

ANTI-INFLAMMATORY ACTIVITY OF FLOWER OF *CNIDOSCOLUS*
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ABSTRACT

Cnidoscolus phyllacanthus, commonly known as faveleira, is a drought-resistant shrub plant. It belongs to the Euphorbiaceae family and thrives in arid regions. The plant has spiny stems, lobed leaves, and produces small white flowers. Its seeds are rich in oil, making them valuable for biodiesel production. Faveleira is also used as fodder for livestock due to its high nutritional content. This study seeks to determine the anti-inflammatory of *Cnidoscolus Phyllanthus* medicinal plant that have been used traditionally to treat inflammation. The present study was undertaken to determine anti-inflammatory of *Cnidoscolus phyllacanthus* using various biochemical parameter. Preparation of Hydroalcoholic extract hot percolation method where obtained % yield as 2.67 % which was brown in color and semi-solid in consistency. Oral Acute toxicity study was evaluated as per OECD guidelines (425) on Wistar albino rats, where three dose levels 100 mg/kg, 200 mg/kg & 300 mg/kg was selected for anti-inflammatory activity. The qualitative phytochemical test was performed for the assessment of the presence of phytoconstituents where the preliminary phytochemical analysis indicated a positive reaction for the presence of coumarins, flavonoids, carbohydrates and anthracene derivatives in Hydroalcoholic extract of *Cnidosolus Phyllacanthus* whereas negative reaction with proteins, amino acids, tannins, alkaloid, and sterols. Anti-inflammatory activity by carrageenan rat paw edema method and Histamine-induced paw edema Method which exhibits anti-inflammatory properties by reducing paw edema in a dose-dependent manner.

KEYWORDS: Flower, Hydroalcoholic extract, *Cnidosolus Phyllacanthus*, Anti-inflammatory.

INTRODUCTION

Herbal medicament is being used in the developing countries for health care. They are standing for their safety, efficacy, and cultural acceptability and also having lesser side effects. The chemical constituents present in herbs, are a division of the physiological functions of living flora and hence they possess better compatibility with the human body.^[1]

Inflammation is a dynamic process that involves well-organized steps of cellular changes within living tissues. Inflammation can from, a few minutes to a few years, Depends on the level of the injury and kind of injury. Acute inflammation is a complex protective reaction triggered in an organism in response to infection, trauma, ischemia, or other noxious stressful stimuli which infectious agents or substances may have caused from their metabolism (Microorganisms and Toxins), as well as by physical agents (Radiation, Burn and Trauma), or chemicals (Caustic substances). India with its biggest repository of medicinal plants in the world may maintain an important position in the production of raw materials either directly for crude drugs or as the bioactive compounds in the formulation of

pharmaceuticals and cosmetics. These are those disorders that cause inflammation as the symptom. Mostly Inflammatory disorders are autoimmune disorders. Autoimmune disorders are in which the inflammation as the symptoms appears by the own body response by immune system. Some auto immune disorders are Gout, Arthritis (Osteoarthritis, Rheumatoid Arthritis (RA), Psoriatic Arthritis) Atherosclerosis, Alzheimer's Colitis Dermatitis Diverticulitis, Irritable Bowel Syndrome (IBS) Systemic Lupus Erythematosus (SLE), Cancer related inflammation.^[2]

Cnidoscolus phyllacanthus, commonly known as faveleira, is an important plant due to its multiple ecological, agricultural, and economic benefits. The plant serves as a nutritious fodder source for livestock, providing essential minerals and proteins. Additionally, its oil-rich seeds have potential for biodiesel production, offering a sustainable alternative to fossil fuels. Traditional medicine utilizes its leaves and extracts to treat inflammation, wounds, and skin infections due to its antioxidant and medicinal properties. Faveleira also supports biodiversity by attracting pollinators such as bees, which aids in maintaining ecological balance. Its

adaptability and resilience to pests reduce the need for chemical fertilizers and pesticides, making it an excellent choice for sustainable agriculture. Furthermore, it plays a role in carbon sequestration, helping mitigate climate change. Due to these diverse benefits, it is a crucial plant for environmental sustainability, economic development, and rural livelihoods. The extracts of *Cnidioscolus* have high inhibition percentage of cell growth. These species are commonly used to treat tumors and inflammation. The roots, bark and latex are used for the treatment of inflammatory processes, genitourinary and in general as antiseptic, dermatologic and ophthalmic. It is also used to treat kidney diseases, urinary infections, contusions, fractures, wounds, warts and hemorrhoids. It is also a good source of proteins, vitamins and minerals.^[3,4]

