



Pharmacological evaluation of anti-inflammatory activity of Pancha Pashana Chendhuram against carrageenan induced paw edema in Rats

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Abstract

The present study investigates the anti-inflammatory activity in ethanolic extract of Pancha Pashana Chendhuram (PPCM) using carrageenan induced paw edema in albino rats. The anti-inflammatory was done by carrageenan induced hind paw edema method using plethysmometer. Indomethacin used as a standard drug. For this activity test groups received Control, Induced 1% Carrageenan (0.1 ml), Standard Indomethacin (40mg/kg) and the PPCM in 10mg/kg and 20mg/kg. The anti-inflammatory activity is more effective in Group V Carrageenan induction with oral administration of PPCM 20mg/kg (40.08%) compared to Group IV Carrageenan induction with oral administration of PPCM 10mg/kg (23.49%). The standard drug indomethacin showed 44.50% inhibition of paw edema. The results suggested that the PPCM has exhibited an effective anti-inflammatory activity mediated via either by inhibition of cyclooxygenase cascade and by blocking the release of vasoactive substances like histamine, serotonin and kinins.

Keywords: Pancha pashana chendhuram, Carrageenan, paw oedema, Antiinflammatory.

Introduction

Inflammation is a protective attempt by the organism to remove the injurious stimuli and to initiate the healing process. Inflammatory diseases are very common throughout the world. Rheumatoid arthritis is one of the oldest known diseases of mankind affecting the majority of population no substantial progress has been made in achieving a permanent cure and different types of rheumatic diseases are a major cause of morbidity of the working force (Sanja *et al.*, 2009). Inflammation results in the liberation of

endogenous mediators like histamine, serotonin, bradykinin, prostaglandins etc. These mediators even in small quantities can elicit pain response. Anti-inflammatory drugs make up about half of analgesics, relieving pain by reducing inflammation as opposed to opioids which affect the central nervous system (Jothibasu and Yogananth, 2009).

Currently, no satisfying drug is available for the treatment of these inflammatory-related diseases. There are two antiinflammatory drugs available in the clinic: corticosteroids and non-steroidal antiinflammatory drugs (NSAID). Despite the fact that corticosteroids and NSAID (non-steroidal antiinflammatory drugs) remain the common choice for the treatment of inflammatory diseases, the usage of these drugs are restricted by their undesirable side effects and the limited potency to reduce the symptoms of inflammation. Moreover, chronic use of corticosteroids anti-inflammatory drugs has been limited as they exhibited a weight gain, osteoporosis and immunosuppressive effects, whereas high dose NSAID medication leads to gastrointestinal tract-related toxicities (Dequeker, 1999). Consequently, the development of novel potential antiinflammatory agents with a desirable side effect is a great of interest.

The use of herbal extracts and nutritional supplements either as alternative or complimentary medicine to the conventional chemotherapy for treatment of anti-inflammatory diseases is well documented in ayurveda and siddha, which is an alternative medicinal system that has been practiced primarily in the Indian subcontinent for 5000 years (Dahankar *et al.*, 2000) inflammatory diseases, including different type of rheumatic disease are a major cause of inhibitory of the working force throughout the world. Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical diseases, though relatively little knowledge about their mode of action is available. There is a growing interest in the pharmacological evaluation of various plants used in Indian traditional systems of medicine. Keeping in view the growing significance of anti-inflammatory related herbal medicines in global market, the present anti-inflammatory study has been carried out on Pancha Pashana Chendhuram in albino rat model

Materials and Methods

Details regarding sample

Pancha Pashana Chendhuram is a classic Siddha herbo-mineral formulation mentioned in Sikitcharathinadeepam.

Drug collection

All the ingredients were obtained from country drug shop, Ramasamy chetti, Parrys Chennai, Tamilnadu, India.

Identification and Authentication

All the raw drugs were identified and authenticated at Central Research Institute (CRI), Chennai, Gunapadam department and Botany department, Govt Siddha Medical College, Arumbakkam, Chennai.

Ingredients of Pancha Pashana Chendhuram (Group I)

P. Thalagam (Arsenic trisulphidium), P. Lingam (Red sulphide of mercury), P. Rasam (Hydrargyrum), P. Ganthagam (Sulphur), P. vellai pasanam (white arsenic), P. manosilai (Red orpiment), P. gandham (Magnetic oxide of iron).

Medicinal plants (Group II)

The medicinal plants Kuppaimeni (*Acalypha indica*), Vетtrilai (*Piper betel*), Paruthi (*Gossyphium hirsutum*), Vellerukkan (*Calotropies proccera*), Thulasi (*Ocimum sanctum*), Uthamani(*Pergulariadaemia*), Poduthalai (*Phyla nodiflora*) were used for the preparation of Pancha Pashana Chendhuram

Purification of Chemicals as per Siddha methods (Sikicharathna Deepam Sarakku Suthi Muraigal)

In the Siddha system of medicine purification of raw drugs is an important procedure in making medicine in order to reduce its toxic effects and to potentiate its therapeutic efficacy. According to the following process each chemical was purified.

