Sudarshan Publication

Available online at S-epub.in/ojs/

THE PHARMA RESEARCH
An International Journal of Pharmacy Research

ANALGESIC AND ANTI PYRETIC ACTIVITY OF METHANOLIC EXTRACT OF *CRESSA CRETICA LINN*

Verma Nishant[1*], Kumar Umesh [1], Jha Keshari Kishore [1], Garg Vipin [2], Singh Ajai Kumar [3]

1. T. M. College of Pharmacy, Teerthanker Mahaveer University, Moradabad (U.P) 244001
2. Department of Pharmaceutical Technology, MIET, Meerut (U.P) 250005
3. Department of Pharmaceutics, Gandhi College of Pharmacy, Karnal (H.R) 132001

ARTICLE INFO

Published on: 15-6-2015
ISSN: 0975-8216

Keywords:
Analgesic; Antipyretic; Paracetamol; Pentazocine; *Cressa cretica*

Corresponding author:
Nishant Verma
Asst. Professor
Department of Pharmaceutical Chemistry, T.M. College of Pharmacy, TMU, Moradabad

ABSTRACT

Cressa cretica (Linn) belonging to family Convolvulaceae. It is commonly known as Rudravanti. It is a holophytic dwarf shrub used in folklore medicine. *Cressa cretica* is traditionally used to treat painful and inflammatory conditions. In the present study, analgesic and antipyretic activities of methanolic extract of *Cressa cretica* at different doses (100 mg/kg, 150 mg/kg, 200 mg/kg) was studied using hot plate, acetic acid induced writhing and yeast induced hyperthermia method. Methanolic extract of *Cressa cretica* showed significant analgesic and antipyretic activities at the dose of 200 mg/kg in all models studied.

INTRODUCTION

Analgesics are primary need of patients to getrid of any kind of pain.[1] Pain is one of the basic symptoms of all human ailments which is a sensorial modality and primarily protective. Analgesics only relieve pain in a particular complaint without affecting its cause.[2] The most eminent analgesics include opiates and NSAIDs, but they are not helpful in all cases due to their adverse effects. Besides pain the fever is another most common symptom of sickness which is caused by increase in the body temperature of an individual at a particular time.[3] Herbal medicines are often used as remedies in these conditions since as a result of poverty medicines may be unaffordable.[4] It is a well-known fact that herbal medicines may be sources of substances with better therapeutic potentials than some currently used orthodox medicines.[5] Herbal medicines are the oldest remedies known to mankind. Herbs had been used by all cultures throughout history but India has one of the oldest, richest and most diverse cultural

living traditions associated with the use of medicinal plants.[6] Medicinal plants are an important therapeutic aid for various ailments. In India, from ancient times, different parts of medicinal plants have been used to cure specific ailments.[7] Cressa cretica (Linn) belonging to family Convolvulaceae. It is commonly known as Rudravanti. It is a holophytic dwarf shrub used in folklore medicine. Cressa cretica (Linn) a halophytic plant which is referred to by the name that reflects the features of Sanjeevani so it is called Sanjeevani. It is mysterious and sought after herbs in India mythology. It's existence and identity are steeped in deep controversy. Cressa cretica (Linn) is a small, dwarf shrub upto 38 cm height. It is start to shoot in the beginning of June. The height of the plant was found to be 7cm-29cm but mostly are of 14cm in hight. The time of fruits and flowers from June to August but during September the plant gradually withers. The plant is distributed throughout India, along sandy shores.[8]

MATERIALS AND METHODS

1.1 COLLECTION AND AUTHENTICATION OF PLANT MATERIAL

Fresh plant of Cressa cretica was collected in the month of June, 2011 from Venkateswara University, Tirupati, Andhra Pradesh, India. The plant was identified and authenticated by Dr. K. Madhava Chetty, Asst. Professor, Department of Botany, Sri Venkateswara University, Tirupati, Andhra Pradesh, India. A voucher specimen has been deposited at the College of Pharmacy, TMU Moradabad (TMU/Consult/2011-12/208) for future reference.

