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Anti-ulcer activity of *Dipteracanthus patulus (Jacq)* leaf extracts

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ABSTRACT: Background: Peptic ulcer disease (PUD) is one of the most common gastrointestinal disorders, having a high rate of morbidity particularly in the population of non-industrialized countries. Peptic ulcer occurs due to an imbalance between the aggressive and the defensive factors. The modern approach to control gastric ulceration is to inhibit gastric acid secretion, promote gastroprotection, block apoptosis, and stimulate epithelial cell proliferation for effective healing. **Objective:** The present study aimed to evaluate the anti-ulcer activity of *Dipteracanthus patulus (Jacq)* leaf extracts. **Method:** Male albino rats with aspirin and ethanol-induced ulcers were divided into four group as control, ranitidine therapy, ethanolic extract of *D. patulus (Jacq)* leaf, and chloroform extract of *D. patulus (Jacq)* leaf respectively for screening of anti-ulcer activity of *D. patulus (Jacq)*. The treatment with extracts of *D. patulus (Jacq)* leaf continued for seven days after which the model was sacrificed to calculate the ulcer index. **Result:** In the aspirin-induced ulcer model, values of ulcer index at the significant level of P<0.0001 were found to be 3.44±0.20 and 3.05±0.12 for the treated group by *D. patulus* ethanol and chloroform extract at a dose of 100 mg/kg respectively. The ulcer index value of test extracts was found to be lowered as compared to both standard and control groups. In the ethanol-induced ulcer model, values of ulcer index at the significant level of P<0.0001 were found to be 2.72±0.09 and 2.97±0.20 for the treated group by *D. patulus* ethanol and chloroform extract at a dose of 100 mg/kg respectively. The ulcer index value of test extracts was found to be lowered as compared to both standard and control groups. **Conclusion:** It could be concluded that the *D. patulus* possessed significant anti-ulcer activity.

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Keywords: Anti-ulcer, Aspirin, *Dipteracanthus patulus*, Ethanol, Ulcer index.

INTRODUCTION:

Peptic ulcer disease is a chronic pathology that affects millions of people worldwide. It is believed that 10 % of the population will develop this condition at some point in their lives. These are lesions observed in the stomach and duodenum extending to the mucosa. *H. Pylori* and NSAID were strongly associated with the development of peptic ulcers [1]. *H. pylori* causes an inflammatory response in the gastric mucosa, with increased production of cytokines and the influx of neutrophils and

macrophages into the gastric mucosa with the release of leukotrienes and reactive oxygen species, which makes the defense of the mucosa and stimulates ulcer formation process [2]. NSAID serves as the risk factor for about 90 % of all ulcers contributing to mucosal damage either by direct mucosal erosion or by inhibition of the cyclooxygenase enzyme [3]. Stress-related mucosal damage (SRMD) is the broad term used to describe the spectrum of pathology attributed to the acute, erosive, inflammatory insult to the upper gastrointestinal tract associated with critical illness [4]. Medicinal plants are considered rich resources of ingredients that can be used in drug development and synthesis of medicines. Plants play a vital role in the development of human cultures around the world. Moreover, some plants are considered an important source of nutrition, and as a result of that plants are recommended for their therapeutic values. "Alternative Medicine" emerges as a better choice as it focuses on therapy with medicinal plants which are effective with minimal side effects. About half a million plants have medicinal values of which many are to be investigated [5]. The plant *Dipteracanthus patulus* belongs to the family *Acanthaceae*. The major phytoconstituents found in *D. patulus* are Tannins, Saponins, Alkaloids, Steroids anthroquinones, Tri-terpenoids, and Flavonoids. The objective of the study is to evaluate the anti-ulcer activity of leaf extract.

Collection and confirmation of plant:

The plant materials used in this study were leaves of *D. patulus (Jacq)* and were collected from the Sengundrapuram, Virudhunagar Dist, Tamil Nadu, India. The plant was authenticated by Dr. Stephen, Department of Botany, American College, Madurai, India.

Preparation of Ethanol and Chloroform extracts of *D. patulus (Jacq)* Leaf

D. patulus Leaves collected were dried at room temperature under shade for 15 days and coarsely powdered. The powdered materials were extracted with ethanol and chloroform. The last traces of the solvent were removed and concentrated to dryness under vacuum using a rotary evaporator. The dried extract was weighed and then kept at -4 ºC until ready for use. The yield of the extract was 66.42 % (w/w) and 33.68 % (w/w). In each experiment, the extract was diluted with water to the desired concentration.