MATERIALS AND METHODS

Preparation of Hydroalcoholic extract of The *Cnidioscolus Phyllacanthus* plant was collected from nearby Guru Ramdas Institute of Science and Technology-Pharmacy, Jabalpur and then identified and authenticated in the Government Science college, Jabalpur (M.P.). The flower was cut down into small pieces, shade dried and powdered to get moderately coarse powder, which is sieved under mesh. About 500gm of dry powder was extracted with using 70% ethanol and 30% water for 18 hrs. Appearance of colorless solvent in the siphon tube was the indication of exhaustive extraction and based on that, further extraction was terminated. The extract was then transferred into the previously weighed empty beaker and evaporated to a thick paste on the water bath, maintained at 50°C to get alcoholic extract.

The qualitative phytochemical test was performed for the assessment of the presence of phytoconstituents such as alkaloids, glycosides, proteins, amino acids, flavonoids, sterols, carbohydrates, phenolic compounds, tannins, acidic compounds and resins.

A suspension formulation of hydroalcoholic extract of *Cnidioscolus Phyllacanthus* extract in 0.5% CMC solution was prepared for further In Vivo studies. 0.5% CMC suspension was prepared by suspending 500 mg of accurately weighed CMC powder in 100 ml of distilled water. 10 ml of vehicle was taken separately. Concentration of prepared extract suspension was 50mg/ml.

The experiment was carried out on Westar albino rats of 4 months, of both sexes, weighing between 120 to 150 gm. They were provided from GRKIST-P, JABALPUR. The animals were acclimatized to the standard laboratory conditions in cross ventilated animal house at temperature 25±2°C relative humidity 44–56% and light and dark cycles of 12:12 hours, fed with standard pellet diet and water *ad libitum* during experiment. The experiment was approved by the institutional ethics committee and as per CPCSEA guidelines.^[5]

Acute inflammation was caused by injecting 0.1 ml of 1 % (w/v) carrageenan in saline into the sub-plantar region of the right hind paw of each rat. The paw volume was measured plethysmometrically at 0 h, 1 h, 2 h, 3h, and 4h after the carrageenan injection. Edema was expressed as mean increase in paw volume relative to control animals. The percentage inhibition of edema was calculated by the following equation: of edema = $100 (1 - V_t/V_c)$, where V_c is the edema volume in the control group and V_t is the edema volume in tested groups

Experimental design

In the experiment, a total of 30 rats was used. The rats were divided into 5 groups comprising of 6 animals in each group.

Group I: Control (Vehicle treated) + inject carrageenan (0.1 ml.).

Group II: Standard, treated with Diclofenac sodium (10 mg/kg, p.o.) + inject carrageenan (0.1 ml.).

Group III: Animals treated with of hydroalcoholic extract of *Cnidioscolus Phyllacanthus*, (100mg/kg p.o.) once daily for seven days + inject carrageenan (0.1 ml.) on 7th day after 1h of last dose.

Group IV: Animals treated of hydroalcoholic extract of *Cnidioscolus Phyllacanthus*, (200mg/kg p.o.) once daily for seven days + inject carrageenan (0.1 ml.) on 7th day after 1h of last dose.

Group V: Animals treated with of hydroalcoholic extract of *Cnidioscolus Phyllacanthus*, (300mg/kg p.o.)