2.2 PREPARATION OF THE EXTRACT

Fresh whole plant along with roots were shade dried reduced to moderately coarse poweder, loaded into soxhlet extractor and was subjected to successive extraction with Petroleum Ether, Benzene, Chloroform, Methanol and distilled water by hot extraction using Soxhlet apparatus at a temperature of 60˚C. The extracts were concentrated under reduced pressure using a rotary vaccuum evaporator to constant weight and preserved in desiccator for further studies.

2.3 ANIMALS

Adult albino wistar rats of both sexes weighing between 200 to 250 gm were used for the study. Also albino mice of both sexes weighing between 20-25gm were used. They were housed in polypropylene cages and fed with standard diet and water ad libitum. The animals were exposed to an alternating 12 h light and dark cycle. All the experimental procedures and protocols involving animals were reviewed by the Institutional Animal Ethics Committee in accordance with the guidelines of CPCSEA.

2.4 DRUGS AND CHEMICALS

Paracetamol (Batch no. RG331, Calpol) was purchased from Glaxo Smith Kline Pharmaceutical Ltd. Mumbai, India. Pentazocine drug (Batch no. 3358, Fortwin) was obtained from Ranbaxy Limited. Aspirin (Batch no. 13007596, Ecosprin) USV Ltd Mumbai. Analytical grade chemicals including various organic solvents (Petroleum Ether, Benzene, Chloroform and Methanol) from Rankem, Pvt. Ltd., New Delhi were used for the successive extraction.

2.5 PRELIMINARY PHYTOCHEMICAL SCREENING

The different extracts were subjected to qualitative phytochemical screening for the identification of the phytoconstituents. While Petroleum Ether, Benzene, Chloroform does not show any appreciable tests for the presence of different phytoconstituents, Petroleum Ether extract showed positive tests for the presence of alkaloids, glycosides, flavonoids. However methanolic extract showed positive tests for alkaloids, glycosides, flavonoids, tannins and phenolic compounds. The plant is used to cure pain and fever, the analgesic and antipyretic activity of the methanolic extract of the plant at different doses levels (100 mg/kg, 150 mg/kg, 200 mg/kg) is being reported here.

2.6 ACUTE TOXICITY STUDY

Acute toxicity study for the extract was carried out according to the method described in the literature.[10] The methanolic extract of Cressa cretica suspended in 5% gum acacia solution in doses of 100-2000 mg/kg was administered orally to animals. The animals were observed continuously for any change in autonomic or behavioral responses for first few hours and at 24 hour interval for a period of 72 hours. At the end of this period the mortality if any in each group was noted.

2.7 ANTIPYRETIC TESTING

Hyperthermia was induced in rats following the method of teotino et al., 1963.[11] Initial rectal temperatures of rats were recorded using a six channel tele-thermometer for 1 min. Rats were made hyperthermic by subcutaneous injection of 20% yeast suspension at a dose of 1 ml/100 gm body weight. When the temperature was at peak (18 hours after yeast injection) the rectal temperature were again recorded. Those animals that showed a rise in rectal temperature of more than 1.2^0 C were used.[12] Different doses of methanolic extract of Cressa cretica were given orally as a suspension prepared in 2% Tween 80 solution. Animals were divided into five groups of six antimals each. First group received 1 ml of 2% Tween 80 solution orally and served as

control. Second, third, fourth and fifth groups received standard antipyretic agent i.e. paracetamol suspension (100 mg/kg), methanolic extract (100 mg/kg), methanolic extract (150 mg/kg), metanolic extract (200 mg/kg) respectively. The rectal temperatures of animals were recorded at 30 minutes intervals for 4 hour following the administration of Tween 80, standard drug and plant extract.[13]

2.8 ANALGESIC ACTIVITY

2.8.1 HOT PLATE METHOD

The hot plate method described by Turner (1965) was followed for the assessment of analgesic activity. Albino mice were introduced to a hot plate maintained at 55 ± 0.5^{0}C. The reaction time to the thermal stimulus was recorded as the time interval from introduction of the animal to the plate until the first lick of the limbs or the first jump of the animals. The test groups received methanolic extract of Cressa cretica at different dose levels prepared as suspension in 2% Tween 80 orally, the standard group received Pentazocine (10 mg/kg, i.p.)[14] and control group received only 1 ml of 2% Tween 80 solution.[15] The reaction times were determined befor and after 30 minutes, 1 hour, 2 hours and 3 hours period with reference to the control group received only vehicle.

