

## **ANTI-INFLAMMATORY AND ANALGESIC ACTIVITIES OF PANCHATIKTA GHRITA**

**Zala Upendra<sup>1</sup>, Vijay Kumar<sup>2</sup>, Chaudhari AK<sup>3</sup>, Ravishankar B<sup>4</sup>, Prajapati PK<sup>5\*</sup>**

1. Reader, Dept. of R.S. & B.K., J.S Ayurveda College, Nadiad, Gujarat, India.
2. Lecturer, Dept. of Pharmacology, PGT-SFC-Cell, Gujarat Ayurved University, Jamnagar, Gujarat, India.
3. Associate Professor, Dept. of R.S. & B.K., IMS, Banaras Hindu University, Varanasi, Uttar Pradesh, India.
4. Director, SDM Research Centre for Ayurveda and Allied Sciences, Udupi, Karnataka, India.
5. Professor & Head, Dept. of R.S. & B.K., I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.

Received: 23-11-2012; Revised: 25-12-2012; Accepted: 27-12-2012

### **Abstract**

Anti-inflammatory and analgesic activities of Murchita and Avartita Panchtikta Ghritha were carried out on Charles foster strain albino rats at the dose of 0.96 ml/kg. The effect on inflammation was studied by carrageenan and formalin induced paw oedema models and compared to control groups. Analgesic activity was evaluated in formalin induced paw licking response (chemically induced pain). The Avartita Panchtikta Ghritha (APG) significantly inhibited carrageenan induced paw oedema, formalin induced paw licking response, however it was failed to suppress the formalin induced paw oedema, whereas Murchita Panchtikta Ghritha (MPG) significantly inhibit formalin induced pain response, however it failed to suppress carrageenan induced paw oedema and formalin induced paw oedema. The results suggest that the APG has significant analgesic and anti-inflammatory potential as reflected by the parameters investigated, while MPG is having only analgesic activity. Thus it can be concluded that APG have significant anti-inflammatory and analgesic activity and can be preferred in the treatment of pain and inflammation.

**Keywords:** Murcchana; Panchatikta; Avartana; Sneha; Carrageenan; Formalin.

### **\*Address for correspondence:**

Prof. Prajapati PK, MD, Ph.D.,  
Professor & Head, Dept. of R.S. & B.K.,  
I.P.G.T. & R.A., Gujarat Ayurved University,  
Jamnagar, Gujarat, India – 361 008.  
E-mail: [prajapati.pradeep1@gmail.com](mailto:prajapati.pradeep1@gmail.com)

### ***Cite This Article***

Zala Upendra, Vijay Kumar, Chaudhari AK, Ravishankar B, Prajapati PK. Anti-inflammatory and Analgesic activities of Panchatikta Ghritha. *Ayurpharm Int J Ayur Alli Sci.* 2012;1(8):187-192.

## INTRODUCTION

Despite progress within medical research during the past decades, the treatment of many serious diseases remains problematic.<sup>[1]</sup> Inflammatory diseases remain one of the world's major health problems.<sup>[2]</sup> Currently, both steroidal anti-inflammatory drugs and non-steroidal anti-inflammatory drugs (NSAIDs) are used in the aid of inflammation. Steroids have an obvious role in the treatment of inflammatory disease, but due to their toxicity, they can only be used over short periods except in very serious cases where the risks are acceptable. Prolonged use of NSAIDs is also associated with severe side effects, notably gastrointestinal hemorrhage.<sup>[3][4]</sup>

Inflammatory diseases are among the most common health problems treated with traditional remedies. Therefore, it is crucial to evaluate the potential of herbal remedies that might serve as leads for the development of potent drugs. A large number of single Indian medicinal plants and their compound formulations are attributed with various pharmacological activities.

Snehakalpana (Medicated oil and ghee preparations) is a pharmaceutical process to get the oleaginous medicinal substance. Ghrita as the Sneha and the five-bitters that is Panchatikta processed to formulate Panchatikta Ghrita.<sup>[5]</sup> The Panchatikta Ghrita was prepared with the help of Panchatikta Kwatha, Kalka and Go-ghrita. Panchatikata Kwatha was prepared by Vasa Patra (Leaves of *Adhatoda vasica* Nees), Nimba twak (Bark of *Azadirachta indica* A. Juss.), Patola panchanga (Whole plant of *Trichosanthes dioica* Roxb.), Guduchi kanda (Stem of *Tinospora cordifolia* (willd) miers. ex HK. F. & Th.) and Kantakari panchanga (Whole plant of *Solanum xanthocarpum* Buru. F.) in a prescribed ratio.<sup>[6]</sup>

Murchhana is to remove the bad smell and impurities etc. type of bad characteristic of crude form of Sneha. By Murchhana Samskara some Sneha gets good smell and color, apart from these, because of Murchhana, Sneha becomes capable to receive more active principle while the preparation of Sneha and also by performing Murchhana Samskara, the potency of the Sneha is enhanced, and Sneha will get the active principle of Murchhana dravyas too. Avartana (repetition of process) is one such technique by which oleaginous medicaments can be reduced into small dosage forms, meanwhile increasing its potency.

