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RESEARCH ARTICLE

EVALUATION OF ANTI-INFLAMMATORY ACTIVITY ON VITEX NEGUNDOLINN

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ABSTRACT

The anti-inflammatory activity of *Vitex Negundo*Linn was studied in carrageenan induced paw edema & cotton pellet granuloma in albino rats. The anti-inflammatory activity of the Ethanolic and aqueous extracts (100 & 200mg/kg) were evaluated against the carrageenan induced paw edema of rats were compared with the standard drug Indomethacin. The extract at the test doses of 100 and 200 mg/kg body weight reduced the paw edema induced by Carrageenan by 48.13% and 57.18% respectively, where as the standard drug showed 61.16. The paw edema suppressant effect was significant (P < 0.05) in the dose of 100&200mg/kg ethanol and aqueous extracts. When compared between control and two extracts, Ethanolic extract shows maximum anti-inflammatory activity. Thus it can be concluded that the extract of *Vittex Negundo*. Linn extract posses the anti-inflammatory activity. **Keywords:** Anti-inflammatory activity, *Vittex Negundo*, paw edema, ethanolic extract, Cotton pellet granuloma.

INTRODUCTION

Vitex Negundo L. (Verbenaceae) is a hardy plant, flourishing mainly in the Indian subcontinent. All parts of the plant from root to fruit possess a multitude of phytochemical secondary metabolites which impart an unprecedented variety of medicinal uses to the plant¹⁻². It is interesting to note that a single plant species used for treatment of a wide spectrum of health disorders in traditional and folk medicine, some of which have been experimentally validated³⁻⁴. The plant is a component of a number of commercially available herbal formulations and has also shown potential as an effective bio-control agent⁵. Employment of techniques such as cell and tissue culture would provide means of rapid propagation, conservation of the plant species, give scope for enhancement of the quality and quantity of the bioactive secondary metabolites occurring in the plant⁶. It is frequently used in folk medicine to treat antiinflammatory, antiarthritis, antioxidant, pesticidel & insecticidal activity, bronchial smooth muscle relaxant activity, antirumatic activity, hepatoprotective, fertility, anti malarial, anti bacterial, filarial, laxative activity, and analgesic activity⁷⁻¹⁰.

MATERIALS AND METHODS

Plant Material

The plant *Vitex Negundo* is collected at the month of septemberin Tirupathi, Chittoordist, A.P. The plant was authenticated by Dr. USHA RANI, Professor of Botany, B.T. College, Madanapalle and the plant specimen herbarium reference no 02/2011 preserved in Sri Krishna Chaithanya College of Pharmacy for future references.

Drugs and reagents

Indomethacin [Marketed Product], n-Hexane, Ethanol, Chloroform, Glacial acetic acid were used in this study.

Preparation of Plant Extracts

The plant *VitexNegundo*. Linnis dried under shade air and coarsely powdered (1kg), extracted by hot continuous extraction for 72hr. The extract is concentrated in vacuum. The extracts are done with solvents [Hexane, Ethyl acetate, Chloroform & Ethanol]. The extract is studied for anti-inflammatory activity¹¹⁻¹² with ethanolic extract of *V.Negundo*Linn, using albino rats of either sex.

Animals

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Adult wistar albino rats (180-230 gms) of either sex are used for pharmacological activities. For this experiment the animals were kept in poly propylene cages (3 per cage) at 25±2°C with relative humidity 45-55% under 12 hr light and dark cycles. All the animals were acclimatized to laboratory condition for a week before use. They were fed with standard animal feed and water *ad libtum*.

Anti-inflammatory activity

Carrageenan Induced Paw Edema

Anti-inflammatory activity was evaluated by using the Carrageenan induced rat paw edema method¹³⁻¹⁴. After 16 hr of fast the rats were divided into four groups of three each.

Group I - served as control group, received 1% CMC w/v.

Group II - served as standard, received Indomethacin (10 mg/kg)

Group III - served as test, received VNE (100 mg/kg)

Group IV - served as test received VNE (200 mg/kg)

After one hour the respective treatment 0.1 ml of 1% w/v Carrageenan suspension was injected subcutaneously into the plantar surface of the right hind paw. The paw volume was measured by using a plethysmometer immediately after 1, 2, 3, 4 h of carrageenan treatment. The anti-

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inflammatory effect of VNE was calculated by following equation ¹⁵.

The anti-inflammatory (%) inhibition = $[(A-B)/A] \times 100$ (ie) = $[(1-B/A)] \times 100$

Where,

A- Represents the paw volume of control group. B- Represents the paw volume of treated group.

Cotton pellet granuloma

Albino rats of either sex with an average weight of 200 g were anaesthetized with ether. The back skin is shaved and disinfected with 70% ethanol. An incision is made in the lumbar region by using a blunted forceps, subcutaneous tunnels are formed and a sterilized cotton pellet 10 mg is placed on both sides in the scapular region. The cotton pellets inserted rats was randomly divided into 4groups of (n=3 rats per group)¹⁶.

Group I - served as control group, received 1% CMC $\mbox{w/v}.$

Group II - served as standard, received Indomethacin (10 mg/kg)

Group III - served as test, received VNE (100 mg/kg)

Group IV - served as test received VNE (200 mg/kg)

On the 8th day cotton pellets was removed, weighed and dried at 60°C for 6 hr.

Then dry weight was calculated after deducting cotton pellet weight was taken as granuloma tissue formation.

The anti-inflammatory (%) inhibition = [(A-B)/A]×100 (ie) = [(1-B/A)] ×100 Where,

A- Represents the wet weight of the cotton pellet.