2.8.2 ACETIC ACID INDUCED WRITHING

Acetic acid induced writhing response in mice Acetic acid solution at a dose of 10 ml/kg (0.6%) was injected i.p. and the number of writhes during the following 15 minutes period was observed. The test groups received methanolic extract of Cressa cretica at different dose levels prepared as suspension in 2% Tween 80 orally, the standard group received Aspirin (10 mg/kg, i.p.) and control group received only 1 ml of 2% Tween 80 solution. Significant reductions in number of writhes by drug treatment as compared to vehicle treatment animals were considered as a positive analgesicresponse. The percent inhibition of writhing was calculated.[16]

$$\% \text{ Inhibition } = \frac{\text{Wc} - \text{WT}}{Wc} x\ 100$$

Where, WC = number of writhes in control group. WT = Number of writhes in test group.

2.9 STATISTICAL ANALYSIS

All the results obtained from various activities, as described above, were analyzed statistically by using Student's t test and $p<0.05$ were considered significant.[17]

3.0 RESULTS

3.1 YIELD OF PLANT EXTRACT

The 130 gm whole plant of Cressa cretica was taken for extraction by Soxhlet apparatus. Total 08.7 gm methanolic extract was obtained. The yield of methanolic extract was found to be 14.94 % w/w.

3.2 PHYTOCHEMICAL TESTING

Phytochemical testing showed that the methanolic extract of Cressa cretica contains alkaloids, glycosides, flavonoids, tannins and phenolic compounds.

3.3 ACUTE TOXICITY

No adverse effect and no mortality were observed in the animals during the period of study, 72 hours up to the dose 2000 mg/kg of methanolic extract of roots of Cressa cretica. Hence, there were no lethal effects in any of the groups. In the study, methanolic extract of Cressa cretica was administered at a dose of 100 mg/kg, which was determined as the most effective dose.

3.4 ANTI-PYRETIC ACTIVITY

The anti-pyretic activity of the methanolic extract of Cressa cretica has been shown in table 1, which showed significant activity at 150mg/kg and 200 mg/kg dose levels. The results were comparable to that of Paracetamol a prototype anti-pyretic drug.

Table.1. Effect of different doses of methanolic extract of Cressa cretica and paracetamol on yeast induced hyperthermia in rats.

	Rectal Temperature (^{0}C)						
	Initial before yeast injection	18 Hrs. after yeast injection	Time after drug administration (hrs)				
			0.5 hrs	1 hrs	2 hrs	3 hrs	4 hrs
Control	35.21± 0.057	36.21± 0.075	36.25± 0.071	36.30± 0.081	36.28± 0.044	36.23± 0.060[a]	35.31± 0.066
Paracetamol (100 mg/kg)	35.15± 0.042	37.25± 0.060	36.34± 0.095[d]	35.70± 0.075[d]	36.33± 0.070[d]	36.81± 0.095[c]	36.10± 0.060[c]
MECC (100 mg/kg)	35.23± 0.084	37.22± 0.036	37.13± 0.042	37.11± 0.064	36.88± 0.065[a]	37.18± 0.070	36.54± 0.075
MECC (150 mg/kg)	35.15± 0.07	37.14± 0.086	36.82± 0.01[c]	35.90± 0.070[d]	36.58± 0.065[d]	36.91± 0.057[b]	36.08± 0.036
MECC (200 mg/kg)	35.21± 0.055	37.26± 0.05	36.60± 0.05[d]	35.83± 0.074[d]	36.41± 0.051[d]	36.71± 0.101[c]	36.04± 0.090

Values are expressed as mean ± S.E.M. (n=6); significance at $p<0.05^a$, $p<0.02^b$, $p<0.01^c$, $p<0.001^d$ as compared to control.

3.5 ANALGESIC ACTIVITY

3.5.1 HOT PLATE METHOD

From the result it can be deduced that the extract has shown dose dependant activity. After administration of the methanolic extracts at all the three dose levels, threr is statistically significant increase in the hot plate reaction time. But the increase is comparable to the standard drug, Pentazocine only at 200 mg/kg dose level (Table 2).

Table 2 Effect of different doses of methanolic extract of Cressa cretica on Hot Plate reaction time in mice.

