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AN EXPERIMENTAL EVALUATION OF PAVATE (*Pavetta indica Linn*) A FOLK MEDICINAL PLANT FOR ANTI INFLAMMATORY AND ANALGESIC ACTIONS IN WISTAR ALBINO RATS-AN VIVO STUDY

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ABSTRACT

Background & Objectives: Plants have played significant role in maintaining human health and improving the quality of human life for thousands of years. *Pavetta indica Linn* is a stout shrub of Rubiaceae family distributed throughout the India and used by the folklore practitioners in various inflammatory conditions. The aim of this study is to evaluate the anti-inflammatory and analgesic actions in wistar albino rats (in vivo). **Methods:** The detailed review of trial drug and disease were carried out. The efficacy of *Pavetta indica Linn* kashaya was evaluated for anti-inflammatory and analgesic action in wistar albino rats. Carrageenan induced paw edema and cotton pellet induced granuloma formation model was used to study anti-inflammatory activity. Hot plate method was used for analgesic activity. **Results & Conclusion:** The test drug *Pavetta indica Linn* was safe even in high dose of 2000mg/kg in acute oral toxicity test. The test drug revealed to be potent anti-inflammatory actions both in terms of acute and chronic studies and also exhibited significant analgesic effect in experimental model indicating the presence of central analgesic activity.

Keywords: Anti-inflammatory; Analgesic; Wistar albino rats; *Pavetta indica Linn*

INTRODUCTION

Ayurveda means the 'the science of life'. The documentation of Ayurveda is referred in Vedas(5000 BC).The Indian science of life is linked with the origin of universe and developed from the various Vedic hymns describing fundamentals/philosophies about the world and life, diseases and medicines. Ayurveda is a natural therapy which has been preserved, developed and now being globalized. Inflammation is the local response of living tissues to injury from any agent which could be microbial, immunological, physical or chemical agents. Cardinal signs of inflammation are rubor, tumor, calor, dolar

and function Laesa[2]. The clinical presentation of Inflammation shows features like pain, swelling and redness. For the treatment of Inflammation and pain non-steroidal anti-inflammatory drugs (NSAID'S) are used. None of the available agents however has appreciable safety profiles on long term use. Often patients turn to the traditional system of medicine for the treatment of chronic inflammatory disorders like osteoarthritis or rheumatoid arthritis. Folk medicine or traditional medicine is one of the natural health care system is being practiced by all human cultures from the beginning of civilization. Traditional/folk medicine which is mostly documented has been handed down orally from one generation to another. Present study is about Pavetta indica Linn belonging to Rubiaceae family. A stout bushy shrub distributed throughout the India and used by folklore practitioners in various disorders like osteoarthritis in the form of decoction of leaves[1]. Local fomentation with the leaves is useful in relieving the pain in case of hemorrhoids. It is also used in the form of paste (*kalka*) in *Agni lepa chikitsa* both internally and externally in folk practice. Though there was work done on anti-inflammatory activity by using alcohol extract, however based on Ayurveda principle by using *kwatha* (decoction) form of this drug has not been carried out so far. Hence to study in detail experimentally research work is being taken up.

Materials and methods

Plant collection & authentication: The drug was collected from near the college premises of M. I. A. M. S Manipal and nearby places of Udupi and is authenticated by K. Gopalakrishna Bhat, (Retd.) Professor, Dept. of Botany, Poornaprajna College, Udupi.

Preparation of *kwata* (Decoction): The leaves of *Pavetta indica Linn* were cleaned properly, dried in shade and coarsely powdered. *Kwata (*decoction) was prepared based on classical reference [3]

- **TED X 1 Group (Decoction of *Pavetta indica Linn* normal dose)**

1part (7.5g) of the drug is added with 16 parts (120ml) of water, boiled and reduced to 1/8th part (15ml). Decoction is allowed to self-cool and used for oral administration.