Animal model:

Adult male albino rats (150 to 200 g) were used in this study. They were maintained in clean, sterile, and polypropylene cages and fed with commercial pellet rat chow (M/S Hindustan Lever Limited, Bangalore, India) and water ad libitum.

Approval:

The study was approved by the Institutional Ethical Committee, which follows the guidelines of the

MATERIALS AND METHOD:

The ethanol, chloroform, and Silica gel were purchased from CDH, New Delhi. The normal saline water was purchased from Baxter, India. The Ranitidine (Aciloc injection) was procured from Cadila, Chennai. Diclofenac sodium (Cofenac) was procured from Cipla, Goa, All other chemicals and reagents used in this research work were of analytical grade and procured from an authorized dealer.

Committee for Control and Supervision of Experimental Animals (CCSEA).

Evaluation of anti-ulcer activity:

Aspirin-induced ulcer model:

Animals were divided into four groups of four animals each. The dosage of drugs was administered by the following as mentioned in Table 2.

The gastric ulcer was induced in each rat by administering aspirin at a dose of 500 mg/kg orally. After 45 min, ethanolic and chloroform extract of *D. patulus* and other drugs were administered for seven days. The animals were sacrificed, and the stomach was excised and cut along the greater curvature, rinsed gently with saline to remove the gastric content and blood clots and the ulcer index was calculated.

Ethanol-induced ulcer model:

The animals were divided into four groups. The dosage of drugs was administered by the following as given in Table 3. The gastric ulcers were induced in rats by administrating absolute ethanol 99 % (1 ml/200 g) orally. After 45 min ethanolic and chloroform extract of *D. patulus* and other drugs were administered for seven days. The animals were sacrificed under anaesthetized conditions the stomach was dissected out and the ulcer index was calculated.

Calculation of Ulcer index and Percentage inhibition:

The following formula calculated the ulcer index (UI):

Ulcer index = 10/x ……(1)

Table 1. Grouped animal models for aspirin-induced ulcer.

Group	Drug administered	Dose (mg/kg)	Route
Group I (Control)	Tween 80	5	Oral
Group II	Standard Ranitidine	30	Oral
Group III	Ethanolic extract of *D.patulus*	100	Oral
Group IV	Chloroform extract of *D.patulus*	100	Oral

Table 2. Grouped animal models for ethanol-induced ulcer.

Group	Drug administered	Dose (mg/kg)	Route
Group I (Control)	Tween 80	5	Oral
Group II	Standard Ranitidine	30	Oral
Group III	Ethanolic extract of *D.patulus*	100	Oral
Group IV	Chloroform extract of *D.patulus*	100	Oral

Table 4. Preliminary phytochemical screening of ethanol and chloroform extract of D. *patulus*.

Sl. No.	Constituents	Ethanol extract of *D. patulus*	Chloroform extract of *D. patulus*
1	Carbohydrate	Absent	Absent
2	Proteins & amino acids	Absent	Absent
3	Flavonoids	Present	Present
4	Alkaloids	Present	Present
5	Tannin	Present	Present
6	Glycosides	Present	Absent
7	Anthroquinone	Present	Present
8	Tri-terpenoids	Present	Present
9	Saponins	Present	Absent
10	Phenol	Absent	Present
11	Steroids	Present	Present

Table 5. Effect of *D. patulus* leaf extracts Aspirin-Induced ulcer.

Sl. No.	Ulcer index	Protection (%)	Treatment	Dose (mg/kg)
1	5.57 ± 0.23	---	Control tween 20	5
2	1.69 ± 0.10**	69. 65	Ranitidine	30
3	3.05 ± 0.12**	45.24	Ethanolic extract	100
4	3.44 ± 0.20	38.24	Chloroform extract	100

Results are expressed as mean + SEM from four observations as compared to the standard group the one-way ANOVA is Graph Pad"s software method, (P< 0.0001) by conventional criteria; this difference is extremely statistically significant.**

Where, x= Total mucosal area/ total ulcerated area

The following formula calculated the percentage inhibition:

$$\text{Inhibition (\%)} = \frac{\text{UI control} - \text{UI treated}}{\text{UI control}} \times 100 \quad \ldots(2)$$