Groups	Dose (mg/kg)	Reaction Time (Seconds)				
		Initial	Time after drug administration (Hrs)			
			0.5 hrs	1 hr	2 hrs	3 hrs
Control		7.63± 0.0260	7.61± 0.0221	7.64± 0.0421	7.66± 0.0137	7.66± 0.0101
Pentazocine	10	7.70± 0.0136	23.64± 0.0793^{d}	30.72± 0.0211^{d}	35.60± 0.0260^{d}	30.63± 0.0490^{d}
MECC	100	7.67± 0.0097	11.96± 0.0390^{d}	15.16± 0.0214^{d}	16.86± 0.0215^{d}	13.00± 0.0316^{d}
MECC	150	7.70± 0.0205	17.96± 0.0490^{d}	21.55± 0.0210^{d}	26.61± 0.0210^{d}	20.51± 0.0281^{d}
MECC	200	7.70± 0.0201^{c}	21.51± 0.0291^{d}	28.61± 0.0170^{d}	33.65± 0.0392^{d}	26.96± 0.0216^{d}

Values are expressed as mean ± S.E.M. (n=6); significance at $p<0.05^{a}$, $p<0.02^{b}$, $p<0.01^{c}$, $p<0.001^{d}$ as compared to control.

3.5.2 ACETIC ACID INDUCED WRITHING

The methanolic extract at dose levels of 100, 150 and 200 mg/kg exhibited 31.51, 66.61 & 80.04 % inhibition of writhing as compared to that of 82.96% inhibition shown by Aspirin. It is quite evident from the result that the extract at 200 mg/kg showed comparable activity to that of Aspirin (Table 3).

Table no 3 Effect of different doses of methanolic extract of Cressa cretica on Acetic acid induced writhing in mice.

S.N.	Groups	Dose (mg/kg)	No. of Writhings (Mean ± SEM)	% Inhibition
1	Control		36.65± 1.325	
2	Aspirin	10	5± 0.386^{d}	80.80
3	MECC	100	25± 0.512^{d}	31.51
4	MECC	150	11.4± 0.498^{d}	66.61
5	MECC	200	6.31± 0.430^{d}	80.04

Values are expressed as mean ± S.E.M. (n=6); significance at $p< 0.05^{a}$, $p< 0.02^{b}$, $p<0.01^{c}$, $p<0.001^{d}$ as compared to control.

2. DISCUSSION

The present study establishes the anti-pyretic and analgesic activities of the methanolic extract of Cressa cretica in the used models. Since antipyretic and analgesic activities are commonly mentioned as characteristic of drugs or compounds which have an inhibitory effect on prostaglandin biosynthesis[18], the yeast induced hyperthermia in rat model was therefore employed to investigate the antipyretic activity of Cressa cretica. It was found that the methanolic extract at the dose of 200 mg/kg showed a significant decrease in rectal temperature similar to that shown by the standard drug paracetamol. This result seems to support the view that the extract has some influence on prostaglandin biosynthesis because prostaglandin is believed to be a regulator of body temperature.[19]

Likewise the analgesic activity of methanolic extract of the plant was evaluated using the hot plate method and writhing test in mice. The hot plate method is useful in detecting centrally acting analgesics[20] where as acetic acid induced writhing method is useful to detect peripheral analgesic effects. Acetic acid, which is used as an inducer for writhing syndrome, causes analgesia by liberation of endogenous substances, which then excite the pain nerve endings. The fact that methanolic extract of Cressa cretica showed analgesic activity in both the studied models, indicate that this effect could be due to the presence of two components, one acting centrally and the other via peripheral route. It was found that the methanolic extract at the dose of 200 mg/kg showed significant analgesic activity, similar to that shown by the standard drug aspirin in acetic acid induced writhing method and pentazocine in hot plate method.From the above results it can be deduced that methanolic extract has shown dose dependent activity. As the phytochemical screening has shown the presence of alkaloids, glycosides, flavonoids, tannins and phenolic compounds in the methanolic extract. Its potent activity may be attributed to the presence of these phytoconstituents. More detailed phytochemical studies are, however, necessary to identify the active principles and exact mechanism of action.

3. ACKNOWLEDGEMENTS

The authors are thankful to Dr. K. Madhava Chetty, Asst. Professor, Department of Botany, Sri Venkateswara University, Tirupati, Andhra Pradesh, India for the identification and authentication of the plant and also to the Department of Pharmaceutical Chemistry, T. M. College of Pharmacy, Teerthanker Mahaveer University, Moradabad for providing research facilities to carry out the work.