B- Represents the dry weight of the cotton pellet.

Statistical Analysis

The results were presented as Mean \pm S.E.M. One way analysis of variance (ANOVA) was followed by Dunnett'st-test for multiple comparisons statistical evaluation. P-values less than 0.05 were considered significant $^{17-19}$.

RESULT AND DISCUSSION

The result of anti-inflammatory activity by Carrageenan induced paw oedema method was tabulated in Table No-1. The extract found to have significant (P < 0.05) antiinflammatory activity in rats. The extract at the test doses 100 and 200 mg/kg body weight reduced the oedema induced by Carrageenan by 48.13% and 57.18 % respectively, where as the standard drug showed61.16% 5 h in table no.1 and figure 1. Thus it can be concluded that the extract of VittexNegundo.Linnextract posses the antiinflammatory activity. It was well known that Carrageenan induced paw edema was characterized by biphasic event with involvement of different inflammatory mediators. In the first phase chemical mediators such as histamine and serotonin play role, while in second phase kinin and prostaglandin were involved. In the present study, results revealed that administration of ethanolic extract of VittexNegundo.Linnreduced the edema starting from the first hour and during all phases of inflammation. The result of anti-inflammatory activity by cotton pellet method was Table Notabulated in 2 and figure

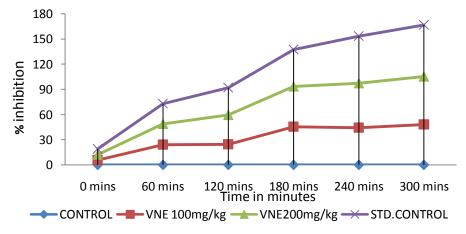


Figure 1: Effect of Vittex Negundo. Linn extract in Carrageenan induced paw oedema in rats

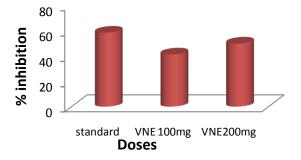


Figure 2: Effect of ethanol extract of VittexNegundo.Linnin cotton pellet granuloma

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Carageenan induced paw oedema (volume in ml) Drug Dose 300 min 0 hr60 min 120 min 180 min 240 min **Control** 0.317 ± 0.0084 0.35 ± 0.0057 0.38 ± 0.0020 0.403 ± 0.0033 0.386 ± 0.003 0.376 ± 0.003 100 0209 ± 0.006 0.266 ± 0.003 0.293 ± 0.003 0.22 ± 0.0057 0.217 ± 0.003 0.195 ± 0.002 **VNE-100** mg/kg (24.67)(45.5)(48.13)(5.9)(24)(44.3) 0.22 ± 0.003 0.263 ± 0.008 0.253 ± 0.003 0.21 ± 0.005 0.181 ± 0.004 0.161±0.007 200 **VNE-200** mg/kg (6.3)(24.85)(34.96)(47.9)(53.1)(57.18)10 0.205 ± 0.003 0.266 ± 0.008 0.263 ± 0.003 0.226 ± 0.006 0.17 ± 0.005 0.146 ± 0.003 STANDARD mg/kg (6.8)(24)(32.39)(43.9)(55.95)(61.16)

Table 1: Effect of VittexNegundo. Linn extract in Carrageenan induced paw oedema in rats (volume in ml)

Table 2: Effect of VittexNegundo.Linn extract in cotton pellet granuloma

Group	Granuloma in mg.		% inhibition	
	Wet weight	Dry weight	For wet weight	For dry weight
Control	135±0.5774	58.33±1.453	-	-
STD	61± 3.055	24± 2.404	54.81	58.62
VNE-100	77.66 ± 3.844	34.66 ± 1.45	37	41.37
VNE-200	69 ±3.214	29 ± 2.309	48.88	50

Effect of VittexNegundoLinnon various biochemical changes in cotton pellet granuloma model VNE treatment significantly decreased the increased level of lysosomal enzymes activity with which transaminase (namely SGOT, SGPT) and alkaline phosphatase. There was an increase evidence that lysosomal enzyme play an important role in development of acute and chronic inflammation ¹⁸. VNE in cotton pellet granuloma,Indomethacin was thestandard drug elucilated inflammatory effect on both excudatory and granulatory phases of Indomethacin. The VNE treated animals inhibited both excudatory as well as granulatory phases of inflammation the degree of anti-inflammatory activity was dose dependent at 100 and 200 mg/kg body weight. Thus the inhibitory activity of VNE shows comparatively significant as that of standard. Most of the anti-inflammatory drugs exerts their beneficial effects by inhibitory either release of lysosomal enzyme or by stabilising the lysosomal membrane which is one of the major event responsible for anti-inflammatory process.

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CONCLUSION

The VittexNegundo Linn extract found to have significant (P < 0.05) anti-inflammatory activity in rats.In the present study, the results revealed that administration of ethanolic extract of VittexNegundo.Linninhibited by edema starting from the first hour and during all phases of inflammation. VNE in cotton pellet granuloma the standard Indomethacin elucilated inflammatory effect on both excudatory and granulatory phases of Indomethacin. The VNE treated orally inhibited both excudatory as well as granulatory phases of inflammation, the degree of anti-inflammatory activity was dose dependent at 100 and 200 mg/kg body weight. Thus the inhibitory activity of VNE shows comparatively significant as that of standard. The results of present study indicates that anti-inflammatory activity further studies on Vittex Negundo. Linn with varying dose level will establish its therapeutic value as well as Mechanism of Action.

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