Therefore different samples of panchatikta Ghrita were prepared in following batches (a) Murchhita and Anavartita (b) Murchhita and Avartita (10 times Avartita) sample of the drug for its quality and same has been evaluated for its effect in the albino rats. For second Avartana, the first Avartita Panchatikta Ghrita is taken in the place of Ghrita and the fresh Panchatikta Kalka and Panchatikta Kwatha are added in the prescribed ratio. Similarly further Avartana were carried by using preceding Ghrita as base.

Panchatikta Ghrita different samples may exhibit different degree of actions because of drug concentration gradient. The formulations studied possess different proportion of Go-Ghrita (cow's ghee), Panchatikta Kalka and Panchatikta Kwatha. Because of their individual properties they are believed to be effective in providing anti-inflammatory and analgesic effects.

## MATERIAL AND METHODS

### Test drug and sample preparation

The raw material (Table 1) of the test formulation were collected from pharmacy attached to the institute and were subjected to pharmacognostical studies in order to evaluate the authenticity. Different samples of test formulation were prepared in the laboratory of

Dept. of Rasa shastra & Bhaishajya Kalpana, I.P.G.T. & R.A, Jamnagar and were used for the experimental study.

**Sample I:** Panchatikta Ghrita according to the classical references – (Sarangdhar.Sam.M. Kh. Cha.9/92) i.e. with Murchhana and without any Avartana. Coded as MPG.

**Sample II:** Panchatikta Ghrita prepared by performing ten Avartana. Coded as APG.

### **Experimental Animals:**

Charles foster strain of albino rats of either sex weighing 220 – 260 g were obtained from the animal house attached to the Pharmacology laboratory, I.P.G.T. & R.A, Jamnagar, maintained on Amrut brand animal pellet feed supplied by Nav Maharashtra Chakan Oil Mills and tap water was given ad-libitum the temperature and humidity was kept at optimum and animals were exposed to natural day night cycles. The experiments were approved by I.A.E.C./II/2004-112/dated 05/08/2004.

### **Dose fixation and schedule:**

The dose fixation for the experimental animals was done on the basis of body surface area ratio by referring to the standard table of Paget and Barnes (1969).<sup>[7]</sup> On this basis, the rat dose was found to be 0.96 ml/kg. The test drugs were administered orally with the help of gastric catheter sleeved to syringe.

### **Anti-inflammatory activity:**

#### **Carrageenan induced rat paw oedema**

The Charles foster strain albino rats were divided into three groups, each group comprising of six animal of either sex. The first group was kept as control in which Go-Ghrita alone (0.96 ml/kg) administered; whereas the second group received Murchhita Panchatikta Ghrita (0.96 ml/kg) and third

group received Avartita Panchatikta Ghrita (ten Avartana) (0.96 ml/kg). The vehicle and test drugs were administered orally to the respective groups for five consecutive days.<sup>[8]</sup>

Initially left hind paw volumes up to the tibio-tarsal articulation were recorded prior to carrageenan injection by using plethysmograph.<sup>[9]</sup> The plethysmograph employed, consisted of 10 ml glass vessel (25 mm × 65 mm) fixed to 2 ml glass syringe through pressure tubing. About 4 ml of mercury was filled in the syringe and the mercury level was adjusted to zero mark on the micropipette. The space between the zero mark and the fixed mark on the glass vessel was filled with water and few drops of teepol. The initial level of fluid was adjusted and set at zero. The paw was immersed in water exactly up to the tibio-tarsal articulation. The increased level of water in the glass vessel was adjusted to the prefixed mark by releasing the pressure of the connected syringe. The level where water and mercury interface in the micropipette was recorded as paw volume.

On fifth day one hour after drug administration oedema was produced by injecting 0.1 ml freshly prepared 1% carrageenan in sterile saline solution to the sub-plantar aponeurosis of the left hind limb. The rats were administered tap water in the dose of 2 ml per 100 g body weight to ensure uniform hydration and hence to minimize variations in oedema formation. Paw volume was recorded three hour after carrageenan injection. Results were expressed as an increase in paw volume in comparison to the initial paw volumes and also in comparison with control group.

