



EVALUATION OF ANTI-INFLAMMATORY AND ANTI-ARTHRITIC ACTIVITY OF KADAMBUR HERBAL VATHA THAILAM

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ABSTRACT

The phytoconstituents obtained from the Kadambur Herbal Vatha Thailam resulted in the identification of volatile oils, fixed oils, alkaloids, tannins, saponins, carbohydrates, flavonoids, phytosterols, phenols, has been proposed. The analysis of the essential oil has also revealed that the presence of various monoterpenoids and sesquiterpenoids. the essential oil, alkaloids, flavonoids, tannins of Kadambur Herbal Vatha Thailam demonstrated a dose dependant anti-inflammatory activity in carrageenan induced rat paw oedema. It has also revealed good activity in cotton-pellet granuloma and adjuvant induced arthritis in model rats.

KEYWORDS: Anti-arthritis activity, anti-inflammatory activity, Essential oils, Flavonoids.

INTRODUCTION

Kadambur Herbal Vatha Thailam was procured as a gift sample from Agasthiar Herbal Physiotherapy Centre, Thanjavur. In this Kadambur herbal Vatha Thailam, mainly contains *Clerodendrum phomides*, *Vitex negundo*, *Delonix elata*, *Cissus quadrangularis*, *Cardiospermum helicabum*, those above are reported to be used as remedial measure against arthritis, anti-inflammatory^[1], rheumatism^[2], swelling^[3], anthelmintic^[4] and anti-spasmodic^[5] activities. Those activities are shown due to the constituent as volatile oil, fixed oil, saponin, flavonoids, tannins are mainly used. Kadambur herbal vatha thailam has been undertaken in present study for its preliminary phytochemical analysis and evaluation for anti-inflammatory and anti-arthritis studies.

MATERIALS AND METHODS

Preparation of Freund's adjuvant^[6]

Kadambur herbal Vatha Thailam was procured as a gift sample from Agasthiar Herbal Physiotherapy Centre, Thanjavur. 25mg of heat killed *Mycobacterium tuberculosis* cell (Being killed at 60°C in 15-20 minutes in the autoclave) was finely ground using a mortar and pestle sufficiently liquid was added thoroughly mixed to make 5mg/ml suspension. This is referred to as Complete Freund's adjuvant. The liquid paraffin is referred in the study of incomplete Freund's. All experimental protocols were reviewed and accepted by the Institutional Animals Ethics Committee (IAEC) No: 265/CPCSEA prior to the initiation of the experiment.

Induction of Adjuvant induced arthritis

Male wistar albino rats weighing about 200-250gm were divided into 6 groups of 6 animals each. The dosage of drugs administered to different groups were as follows, Group 1 serves as control, and received 0.5ml normal saline, Group 2 received 0.5ml (200mg/kg) of kadambur Herbal Vatha Thailam, Group 3 received indomethacin (2.5mg/kg). All drugs were administered orally. Adjuvant arthritis was induced by subcutaneous injection of Complete Freund's adjuvant (0.1ml) into plantar tissue of the left hind paw of each rat. Rats in this group were known as inflamed control group. The non-inflamed control group consists of rats injected with 0.1ml of liquid paraffin. The test consists of Complete Freund's adjuvant injected rats challenged with doses of the test drugs administered orally 2 hours before the induction of arthritis. The drug administrations were continued as the same time of the day for 19 more days. Development of adjuvant induced swelling in the paws of both the injected and non-injected limbs of each rat were monitored daily as the percentage increase in paws.

Carageenan induced oedema in rats^[7,8]

Oedema was induced by injecting 0.1ml of carageenan (1%w/v) in distilled water into the sub plantar region of the left hind paw after one-hour (oral) drug administration. One group was used as a control and given only vehicle (2%w/v) Tween 80. The volume of paw was measured with the volume differentiated meter Model 7140 UGO Basile after 0, 60, 120, 180, 240, 360, 480 minutes of carageenan injection. Results were determined as the percentage inhibition of oedema

compared with control group. Cotton pellet granuloma in rats^[9] -Autoclaved cotton pellets 50±1mg were implanted by subcutaneous incision on the back under ether anesthesia. Drugs were administered daily orally for 7 days. Animals were sacrificed on day 7 and the granuloma was dried in an oven at 60°C and weighed to determine the percentage inhibition of granuloma.

Statistical analysis

Data are expressed as Mean±S.D statistical analyses were carried out using One-way ANOVA followed Tukey's post hoc tests. One-way ANOVA was to examine the mean differences in paw oedema between the groups. Data were analysed using the SPSS (Version 12.0, Chicago, USA) and p value was set <0.05 for all analysis.

RESULTS

The preliminary phytoconstituents of Kadambur herbal Vatha Thailam revealed the presence of essential oil.

Herbal oil, tannin, alkaloids, flavonoids. The Kadambur Herbal Vatha Thailam demonstrated a dose related anti-inflammatory activity in acute model of inflammation. In oral dose of 0.5ml/kg it showed a extend of 65.09% ($p<0.001$) inhibition (Table 3) where as standard drug indomethacin in dose of 2.5mg/kg produced 74.9±3.9 inhibition after 24 hours ($p<0.001$). The phytoconstituents in cotton pellet granuloma (a chronic model of inflammation) when administered orally in the dose of 1mg/kg showed 30.93% inhibition of granuloma was shown by standard indomethacin 2.5mg/kg dose ($p<0.001$) (31.95%). The essential oil in Freunds adjuvant induced arthritis in rats at the oral dose of 0.5mg/kg inhibit rat paw oedema by 37.65% after 27 days where as indomethacin 2.5mg/kg produced inhibited 55.24% inhibition of paw oedema after 21 days ($p<0.001$).