- **TED X 2 Group (Decoction of *Pavetta indica Linn* double dose)**

1part (10g) of the drug is added with 16parts (160ml) of water, boiled and then reduced to 1/8th part (20ml). This decoction is allowed to self-cool and used for oral administration.

Acute toxicity study of drug: Acute toxicity test to determine the LD_{50} dose of the Pavetta indica Linn kwata. Test formulation was administered through oral route at different dose level in 5 different rat as 175mg/kg, 550mg/kg, 2000mg/kg, 2000mg/kg, 2000mg/kg, all the animals were observed at ½, 1, 2, 3,4, 24,48hrs dosing and there after daily once for mortality during the entire period of study (i.e.14 days). Maximum tolerated dose was calculated by employing OECD 425 guidelines with AOT software[4]. The LD_{50} value was found to be 2000mg/kg.

Experimental animals: Albino rats of Wistar strain of either sex between 160-280g were obtained from animal house attached to department of Pharmacology, SDM Research Centre Udupi, India. The experimental protocol was approved from the institutional ethical committee under the reference no SDMCRA/IAEC/MU-DG-29/05/2017. The animals were fed with normal diet and water *ad libitum* throughout the study. The animals were marked with the help of picric acid to permit individual identification and kept in their cages for 7 days prior to the start of dosing to allow for acclimatization to the laboratory conditions.

Animal grouping: Wistar albino rats of either sex weighing 160-280g, 24 rats were divided into 4 different groups, six in each group. Control group were administered with normal tap water. The standard groups were administered with Diclofenac sodium[5] 100mg/kg and Dexamethasone[6] 1000µg/kg for analgesic and anti-inflammatory study respectively. The test groups were administered with normal and double dose of kwata.

Procedure for testing acute inflammatory activity

Carrageenan induced hind paw oedema test in rats were carried out by method of Winter et al[7]. Acute inflammation was produced by injecting 0.1ml of 1% Carrageenan solution into sub plantar surface of rats hind paw. The group specific drugs were administered 1 hour before the Carrageenan injection. The volume up to the tibiotarsal articulation was measured by using a Plethysmometer at basal, 2h, 3h and 6h after Carrageenan injection and expressed as ± SEM (n=6). Average volume for trial group and standard group were compared for statistical significance with those of control group.

Procedure for testing chronic inflammatory activity

Cotton pellet induced granuloma formation test in rats were carried out by Garcia et al method[8]. The rats were anaesthetized with ether. Dorsum would be

shaved and swabbed with 70% (v/v) alcohol. Midline incision of 1 cm would be made in the intrascapular region. A small tunnel would be made on either side of the incision with the help of small blunt forceps. One sterile cotton pellet weighing 100 mg (prepared by rolling a cotton piece of 100 mg and sterilized by autoclaving for 30 minutes under 15 lbs. pressure) would be inserted per tunnel and closed the incision with interrupted sutures after expelling the air from the tunnel. Group 1 was treated with tap water and considered as control group. Group II and III were administered with the test drug normal and double dose group of *kwata* respectively for 7 consecutive days starting from the day of implantation. The fourth group would be taken as standard and administered with the standard drug Dexomethasone (1000µg/kg p.o). The rats were sacrificed on 8th day the blood collected from the orbital plexus of rat from that serum was separated and analyzed for the determination of C-reactive protein and dissected for collection of thymus, spleen, adrenal glands and cotton pellets would be removed and cleaned of extraneous tissue and dried by placing them in a hot air oven overnight at 80°C and then weighed. The difference between the initial weight and the final weight of the pellet after drying would be taken as the granuloma tissue weight. The result would be expressed as mg granulation tissue formed per 100 g body weight .The weight of adrenal glands, spleen and thymus were noted. Further these organs were placed in a clean glass bottle containing 10% formalin and sent to the pathology laboratory for histopathological investigations.

