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Research Article

Antiinflammatory Activity of Stem and Leaf of *Myxopyrum serratulum* A.W. Hill (Oleaceae)

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ABSTRACT

In the present study antiinflammatory activity of ethanol extracts of stem and leaf of *Myxopyrum serratulum* were evaluated by carrageenan induced rat paw edema model in rats. Preliminary phytochemical analysis of ethanol extracts of stem and leaf showed the presence of alkaloid, catechin, coumarin, flavonoid, tannin, saponin, steroid, phenol, glycoside, terpenoid and xanthoprotein. Maximum inhibition (86.09%) was obtained at the dose of 400 mg kg^{-1} of *M. serratulum* stem after 3 hours of drug treatment in carrageenan induced paw edema, whereas, indomethocin produced 85.37% of inhibition. The present study suggests that *M. serratulum* stem and leaf extracts possess strong antiinflammatory property so it has immense scope as an effective source to develop drug for the treatment of inflammatory related diseases.

Keywords: *Myxopyrum serratulum*, carrageenan, GC-MS, inflammation.

INTRODUCTION

Inflammation is a normal, protective response to tissue injury caused by physical trauma, noxious chemicals or microbiological agents. There are two types of inflammation which are acute inflammation and chronic inflammation. Acute inflammation is associated with increased vascular permeability, capillary infiltration and emigration of leukocytes. Chronic inflammation is associated with infiltration of mononuclear immune cells, macrophages, monocytes, neutrophils, fibroblast activation, proliferation and fibrosis[1]. The classic signs of inflammation are local redness, swelling, pain, heat and loss of function[2]. It is believed that current drugs available such as opoids and non-steroidal antiinflammatory drugs (NSAIDS) are not useful in all cases of inflammatory disorders, because of their side effects and potency[3]. The study of plants that have been used traditionally for curing inflammation is still fruitful and logical research strategy in the source of new antiinflammatory drugs[4]. Research on the biological activities of plants during the past two centuries has yielded compounds for the development of modern drugs[5]. *Myxopyrum serratulum* (Oleaceae) commonly known as "Chaturamulla" is a large woody climbing shrub. The leaves are astringent, acrid, sweet, thermogenic, anodyne, febrifuge and tonic. They are useful in vitiated conditions of kapha and vata, cough, asthma, rheumatism, cephalalgia, nostalagia, consumption, fever, otopathy, neuropathy and cuts and wounds[6]. Taking into consideration of the medicinal importance of this plant, the ethanol extracts of stem and leaf of *M. serratulum* were evaluated for their antiinflammatory activity. However, no data are available in the literature on the antiinflammatotry activity of *M. serratulum*. This study was therefore undertaken to evaluate the effect of ethanol extracts of stem and leaf of *M. serrulatum* on antiinflammatory activity in carrageenan induced rat paw edema.

MATERIALS AND MEDTHODS

The stem and leaf of *Myxopyrum serratulum* were collected from Pechiparai, Kanyakumari District, Tamil Nadu. The collected samples were cut into small fragments and shade dried until the fracture is uniform and smooth. The dried plant material was granulated or powdered by using a blender and sieved to get uniform particles by using sieve No. 60. The final uniform powder was used for the extraction of active constituents of the plant material.

Preparation of plant extract for antiinflammatory activity

The dried stem and leaf of *Myxopyrum serratulum* were powdered in a Wiley mill. Hundred grams of stem and leaf 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 preliminary phytochemical screening and 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 ± 2^0C) 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

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Table 1: Effect of *Myxopyrum serratulum* stem and leaf extracts on the Percentage inhibition of Carrageenan induced paw edema

Treatment Groups	Edema volume (ml)					% Inhibition after 180 min
	Dose mg/kg	0 min	60 min	120 min	180 min	
Group-I	Normal saline	53.18±1.64	92.65±2.84	121.64±3.91	154.88±4.15	-
Group-II	100mg/kg	43.81±0.98	42.16±1.92**	38.22±2.16***	36.13±1.62***	76.67
Group-III	200 mg/kg	39.16±1.13	38.21±1.16***	35.21±1.08***	32.65±1.93***	78.91
Group-IV	400 mg/kg	35.84±1.36	28.16±0.91***	23.16±0.96***	21.54±1.52***	86.09
Group-V	100mg/kg	40.54±1.36*	58.16±1.56**	46.84±1.91***	31.62±1.16***	78.36
Group-VI	200 mg/kg	38.16±1.39*	50.63±1.39**	39.27±1.67***	26.18±1.94***	82.08
Group-VII	400 mg/kg	34.84±0.93**	43.16±1.18**	34.16±1.13***	22.56±1.39***	84.56
GroupVIII	10 mg/kg	36.86±1.92	27.15±0.84***	24.16±0.74***	22.65±1.16***	85.37