Rasam (Mercury):

35 gms of Mercury is triturated with brick powder and turmeric powder for one hour respectively and washed with water. Then the Mercury is boiled with the juice of Kuppai maeni (1.3 litres) until it is detoxified.

Gandhagam (Sulphur):

Sulphur is placed in an iron spoon. A small quantity of cow's butter is added and the spoon is heated till the butter melts; this mixture is immersed in inclined position in cow's milk. This procedure is repeated for 30 times to get purified Sulphur. Each time, fresh milk is to be used.

Lingam (Red sulphide of mercury):

Lime juice, Cow's milk and the Indian Acalypha juice are mixed in equal proportion and allowed to fuse Cinnabar so as to get it in a consolidated potency state.

Manosilai (Arsenic di sulphide):

Manosilais buried in limestone and poured by donkey urine to get purified.

Thalagam (Arsenic tri sulphide):

Thalagam is buried in limestone and poured by donkey urine to get purified.

Vellai padanam(Arsenic sulphide):

35gms of vellai padanam is triturated with lime juice 105gms, repeat for 7 times, until it is detoxified.

Gaandham (Magnetic oxide of iron):

Gandham is soaked in lime juice, soured butter milk, soured kaadi, for each 3days then dried in sunlight.

Preparation of pancha pashana chendhuram (INT)

All the group-I drugs are grinded with group-II HERBAL JUICES for 12 hours and made into small poultices (Villai). Then it is to be dried in the sun shade. The dried poultice is covered with piper betel leaf. The covered poultices are placed in mud plate and closed with same size of another mud plate, and is sealed with seven layers of mud pasted cloth. Then the contents are ignited to Deepakini (Mild flame) for 12 hrs. Then the

collected chendhuram is again subjected to heat for 8 hrs and honey is used as a vehicle. The end product is grinded as a fine powder and preserved in an air tight container.

Anti-inflammatory activity

The experimental setup was to study the anti-inflammatory property of Pancha pashana chendhuram against Carrageenan induced paw edema. Inflammation is a tissue reaction to infection or irritation due to a foreign substance. For the experiment, the animals were divided into 5 groups with 6 animals in each group.

- Group-I (control) received 3% gum acacia 10 ml/kg p.o.
- Group-II (Carrageenan) received 0.1ml of 1% w/v suspension of carrageenan S.C
- Group-III (standard) received Indomethacin 40 mg/kg p.o.
- Group-IV (Test-1) received PPCM 10mg/kg p.o.
- Group-V (Test-2) received PPCM 20mg/kg p.o.

All the drugs were administered orally and the volume of medicaments kept constant at 10 ml/kg body weight of the animals it was administered orally to rats 1 hr before subcutaneous injection of carrageenan. After 1 hr 0.1ml of 1% w/v suspension of carrageenan was injected into sub-plantar region of the left hind paw to all the groups. The paw volume was measured at 1, 2, 3, 4, and 5 hr using Plethysmometer (Model 7150 UGO Basile, Italy). Edema was expressed as the mean increase in paw volume relative to control animals.

Results and Discussion

Inflammation is a common phenomenon and it is a reaction of living tissues towards injury. Steroidal anti-inflammatory agents will lyse and possibly induce the redistribution of lymphocytes, which cause rapid and transient decrease in peripheral blood lymphocyte counts to affect longer term response (Das et al., 2012)

The effect of PPCM 10 and 20 mg/kg *p.o* were studied in albino rats by observing its anti-inflammatory activity induced by Carrageenan. Carrageenan induced inflammation is a useful model for the estimation of anti-inflammatory effect. The development of oedema in the paw of the rat after the injection of Carrageenan is due to the release of histamine, serotonin and prostaglandin (Georgewill *et al.*, 2010 and Georgewill, 2010)

Table 1 showed that the extract exhibited statistically significant in doses of 10 and 20 mg/kg within 1 hr of administration of PPCM. The effect of PPCM on carrageenan-induced rat paw edema at different hours of study was compared to that of control and standard drug (Indomethacin) for the evaluation of anti-inflammatory activity on the basis of percent inhibition of paw edema volume.