Procedure for testing chronic inflammatory activity:

Eddy's hot plate method

The analgesic activity of Pavetta indica Linn was assessed using hot plate method of Eddy and Leimbach (1953)[9]. The temperature was maintained at 55 ± 0.2^0C. This is hot enough to cause discomfort without tissue damage. Animals licked their paw and jumped as an indication of pain. These rats were treated with as follows; control group received normal water. The test groups were treated with normal and double dose of *kwata*. The standard group received Diclofenac sodium 100mg/kg by the oral route. The time taken by the animal to lick the fore or hind paw or jump out of the plate was takes as the latency time.

Statistical Analysis:

All data were expressed as Mean±S.D. The statistical analysis was done by One way ANOVA (Tukey) followed by Dunnett`s Multiple Comparison `t` test as post- HOC test using Graph pad InStat version 3.00 for Windows 98, Graph pad Software, San Diego, California, USA. P-value considered as significant are indicated by "*" and "**" for $p<0.05$ and $p<0.01$ respectively

RESULTS

Table 1: The effect of *Pavetta indica kwata* on percentage increase of paw oedema at different time interval

	Mean ±SEM Percentage increase of paw oedema at different time interval				
Group	Basal	1st hour	3rd hour	6th hour	24th hour
Control	0.771±0.013	0.985±0.052	1.26±0.084**	1.138±0.086**	1.098±0.049**
Standard group	0.703±0.020	0.743±0.016	0.746±0.026	0.745±0.035	0.715±0.019
TED	0.798±0.024	1.111±0.021**	1.31±0.047**	1.125±0.065**	0.931±0.060
TED X 2	0.736±0.028	0.933±0.055**	1.011±0.050**	1.155±0.105**	0.801±0.043

Data: MEAN ± SEM.

**p < 0.01 in comparison to control group.

The TED and TED X 2 administered groups showed there was extremely significant increase in paw volume in test group of 1st hour, 3rd hour and 6th hour and non-significant increase in 24th hour when compared to basal volume of same group.

Table 2: Effect of *Pavetta indica kwata* on cotton pellet implanted Granuloma formation

Group	Mean ± SEM: Granuloma tissue weight (mg)/100g body weight	% Change
Control	0.292 ± 0.043	-
Standard	0.227 ± 0.020	22.26 ↓
TED	0.394 ± 0.039	34.93↑
TED X 2	0.179 ±0.027	38.69↓

Data: MEAN ± SEM.
The data shows there was decrease in cotton pellet implanted granuloma formation in standard group and TED X 2 group but increase in TED group when compared to control group, the observed decrease and increase was found to be statistically non-significant.

Table 3: Effect of *Pavetta indica kwata* on weight of Spleen in cotton pellet implanted rats

Group	**Mean ± SEM Spleen weight (g)**	**% Change**
Control	1.185± 0.156	-
Standard	0.63 ± 0.069**	46.83 ↓
TED	0.648 ± 0.088**	45.31 ↓
TED X 2	0.956 ± 0.083	19.32 ↓

Data: MEAN ± SEM.
The data shows there was decrease in weight of spleen in standard group and TED group when compared to control group, the observed decrease was found to be statistically very significant. The data shows there was decrease in weight of spleen in TED X 2 group when compared to control group, the observed decrease was found to be statistically non- significant.

Table 4: Effect of *Pavetta indica kwata* on weight of Thymus in cotton pellet implanted rats

Group	Mean ± SEM Thymus weight (g)	% Change
Control	0.633 ± 0.040	-
Standard	0.50 ± 0.022	21.01 ↓
TED	0.52 ± 0.043	17.85 ↓
TED X 2	0.57 ± 0.115	9.95 ↓

The data shows there was decrease in weight of Thymus in standard group, TED group and TED X 2 group when compared to control group, the observed decrease was found to be statistically non-significant.

