

Anti-Inflammatory activity of whole plant of *Cynoglossum Zeylanicum* (VAHL Ex Hornem) Thunb. Ex. Lehm

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Introduction

Inflammation is a bodily response to injury, infection or destruction characterized by heat, redness, pain, swelling and disturbed physiological functions. Inflammation is a normal protective response to tissue injury caused by physical trauma, noxious chemical or microbial agents. It is the body response to inactivate or destroy the invading organisms, to remove the irritants and set the stage for tissue repair. It is triggered by the release of chemical mediators from injured tissue and migrating cells. [1] The commonly used drug for management of inflammatory conditions are non-steroidal anti-inflammatory drugs, which have several adverse effects especially gastric irritation leading to formation of gastric ulcers.[2] The attention of pharmacologists throughout the world has been focused on finding out safer and potent anti-inflammatory drug. The natural products today symbolize safety in contrast to the synthetic

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ABSTRACT

In the present study, *Cynoglossum zeylanicum* whole plant was extracted with ethanol and evaluated for anti-inflammatory activity in rats using a carrageenan induced paw edema method. Ethanol extract exhibits potent anti-inflammatory activity at 150mg/Kg 3rd hr after administration is compared with reference standard drug, Indomethacin. Observed pharmacological activity in the present study provides scientific validation of ethnomedicinal use of this plant in treating acute inflammation.

Keywords: *Cynoglossum zeylanicum*; Anti-inflammatory; Edema.

drugs that are regarded as unsafe to humans and environment. So, people are returning to the natural products with the hope of safety and security.[3] Plant extracts were reported for the biological activities such as antidiabetic, hepatoprotective and antitumor activities.[4-6] However, so far there is no systematic study on anti-inflammatory activity has been reported in the literature. Hence, the present study focuses on evaluating the anti-inflammatory activity if whole plant of *Cynoglossum zeylanicum*.

Materials and Methods

Plant Material:

The whole plants of *Cynoglossum zeylanicum* (Vahl Ex Hornem) Thunb. Ex. Lehm. were collected from Kodagiri, Nilagiri Biosphere Reserve, Western Ghats, Tamil Nadu and identified by the Botanical Survey of India, Coimbatore. A voucher specimen was retained in Ethnopharmacology Unit, Research Department of Botany, V. O. Chidambaram College, Tuticorin for further reference.

Preparation of plant extract for anti-inflammatory activity:

The dried whole plants of *Cynoglossum zeylanicum* were powdered in a Wiley mill. Hundred grams of

plant powder was packed in a Soxhlet apparatus and extracted with ethanol. The ethanol extract was concentrated in a rotary evaporator. The concentrated ethanol extract was used for anti-inflammatory activity.

Animals:

Adult Wistar Albino rats of either sex (150-200g) were used for the present investigation. Animals were housed under standard environmental conditions at temperature (25±2°C) and light and dark (12:12 h). Rats were fed with standard pellet diet (Goldmohur brand, MS Hindustan lever Ltd., Mumbai, India) and water *ad libitum*.

Acute toxicity study:

Acute oral toxicity study was performed as per OECD-423 guidelines (acute toxic class method), albino rats (n=6) of either sex selected by random sampling were used for acute toxicity study (OECD, 2002). The animals were kept fasting for overnight and provided only with water, after which the extracts were administered orally at 5mg/kg body weight by gastric intubations and observed for 14 days. If mortality was observed in two out of three animals, then the dose administered was assigned as toxic dose. If mortality was observed in one animal, then the same dose was repeated again to confirm the toxic dose. If mortality was not observed, the procedure was repeated for higher doses such as 50, 100 and 2000 mg/kg body weight.