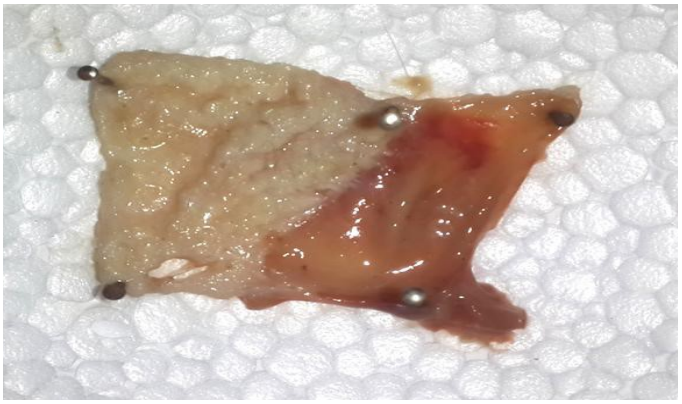

Fig 1. Control - Aspirin-induced ulcer model.

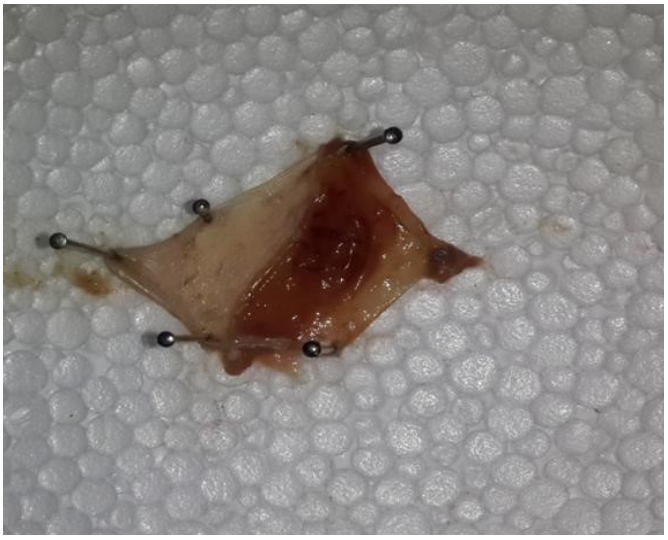

Fig 2. Ethanolic extract of *D. patulus (Jacq)* 100 mg/kg treated - Aspirin-induced ulcer model.

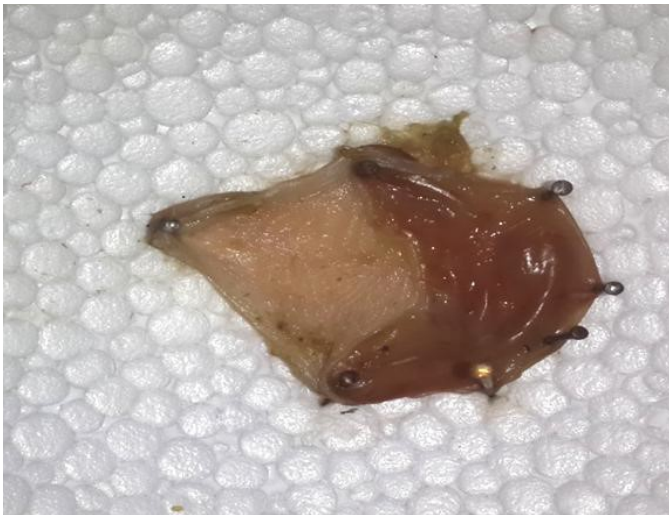

Fig 3. Chloroform extract of *D. patulus(Jacq)* 100 mg/kg treated- Aspirin-induced ulcer model.

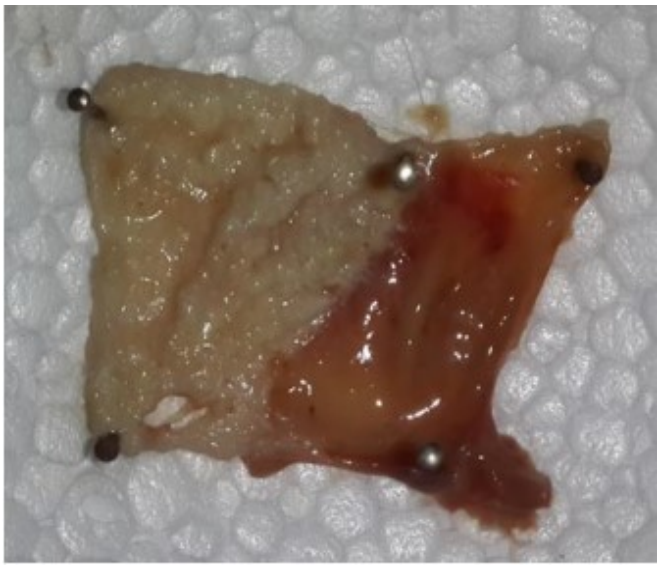

Fig 4. Control - Aspirin-induced ulcer model.