Table 5: Effect of *Pavetta indica kwata* on weight of Adrenal gland in cotton pellet implanted rats

Group	Mean ± SEM Adrenal gland weight(g)	% Change
Control	0.106 ± 0.013	-
Standard	0.092 ± 0.015	13.20 ↓
TED	0.111 ± 0.013	5.28 ↑
TED X 2	0.11 ± 0.006	3.77 ↑

Data: MEAN ± SEM.
The data shows there was decrease in weight of Adrenal gland in standard group when compared to control group, the observed decrease was found to be statistically non-significant. The test drug TED and TED X 2 administered groups showed increase in weight of Adrenal gland when compared to control group, the observed increase was found to be statistically non-significant.

Table 6: Effect of Pavetta *indica kwata* on serum C – Reactive Protein level in cotton pellet implanted rats

Group	Mean ± SEM C-Reactive Protein level	% Change
Control	0.023 ± 0.002	-
Standard	0.090 ± 0.031*	293.04 ↑
TED	0.033 ± 0.006	43.47 ↑
TED X 2	0.038 ± 0.020	65.21 ↑

Data: MEAN ± SEM.
*$p < 0.05$ in comparison to control group.
The data shows there was increase in C – reactive protein in standard group and when compared to control group, the observed increase was found to be statistically significant. The data shows there was increase in C – reactive protein in TED and TED X 2 group when compared to control group, the observed increase was found to be statistically non-significant.

Table 7: The effect of *Pavetta indica kwata* on paw lick/jump response at different time interval.

	Mean ±SEM Pain threshold at different time interval					
Group	60mins	90mins	120mins	180mins	240 mins	24 Hour
Control	6.265± 1.156	8.696 ± 1.007	12.701±2.341	10.47 ± 1.925	8.051 ± 0.705	7.63 ± 0.95
Standard group	12.325±2.254 *	16.448 ± 1.408	13.763±1.909	11.538 ±2.217	12.608 ±2.863	13.84 ± 4.83
TED	8.745 ± 0.872	11.315 ± 1.661	15.458±1.196	19.775 ±2.823	17.575 ± 4.114	13.37 ±2.70
TED X 2	13.55±1.970*	31.435±9.069**	37.88 ± 6.980 **	48.803±12.107**	36.206±5.693**	7.716 ± 1.570

Careful analysis of the results indicate that in comparison to initial values pain threshold was found to be elevated at 60 min after test drug administration and continued till 240 min in the TED X 2 administered group. This may indicate that the effect of test drug in double dose exhibit significant central analgesic activity compared to standard drug.

DISCUSSION

Inflammation comprises of three phases namely acute, sub-acute and chronic. In acute inflammation due to change in small blood vessels, fluid and granulocytic cells accumulate at the site of injury. The initial phase of the oedema is due to the release of histamine and serotonin. The second accelerating phase of swelling is due to the release of prostaglandin and lysosome synthesis or activity. In the present study among the two dosage forms (TED and TED X 2) of *Pavetta indica kwata*, double dosage (TED X 2) form may significantly inhibited both the phases of carrageenan induced paw oedema, while single dosage (TED) form inhibited only to non-significant manner.

The cotton pellet granuloma method has been widely employed to assess the transudative, exudative and proliferative components of chronic inflammation. The fluid absorbed by the pellet greatly influences the wet weight of the granuloma and dry weight correlates well with the amount of granulomatous tissue formed. In present study TED X 2 dose showed moderate and statistically non-significant decrease in dry cotton pellet weight in comparison to standard group. This indicates that the test drug in double dose may have moderate anti-proliferative activity. But TED (Single) dose did not showed any significant impact as compared to TED X 2 this may indicate that there is no effect of TED dose in reducing the synthesis of **t**proteins, collagen and infiltration of macrophages. Another important point to be noted that earlier study was done on inflammatory activity by using methanol extract was evaluated against several models of inflammation such as carrageenan, histamine and dextran induced pedal inflammation in rats. The extract showed 48.41%, 41.10% and 24.22% inhibition respectively [10]. But in present study *kwata (*decoction) showed 77.53% decrease in paw oedema.

