

Antiinflammatory Activity of Whole Plant of *Catharanthus pusillus* (Apocynaceae)

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ABSTRACT

This study was intended to evaluate the antiinflammatory activity of the whole plant of *Catharanthus pusillus*. The antiinflammatory activity was carried out by using carrageenan induced paw edema. The ethanol extract of whole plant of *Catharanthus pusillus* was administered at different doses such as 150, 300 and 500 mg/kg body weight and the study was compared with standard drug indomethacin (10mg/kg). The extract exhibited significant anti-inflammatory activity, which supports the traditional medicinal utilization of this plant.

Keywords: *Catharanthus pusillus*, Anti-inflammatory, Carrageenan

INTRODUCTION

Inflammation is a complex process, which is frequently associated with pain and involves occurrences such as, the increase of vascular permeability, increase of protein denaturation and membrane alteration. When cells in the body are damaged by microbes, physical agents or chemical agents, the injury is in the form of stress. Inflammation of tissue is due to response to stress. It is a defensive response that is characterized by redness, pain, heat, and swelling and loss of function in the injured area. Loss of function occurs depends on the site and extent of injury. Since inflammation is one of the body's nonspecific internal systems of defense, the response of a tissue to an accidental cut is similar to the response that results from other types of tissue damage, caused by burns due to heat, radiation, bacterial or viral invasion¹. The attention of pharmacologist throughout the world has been focused on finding out safer and potent antiinflammatory drug. The natural products today symbolize safety in contrast to the synthetic 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. *Catharanthus Pusillus* belonging to family Apocynaceae is known with various names in India and all over the world. It is widely used as various treatments of diseases and traditionally used as herbal medicine². The roots, leaves and latex of these plants are used to treat skin and liver diseases, leprosy, dysentery, worms, ulcers, tumor and ear aches. The leaf powder of *C. pusillus* were mixed with coconut oil and used to treat the antidandruff activity and also used to kill the lice³. However, so far there is no systematic study on antiinflammatory activity of *C. pusillus* has been reported in the literature. Hence the present study focuses on evaluating the antiinflammatory

activity of whole plant of *Catharanthus pusillus*. To our knowledge no report on the effect of this plant on experimental inflammation. This study was therefore undertaken to evaluate the effect of ethanol extract of the whole plant of *Catharanthus pusillus* on antiinflammatory activity in carrageenan induced rat paw edema.

MATERIALS AND METHODS

Collection of plant material

The whole plant of *Catharanthus Pusillus* were collected from Pechiparai, Kanayakumari District, Tamil Nadu. With the help of local flora, voucher specimens were identified and preserved in the Ethnopharmacology unit, Research department of Botany, V.O.Chidambaram College, Thoothukudi, Tamil Nadu for further references.

Preparation of plant extract for antiinflammatory activity

The dried whole plant material of *Catharanthus Pusillus* was powdered in a Wiley mill. Hundred grams of whole 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 antiinflammatory 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 was performed by following OECD-423 guidelines (acute toxic class method), albino rats (n=6) of either sex selected by random sampling were used for acute toxicity study⁴ (OECD, 2002). The animals were

Table 1: Effect of CP extract on the Percentage inhibition of Carrageenan induced paw edema

Treatment Groups	Dose mg/kg	Edema volume (ml)				% Inhibition after 180 min
		0 min	60 min	120 min	180 min	
CONTROL (Group-I)	Normal saline	31.15±1.18	73.18±2.16	98.18±2.50	138.60±2.94	
Group-II	150 mg/kg	30.60±1.17	61.60±1.96ns	58.16±1.33**	40.16±1.65***	71.02
Group-III	300 mg/kg	32.50±1.68ns	56.15±1.35*	44.50±0.98***	26.15±1.26***	81.13
Group-IV	500 mg/kg	36.10±1.50*	40.50±2.16***	31.60±1.12***	20.15±1.36***	85.46
Indomethacin (Group-V)	10 mg/kg	31.40±1.65ns	39.60±1.30***	30.80±1.56***	19.84±1.30***	85.68

Each Value is SEM ± 5 individual observations * P < 0.05 ; ** P<0.01 *** P<0.001, Compared paw edema induced control vs drug treated rats

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 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.

Antiinflammatory 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)

Group II - Ethanol extract of *C. Pusillus* whole plant 150 mg/kg body weight.

Group III- Ethanol extract of *C. Pusillus* whole plant at 300mg/kg body weight.

Group IV- Ethanol extract of *C. Pusillus* whole plant at 500mg/kg body weight.

Group V – Indomethacin (10 mg/kg, p.o).

All the drugs were administered orally. Indomethacin served as the reference standard antiinflammatory 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. Percentage inhibition was calculated using the formula;

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

Where, V_t the percentage represents the percentage difference in increased paw volume after the administration of test drugs to the rats and V_c represents 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 0.001, 0.01 and 0.05 was taken as significant

RESULTS AND DISCUSSION

The plant extract did not exhibit any mortality upto the dose level of 2000 mg/kg. So the extract safe for long term administration. The present study of antiinflammatory activity of ethanol extract of *C.pusillus* against carrageenan induced paw edema shows that the extracts have significant effect on inflammation and markedly reduced the swelling. The percentage reduction in the paw volume in the group of animals treated with *C.pusillus* extract 150mg was 71.02% ,300mg/kg was 81.13% and for the 500mg/kg was 85.46% at 3 hours. It shows that the plant extract have significant ($P < 0.001$) antiinflammatory effect and the results were compared with indomethacin 10mg/kg and show percentage paw volume reduction of 85.68 %. The acute model of inflammation, upon challenge by phlogistic stimuli, ethanol extract of *C.pusillus* whole plant showed significant ($P < 0.001$) anti-inflammatory activity. The edema and inflammation induced by carrageenan is shown to be mediated by histamine and 5-HT during 1h, after which increased vascular permeability is maintained by the release of kinins upto 3h, the mediators appear to be prostaglandins, the release of which is closed associated with the migration of leucocytes into the inflamed site^{5,6}. The carragennan induced paw edema model in rats is known to be sensitive to cyclooxygenase(COX) inhibitors and has been used to evaluate the effect of non-steroidal and anti-inflammatory agents⁷. Therefore our results suggest that the inhibitory effect of the ethanol extract of *C.pusillus* whole plant of carrageenan induced paw edema may be due to the suppression of the release of mediators including histamine, resotonin, bradykinins and prostaglandins responsible for the first and the second phase of acute inflammation induced by carrageenan. These are also evidences that compounds inhibiting the carrageenan induced edema are effective in inhibiting the enzyme cyclooxygenases⁸. Based on these reports, the inhibitory effect of *C.pusillus* extract on carragennan induced inflammation could be mediated via this mechanism. According to the present study, it can be concluded that the ethanol extract of *C.pusillus* whole plant possesses

antiinflammatory effect. Further investigations are required to isolate the active principles present in the extract and to determine their exact mechanism of action.

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