RESEARCH ARTICLE

Evaluation of analgesic activity of *Dahlia pinnata* leaf extracts in Swiss albino mice

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ABSTRACT

Dahlia pinnata Cav. (Asteraceae), commonly known as Dalia in Bengali, is a bushy, herbaceous ornamental plant indigenous to Mexico and widely cultivated in different regions of India. The present study assessed three different successive solvent extracts from *D. pinnata* leaf for their analgesic potential by acetic acid induced writhing assay in Swiss albino mice. All the test extracts exhibited significant analgesic activity. The methanol extract was found to be the most potent followed by the chloroform and petroleum ether extracts respectively. The present preliminary study demonstrated marked analgesic activity of *D. pinnata* leaf in Swiss albino mice.

Keywords: Analgesic, *Dahlia pinnata*, writhing, polyphenols, leaf.

INTRODUCTION

Traditional or herbal medicine worldwide is being re-evaluated by extensive research on different plant species and their therapeutic principles. The major merits of traditional medicine seem to be their perceived efficacy, low incidence of serious adverse effects and comparatively low cost.

Dahlia pinnata Cav. (synonym: *Dahlia variabilis* (Willd.) Desf.) (Family: Asteraceae) is a bushy, herbaceous ornamental plant indigenous to Mexico, Central America and Colombia. The dahlia is the national flower of Mexico. It is also widely cultivated in Indian gardens (commonly known as Dalia in Bengali) in winter season for its bright colourful flowers. It is a good source of flavonoids and anthocyanins. Its flowers can be regarded as a source of natural dye. [1, 2] However, studies on its leaf have not been reported still now. Therefore, in the present investigation we attempted the analgesic assessment of successive petroleum ether, chloroform and methanol extracts from *D. pinnata* leaf.

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MATERIALS AND METHODS

Plant material

The mature leaves of *Dahlia pinnata* Cav. (synonym: *Dahlia variabilis* (Willd.) Desf.) (Family: Asteraceae), were collected during the month of December 2012 from Dumdum, Kolkata, West Bengal, India. The plant material was taxonomically identified by Dr. V. P. Prasad of the Central National Herbarium, Botanical Survey of India, Howrah, West Bengal, India. The voucher specimen (CNH/10/2013/Tech.II/960) was maintained in our research laboratory for future reference. The plant material was shade-dried with occasional shifting and then powdered with mechanical grinder, passing through sieve no. 40, and stored in an air-tight container.

Preparation of plant extracts

The dried powdered material was first extracted (percolation) with petroleum ether (60-80°C), the percentage extractive value was 2.0 % w/w. The marc thus obtained was further extracted (percolation) successively with chloroform and methanol for 72 h. The solvent was distilled off under reduced pressure resulting in semisolid mass that was vacuum dried to yield the dry extracts and the percentage extractive values were accordingly 3.5% w/w and 12% w/w respectively. The preliminary phytochemical analysis was performed for all three extracts to identify the phytoconstituents present in the extracts. [3]

Drugs and chemicals

Acetyl salicylic acid (aspirin) and glacial acetic acid were from Sigma-Aldrich Chemical Corp. (St. Louis, MO, USA). All other chemicals and reagents were of analytical grade obtained commercially.

Experimental animals

Adult Swiss albino mice of either sex weighing 20 ± 2 g were procured from registered breeders (Reeta Ghosh & Co., Kolkata, India) and maintained under standard laboratory conditions (temperature 25 ± 2°C with dark and light circle 14/10 h). They were allowed free access to standard dry pellet diet (Hindustan Lever, Kolkata, India) and water *ad libitum*. The mice were acclimatized to laboratory condition for 10 days before commencement of the experiment. All experimental procedures were reviewed and approved by the Institutional Animal Ethics Committee.

Analgesic evaluation: acetic acid-induced writhing test

Swiss albino mice were divided into five 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.*) which served as reference. Groups III, IV and V received the petroleum ether, chloroform and methanol extracts at the doses of 150 mg/kg b.w., *p.o.* respectively. Thirty minutes after aspirin and extracts administration, group II to V received acetic acid (1% v/v, 10 ml/kg b.w., *i.p.*) and writhing reflex was noted for the period of 15 minutes. [4, 5] The mean writhing scores in each group were calculated and expressed the percentage of protection using the following formula:-

(Control mean - Treated mean/ Control mean) ×100 %.

Statistical analysis

The data are represented as mean ± standard error of mean (SEM). Degree of significance was assessed by Student's '*t*' test.

RESULTS AND DISCUSSION

Preliminary phytochemical studies revealed the presence of triterpenoids and steroids in the petroleum ether extract; triterpenoids, steroids and alkaloids in the chloroform extract; and triterpenoids, steroids, alkaloids, saponins, polyphenolic compounds and carbohydrates in the methanol extract of *D. pinnata* leaf.

The analgesic activity of *D. pinnata* leaf extracts was evaluated by acetic acid induced writhing method in mice to assess peripheral (non-narcotic) type of analgesic activity. [6] Acetic acid induced writhing is chemically induced nociception by intraperitoneal injection of dilute acetic acid solution to mice. The chemical agents can produce nociceptive reactions in mice. Intra-peritoneal injection of phenyl para quinone, bradykinin or dilute acetic acid (1-3% v/v) produces pain reaction that is characterized as writhing response. Constriction of abdomen, turning of trunk (twist) and extension of hind limbs (at least one) are considered as writhing reaction to chemically induced pain. [6, 7]

Acetic acid induced writhing test is known as a visceral pain model nociception. Several mediators like kinins, acetylcholine, substance P, calcitonin-gene-related peptide and prostaglandins (PG) take part in visceral pain model nocicepotion and transmission of the nociception from the viscera. In this test both central and peripheral analgesics are detected. Analgesics of narcotic (central) e. g. morphine, pentazocin, pethidine etc and non-narcotic (peripheral) type, e. g. aspirin, ibuprofen, indomethacin etc can inhibit the writhing response in mice. [7-9] The results of the present study are in agreement with those of similar other recent findings. [10, 11]

Present results indicated that the methanol extract of *D. pinnata* leaf was the most potent followed by the chloroform and petroleum ether extracts respectively (Table 1). Preliminary phytochemical studies revealed the presence of polyphenols in the methanol extract. Polyphenols are well known natural products

possessing several important pharmacological activities. [12] The maximum analgesic effect exhibited by methanol extract may be due to the presence of polyphenols in it.

Based on the results obtained from the present preliminary investigation, it can be inferred that all the test extracts had effective peripheral analgesic actions. The present preliminary study confirms marked analgesic activity of *Dahlia pinnata* leaf in Swiss albino mice. Further studies are presently necessary to confirm the identity of the bioactive principles responsible for these actions.

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Table 1: Analgesic effect of *D. pinnata* leaf extracts on acetic-acid induced writhing in mice.

Treatments	Dose	Number of writhes	% Protection
Acetic acid (1% v/v)	10 ml/kg	52.83 ±1.400	-
Acetic acid + Aspirin	100 mg/kg	17.26 ±1.606*	67.32
Acetic acid + Pet. ether extract	150 mg/kg	24.21 ±1.561*	54.17
Acetic acid + Chloroform extract	150 mg/kg	19.15±1.572*	63.75
Acetic acid + Methanol extract	150 mg/kg	17.56±1.291*	66.76

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

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