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COMPARATIVE ANTI-INFLAMMATORY ACTIVITY OF *PANCHATIKTA GHRTA* PREPARED BY THREE DIFFERENT CLASSICAL METHODS AGAINST CARRAGEENAN-INDUCED INFLAMMATION RATS.

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ABSTRACT

Background: *Panchatiktaghrta* is an Ayurvedic medicated herbal *ghrita* formulation mentioned in classics for many diseases particularly for skin disorders and related inflammation. According to Sastra, the ingredients of *Panchatiktaghrta* formulation possess more *tikta*, *kasaya* and *kusthaghana* and *krimighna* properties. Various methods are described in texts for preparation of *Panchatiktaghrta* formulation. Therefore, in the present study *Panchatiktaghrta* prepared by three different methods namely 1.Sharangadara Samhita (Murchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) 2.Bhaishajyaratnavali (Murchita Ghrita, Triphala Kalka, Panchathikta Kwatha) *3.* Sharangadara Samhita (Amurchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) and evaluated for its anti-inflammatory activity against carrageenan-induced inflammation in rats. **Materials & methods:** Comparative efficacy of three different formulations of *Panchatiktaghrta* formulations were assessed against paw oedema-induced by injecting 0.1ml of 1% (w/v)carrageenan in normal saline into the plantar aponeurosis of left hind limb of rats. Percentage increase in paw volume in comparison to the initial volumes were noted for each rat in test drug administration group and compared with control group for anti-inflammatory activity. **Results:** All three *Panchatiktaghrta* formulations produced

significant suppression of carrageen an-induced hind paw oedema in rats. *PanchatiktaGhrta* prepared by. Sharangadara Samhita (Murchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) produced better suppression of paw oedema (83.17%) followed by *PanchatiktaGhrta* prepared by Bhaishajyaratnavali (Murchita Ghrita, Triphala Kalka ,Panchathikta Kwatha) (68.27%) and. Sharangadara Samhita (Amurchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) (62.23%) in comparison to control group. **Conclusion:** From the present study, it is concluded that *PanchatiktaGhrta* has an anti-inflammatory activity in experimental model of rats which may attribute to its role in *kusthaghana.*

KEYWORDS: Panchatikta ghrita, Sharangadara Samhita, Bhaishajyaratnavali, Anti-inflammatory Activity, Carrageenan etc.

INTRODUCTION

Inflammation is defined as the local response of living mammalian tissues to injury due to any agent. It is a body defense reaction in order to eliminate or limit the spread of injurious agent, followed by removal of the necrosis cells and tissues.[1] Inflammation is characterized in acute phase by increased blood flow and vascular permeability along with the accumulation of fluid, leukocytes and inflammatory mediators such as cytokines. In the sub acute/chronic phase it is characterized by the development of specific humeral and cellular immune responses to pathogens present at the site of tissue injury.[2] Many drugs are available which help to reduce inflammation. Risks include upper gastrointestinal bleeding and perforation associated with non-steroidal anti-inflammatory drugs (NSAIDs) and physical dependence and abuse liability developed by opiates.[3] These problems have led to a search for potent anti‑inflammatory drugs from plant sources as alternatives to NSAIDs.

Medicinal plants play a vital role as source of raw materials for both the modern as well traditional systems of medicine.[4] Plant medicines are great importance in the primary healthcare in many developing countries. In Ayurveda, utilizing a large number of medicinal plants were used for the treatment of human diseases.[5] The medicinal plants occupied their exclusive place in human life. It provides more information about the use of plants or plant parts as medicine.[6,7] *Pancatiktaghrta* is an Ayurvedic medicated herbal *ghrita* formulation mentioned in classics for many diseases particularly for skin disorders and related inflammation. *Pancatiktaghrta* is an Ayurvedic formulation of a *Sneha Kalpana*. It is a form of secondary *Kalpana* having aim of this arrangement is mass transfer of the aqueous and

lipid-soluble active principles of all treated herbal drugs and material of animal and mineral origin, if any, in accordance of established formulae quoted in authoritative text books of Ayurveda which should serve therapeutic objectives as per indications of the classical treatise of Ayurveda.[8] *Sneha Siddha* (fat soluble) drugs have better pharmacokinetic action (ADME) in comparison to other dosage forms because of the lipoid nature of the biomembranes, as lipid soluble substances readily permeate into the cells.[9] *Sanskara* is a main important term in *Snehakalpana* which imparts qualitative improvement in *sneha*.[10,11]

According to *Sastra,* the ingredients of *Pancatiktaghrta* formulation possess more *tikta*, *kasaya* and *kusthaghana* and *krimighna* properties. Various methods described in texts for preparation of *Pancatiktaghrta* formulation. Therefore, in the present study *Pancatiktaghrta* prepared by three different methods namely 1.Sharangadara Samhita (Murchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) 2. Bhaishajyaratnavali(Murchita Ghrita, Triphala Kalka, Panchathikta Kwatha) *3.* Sharangadara Samhita (Amurchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha) and evaluated for anti-inflammatory activity against carrageenan-induced inflammation in rats.

MATERIALS AND METHODS

Plant materials and drug

Ingredients of the *Pancatiktaghrta are, Triphala, Haridra, Musta, Nimbu swarasa* were supplied by the Pharmacy, Gujarat Ayurved University, Jamnagar. Vasapatra, Nimbatwak, *Guduchikanda, Kantakari pancanga* were collected by the scholar himself from the areas surrounding Jamnagar city region, where as *Patolapancanga* was collected from Surat district. The *Pancatiktaghrta* was prepared by the scholar himself in R.S.&B.K. Department, IPGT&RA, Jamnagar.