#### **Formalin induced paw oedema**

The test condition and grouping were similar to carrageenan induced paw oedema as mentioned above. Pedal inflammation was induced by injecting 0.1 ml of 3 % formalin solution below the plantar aponeurosis of the

right hind paw of the rats. The paw volume was recorded immediately prior to drug administration (0 h) and then at 24 then 48 hrs after formaldehyde injection. Results were expressed as an increase in paw volume in comparison to the initial paw volumes and also in comparison with control group.<sup>[10]</sup>

### **Analgesic activity**

#### **Formalin induced hind paw licking**

Animal grouping and test drug administration are similar to carrageenan induced paw oedema model. Pain response was induced by injecting 0.1 ml of 3% formalin in distilled water in subplantar region of right hind paw and the duration of paw licking as an index of nociception was counted in periods of 0 to 10 minutes (Early phase) and 20 to 30 minutes (Late phase).<sup>[11]</sup>

### **Statistical analysis**

Data are expressed as Mean  $\pm$  SEM (standard error of mean). Statistical evaluation was carried out by unpaired student 't' test. Statistical significant is expressed as \*P<0.05, \*\*P<0.01, \*\*\*P<0.001.

## **RESULTS**

#### **Effect on carrageenan induced paw oedema**

Table 2 shows carrageenan induced paw oedema. Avartita Panchatikta Ghrita group at the dose of 0.96 ml/kg has significant reduction in carrageenan induced paw oedema (P<0.05) at 3 hour when compared to control group, however Murchhita Panchatikta Ghrita failed to suppress oedema formation induced by carrageenan.

#### **Effect on formalin induced paw oedema**

Table 3 shows formalin induced paw oedema. Both the test drugs did not suppress the

formalin induced paw oedema at both 24 and 48 hour time interval.

#### **Formalin induced paw licking**

Table 4 shows formalin induced paw licking response. Both Murchhita Panchatikta Ghrita and Avartita Panchatikta Ghrita group shows significantly decrease in paw licking response at both early phase (P<0.001) and late phase (P<0.01), however licking response of Avartita Panchatikta Ghrita at late phase was less (P<0.05) in comparison to Murchhita Panchatikta Ghrita.

## **DISCUSSION**

Carrageenan induced inflammation in rats is one of the most suitable acute model to screen anti-inflammatory agents. The intraplantar infection of carrageenan in rats leads to paw edema. Its first phase (Up to 3 hrs after injection of carrageenan) results from the concomitant release of mediators: histamine, serotonin and kinins on the vascular permeability. The second phase is release of prostaglandins.<sup>[12]</sup> Among the two forms of test formulation APG produced a considerable suppression of oedema formation against carrageenan induced paw oedema in rats. The observed effect may be due to inhibition of phlogistic mediators, antagonizing their interaction with their respective receptors or it may be due to general mechanism like increasing the membrane stability in the cell. The formalin-induced inflammation in the rats foot may be conveniently divided into two parts, the first involving 5-hydroxytryptamine as mediator and the second mediator which is unrelated to 5-hydroxytryptamin.<sup>[13]</sup> Both the test formulation did not produce any significant effect on either phase of the formaldehyde injection induced algogenic effect. It shows that both the form of test formulation is not having 5-HT suppression activity.

**Table 1: Formulation composition of Murchhita Panchatikta Ghrita and Avartita Panchatikta Ghrita**

Ingredients	MPG	APG
1. Kalka	900g	9 kg
a) Vasapatra (Fresh leaves)	180g	1.8 kg
b) Guduchikanda (Fresh stem)	180g	1.8 kg
c) Nimba Twak	180g	1.8 kg
d) Kantakari Panchanga	180g	1.8 kg
e) Patola Panchanga	180g	1.8 kg
2. Go-ghrita (Murchhita Ghrita)	3.6 kg	3.6 kg
3. Panchatikta Kwatha	14.4 ltr	144 ltr

**Table 2: Effect on carrageenan induced hind paw oedema**

Group	Percentage increase in paw volume After 3 hrs.	Percentage inhibition
Control	65.91 ± 04.03	--
MPG	78.33 ± 11.57	18.84 ↑
APG	42.33 ± 09.58*	35.77 ↓

Data: Mean ± SEM, ↑-Increase, ↓-Decrease, \*P<0.05 (comparison to control group, Unpaired t test)

**Table 3: Effect on formalin induced hind paw oedema**

Group	Percentage increase in paw volume			
	After 24 hrs.	Percentage inhibition	After 48 hrs.	Percentage inhibition
Control	69.33 ± 08.13	--	73.03 ± 10.07	--
MPG	79.83 ± 05.76	15.14↑	77.65 ± 06.05	6.32↑
APG	98.65 ± 08.63*	42.23↑	71.75 ± 05.30	1.75↓

