Analgesic and Locomotor Activity of Extracts of *Cleome rutidosperma* DC

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Crude methanol, chloroform and petroleum ether (60-80°) extracts of *Cleome rutidosperma* were investigated for analgesic (narcotic and non narcotic) and locomotor activity in mice at a dose of 100 mg/kg administered orally. All these extracts showed significant analgesic and depressed locomotor activity when compared to control and standard drug treatment with morphine sulphate, aspirin and chlorpromazine, respectively.

Cleome rutidosperma (family: Capparidaceae) is a plant with no known traditional use. This plant is a common weed in waste ground and grassy places. The plant grows to about 0.5 m or taller. Leaves are compound with 3 leaflets. Flowers are purple and found singly. Fruits are long narrow capsules and when ripe split into two, scattering many small, round and blackish seeds (http://www.science.edu.sg/ssc/wildflowers/catswhiskerfamily.jsp).

Although there is no information regarding the medicinal use of this plant, thorough literature survey indicated however, that for other plants of the genus Cleome, the leaves are stimulant while the roots are stimulant, antiscorbutic and anthelmintic and the seeds are rubifacient, vesicant, anthelmintic and carminative[1],. Thus, it is reasonable to expect that this plant might have some similar or other kinds of biological activity. In the present study we investigated the analgesic and locomotor activity of various extracts of *Cleome rutidosperma*.

The whole plant of *Cleome rutidosperma* was collected from North-24 Parganas district of West Bengal during September 2002 and authenticated at the Herbarium, Botanical Survey of India, Shibpur, Howrah. First the plants were washed thoroughly with ordinary water and then with distilled water. Then they were dried under shade and the dried aerial parts were cut and ground in a mixer to obtain the plant powder. This plant powder was divided into 3 equal portions and each portion was subjected to extraction in a Soxhlet separately with methanol, chloroform and petroleum ether (60-80°). The extracts were concentrated to dryness under reduced pressure. All the extracts were preserved in a desiccator for further studies. The test doses of each extract were prepared as a suspension with Tween-80 (1%) in distilled water to get the desired concentration of the extract.

The analgesic activity was evaluated by both tail flick method[2-4] and acetic acid-induced writhing method[2,3,5] to ascertain narcotic and nonnarcotic type of activity, respectively. The experimental protocols have been approved by the Institutional Animals Ethics Committee. In the tail flick method[2-4], Swiss mice of either sex (20-25 g) were randomly distributed into five groups consisting of six animals in each group. The first group served as control and the animals of that group were administered vehicle Tween-80 (1%) orally. The second groups of animals were administered morphine sulphate at a dose of 5 mg/kg, intraperitoneally. The animals of the third, fourth and fifth group were treated with methanol, chloroform and petroleum ether extract, respectively at a dose level of 100 mg/kg, orally. The reaction time was noted at 20, 40 and 60 min time intervals, after drug administration. The percent inhibition of tail flick response measured as time to tail flick was calculated using the formula, % protection=$(1-W_t/W_c)x100$, where W_t and W_c are the mean values of the time to tail flick in the test and control groups, respectively. The results are recorded in the Table 1.

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TABLE 1: ANALGESIC ACTIVITY OF VARIOUS EXTRACTS OF *C. RUTIDOSPERMA* BY TAIL FLICK METHOD

Group	Dose (mg/kg)	Mean time(s)±S.E[8]		
		20 min	40 min	60 min
Vehicle Control	-	1.7±0.16	1.8±0.15	1.7±0.12
Morphine sulphate	5	8.7±1.14*	14.6±1.32*	15.9±0.68*
Petroleum ether	100	6.6±0.95*	9.4±0.75*	9.9±1.04*
Chloroform	100	4.0±0.7	4.4±0.51*	5.3±0.51*
Methanol	100	4.0±0.47	6.2±0.59*	6.9±0.73*

*Indicates significant difference at P<0.001 when compared to control.

The data were analysed using student's 't' test and the level of significance was set at P<0.001.