Animals

Charles Foster albino rats weighing 200±20 g of either sex were obtained from Animal house. They were housed in standard polypropylene cages and kept under controlled room temperature, relative humidity and 12h light -dark cycle. The animals were fed with standard laboratory diet of “Amrut” brand rat pellet and drinking water *adlibitum*. Food was withdrawing 12h before and during the experimental hours. The experimental protocol was approved by Institutional Animal Ethics Committee as per CPCSEA guidelines.

Dose

The oral dose of the test formulations was calculated by extrapolating the human dose to rat dose (450 mg/kg) based on the body surface area ratio by referring to the standard table of Paget and Barnes.[12] The test drug was administered orally by the oral catheter.

Experimental design

Charles Foster albino rats weighing between 200±20 g of either sex were randomly divided in to four groups each consisting of six rats. Group (I) was kept as control group which received equal quantity of simple cow's ghee as vehicle. Group (II) to (IV) received *PancatiktaGhrta* prepared by Murchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha (**Sample A**), Murchita Ghrita, Triphala Kalka, Panchathikta Kwatha (**Sample B**) and Amurchita Ghrita, Panchathikta Kalka, Panchathikta Kwatha (**Sample C**).

The inhibitory effect of *Panchtiktaghrta* on carrageenan-induced edema was evaluated by using standard method.[13] The test drug was administered at a dose of 450 mg/kg bodyweight for seven consecutive days, while in the control group equal volume of simple cow's ghee was administered.On eighth day, initially left hind paw volumes up to the tibio-tarsal articulation were recorded by using plethysmograph. The test drug was administered in accordance with body weight of rats. The rats were administered 2ml/100gm body weight of drinking water to ensure uniform hydration and hence, to minimize variations in oedema formation. One hour after administration of the test drug, paw oedema was induced by injecting 0.1ml of 1% (w/v) carrageenan aqueous suspension in normal saline subcutaneously into the plantar aponeurosis of left hind limb. The paw volume was again recorded after three hours after carrageenan injection by using a plethysmograph. Results were expressed as percentage increase in paw volume in comparison to the initial volumes and also in comparison with control group. If the percentage in paw volume is significantly less in test drug administered group in comparison to control group, then the drug was considered to possess anti-inflammatory activity.[14]

Statistics

The data were expressed as mean ± standard error of mean (SEM). The significance of differences among the groups was assessed using one way analysis of variance (ANOVA) with Dunnet's multiple t test. P value less than 0.05 was considered as statistically significant and $P<0.001$ was considered statistically highly significant.[15]

RESULTS AND DISCUSSION

Carrageenan-induced acute inflammation in rats represents a classical model for studying the acute inflammation and used to study anti-inflammatory activity of test drugs. The model employed was carrageenan-induced hind paw oedema representing acute inflammation characterized by fluid and cell exudation. These tests were carried out mainly to provide experimental basis to the clinical application of test drug in psoriasis. Data on the effect of test drug on 1% carrageenin induce hind paw oedema in rats have been summarized in Table-1. Test drug produced significant suppression of carrageenan-induced hind paw oedema in rats. The suppression was 68.27% with sample-A, 83.17% with sample-B and 62.23% with sample-C. All the three test samples produced significant anti inflammatory activity in the hind paw oedema test indicating that, they possess significant anti-inflammatory activity.

Table1. Effects of *PancatiktaGhrta* on carrageenan-induced hind paw oedema in albino rats

Groups	Dose (mg/kg)	Percentage increase in paw volume	
		24 hours	48 hours
Control	-	92.10±19.88	-
Panchtiktaghrta- A	450	29.21±10.70*	68.27
Panchtiktaghrta-B	450	15.30±7.00**	83.17
Panchtiktaghrta-C	450	34.79±13.92*	62.23

* P<0.05, **P<0.01when compared to control group (Annova followed by Dunnett'smultiple 't' test)

Pancatikta Ghrta prepared by *Ghrta Murchana* with panchathikta kalka produced better suppression of paw oedema followed by *PancatiktaGhrta* prepared by *Ghrta Murchana* and *Triphalakalka* and without *Ghrta Murchana* and *Triphalakalka* in comparison to control group. However, the difference between these groups did not reach statistically significant levels. The carrageenan‑induced rat paw edema is a biphasic process and is produced by the release of many vasoactive inflammatory mediators.[16] The early phase is mediated by histamine and serotonin and the late phase by the release of Kinins and prostaglandins.[17] It is possible that the active principle contained in the test drugs may be modulating the formation and release of the above mentioned phlogistic mediators. *Pancatikta ghrta* formulation possesses more *tikta* and kasaya which may attribute to its *kusthaghana* and *krimighna* properties.

From the present study, it is concluded that *Pancatikta Ghrta* has anti-inflammatory activity in experimental model of rats which may attribute to its role in *kusthaghana* and *krimighna* properties. *Pancatikta Ghrta* prepared by *Ghrta Murchana with pancha tikta kalka showed* better anti-inflammatory activity followed by *Pancatikta Ghrta* prepared by *Ghrta Murchana* and *Triphala kalka* and without *Ghrta Murchana* and *Triphala kalka.*

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