Data: Mean ± SEM, ↑-Increase, ↓-Decrease, \*P<0.05 (comparison to control group, Unpaired t test)

**Table 4: Effect on formalin induced paw licking**

Group	Number of paw lickings			
	0-10 min	Percentage inhibition	20-30 min.	Percentage inhibition
Control	32.83 ± 3.74	--	30.83 ± 4.06	--
MPG	09.00 ± 2.11***	72.59 ↓	12.00 ± 2.59**	61.08 ↓
APG	09.16 ± 1.93***	70.10 ↓	18.33 ± 1.27**	40.54 ↓

Data: Mean ± SEM, ↓-Decrease, \*\*P<0.01, \*\*\*P<0.001 (comparison to control group, Unpaired t test)

When formalin is injected subcutaneously into the paw, it produces intense pain reaction. The effect is seen in two phases. The initial phase last for 0-10 min of formalin injection; it is supposed to be mediated through modulation of neuropeptides.<sup>[14]</sup> The second phase which is observed 20-30 min of formalin injection is supposed to be mediated through release of inflammatory mediators like prostaglandin etc. Both the forms of test formulation significantly decrease the paw licking

response at initial phase (0-10 min), however APG treated group fails to decrease paw licking response at second phase (20-30 min).

## CONCLUSION

The results presented here demonstrate that Avartita Panchatikta Ghrita inhibit pain and inflammation with an interesting analgesic and anti-inflammatory activity profile. The Murchhita Panchatikta Ghrita is having only

analgesic activity. Based on the above it can be mentioned that Avartana procedure improves the activity profile of the panchatikta Ghrita.

## REFERENCES

1. Bohlin L. Structure activity studies of natural products with anti-inflammatory effects. In: Hostettmann K, editor. Phytochemistry of plants used in traditional medicine. Oxford: Clarendon Press 1995; 137-161.
2. Yesilada E, Uston O, Sezik E, Takaishi Y, Ono Y, Honda G. Inhibitory effects of Turkish folk remedies on inflammatory cytokines: interleukin-1 and tumor necrosis factor. J Ethnopharmacol 1997; 58:59-73.
3. Robert A, Hancher AJ, Lancaster C, Nezamis JE. Prostacyclin inhibits enteropooling and diarrhea. In: Vane JR, Bergstrom S, editor. Prostacyclin. New York: Raven Press 1979, p.147-158.
4. Miler TA. Protective effects of prostaglandins against gastric mucosal damage: Current knowledge and proposed mechanisms. Am J Physiol 1983; 245: 601-623.
5. Sarngadhara. Sharangadhara Samhita. Himasagara Chandra murty, editor. 2<sup>nd</sup> ed. Varanasi: Chaowkhamba Sanskrit Series Office; 2007. Madhyam khanda 9/92.p.212.
6. Ibid. 9/4-5.p.199.
7. Paget GE, Barnes JM. Evaluation of drug activities, pharmacometrics (Vol. 1). Lawrance DR, Bacharch AL, editors. New York: Academic press; 1969, p.161.
8. Winter CA, Risely EA, Nuss GW. Carrageenan induced edema in hind paw of the rat as assay for anti-inflammatory drugs. Proc Soc Exp Bio Med 1962; 111:544-547.
9. Bhatt KR, Mehta RK, Srivastava PN. Simple methods of recording anti-inflammatory effect on rat paw oedema. Indian J Physiol Pharmacol 1977; 21: 399-400.
10. Roy A, Gupta JK, Lahiri SC. Further studies on anti-inflammatory activity of two potent indan-1-acetic acids. Indian J Physiol Pharmacol 1982; 26: 207-214.
11. Hunskar S, Hole K. The formalin test in mice, dissociation between inflammatory and non-inflammatory pain. Pain 1987; 30:103-114.
12. Di Rosa M, Giroud JP, Willoughby DA. Studies on the mediators of the acute inflammatory response induced in rats in different sites by carrageenan and turpentine. J Pathol 1971; 104: 15-29.
13. Northover BJ, Subramanian G. Some inhibitors of histamine-induced and formaldehyde-induced inflammation in mice. Brit J Pharmacol 1961; 16: 163-169.
14. Fernando TRGW, Ratnasooriya WD, Deraniyagala SA. Antinociceptive activity of aqueous leaf extract of *Tetracera sarmentosa* L. in rats. Phcogres 2009; 1(6):381-386.

Source of Support: Nil

Conflict of Interest: None Declared