ANTI-INFLAMMATORY ACTIVITY OF *NYCTANTHES ARBOR-TRISTIS* FRUIT IN RODETNS

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Summary

Water soluble fraction of ethanol extract of fruits of *Nyctanthes arbor-tristis* Linn (NATEF) was pharmacologically validated for its anti-inflammatory properties in experimental animals using various models such as Carrageenan-induced paw oedema and Cotton pellet induced granuloma in rats for anti-inflammatory activity. Extract was given orally at two different dose levels (250 and 500 mg/kg) once daily for three consecutive days, while indomethacin was administered as positive control. Studies have shown that the fruit extract has activity to prevent inflammation in the rodents. The anti-inflammatory effect was dose dependent and found to be statistically significant as compared to the control.

Keywords: Anti-inflammatory, *Nyctanthes arbor-tristis*

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Introduction

Inflammation is defined as local response of living mammalian tissue to injury due to any agent. Inflammation manifests usually in form of painful swelling associated with some changes in skin covering the site (1). Since his existence on planet, man has been dependent on nature for curing various body diseases. From ancient civilization various parts of different plants were used to eliminate pain, control suffering and counteract disease. Most of the drugs used in primitive medicine were obtained from plants and are the earliest and principal natural source of medicines (2, 3, and 4). *Nyctanthes arbor-tristis*, (Fam. Oleaceae) is commonly known as Parijatham, Harsinghar and Night Jasmine. The leaves, flowers, seeds and bark of *Nyctanthes arbor-tristis* are widely used in traditional remedies and folkloric medicines in India. Widely distributed throughout India and also cultivated in gardens for its fragrant flowers (5, 6). The 50% ethanolic extract of the seeds, leaves, roots, flowers and stem of the plant has been proved to posses antiamoebic (7) and anti allergic properties (8).The arbortristoside A isolated from the seeds found to have antitumor activity (9). Many iridoid glycosides have been isolated from the leaves and seeds of the plant. These include arborside A, arborside B and arborside C (10). The seeds are used as anthelmintics and in alopecia. It is antibilious and an expectorant, and is also useful

in bilious fevers (11). The powdered seeds are used to cure scurfy affections of scalp, piles and skin diseases (12). Earlier, we have reported the anxiolytic activity (13), antidepressant activity (14) and anti-aggressive activity (15) of leaf extract and analgesic activity of fruit extract (16) of *Nyctanthes arbor- tristis*. The present study was undertaken to evaluate the anti-inflammatory activity in fruit extract of *Nyctanthes arbor- tristis.*

Material and Method

Preparation of plant extracts

The fruits of *Nyctanthes arbor-tristis* were collected from the local garden of Lucknow, India. The plant material was identified and authenticated taxonomically at National Botanical Research Institute, Lucknow. A voucher specimen (LWG accessions No. 94392) of the collected sample was deposited in the institutional herbarium for future reference. The powdered fruits of *Nyctanthes arbor-tristis* (5 kg) were passed through S.S. sieve (20 mesh) before extraction. Plant material was successively extracted with ethanol (50%) in soxhlet apparatus. The crude extract obtained was concentrated in a rotary evaporator under reduced pressure and freeze dried to yield 10.8% w/w. Water soluble fraction of this extract (NATEF) was taken for the study.

Animals

Adult albino rats (150-180g) and Wister mice (25-35g) of either sex were obtained form the Animal House of the Institute and were randomly distributed into different experimental groups. The rats were housed in groups of six in polypropylene cages at an ambient temperature of 25 ± 10^0C and 45-55% RH with a 12:12 h light /dark cycle. Animals were provided with commercial food pellets and water ad libitum. All studies were performed in accordance with the guide for the care and use of laboratory animals.

Drug treatment

In the acute toxicity study no deaths were observed during the period at the doses tested up to 2000 mg/kg. Hence, the NATEF was administered orally at two different dose levels (250 and 500 mg/kg) once daily for three consecutive days. Control group of animals received suspension of 1% CMC in distilled water. Standard drug was administered intraperitoneally to rodents 30 min. before experiments for comparison. Experiments were conducted on day 3, one hour after the last drug administration.

Safety evaluation

NATEF was administered to 10 mice and 10 rats in a dose of 2g/Kg p.o. and observations were made for gross behavioral changes such as locomotion, rearing, respiration, tremors, passivity, righting reflex, lacrimation and mortality for 14 days (17).

Assessment of anti-inflammatory activity

The two most widely used rodent models were chosen to evaluate the effect of NATEF on anti-inflammatory behavior such as, Carrageenan-induced paw oedema and Cotton pellet induced granuloma in rats.

Carrageenan-induced paw oedema in rats

Male albino rats were injected with 0.1 ml of a 1% carrageenan solution in saline into the sub-planter region of the left hind paw. The paw was marked with ink at the level of the lateral malleolus and immersed in mercury up to this mark. The paw volume was measured before and 1, 2, 3, 4, and 6 h after the injection of carageenan by the mercury displacement method plethysmographically. The oedema volume was determined and expressed as percentage swelling, compared with the initial hind paw volume of each rat (18).