In Histamine-induced paw edema Method, Paw edema was induced after 1 h by sub plantar administration of 0.1 ml histamine (1 µg/ml) on the right hand paw. The linear paw circumference were measured initially and after every 1 hr up to 4 h. Test groups were treated with the different doses of ethanol extract, standard group with diclofenac sodium (10 mg/kg, p.o.) and control group with vehicle prior to the histamine administration.^[6] The inhibition of inflammation was calculated using the following formula:

$$\text{Percentage inhibition} = (V_c - V_t)/V_c \times 100$$

Where, V_t is the paw volume of test group and V_c is the paw volume of control group

In the experiment, a total of 30 rats was used. The rats were divided into 5 groups comprising of 6 animals in each group.

Group I: Control (Vehicle treated) + inject histamine (0.1 ml.).

Group II: Standard, treated with Diclofenac sodium (10 mg/kg, p.o.) + inject histamine (0.1 ml.).

Group III: Animals treated with hydroalcoholic extract of *Cnidioscolus Phyllacanthus*, (100mg/kg p.o.) once daily for seven days + inject histamine (0.1 ml.) on 7th day after 1h of last dose.

Group IV: Animals treated with hydroalcoholic extract of *Cnidioscolus Phyllacanthus*, (200mg/kg p.o.) once daily for seven days + inject histamine (0.1 ml.) on 7th day after 1h of last dose.

Group V: Animals treated with hydroalcoholic extract

of *Cnidosolus Phyllacanthus*, (300mg/kg p.o.) once daily for seven days+ inject histamine (0.1 ml.) on 7th day

Statistical analysis

All the values are expressed as mean standard error of mean (S.E.M.) and analyzed for ANOVA and potshot Turkey-Kramer Multiple Comparisons Test by employing statistical software, Graph Pad Instate 3. Differences between groups were considered significant at $P < 0.05$ levels.

RESULT AND DISCUSSION

Hydroalcoholic extraction of *Cnidosolus Phyllacanthus* flower was performed where the solvent was completely removed under reduced pressure and a semisolid mass was obtained %yield as 2.67 % which was brown in color and semi-solid in consistency.

The preliminary phytochemical analysis indicated a positive reaction for the presence of coumarins, flavonoids, carbohydrates and anthracene derivatives in Hydroalcoholic extract of *Cnidosolus Phyllacanthus* whereas negative reaction with proteins, amino acids, tannins, alkaloid, and sterols.

Acute inflammation was caused by injecting 0.1 ml of 1 % (w/v) carrageenan in saline into the sub-plantar region of the right hind paw of each rat. The paw volume was measured plethysmometrically at 0h, 1h, 2h, 3h, and 4h after the carrageenan injection. Edema was expressed as mean increase in paw volume relative to control animals. The percentage inhibition of edema was calculated by the following equation: % inhibition of edema= $100 (1 - V_t/V_c)$, where V_c is the edema volume in the control group and V_t is the edema volume in tested group: [Winter *et al.*, 1962].

Table No. 1: Effect of Hydroalcoholic extract of *Cnidosolus Phyllacanthus* on carrageenan induced paw edema in rats.

Groups	Paw volume (mm) (Mean±SEM)					% Inhibition
	0hr	1 hr	2 hr	3hr	4hr	
I	0.24±0.02	0.41±0.03	0.58±0.03	0.63±0.02	0.6±0.02	-
II	0.25±0.03	0.33±0.02	0.43±0.02a**	0.44±0.04a***	0.37±0.03a***	38.33
III	0.22±0.02	0.39±0.02	0.52±0.04b**	0.55±0.03a**,b**	0.43±0.03a***	28.33
IV	0.23±0.03	0.37±0.03	0.48±0.03 a**	0.53±0.03a***,b*	0.41±0.02a***	31.66
V	0.23±0.03	0.34±0.04	0.45±0.02 a**	0.5±0.01a***	0.4±0.04a***	33.33

All values are mean ± SEM, n = 6. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ a- Significance difference as compared to control (group-I). b- Significant difference as compared to standard (Group II)

In the experiment, a total of 30 rats was used. The rats were divided into 5 groups comprising of 6 animals in each group.

Group I: Control (Vehicle treated) + inject carrageenan (0.1 ml.).

Group II: Standard, treated with Diclofenac sodium (10 mg/kg, pod.) + inject carrageenan (0.1 ml.).

