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EVALUATION OF ANALGESIC ACTIVITY OF SIDDHA HERBAL DRUG *KEELVAYU NIVARANA CHOORANAM* BY ACETIC ACID INDUCED WRITHING RESPONSE METHOD

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ABSTRACT

The Siddha herbal formation *Keelvayu Nivarana Chooranam* (KVNC) was studied for its in vivo analgesic activity by acetic acid induced writhing response method in Wister Albino Rats. KVNC has four ingredients which includes *Nannariverpattai Chooranam (Hemidesmus indicus*), *Parangipattai Chooranam* (*Smilax chinensis*), *Seemai Amukara Chooranam (Withania somnifera), Chittaraththai Chooranam (Alpinia officinarum*) and mentioned in Siddha literature for treating *Keelvatham* (Arthritis). Pain is an unpleasant sensation localized to a part of the body and can be debilitating. People are looking for treatment with cost effective and less adverse drug reactions. This study was aimed to compare the test drug with the standard drug. Wistar albino rats were divided into four groups (n = 6). Group I received acetic acid, Group II received aspirin (100 mg/kg b.w.p.o.) Group III and IV received KVNC at the doses of 100 mg/kg and 200 mg/kg b.w., p.o. respectively. Five minutes after the acetic acid injection, the number of times that each animal presented abdominal constriction was counted for 15 consecutive minutes. Percentage of inhibition in Group II (Standard drug) shows 84.85%, Group III (KVNC 100mg) shows 75.76%, Group IV (KVNC 200mg) shows 78.79%. The Drug KVNC exhibited potent analgesic effects in animal model.

KEYWORDS: *Analgesic activity, Siddha drug, Keelvayu Nivarana Chooranam, Writhing test.*

1. INTRODUCTION

In Siddha system of medicine, there are various medicines used for painful conditions. The word "Siddha" not only denotes simplicity, uniqueness, ancientness, nobility, truth and purity but includes all these senses and stands a unique, lofty entity. "Siddhi" refers to a yogic state. "Siddhars" are said to be the yogis, having lived a complete life.[1] *Keelvayu Nivarana Chooranam* (KVNC) is a compound drug mentioned in the literature The Pharmacopoeia of Siddha Research Medicine for treating *Keelvatham* (Arthritis). Arthritis causes pain and inflammation, making disability. Pain is an unpleasant sensation localized to a part of the body and can be debilitating. Pain has a major impact on quality of life, sleep and socio-economic factors, such as one's ability to adequately perform at work. Many chronic pain sufferers wind up taking huge amounts of analgesic drugs. Analgesic drug products that are used widely are opioids, acetaminophen and the Non-Steroidal Anti-Inflammatory Agents (NSAIDs), all of which have serious, potential toxicities, even when used in therapeutic doses.[2]

In this condition patients suffer physically and mentally. So obviously they are in the period of expecting an alternative medicine. While in this situation, they are so many medicines are available in Siddha system of medicine which was all given by the Siddhars to help us to live a disease free life. The Author has chosen the Siddha herbal drug "*KEELVAYU NIVARANA CHOORANAM*" and the present study is carried out to validate the analgesic activity of KVNC for treating painful diseases like arthritis.

2. MATERIALS AND METHODS

2.1 Drug Selection[3]

Keelvayu Nivarana Chooranam (KVNC) was taken as a compound drug from the literature The Pharmacopoeia of Siddha Research Medicine (chapter 2-14)

2.2 Ingredients

Nannariverpattai Chooranam (Hemidesmus indicus)- 116 g
Parangipattai Chooranam (*Smilax chinensis*) - 116 g
Seemai Amukara Chooranam (Withania somnifera)- 116 g
Chittaraththai Chooranam(Alpinia officinarum) - 58 g

2.3 Source of Collection

All the raw drugs were bought from Ramasamy chetty country drug shop at Parry's corner, Chennai, Tamilnadu, India.

2.4 Identification and Authentication of the drug

All the raw drugs were identified and authenticated by the *Gunapadam* experts in Government Siddha Medical College, Arumbakkam, Chennai – 106. The specimen sample of all the herbs have been preserved in PG *Gunapadam* department individually for future reference. Ref No: GSMC/PGGM/014-017/2014-2017.