Cotton pellet induced granuloma in rats

Sub acute inflammation was produced by cotton pallet induced granuloma in rats. Sterile cotton (50+1 mg) soaked in 0.2 ml of distilled water containing penicillin (0.1mg) and streptomycin (0.13 mg) was implanted subcutaneously bilaterally in exilla under the ether anesthesia. The animals were sacrificed and then dry weight was taken. The weight of the cotton pallet before implantation was compared from the weight of the dried, dissected pellets (19).

Statistical analysis

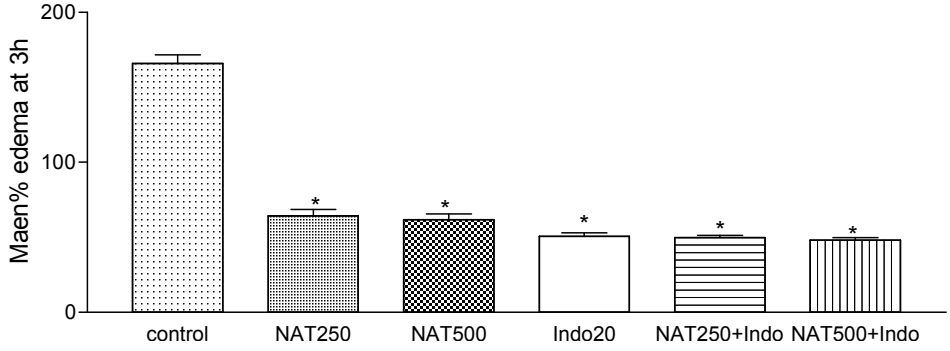
The values were represented as mean ± S.E.M. for six rats. Analysis of variance (ANOVA) test was followed by individual comparison by Newman–Keuls test using Prism Pad software for the determination of level of significance.

Result and Discussion

Acute inflammatory response is characterized by an increase in vascular permeability and cellular infiltration leading to oedema formation, as a result of extravasation of fluid and proteins and accumulation of leukocytes at the inflammatory site. Carrageenan-induced rat paw oedema is a widely used test to determine the anti-inflammatory activity, and it has been fully characterized in the past (20, 21, 22, 23).This study demonstrated that NATEF extract was effective in animal models of acute inflammation (Figure-1). Among the many methods used for screening of anti-inflammatory drugs, Carrageenan-induced paw oedema is widely used for determining the acute phase of the inflammation. Histamine, 5-hydroxytryptamine and bradykinin are the first detectable mediators in the early phase of carrageenan-induced inflammation (24), whereas prostaglandins are detectable in the late phase of inflammation (25). In the cotton pellet induced granuloma models of sub-acute inflammation, NATEF extract significantly reduced the weight of granulation tissue and potentiated the anti-inflammatory activity of indomethacin (Figure-2). This method has been useful for evaluation of steroidal and non-steroidal anti-inflammatory drugs (26). It has been considered that cotton pellet induced granuloma is closely related to the formation of antibodies. The water soluble fraction of

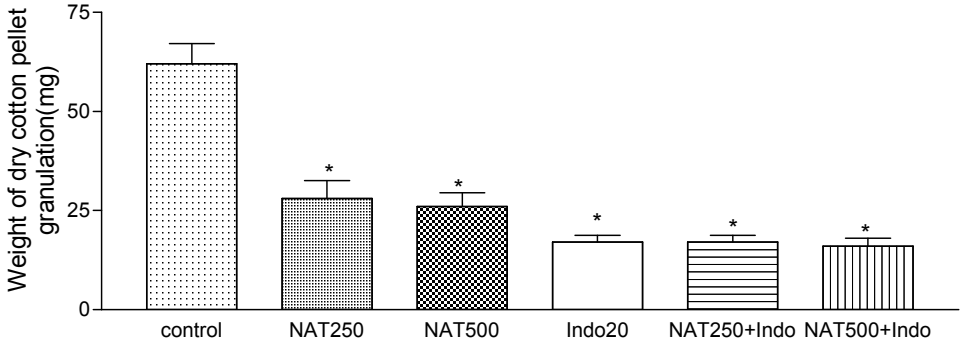
ethanolic extract of *Nyctanthes arbor-tristis* at a dose level of 250 and 500 mg/kg showed a significant inhibitory effect on granuloma formation. This study revealed that the extract was active against the inflammation induced by a foreign body however this effect of extract was less pronounced than that of indomethacin.

Figure 1: Effect of *Nyctanthes arbor - tristis* Fruit Extract on Carrageenan Induced Acute Paw Oedema in Rats



Data are given as mean ± S.E.M. (n = 12). $^{*}P < 0.001$ compare to control

Figure 2: Effect of *Nyctanthes arbor - tristis* Fruit Extract on Cotton Pellet Induced Granuloma in Rats.



Data are given as mean ± S.E.M. (n = 12). $^{*}P < 0.0001$as compared to control

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