Group III: animals treated with Hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (100mg/kg pod.) once daily for seven days+ inject carrageenan (0.1 ml.) on 7th day

after 1h of last dose.

Group IV: animals treated with Hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (200mg/kg pod.) once daily for seven days+ inject carrageenan (0.1 ml.) on 7th day after 1h of last dose.

Group V: animals treated with hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (300mg/kg pod.) once daily for seven days+ inject carrageenan (0.1 ml.) on 7th day after 1h of last dose.

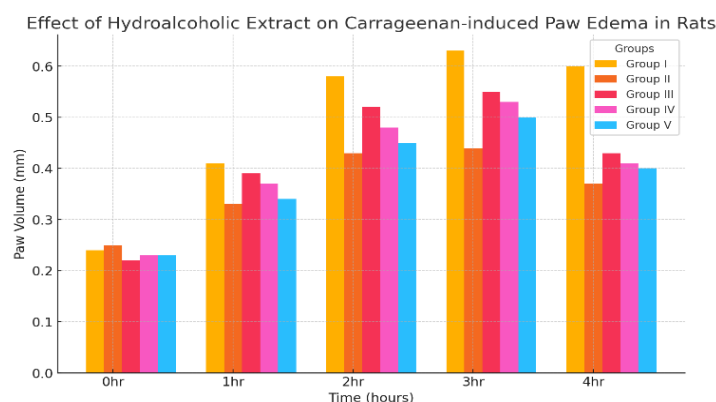


Figure 1: Effect of hydroalcoholic extract *Cnidosolus Phyllacanthus* on carrageenan-induced Paw Edema in Rats.

Observations

1. Group I (Control) showed a progressive increase in

paw volume, reaching its peak at 3 hours (0.63 mm) and slightly reducing at 4 hours (0.6 mm).

2. Group II (Standard Drug) exhibited a significant reduction in paw volume compared to the control, with maximum inhibition (38.33%) at 4 hours.
3. Groups III, IV, and V (Test Groups) showed a dose-dependent reduction in paw edema, indicating the anti-inflammatory potential of the extract:
 - Group III: Moderate reduction (28.33% inhibition)
 - Group IV: Slightly better response (31.66% inhibition)
 - Group V: Highest response among test groups (33.33% inhibition)
4. Overall Trend: The hydroalcoholic extract significantly reduced paw edema, with Group V showing results closest to the standard drug.

The hydroalcoholic extract of *Cnidosolus Phyllacanthus* exhibits anti-inflammatory properties by reducing paw edema in a dose-dependent manner. The results suggest its potential therapeutic value in inflammation management.

(ii) Effect of Hydroalcoholic extract of *Cnidosolus Phyllacanthus* on Histamine induced paw edema in rats

Paw edema was induced after 1 h by sub plantar administration of 0.1 ml histamine (1 µg/ml) on the right hand paw (Amman R *te al.*, 1995). The linear paw circumference were measured initially and after every 1 hr up to 4 h. Test groups were treated with the different doses of ethanolic extract, standard group with diclofenac sodium (10 mg/kg, *p.o*) and control group with vehicle prior to the histamine administration. The inhibition of inflammation was calculated using the following formula:

$$\text{Percentage inhibition} = (V_c - V_t) / V_c \times 100$$

Where, V_t is the paw volume of test group and V_c is the paw volume of control group

In the experiment, a total of 30 rats was used. The rats were divided into 5 groups comprising of 6 animals in each group.

Group I (Vehicle treated) + inject histamine (0.1 ml.).

Group II Standard, treated with Diclofenac sodium (10 mg/kg, *p.o.*) + inject histamine (0.1 ml.).

Group III animals treated with hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (100mg/kg *p.o.*) once daily for seven days+ inject histamine (0.1 ml.) on 7th day after 1h of last dose.

Group IV animals treated with Hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (200mg/kg *pod.*) once daily for seven days+ inject carrageenan (0.1 ml.) on 7th ay after 1h of last dose.