Anti-inflammatory activity

Carrageenan induced hind paw edema:

Albino rats of either sex weighing 150-200 grams were divided into four groups of six animals each. The dosage of the drugs administered to the different groups was as follows. Group I - Control (normal saline 0.5 ml/kg), Group - II, III and IV - *Cynoglossum zeylanicum* (50,100 and 150 mg/kg, p.o.), Group V - Indomethacin (10 mg/kg, p.o.). All the drugs were administered orally. Indomethacin served as the reference standard anti-inflammatory drug.

After one hour of the administration of the drugs, 0.1 ml of 1% W/V carrageenan solution in normal saline was injected into the sub plantar tissue of the left hind paw of the rat and the right hind paw was served as the control. The paw volume of the rats were measured in the digital plethysmograph (Ugo basile, Italy), at the end of 0 min., 60min., 120min., 180min., 240min., 360min., and 480min. The percentage increase in paw edema of the treated groups was compared with that of the control and the inhibitory effect of the drugs was studied. The relative potency of the drugs under investigation was calculated based upon the percentage inhibition of the inflammation.

$$\text{Percentage Inhibition} = [(V_c - V_t) / V_c] \times 100$$

Where,

V_t= Percentage difference in increased paw volume after the administration of test drugs to the rats

V_c= Difference of increased volume in the control groups

Statistical analysis

The data were analyzed using student's t-test statistical methods. For the statistical tests a *p* values of less than 0.01 and 0.05 was taken as significant.

Result and Discussion

The plant extracts did not exhibit any mortality upto the dose level of 2000mg/Kg. So, the extracts safe for long term administration. The ethanol extract of *C. zeylanicum* whole plant at the doses of 50, 100 and 150 mg/Kg decrease the edema significantly (*p*<0.001) by 66.81%, 74.72% and 80.53% respectively at 3rd h after administration of the extract (Table 1) when compared to the control group. The effect was compared to the activity produced by standard drug Indomethacin (79.57%). The dose dependent inhibition of edema was observed with ethanol extract of *C. zeylanicum* whole plant treatment.

Carrageenan-induced edema has been commonly used as an experimental animal model for acute inflammation and is believed to be biphasic. The early

phase (1 to 2h) of the carrageenan model is mainly mediated by histamine, serotonin and increased synthesis of prostaglandins in the damaged tissue surroundings. The late phase (3 h) is sustained by prostaglandin release and mediated by bradykinin, leukotrienes, polymorphonuclear cells and prostaglandins produced by tissue macrophages.[7-8] Prostaglandin-E₂, a powerful vasodilator, synergizes with other inflammatory vasodilators such as histamine and bradykinin and contributes to redness and increased blood flow in areas of acute

inflammation. The significant ($p < 0.001$) suppressive activity of the ethanol extract of *C. zeylanicum* whole plant in late phase shows its potent anti-inflammatory effect. This result is quite similar to the one observed for indomethacin at 10 mg/Kg, which inhibited 79.57%. Therefore, it is suggested that the mechanism of action of the extract may be related to histamine and prostaglandin synthesis inhibition. Further studies will be carried out to isolate and characterize anti-inflammatory chemical constituents present in the methanolic extract of this plant.

Table 1: Effect of CZW extract on the Percentage inhibition of Carrageenan induced paw oedema

Treatment Groups	Dose mg/kg	Oedema volume (ml)				% Inhibition after 180 min
		0 min	60 min	120 min	180 min	
CONTROL (Group-I)	Normal saline	24.68±1.74	88.54±1.63	104.54±2.19	126.34±1.91	-
Group-II	50 mg/kg	32.14±1.13	56.39±1.74	50.11±1.36	41.93±1.08***	66.81
Group-III	100 mg/kg	30.54±1.73	58.16±1.58	42.69±1.07	31.93±1.81***	74.72
Group-IV	150 mg/kg	31.95±1.26	65.14±2.56	30.93±1.14	24.59±1.93***	80.53
Indomethacin (Group-V)	10 mg/kg	28.55±1.17	64.93±1.18	32.84±1.93	25.81±1.33***	79.57

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