Each Value is SEM ± 6 individual observations * P < 0.05 ; ** P<0.01 *** P<0.001,Compared paw edema induced control vs drug treated rats

(n=6) of either sex selected by random sampling were used for acute toxicity study[7]. The animals were kept fasting for overnight and provided only with water, after which the extracts were administrated 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 administrated 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 six weighing 150-200 grams were divided into eight 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 – ethanol extract of *M. serratulum* stem (100 mg/kg, 200 mg/kg and 400 mg/kg, p.o.), Group V, VI and VII – ethanol extract of *M. serratulum* leaf (100 mg/kg, 200 mg/kg and 400 mg/kg, p.o.) and Group VIII- 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;

Percentage inhibition = $[(Vc-Vt)/Vc] \times 100$

Where, Vt the percentage represents the percentage difference in increased paw volume after the administration of test drugs to the rats and Vc 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

The photochemical screening of ethanol extracts of stem and leaf of *M. serratulum* revealed the presence of alkaloid, catechin, coumarin, flavonoid, tannin, saponin, steroid, phenol, glycoside, terpenoids and xanthoprotein. Acute toxicity revealed the non-toxic nature of the ethanol extracts of stem and leaf of *M. serratulum*. In the present study, the antiinflammatory activity of ethanol extracts of stem and leaf of *M. serratulum* were assayed in albino rats using carrageenan induced rat paw edema method. Table 1 shows the antiinflammatory activity of ethanol extracts of stem and leaf of *M. serratulum* significantly inhibited the rat paw edema at 3rd hour post carrageenan were 76.67%, 78.91% and 86.09% for 100 mg kg^{-1}, 200 mg kg^{-1} and 400 mg kg^{-1} of stem extract and 78.36%, 82.08% and 84.56% for 100 mg kg^{-1}, 200 mg kg^{-1} and 400 mg kg^{-1} of leaf extract respectively. The results were compared with indomethacin of 10 mg/kg, which shows paw reduction of 85.37%.

DISCUSSION

Carrageenan-induced inflammation is useful to detect antiinflammatory agents[8]. The development of edema in the paw of the rat has been described by Vinegar[9] as a biphasic event. The initial phase is atributed to the release of histamine and serotonine[10]. The second, accelerating, phase of swelling is due to release of prostaglandin like substance[9]. It has been reported that the second phase of edema is sensitive to both clinically useful steroidal and non-steroidal antiinflammatory agents[9,8] and they are related to COX inhibition, specially COX-2. Ethanol extracts of stem and leaf of *M. serratulum*, at a dose of 400 mg/kg, i.p. showed antiinflammatory activity comparable to that induced by indomethacin.

5-Hydroxymethylfurfural, Pyrazolidine-3,5-dione, 4-phenyl-, Methyl trans-4-methylcinnamate, 4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol, 3,7,11,15-Tetramethyl-2-hexadecen-1-ol, 1-(2-Hydroxy-ethyl)-2-

methyl-1H benzoimidazole-5-carboxylic acid methyl ester, Isoquinoline, 1,2,3,4-tetrahydro-1-allyl-6,7-dimethoxy-3,3-dimethyl-, Phytol, 9,12-Octadecadienoic acid (Z,Z)-, 9-Octadecenoic acid, (E)-, Oleic Acid, Betulin, Quinoline, 3-dodecyl-2-methyl-4-[(4methoxyphenyl)methoxy]-, dl-α-Tocopherol, 9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, acetate, (3β,4α,5α)-, 1H-Indol-4-ol, Phytol, acetate, Linoleic acid ethyl ester, Stigmast-4-en-3-one, 9,10-Secocholesta-5,7,10(19)-triene-3,25,26-triol, (3β,5Z,7E)-, were reported in the ethanol extracts of *M. serratulum* by GC-MS analysis this compounds may have the role in antiinflammatory effect. Further study will be carried out to isolate and characterize other antiinflammatory chemical constuents present in the extract of the plant.

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