Table 1: Anti-inflammatory activity of Ayurvedic drugs against Cotton Pellet Granuloma in rats.

Groups	% INCREASE IN PAW THICKNESS (MEAN ± S.E)						Percentage reduction in Paw volume
	POST INSULT TIME OF ASSAY IN MINUTES						
	0	60	120	180	240	360	
VEHICLE	44.3±2.2	51.7±3.9	64.3±5.2	78.0±6.7	88.3±4.9	89.6±7.1	-
INDOMETHACIN	35.8±2.0	43.6±2.7	51.8±1.8	54.2±3.7	55.6±4.3	58.4*±5.3	34.82%
KHVT	38.9±3.7	45.3±3.9	52.6±2.5	50.5±5.4	56.7±4.3	54.5*±3.8	39.17%
*P<0.001 P Vs Standard (n=6)							

Table 2 –Anti-inflammatory activity of Ayurvedic drugs against Carageenan induced Paw oedema in Albino rats.

Table 2 Anti-inflammatory activity of standard drug against carageenan paw oedema in Rats.							
% INCREASE IN PAW THICKNESS (MEAN ± S.E)							
Groups	POST INSULT TIME OF ASSAY IN MINUTES						Percentage reduction in <u>paw</u> volume
	0	60	120	180	240	360	
VEHICLE	65.5±5.4	72.7±6.4	84.9±5.9	98.6±8.9	108.0±7.3	109.6±7.7	-
INDOMETHACIN	55.8±3.5	63.6±4.9	70.6±5.4	72.66.1	72.8±4.4	74.6*±7.1	31.93%
KHVT	60.2±3.9	66.6±3.8	72.8±3.5	74.5±4.1	76.7±2.8	74.5*±4.5	32.02%
*P<0.001 P Vs Standard (n=6)							

Table—3 Anti-arthritis activity of Kadambur Herbal Vatha Thailam against Freund's adjuvant induced Arthritis.

Treatment	%INCREASE IN PAW VOLUME ON INJECTED PAW (MEAN±S.E)									
	POST INSULT TIME IN ASSAY MINUTES									
	1	3	5	7	9	11	13	15	17	19
Control (inflamed)	102.5±19.8	210.7±14.3	195.8±10.9	180.48.9	200.3±15.6	205.7±11.9	201.5±66.2	260.2±17.8	266.3±20.3	268.7±1
Control (Non inflamed)	12.3±1.0	28.5±2.2	45.3±3.1	25.7±1.3	25.1±1.9	28.3±2.1	22.5±1.8	20.6±1.3	20.3±1.1	20.5±1.7
Indomethacin (5mg/kg)	70.84.3	88.9±5.6	73.1±4.2	78.3±5.1	88.2±2.8	93.8±6.1	99.5±5.6	103.9±7.3	107.1±8.6	113.5*±5.3
KHVT	90.7±2.3	137.5±7.8	110.2±8.8	113.4±7.7	123.7±6.3	131.5±6.3	133.8±8.7	148.4±11.8	150.6±11.7	146.3*±0.9
*P<0.00 P Vs Standard (n=6)										

Table-4 Anti-arthritis activity of Kadambur Herbal Vatha Thailam against Freund's adjuvant induced Arthritis.

Treatment	%INCREASE IN PAW VOLUME ON NON INJECTED (MEAN±S.E. n=6)						
	POST INSULT TIME OF ASSAY MINUTES						
	7	9	11	13	15	17	19
Control (inflamed)	18.7±1.3	35.3±2.1	48.7±1.9	88.9±3.2	123.3±12.3	178.9±12.3	185.715.3
Control (Non inflamed)	0	0	0	0	0	0	0
Indomethacin (2.5mg/kg)	10.8±0.7	24.3±1.3	32.7±2.3	43.5±2.9	67.1±3.3	67.1±3.3	123.4*±9.7
KHVT (200mg/kg)	10.9±0.5	22.8±1.3	28.4±2.3	53.7±1.9	77.4±3.3	77.4±3.3	128.5*±8.3
*P<0.001 P Vs Standard n=6							

DISCUSSION

The phytoconstituents of Kadambur Herbal Vatha Thailam have shown significant anti-inflammatory activity in acute as well as chronic model and also remarkable anti-arthritis activity in adjuvant-induced arthritis. Oedema, which develops after carageenan injection, is a biphasic event.^[10-11] The initial phase is attributed to the release of histamine and serotonin. The oedema maintained between the first and second phase is due to kinin like substance. It has been reported that the second phase is said to be promoted by prostaglandin like substance. It has been reported that the second phase of oedema is sensitive to drugs like hydrocortisone, phenylbutazone and indomethacin. In conclusion, remarkable anti-inflammatory and anti-arthritis activity of phytoconstituents of kadambur herbal vatha thailam could be correlated to other species like *S.callosus*,

S.hyeneanus and *S.xiocephala* for the treatment of various inflammatory conditions.

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