2.5 Purification of the drugs

All the drugs mentioned here were purified as per the Siddha literature.

i. *Nannariverpattai* were washed in the running tap water to remove the soil andimpurities.
ii. *Parangipattai* was dried and powdered and then it was purified by *Pittaviyal* method (steam cooking in milk). A mud pot was taken and it was half filled by milk and half filled by pure water. The mouth of the pot was sealed by a cloth. This *chooranam* then placed over the cloth and the pot was heated. The same drug was later dried and powdered then sieved again.
iii. *Amukara* was dried and powdered and then it was purified by *Pittaviyal* method (steam cooking in milk). A mud pot was taken and it was half filled by milk and half filled by pure water. The mouth of the pot was sealed by a cloth. This *chooranam* then placed over the cloth and the pot was heated. The same drug was later dried and powdered then sieved again.
iv. *Chittaraththai* were washed in the running tap water to remove the soil and impurities.

2.6 Preparation of the trial drug – *Keelvayu Nivarana Chooranam*

2.6.1 Procedure

All the above purified ingredients were powdered in an iron mortar separately and it was sieved by a cotton cloth. Then these powders were mixed together and bottled up. It was labeled as *Keelvayu Nivarana Chooranam* (KVNC).

2.6.2 Purification of the *Chooranam*: Steaming process (*Pittaviyal murai*)

The *Keelvayu Nivarana Chooranam* was purified by *pittaviyal* method (steam cooking in milk) as per Siddha classical literature. A mud pot was taken and it was half filled by milk and half filled by pure water. The mouth of the pot was sealed by a cloth. This chooranam then placed over the cloth and the pot was heated. After this process the drug was later dried and powdered then sieved again. It was used for the further study.

2.6.3 Storage of the drug

The prepared test drug was stored in a clean, air tight glass container. The contents were inspected frequently to avoid moisture and insects.

2.7 Evaluation of Analgesic activity - Acetic acid induced Writhing test

2.7.1 Test Animals and Test Conditions

Wistar albino rats (150-200gm) were obtained from Wistar albino rats (150-200gm) were obtained from TANUVAS, Madhavaram, and Chennai. All the animals were kept under standard environmental condition (22±3°C). The animals had free access to water and standard pellet diet (Sai meera foods, Bangalore).

2.7.2 Preparation of animals

The animals are randomly selected, marked to permit individual identification, and kept in their cages for at least 7 days prior to dosing to allow for acclimatization to the laboratory conditions.

2.7.3 Procedure - Acetic acid induced Writhing test

Wistar albino rats were divided into four groups (n = 6). Group I received acetic acid (1% v/v, 10 ml/kg b.w., i.p.) and writhing reflex was noted for the period of 15 minutes. Group II received aspirin (100 mg/kg b.w.p.o.) Group III and IV received KVNC at the doses of 100 mg/kg and 200 mg/kg b.w., p.o. respectively. 30 min after aspirin and KVNC administration, group II and III received acetic acid (1% v/v, 10 ml/kg b.w., i.p.) and writhing reflex was noted for the period of 15 min. The animals were previously treated, by oral administration (p.o.) with *KeelvayuNivarana Chooranam* 1 h before the stimulation with acetic acid. Control animals received the same volume of vehicle. Five minutes after the acetic acid injection, the number of times that each animal presented abdominal constriction was counted for 15 consecutive minutes.

3. RESULTS AND DISCUSSION

This present study aimed to validate the Analgesic effect of KVNC on acetic acid induced writhing in rats. Writhing method is the most common test for evaluating the analgesic efficacy of drugs/compound in rodents. Abdominal constrictions in rats were caused by the intraperitoneal injection of acetic acid. The abdominal constriction response induced by glacial aceticacid is a sensitive procedure to establish peripherally acting analgesics. This response is thought to involve local peritoneal receptors.[4]

The number of writhing observed during a 20 min period in control group was 33.53±0.87. It was also observed that animals in test group showed delayed onset of writhes (after 10min) as compared to other groups in which onset of writhes was within 5 min. The *Keelvayu Nivarana Chooranam* (100 mg/kg, p.o.) showed the significant ($P<0.001$) reduction in the number of writhes induced by acetic acid. Aspirin significantly reduced the

number of writhes (P<0.001). There was a significant, inhibition of pain response in rats.

