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ANALGESIC AND ANTI-INFLAMMATORY ACTIVITY IN SIDDHA DRUG FORMULATION KAALAKODI RASAM

Dr. S. Thanikaiselvi*[1] and Dr. R. Antony Duraichi[2]

[1]PG Scholar, Department of Gunapadam, Govt. Siddha Medical College, Palayamkottai.
[2]Lecturer, Department of Gunapadam, Govt. Siddha Medical College, Palayamkottai.

***Corresponding Author: Dr. S. Thanikaiselvi**
PG Scholar, Department of Gunapadam, Govt. Siddha Medical College, Palayamkottai.

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ABSTRACT
Siddha system is the ancient system of heeling and it's based on combination of medical practices and preventive methods, as well as alchemy and mysticism. Herbo metallic preparations play an important role in traditional system of medicine. One such effective & safe herbo metallic formulation is kaalakodi rasam (KKR). As indicated in Siddha literature KKR is a herbo metallic formulation treated for osteoarthritis. **Aim:** To study of analgesic and anti-inflammatory activity in the siddha drug formulation Kaalakodi rasam. **Materials and methods:** The analgesic and anti-inflammation activities evaluation were done by Acetic acid Induced Writhing Test method in Swiss albino mice and Carrageenan induced acute hind paw oedema method on Wister albino rats, respectively. Doses of different proportions 100mg, 200mg of KKR powder suspension were given to the animals for a stipulated period of time. **Results and conclusion**: KKR having significant analgesic activity against the acetic acid-induced writhing in mice and anti-inflammatory activity against the carrageenan -induced paw oedema in rats.

KEYWORDS: Kaalakodi rasam, analgesic and anti-inflammatory study, siddha, osteoarthritis.

INTRODUCTION
Siddha system is the ancient system of heeling and it's based on combination of medical practices and preventive methods, as well as alchemy and mysticism. Other system of medicines is gives priority to herbal preparation for treating disease but our Siddha medicines preparation we are using combination of plants, metals, and minerals. Herbo-metallic preparations play an important role in traditional system of medicine. While such preparation are held to be safe, effective in small doses, when prepared and used following specific guidelines of Siddha text.[1]

One such effective & safe herbo-metallic formulation is kaalakodi rasam. In Siddha literature KKR is a herbo-metallic formulation treated for osteoarthritis. Osteoarthritis (OA) is a disease of cartilage degradation, which results pain in major joints, especially in knee joint. Globally OA ranks eighth in all diseases and covers around 15% proportions among all musculoskeletal problems. Clinical symptoms and radio-diagnosis are the basis of diagnosis used for OA characterization. India has higher proliferative rate of OA among world and expected to be at top rank in chronic diseases till 2025.[2]

AIM AND OBJECTIVE
To study of analgesic and anti-inflammatory activity in the siddha drug formulation Kaalakodi rasam.

MATERIAL AND METHODS
Collections of drugs
The drugs were collected from reputed raw drug shop in Madurai and Nagerkoil. These drugs were analysed and authenticated by Government Siddha Medical College, Palayamkottai.

Method of preparation of KKR
The KKR drug was prepared under siddha literature Chikitcha Rathnam Deepam Ennum Vaidhya Chinthamani by kannusaamy pillai. First make gajili with Purified rasam (Hydragirum) & ganthagam (sulphur). Then fried thalagam (Trishulphate of arsenic), lingam (Red sulphide of mercury), naabi (aconitum napellus root), naervalam (croton tiglum seeds), sukku (zingiber officinale root), arisithipilli (piper longum fruit), milagu (piper nigrum fruit), kadukai (terminalia chebula fruit) thaanrikaai (terminalia bellirica fruit), nelli (phyllanthus emblica fruit), nerunjil (tribulus terres fruit) are in a pan and make it as powder. Then mix with Alli kilangu juice (nymphaea nouchalli fruit) and grind in a kalvam for 3 samam (9hrs). Then making pills as 130 mg

size and dried in shadow and shut in a ceramic container.[3]

Selection of animal species

Animals were obtained from the King Institute of Preventive medicine, Guindy, Chennai. The rats were used after obtaining Institutional Animal Ethical Committee clearance bearing the IAEC approval No. IAEC/XLIV/32/CLBMCP/2014 They were kept in C.L. Baid Metha College of Pharmacy, Thoraipakkam Chennai.

Housing and Feeding conditions

Animals were fed with a balanced standard pellet diet procured from Sai Meera Foods, Bangalore and maintained under standard laboratory conditions, providing 19-25^{0}C temperature, relative humidity 30-70% standard light cycle (12 hour light, 12 hour dark) and water Ad-libitum. Unlimited supply of drinking water and conventional lab diets were adopted. Animals were kept in individual cages with raised floors of wide mesh to prevent coprophagy and numbered. Animal welfare guidelines were observed during the maintenance period and experimentation.

Preparation of animals

The animals were randomly selected, marked to permit individual identification, and kept in their cages for 5 days prior to dosing to allow for acclimatization to the laboratory conditions.

Preparation of Doses

The powdered form of kaalakodi rasam was mixed uniformly in 2% CMC and made into uniform suspension to achieve 200mg/ml as main stock solution and used in this study and was found suitable for dose accuracy.

ANALGESIC ACTIVITY

Analgesia is defined as a state of reduced awareness to pain, and analgesics are substances, which decrease pain sensation (pain - killers) by increasing threshold of painful stimuli. The commonly used analgesics are Aspirin, Paracetamol (non - narcotic type) and Morphine (narcotic type). Painful reaction in experimental animals can be produced by applying noxious (unpleasant) stimuli such as (i) thermal (radiant heat as a source of pain), (ii) chemical (irritants such as acetic acid and bradykinin) and (iii) physical pressure (tail compression).[4] In the present study the attempt has been focused to evaluate the Analgesic activity of kaalakodi rasam against Acetic acid induced writhing response in Swiss albino mice.