In histopathological examination of the spleen, thymus and adrenal gland sections tissue architecture was not affected significantly in any of the control, test drug and standard drug administered groups. It clearly indicates they do not have lymphophenic activity. In TED X 2 dose administered group Inflammation was reduced compared to TED and control group. The effect was much better in TED X 2 administered group. This indicates that the test formulation has better anti-inflammatory effect.

The hot plate method is a selective model for studying the central analgesic activity. In this model peripheral analgesic mechanisms are not involved. The effect of test drug in double dose exhibit significant central analgesic activity compared to standard drug.

Probable mode of action

The test drug *Pavetta indica Linn* is predominant with *tikta rasa, laghu* and *ruksha guna, sita virya, katu vipaka*[11]. This unique combination will be beneficial in pacifying the aggrevated *vata* and *kapha doshas*, which is beneficial in breaking the pathogenesis of inflammation.

In addition the test drug possesses activities like *Vedana stapana* (Analgesic), *Shothahara* (Anti-Inflammatory), *Kamala* (Jaundice), *Kandu* (Itching), *Visphota* (Blister), which are again beneficial in pacifying the symptoms of inflammation.

Considering all these activities it can be said that *Pavetta indica Linn* is a beneficial in pathological conditions where inflammation is the predominant one.

CONCLUSION

1. The test drug *Pavetta indica Linn* was safe even in high dose of 2000mg/kg in acute oral toxicity test.

The test drug revealed to be potent anti-inflammatory actions both in terms of acute and chronic studies. And also exhibited significant analgesic effect in experimental model indicating the presence of central analgesic activity.

2. Thus provides an evidence for the presence of the desired activity in experimental animals, which can be offered, has the pharmacological basis for the clinical efficacy of the drug in the management of pain and inflammation in arthritic disorders.

REFERENCES

1. Kirtikar. K. R .and Basu.B.D.Indian medicinal plants; Published by- International book distributors; vol-2; Edition-2nd; Dehradun; 2006; Pp 1592, Page no:1291-1292
2. Harsh Mohan; Text book of Pathology; Jaypee Brothers; New Delhi; 2015; Pp-954; Page no-116
3. Murthy Srikanth; Sarangadhar Samhita; Choukhambha Orientalia; Varanasi; 6th Edition; 2006; Pp-250; Page no-56
4. OECD Guideline for Testing of Chemicals (Internet). [Updated 2001 December 17; cited 2019 April 3]. Available from: www.oecd.org/chemicalsafety/testing/oecdguidelinesforthetestingofchemicals.htm
5. Satoskar Bhandarkar.S.D, Ainapure. S.S; Pharmacology and Pharmacotherapeutics; Popular Prakashan; Mumbai; 17th Edition; 2001; Pp-1111; Page no-166,174
6. Satoskar Bhandarkar.S.D, Ainapure. S.S; Pharmacology and Pharmacotherapeutics; Popular Prakashan; Mumbai; 17th Edition; 2001; Pp-1111; Page no-928
7. Winter C.A, Risley E.A, Nuss G.W, (1962); Proc. Soc. Exp. Biol; 111,544
8. Garcia Ranganell B.J, Bells L, Sanchez M.S, Form J.(1980) Arzneim-Forsch; 30(II); 1091
9. Niraltha G Sowmya: Experimental Evaluation of Anti-Inflammatory, analgesic and anti-inflammatory in chloroform extract of eight plants; Bangalore; Rajiv Gandhi University of Health Sciences: 2013-2014
10. Mandal C Subhash; Evaluation of anti-inflammatory potentials of pavetta indica Linn, leaf extracts; Phytotherapy research, vol-17 .2003.
11. The Ayurvedic Pharmacopoeia of India, part-I, vol-VI, Edition-1st 2008 New Delhi; Pp-426; Page no-122,123.

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