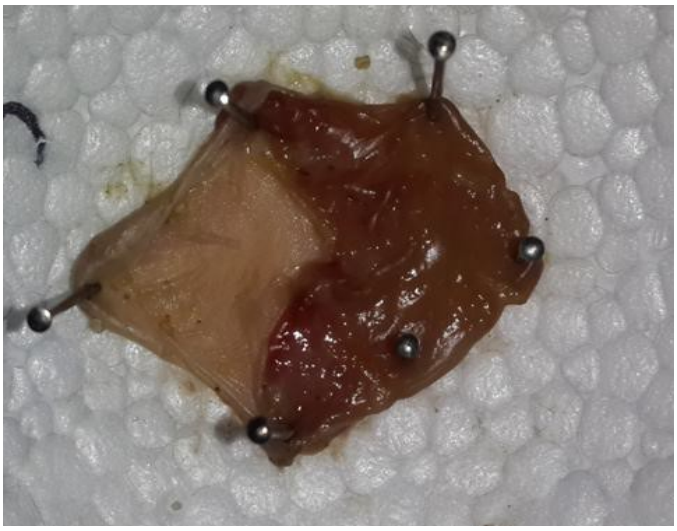

Fig 6. Chloroform extract of *D. patulus (Jacq)* 100 mg/kg treated- Ethanol induced ulcer model.

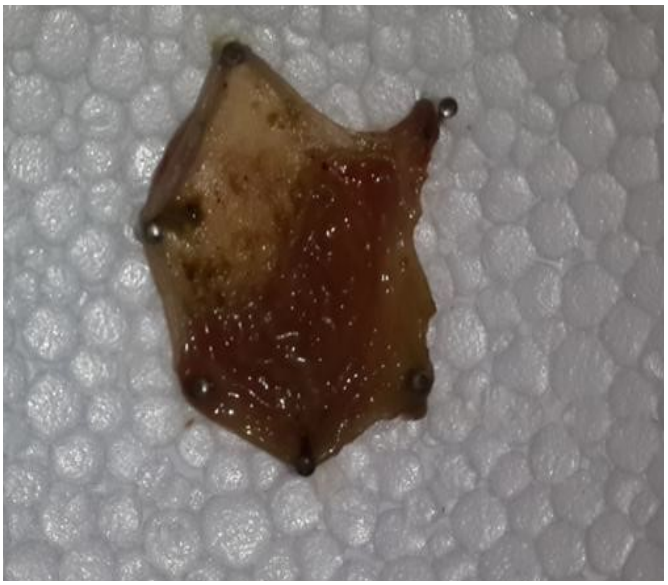

Fig 5. Ethanolic extract of *D. patulus (Jacq)* 100 mg/kg treated- Ethanol induced ulcer model.

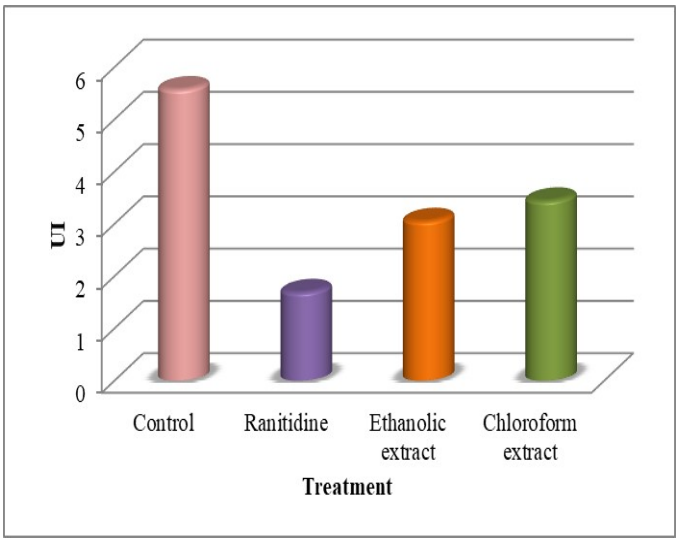

Fig 7. Ulcer index (UI) of Alcohol induced ulcer model.