REFERENCES

1. Semwal, D.K., Semwal, R.B., Semwal, R., Jacob, V., Singh, G., Analgesic and antipyretic activities of Gindarudine, a morphine alkaloid from Stephania glabra, Current Bioactive Compounds, 2011, (7), 214-217.
2. Mate, G.S., Naikwade, N.S., Chowki, C.S.A.A., Patil, S.B., Evaluation of anti-nociceptive activity of Cissus quadrangularis on albino mice, Int. J. Green Pharm., 2008, (2), 118-121.
3. Wolf, S.M., Fauci, A.S., Dale, D.C., Unusual etiologies of fever and evaluation. Annu. Rev. Med., 1975, (26), 277-281.
4. Soucat, A., Levy, B.D., De, B.X., Gbedonou, P., Lamarque, J.P., Bangoura, o., Camara, O., Gandaho, T., Ortiz, C., Kaddar, M., Knippenberg, R., Affordability, cost effectiveness and efficiency of primary health care: the bamako initiative experience in benin and guinea. Int. J. Health Plan Mgt, 1997, (12), S81-S108.
5. Wang, Y., Wang, X.,, Cheng, Y., A computational approach to botanical drug design by modeling quantitative composition activity relationship. Chem. Biol. Drug Design, 2006, 968), 166-172.
6. Montbriand M. J., Senior and health-professionals mismatched perceptions and communication about prescription and non-prescription medication, Canadian Journal on Aging, (2000), Vol.-19, 35–56.
7. Gordon M. C., David, J. N., Natural product drug discovery in the next millennium, Pharm Biol., (2001), Vol.-139, 8-17.
8. Verma, N., Jha, K. K., Sharma, R., Chaudhary, S., Singh, A. K., Kumar, A., Phytochemistry biological properties, phytochemistry and traditional uses of rudravanti (Cressa cretica): a review, Pharmatutor magazine 2014, 2(8), 154-161.
9. Litchfield, J.T., Wilcoxon, F., A simplified method of evaluating dose-effect experiments, J. Pharmacol. Exp. Ther., 1949, (96), 99-135.
10. Teotino, U.M., Friz, L.P., Gandini, A. Bella, D.D., Thiodirevative of 2,3-dihydro - 4H - 1,3-benzoxazin-4-one synthesis and pharmacological properties. J. Med. Chem. 1963, (6), 248-250.
11. Taesotikul T., Panthong A., Kanjanapothi D., Verpoorte R., Scheffer J.J.C. Antiinflammatory, antipyretic and antinociceptive activities of Tabernaemontana pandacaqui Poir. J. Ethanopharmacol 2003, (84) 31-35.
12. Narayanan N., Thirugnanasambantham P., Viswanathan S., Vijayasekaran V., Sukumar E. Antinociceptive, ant-inflammatory and antipyretic effects of

ethanol extract of Clerodendron serratum roots in experimental animals. J. Ethanopharmacol 1999, (65), 237-241.

13. Puchchakayala G., Podili Laxmi, Bobbala Diwakar et al. Antinociceptive and anti-inflammatory effects of Cleome chelidonni linn. roots in experimental animals. Phcog. Mag. 2008, 4(13) 32-36.
14. Verma, N., Chaudhary, S., Garg, V., Tyagi, S., Anti inflammatory and analgesic activity of methanolic extract of stem bark of Ficus religiosa, International journal of pharma professional's research, 2010, 1(1), 145-147.
15. Kulkarni S. K. Handbook of Experimental Pharmacology. Vallabh Prakashan, New Delhi, 2nd ed., 1993, 82-87.
16. Vane J. R. The evaluation of non-steroidal anti-inflammatory drug and their mechanism of action. Drug 1987, (33), 8-27.
17. Milton A. S. Prostaglandins and fever. Trends in Pharmacological Sciences 1982, (40), 490-492.
18. Radhakrishnan R., Zakaria M.N.M., Islam M. W. et al. Analgesic and Anti-Inflammatory Activities of Teucrium stocksianum. Pham. Bio. 2001, 39 (6): 455-459.