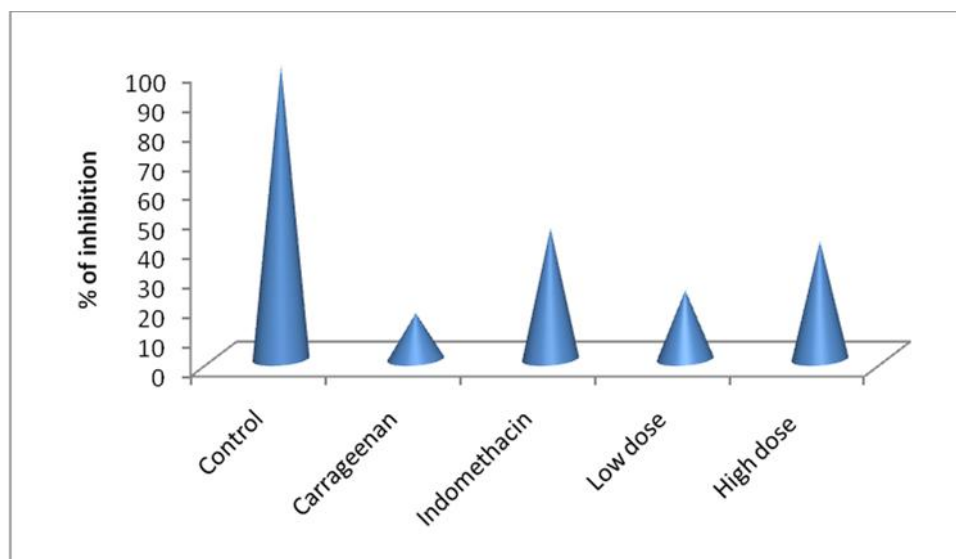
Table 1: Effect of PPCM on anti-inflammatory activity on carrageenan rat oedema

Group	Dose	Initial paw volume	Change in paw edema mm at different time intervals				
		0hr	1 hr	2hr	3hr	4hr	5hr
I	Control	1.20 ± 0.14	1.20±0.14	1.20±0.14	1.20±0.14	1.20±.14	1.20±0.14
II	Carrageenan	1.21± 0.17	1.91 ± 0.21	2.27 ± 0.02	2.37 ± 0.14	2.48 ± 0.18	2.62 ± 0.17
III	Indomethacin	1.01± 0.06	2.10 ± 0.26	1.56 ± 0.15	1.47 ± 0.05	1.34 ± 0.18	1.25 ± 0.16
IV	Low dose	1.17 ± 0.13	1.39 ± 0.26	1.40 ± 0.07	1.32 ± 0.13	1.26 ± 0.12	1.22 ± 0.13
V	High dose	1.02 ±0.20	1.60 ± 0.07	1.75 ± 0.25	1.52 ± 0.46	1.39 ± 0.28	1.27 ± 0.26

Here in the control group there is no difference between the initial Paw Volume and the Paw Volume of each hour after induction. The Group II is carrageenan induced which will show an elevated level of paw volume in each hour. At the end of the 5th hr the paw volume is higher than the Initial Paw Volume. In Group III the Standard Indomethacin is intraperitoneally received which gives low paw volume in each hr (1st to 5th hr) Finally at the end of 6th hr paw volume shows least value. The Group IV the Carrageenan is subcutaneously induced along with the oral administration of PPCM of 10mg/kg/*p.o*. Here the 1st and 2nd hr shows elevated values of Paw Volume. After that the values were lowered in 3rd, 4th and 5th hrs respectively. In the same way

Group V Carrageenan induction with oral administration of PPCM of 20mg/kg/*p.o* is given, which show increased values of paw volume in 1st and 2nd hour after it gave low paw volume at the end of the 5th hr.

In the present study 10 mg/kg b.wt. and 20mg/kg. b.wt of PPCM significantly reduced the carageenan induced paw oedema inflammation as compared with that of the standard drug, indomethacin. The PPCM of 20 mg/kg b.wt and 10 mg/kg b.wt produced remarkable percentage of inhibition (40.08% and 23.49%, respectively) and was low as compared to the reference drug (44.50%) (Figure. 1).

Fig 1: Percentage of inhibition of paw volume

Several herbal plants constituents have demonstrated anti-inflammatory properties (Vijaya *et al.*, 2013 and David Wilson *et al.*, 2014). Indomethacin showed more or less uniform inhibition of edema in early intermediate and later phases. Both PPCM doses showed also more or less significant inhibition of carrageenan induced paw edema in early phases while significant inhibition at later phases. Acute inflammation induced by carrageenan results from cell damage, which provokes the production of endogenous mediators, such as, histamine, serotonin, prostaglandins, and bradykinin. It is well known that inhibition of edema induced by carrageenan in rats is one of the most suitable test procedures to screen anti-inflammatory agents as it closely resembles human arthritis. Hence, the present result indicates the efficacy of 10 and 20 mg/kg b.wt of PPCM as an efficient therapeutic agent in acute anti-inflammatory conditions.

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