Grouping of animals

The animals were divided into four groups each containing six animals

Group I: served as untreated control and received vehicle (1ml/kg p.o)

Group II: served as standard group and received Diclofenac sodium (25mg/kg. p.o)

Group III: animals were treated with 100 mg/kg of Kaalakodi rasam

Group IV: animals were treated with 200 mg/kg of Kaalakodi rasam

Acetic acid Induced Writhing Test (Chemical Stimulation)

The analgesic activity of the samples was evaluated using acetic acid induced writhing method in mice. In this method, acetic acid is administered intra-peritoneal to the experimental animals to create pain sensation. As a standard control, any standard NSAID drug can be used. In the present study diclofenac sodium was used to serve the purpose. The Kaalakodi rasam was administered orally in two different doses (100, 200 mg/kg body weight) to the mice after an overnight fast. Test samples and vehicle were administered orally 30 minutes prior to intra peritoneal administration of 0.7% v/v acetic acid solution (0.1ml/10g) but diclofenac sodium was administered 15 minutes prior to acetic acid injection. Then the animals were placed on an observation table. Each mouse of all groups observed individually for counting the number of writhing they made in 15 minutes commencing just 5 minutes after the intra peritoneal administration of acetic acid solution.

Full was not always accomplished by the animal, because sometimes the animals started to give writhing but they did not complete it. This incomplete writhing was considered as half writhing. Accordingly, two half-writhing were taken as one full writhing.[4] The number of writhes in each treated group was compared to that of a control group while diclofenac sodium (25mg/kg) was used as a reference standard (positive control).

The percentage protection against abdominal writhing was used to assess the degree of analgesia and was calculated using the formula.[5]

$$\% \text{ inhibition: } \frac{\text{Mean control group} - \text{Mean treated group}}{\text{Mean of control group}} \times 100$$

ANTI-INFLAMMATORY STUDIES

Everyone have has personal experience of inflammation and pain. The classic signs of inflammation have long been recognized; the tissues become red, swollen, tender, or painful, there is local heat and the patient may be febrile. Inflammation can be categorized mainly as Chronic and Acute inflammatory disease.[7]

Acute and chronic inflammatory diseases are still one of the most important health problems in the world. Although several agent known to treat inflammatory disorders, their prolonged use often leads to gastric intolerance, bone marrow depression, water and salt retention. For this reason there is a need to find and develop new anti-inflammatory drugs with low side effects.[6]

Formaldehyde, Dextran, Carrageenan, histamine, and other inflammagen induced inflammation model are frequently used in screening for the anti-inflammatory activity of new compounds, where implantation off foreign body under the skin is often used to investigate the effects of drugs on the proliferative phase of the chronic inflammation model.[5]

In the present study the attempt has been focused to evaluate the anti-inflammatory activity of kaalakodi rasam**.** Using carrageen induced paw-oedema in rats as a model. For comparison purpose, indomethacin is taken as a reference compound.

Carrageenan induced paw oedema in rats

Anti-inflammatory activity of Kaalakodi rasam was assessed by carrageenan paw oedema.[8] Rats were divided into 4 groups (n = 6). Animals of all the groups injected with 0.1 ml of carrageenan in 0.9% normal saline, under the plantar aponeurosis of the right hind paw.

1. Group-I: animals (carrageenan control) received vehicle 30 min prior to administration of carrageenan injection.
2. Group-II: animals the standard reference group was given p.o. aqueous solution of Indomethacin (5mg/kg), 30 min prior administration carrageenan injection.
3. Group-III: animals received 100mg/kg of Kaalakodi rasam 30 min prior to administration of carrageenan injection.
4. Group- IV: animals received 200mg/kg of Kaalakodi rasam 30 min prior to administration of carrageenan injection.

The paw volume was measured using plethysmograph immediately after 1 hour of injection again at 2, 3 and 4th hour eventually after treatment. The mean volume was compared with control group. Inhibion of swelling is compared with that of control group.[4]

RESULT

Acetic acid Induced Writhing Test

Table - I

Treatment	Dose	No of writhing (mean ±S.E.M)	Inhibition (%)
Control	Saline	34.88± 1.86	-
Diclofenac sodium	25mg/p.o	6.98± 4.16*	80.1
Kaalakodi rasam	100mg	17.42 ± 0.57*	50.1
Kaalakodi rasam	200mg	10.45 ± 0.62*	70.45

Carrageenan induced paw oedema in rats

Table - II

Group	Dose mg/kg	Mean Paw volume in ml			
		1hr	2hr	3hr	4hr
Control	Vehicle	1.38± 0.064	1.42± 0.005	1.48± 0.009	1.50± 0.003
Indomethacin	10 mg/kg	1.36± 0.061*	1.30± 0.034*	1.23±0.077**	1.05± 0.060**
Kaalakodi rasam	100 mg/kg	1.34± 0.020	1.33± 0.015*	1.25± 0.066*	1.16± 0.049*
Kaalakodi rasam	200 mg/kg	1.31± 0.043	1.30± 0.010	1.22± 0.078	1.07± 0.061**

DISCUSSION

KKR possess significant analgesic and anti-inflammatory potential as evidenced from table I and table II. Thus Kaalakodi rasam having analgesic activity against the acetic acid-induced writhing in mice and anti-inflammatory activity against the carrageenan - induced paw oedema in rats. These findings support the use of KKR in traditional system of medicine for the management of pain and inflammatory conditions.

CONCLUSION

Kaalakodi rasam having significant analgesic activity against the acetic acid-induced writhing in mice and anti-inflammatory activity against the carrageenan -induced paw oedema in rats.

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