The nonnarcotic analgesic activity was evaluated against acetic acid-induced writhing in mice. In this method, Swiss mice of either sex of weight between 20-25 g were randomly distributed in five groups each consisting of six animals. The first group served as control and the animals of that group were administered 3% v/v acetic acid at a dose of 1 ml/100 g intraperitoneally. The onset of writhing was noted and the number of writhings was recorded for a period of 10 min for each animal of the group. The second group of animals was administered aspirin at a dose of 25 mg/kg, intraperitoneally and 15 min later, the animals of that group were administered acetic acid. The onset and the number of writhing response were observed. The animals of the third, fourth and fifth group were treated with methanol, chloroform and petroleum ether extract, respectively at a dose level of 100 mg/kg, orally, and the acetic acid-induced writhings were recorded as described for groups 1 and 2. Percent protection against acetic acid-induced writhing was calculated using the formula, % protection=$(1-W_t/W_c)\times100$, where W_t and W_c are the mean values of number of writhing in the test and control groups, respectively. The results are recorded in the Table 2. The data were analysed using student's 't' test and the level of significance was set at P<0.001.

TABLE 2: ANALGESIC ACTIVITY OF VARIOUS EXTRACTS OF *C. RUTIDOSPERMA* BY ACETIC ACID-INDUCED WRITHING METHOD

Group	Dose (mg/kg)	Number of Writhings Mean ± S.E[8]
Vehicle Control	—	56.7±0.88
Aspirin	25	36.5±1.52*
Petroleum ether	100	27.2±1.95*
Chloroform	100	42.2±1.62*
Methanol	100	30.2±2.17*

*Indicates significant difference at P<0.001 when compared to control.

The CNS depressant activities of the extracts were evaluated by studying locomotor activity of mice using an actophotometer[2,6,7]. In this method, Swiss mice of either sex (20-25 g) were randomly distributed in four groups of six animals each. Animals of the first group were placed individually in the activity cage for 10 min and the activity was monitored. Then the animals were given chlorpromazine 3 mg/kg, intraperitoneally and were tested again for activity 30 min after administration. The animals of the second, third and fourth group were treated with methanol, chloroform and petroleum ether extract, respectively at a dose of 100 mg/kg, orally and tested similarly. Percent decrease in activities

TABLE 3: LOCOMOTOR ACTIVITY OF VARIOUS EXTRACTS OF *C. RUTIDOSPERMA*

Group	Dose (mg/kg)	Average % change in activity ± S.E.[7]
Chlorpromazine	3	80.6±1.07*
Petroleum ether	100	54.9±3.26*
Chloroform	100	42.4±1.35*
Methanol	100	64.4±3.13*

*Indicates significant difference at P<0.001 when compared to control.

were calculated for each animal using the formula, percent decrease in activity=$(1-W_a/W_b)\times100$, where W_a and W_b are average activity scores before and after drug administration, respectively and the average decrease in activity was calculated for all groups. The results are presented in Table 3. The data were analysed using student's 't' test (paired) where readings of the animals before drug administration served as control and the level of significance was set at $P<0.001$.

In the tail flick method[2-4], it was found that all the extracts showed significant narcotic analgesic activity. The activity was found to be maximum for petroleum ether extract and minimum for chloroform extract. The activities were about 30-60% of that of morphine sulphate. Similar results were obtained from acetic acid-induced writhing test also, showing significant activity for all extracts. The activity was maximum for petroleum ether extract and minimum for chloroform extract and the activity was comparable to that produced by standard aspirin.

In activity evaluation study, it was found that all three extracts significantly depressed the locomotor activity, which was found to be slightly lower than that produced by the standard, chlorpromazine. Here also, the activity was found to be maximum for petroleum ether extract and minimum for chloroform extract. Thus, it can be concluded that, on preliminary screening of crude extracts of *Cleome rutidosperma*, it was found that petroleum ether extract exhibited analgesic and locomotor depressant activities followed by methanol and chloroform extracts, which possessed these activities to a lesser extent.

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Spectrophotometric Method for the Determination of Lacidipine in Tablets

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A simple and sensitive spectrophotometric method has been developed for the determination of lacidipine in bulk and tablets. The method is based on the reaction of lacidipine with ferric chloride, potassium ferricyanide and hydrochloric acid to form a bluish green colored chromogen with an absorption maximum of 740 nm. Beer's law was obeyed in the range of 0-10 μg/ml. The proposed method has been successfully applied to the analysis of the bulk drug and its dosage forms. Statistical comparison of the result with that obtained with reported method showed good agreement and indicated no significant difference in precision. This method does not require any extraction or heating.

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Lacidipine is chemically 4-{2-[3-(1,1-dimethylethoxy)-3-oxo-1-propenyl phenyl]-1,4-dihydro-2,6-dimethyl-3,5-pyridine dicarboxylic acid}diethyl ester[1]. Reported methods of