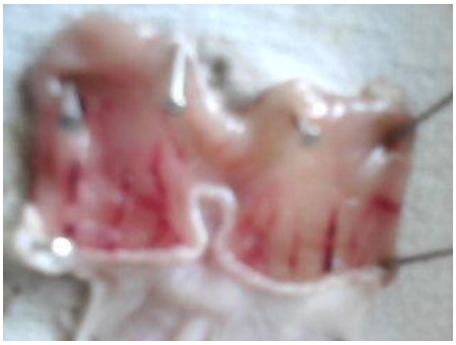
**Table 2: ulcer index ,volume of gastric juice, gastric acidity, pH of gastric juice giving different treatment**

| Treatment | Dose (mg/kg) | Animal no. | Body wt in gm | Ulcer index | Volume of gastric juice | pH of gastric juice | Gastric acidity(milli equivalents/100g) |
|---|---|---|---|---|---|---|---|
| Normal | 0 | 1 | 200 | 0 | 0.5 | 8.9 | 20 |
| | | 2 | 170 | 0.5 | 0.8 | 9.3 | 19 |
| | | 3 | 175 | 0.5 | 0.7 | 8.4 | 17 |
| | | 4 | 150 | 0 | 0.4 | 8.9 | 21 |
| | | 5 | 160 | 0 | 0.4 | 9.5 | 19 |
| | | 6 | 190 | 0 | 0.6 | 8.4 | 20 |
| | | Mean | 174.1667 | 0.1666 | 0.5666 | 8.9 | 19.3333 |
| Ethanol induced | 1 ml | 1 | 170 | 4 | 2 | 4.8 | 40 |
| | | 2 | 160 | 3 | 1.8 | 5.6 | 41 |
| | | 3 | 190 | 4 | 1.7 | 5.9 | 39 |
| | | 4 | 150 | 4 | 1.9 | 5.4 | 41 |
| | | 5 | 140 | 3 | 1.8 | 5.1 | 39 |
| | | 6 | 170 | 4 | 2 | 5.3 | 40 |
| | | Mean | 163.3333 | 3.6666 | 1.8666 | 5.35 | 40 |
| extract:1 (pet.ether) | 150 mg/kg | 1 | 150 | 0.5 | 0.5 | 7.9 | 25 |
| | | 2 | 190 | 0.5 | 0.9 | 8.5 | 24 |
| | | 3 | 180 | 1 | 0.6 | 8.3 | 25 |
| | | 4 | 160 | 0.5 | 0.4 | 7.9 | 26 |
| | | 5 | 190 | 1 | 0.5 | 8.1 | 24 |
| | | 6 | 180 | 0.5 | 0.5 | 7.4 | 25 |
| | | Mean | 175 | 0.6666 | 0.5666 | 8.0166 | 24.8333 |
| extract:2 (benzene) | 150 mg/kg | 1 | 170 | 1 | 0.9 | 7.5 | 28 |
| | | 2 | 150 | 1.5 | 0.8 | 6.5 | 29 |
| | | 3 | 170 | 1.5 | 0.7 | 6.1 | 27 |
| | | 4 | 160 | 0.5 | 1.1 | 7.6 | 29 |
| | | 5 | 140 | 0.5 | 1.2 | 6.5 | 28 |
| | | 6 | 200 | 1 | 0.9 | 6.9 | 29 |
| | | Mean | 165 | 1 | 0.9333 | 6.85 | 28.3333 |
| Ranitidine | 80 mg/kg | 1 | 160 | 1 | 0.5 | 8.9 | 16 |
| | | 2 | 170 | 0.5 | 0.4 | 8.8 | 19 |
| | | 3 | 160 | 0.5 | 0.5 | 8.1 | 18 |
| | | 4 | 170 | 0.5 | 0.6 | 7.9 | 16 |
| | | 5 | 190 | 1 | 0.4 | 8.9 | 17 |
| | | 6 | 180 | 0.5 | 0.3 | 8.5 | 16 |
| | | Mean | 171.6667 | 0.6666 | 0.45 | 8.5166 | 17 |

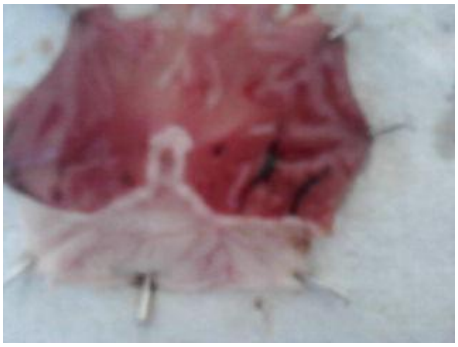
glandular portion of rat stomach which appeared as elongated bands of thick, black and dark red lesions. Pet ether and benzene extract has shown significant ulcer index of0.6 and 1 with the dose of 150mg\kg respectively in comparison to control, ranitidine as reference standard drug was reduction of ulcer.

The aetiology of peptic ulcer is unknown in most of the cases, yet it is generally accepted that it results from an imbalance between aggressive factors and the maintenance of mucosal integrity through the endogenous defence mechanisams.to regine the balance, different therapeutic agents are used to boost the mucosal defence mechanisms by increasing mucosal production ,stabilising the surface epithelial cells or interfering with the prostaglandin synthesis.