Table 6. Effect of *Dipteracanthus patulus* leaf extracts Ethanol-Induced Ulcer.

Sl. No.	Ulcer index (UI)	Protection (%)	Treatment	Dose (mg/kg)
1	5.75 ± 0.08	--	Control tween 20	5
2	1.54 ± 0.13**	73.21	Ranitidine	30
3	2.72 ± 0.09**	52.69	Ethanolic extract	100
4	2.97 ± 0.20	48.34	Chloroform extract	100

Results are expressed as mean ± SEM from four observations as compared to standard group the one-way ANOVA is Graph Pad"s software method, (P< 0.0001) by conventional criteria; this difference is extremely statistically significant.**

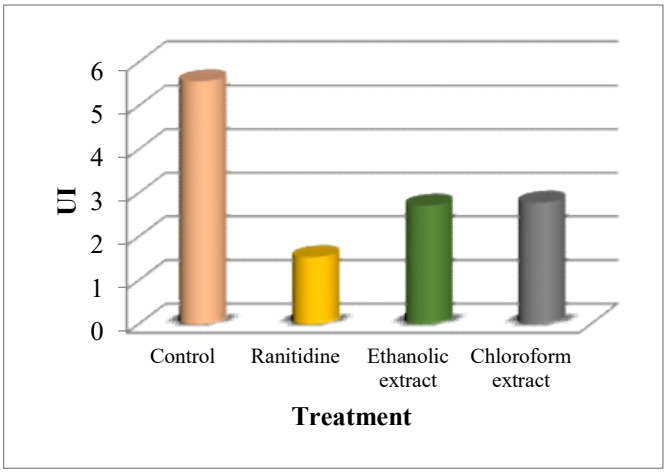

Fig 8. Ulcer index (UI) of Alcohol induced ulcer model.

RESULTS AND DISCUSSION:

The extracts of the *D. patulus* has significantly protected the gastric mucosa against ethanol challenge as shown by reduced values of ulcer index as compared to control group signifying its potent cytoprotective effect. It was evaluated that *D. patulus* extracts can suppress gastric damage induced by aggressive factors. The preliminary phytochemical studies revealed the presence of flavonoids in ethanolic extract of *D. patulus.*

In this study, we observed that *D. patulus* provides significant anti-ulcer activity against gastric ulcers in rats. Both extracts produced a significant ($p<0.001$) anti-ulcer activity. The active compounds would be able to stimulate mucus, bicarbonate and the prostaglandin secretion and counteract with the deteriorating effects of reactive oxidants in gastrointestinal lumen [6].

In the NSAIDs-induced gastric ulcer model, the ulcers were induced in rats by the administration of Aspirin (500 mg/ kg p.o). In the present study, in gastric ulcer model induced by Aspirin (500 mg/kg p.o) in rats the values of ulcer index were reduced in the treated group by *D. patulus* ethanol extract 100 mg/kg (3.05±0.12) and chloroform extract 100 mg/kg (3.44±0.20) there was a significant reduction in the ulcer index (**P<0.0001) as standard compared to control group [7].

Ethanol-induced ulcers major in the glandular part of the stomach was reported to stimulate the formation of leukotriene C4 (LTC4), mast cell secretory products, and reactive oxygen species resulting in the damage of rat gastric mucosa. In the present study, in gastric ulcer model induced by Ethanol (1 ml/kg, p.o.) in rats the values of ulcer index were reduced in the treated group *D. patulus* ethanolic extract 100 mg/kg (2.72±0.09) and chloroform extract 100 mg/kg (2.97±0.20), there was a significant reduction in the ulcer index (**P<0.0001) as standard compared to control group [8].

CONCLUSION:

Worldwide, there is a progressive tendency in favour of traditional and integrative health sciences both in research and clinical practice. Numerous factors are concerned in the ulcerogenesis, and gastric mucosal damage induced by different models active in the present study relating, mucosal damage induced by non-steroidal anti-inflammatory drugs and excessive free radical production. In the conclusion, the Chloroform and ethanolic extract of *D. patulus* leaf that potentiated the anti-ulcer activity. Therefore, further studies will be focused on the preliminary phytochemical studies responsible for the observed active constituents have to be isolated and identified.

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