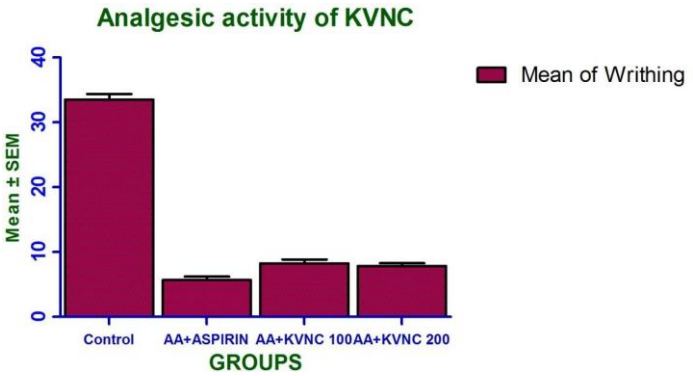
The Acetic acid -induced writhing response is believed to be produced by the liberation of endogenous substance, notably metabolites of the arachidonic cascade. Hence, Acetic acid causes analgesia by liberating endogenous substances including serotonin, histamine, bradykinin which stimulates pain nerve endings. Local peritoneal receptors are postulated to be partly involved in the abdominal constriction response.[5] The method has been associated with prostanoids in general, i.e. increased levels of PGE_2 and $PGF_2\alpha$ in peritoneal fluids as well as lipoxygenase products.[6]

Table No. 1: Analgesic effect of KVNC on acetic acid induced writhing in rats.

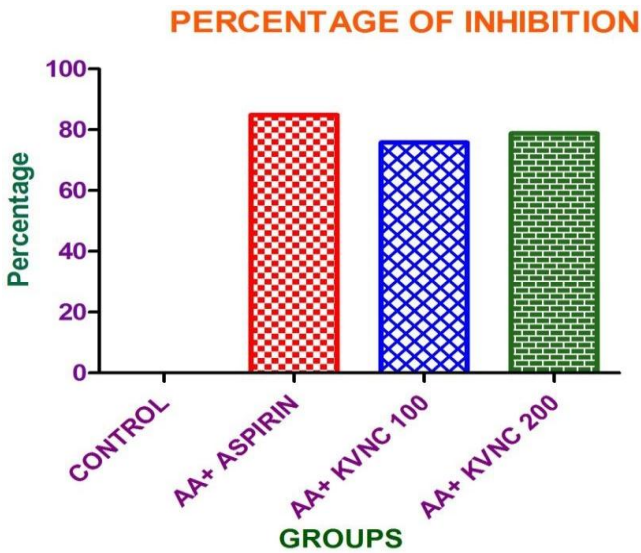
Groups	Treatment	Dose	Mean of writhing ± SEM (sec)	% of Inhibition
Group I (control)	Acetic acid (1% v/v)	10 mg/kg	33.53±0.87	_
GroupII (standard)	Acetic acid + Aspirin	100 mg/kg	5.68±0.53***	84.85%
Group III	Acetic acid + KVNC	100 mg/kg	8.25±0.60***	75.76%
Group IV	Acetic acid + KVNC	200 mg/kg	7.85±0.42***	78.79%

*Values are mean ± SEM (n = 6 Dunnett' test). ***p< 0.001 when compared to control.*

Percentage of inhibition in Group II (Standard drug) shows 84.85%, Group III (KVNC 100mg) shows 75.76%, Group IV (KVNC 200mg) shows 78.79%. Group IV which had been treated with 200 mg of KVNC shows very close percentage of inhibition to Group II standard drug. Therefore, the *Keelvayu Nivarana Chooranam* might inhibit the synthesis and/or release of these endogenous substances. Significant reduction in abdominal constriction compared with vehicle treated animals was considered as antinociceptive response. So it can be concluded that the Siddha drug *Keelvayu Nivarana Chooranam* possess excellent peripheral analgesic property which is equipotent to standard drug used in this study.



Graph No. 1. Analgesic activity of KVNC.



Graph No.2. Percentage inhibition of KVNC.

4. CONCLUSION

The present study suggests that the Siddha poly herbal formulation has significant percentage of inhibition that shows marked analgesic activity. The drug may have the phyto constituents which inhibit enzymes for reducing analgesic conditions. Further clinical studies are needed for patients suffering from painful conditions and inflammation diseases requiring analgesics.

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