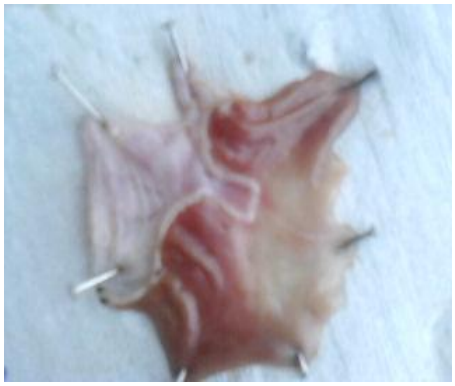
Ethanol induced gastric ulcer was employed to study the cytoprotective effect of the extracts. Ethanol induced gastric lesion formation may be due to stasis in gastric blood flow which contributes to the development of the haemorrhage and necrotic aspects of tissue injury. Alcohol rapidly penetrates the gastric mucosa apparently causing cell and plasma membrane damage leading to increased intra cellular membrane to sodium and water. The massive intracellular accumulation of calcium represents a major step in the pathogenesis of gastric mucosal injury. This leads to cell death and exfoliation in the surface epithelium .the extracts shows protection against characteristics lesions produced by ethanol administration on this antiulcer effect of PECG may be due to both reductions in gastric acid secretion and gastric cytoprotection .



**benzene extract**

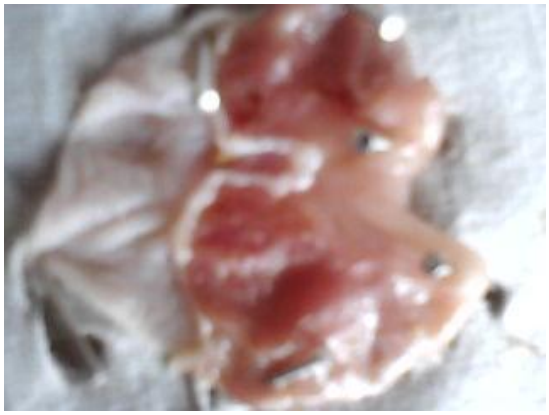


**ethanol induced ulcers**



**Normal**

**pet ether extract**



**Ranitidine**

**Figure :1 Effect of different extract in Gastric Ulcers**

## CONCLUSION

The results of the Pharmacognostical characters of the leaves of C.gynandra such as the microscopic features may help in laying down micro-morphological standards as per WHO guide lines for authentication of the leaf drug. Adulterants if any, can be easily identified using these parametars. Further this study also implies that dietary poly phenolic phyto chemical especially the flavonoid, saponins accumulated in leaves may supply substantial anti-ulcer agents, which in turn, may inhibit prevent or retard the development of several chronic diseases and there by provide health promoting effect.

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27. Elizabeth H. Flora Sicolani Todlana Sat May 15 14:10:56 2004. Cleome gynandra is an adible plant found in Zimbabwe mostly, during rain season. This wild plant is eaten fresh as relish and may be cooked and dried for cunsumption during dry seasons. It is called "ULUDE" in Zulu or Ndebele. It tastes very bitter originally, but boiling it with vegetable oil, tomatoes and a pinch of salt makes is a good relish. Author's name: Flo Todlana
28. F Lakor Sun Mar 12 2006 Flora Sicolani Todlana-Lakor Cleome gynandra is an adible plant found in Zimbabwe mostly, during rain season. This wild plant is eaten fresh as

relish and may be cooked and dried for consumption during dry seasons. It is called "ULUDE" in Zulu or Ndebele. It tastes very bitter (highly alkaloidal) originally, but cooking/boiling it with vegetable oil, tomatoes/onions and a pinch of salt makes is a good relish. Author's name: F Todlana-Lakor.

29. Elizabeth H. Thu Mar 27 2008. Cleome gynandra is a well known and used edibla plant for the majority people in namivbia, the Wambo, living in their reagon in the NOth called Owambo. It is called in their languga eOshiwambo "Ombidi" and prepared like spinach. In English the Wambo call the plant "wild spinach".
30. Elizabeth H. Loma Fefe Sigogo. Wed May 21 2008. Flora Sicolani Todlana-Lakor Cleome gynandra is an adible plant found in Zimbabwe mostly, during rain season. This wild plant is eaten fresh as relish and may be cooked and dried for consumption during dry seasons. It is called "ULUDE" in Zulu or Ndebele. It tastes very bitter (highly alkaloidal) originally, but cooking/boiling it with vegetable oil, tomatoes/onions and a pinch of salt makes it a good relish. Author's name: F Todlana.
31. Melenia Khuzwayo Tue Jul 22 2008. Ulude can not be classified for any medical reasons or use. Class = Food.Rate This Plant -Please rate this plants for how successful you have found it to be. You will need to be logged in to do this. Our intention is not to create a list of 'popular' plants but rather to highlight plants that may be rare and unusual and that have been found to be useful by website users. This hopefully will encourage more people to use plants that they possibly would not have considered before
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