Group V animals treated with hydroalcoholic extract of *Cnidosolus Phyllacanthus*, (300mg/kg *pod.*) once daily for seven days+ inject carrageenan (0.1 ml.) on 7th day after 1h of last dose.

Table No. 2: Effect of Hydroalcoholic extract of *Cnidosolus Phyllacanthus* on histamine induced paw edema in rats.

Groups	Paw volume (mm) (Mean±SEM)					% Inhibition
	0hr	1 hr	2 hr	3hr	4hr	
I	0.3±0.02	0.5±0.02	0.65±0.03	0.68±0.03	0.66±0.03	-
II	0.32±0.03	0.42±0.01	0.47±0.04a***	0.48±0.02a***	0.41±0.03a***	37.87
III	0.31±0.01	0.46±0.03	0.55±0.03a**	0.6±0.02b***	0.5±0.04a***,b*	24.24
IV	0.33±0.03	0.45±0.01	0.54±0.02 a**	0.55±0.01a***	0.46±0.01a***	30.3
V	0.32±0.02	0.44±0.02	0.51±0.03a***	0.52±0.03a**	0.45±0.01a***	31.81

All values are mean ± SEM, n = 6. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, a- Significance difference as compared to control (group-I). b- Significant difference as compared to standard (Group II)

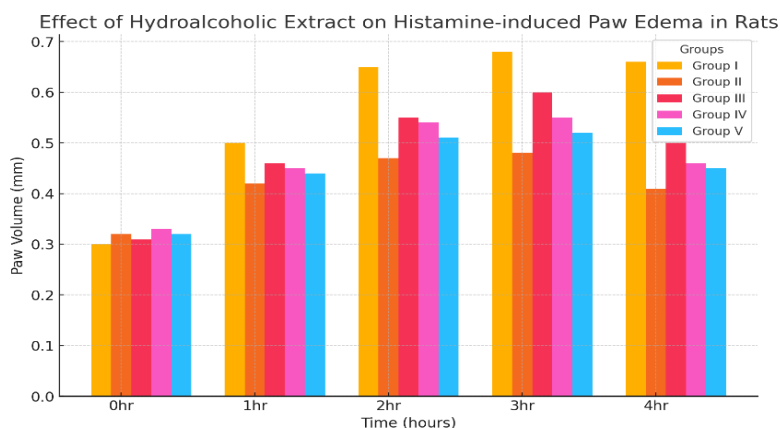


Figure 2: Effect of Hydroalcoholic extract of *Cnidosolus Phyllacanthus* on Histamine induced paw edema in rats.

Here is the bar diagram representing the effect of the hydroalcoholic extract on histamine-induced paw edema in rats.

Key Observations

1. Group I (Control) showed a steady increase in paw volume, peaking at 3 hours (0.68 mm) and slightly reducing at 4 hours (0.66 mm).
2. Group II (Standard Drug) exhibited significant inhibition of edema (37.87%), with paw volume notably lower than the control group at all time points.
3. Test Groups (III, IV, and V) demonstrated a dose-dependent reduction in paw edema, confirming the anti-inflammatory effect of the extract:
 - Group III: Moderate reduction (24.24% inhibition)
 - Group IV: Better response (30.3% inhibition)
 - Group V: Highest effect among test groups (31.81% inhibition)
4. Overall Trend: The hydroalcoholic extract significantly reduces paw edema, with Group V showing effects closest to the standard drug.

The hydroalcoholic extract of *Cnidosolus Phyllacanthus* effectively reduces histamine-induced inflammation, demonstrating anti-inflammatory potential in a dose-dependent manner.^[6,7,8]

CONCLUSION

In conclusion hydroalcoholic extract of *Cnidosolus Phyllacanthus* possess significant anti-inflammatory action as dose dependent manner. But maximum effect was observed at dose 300mg/kg. The results suggest its potential therapeutic value in inflammation management. These results support the potential traditional use of the plant in folk medicine. At present, there are no reports on investigation to identify the active components present in flower of hydroalcoholic extract of *Cnidosolus Phyllacanthus*. Further investigations are anticipated to identify the active components and lead to their further clinical use.

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