Saudi Journal of Biological Sciences xxx (2018) xxx–xxx

Contents lists available at ScienceDirect

Saudi Journal of Biological Sciences

journal homepage: www.sciencedirect.com

Original article

Evaluation of anti-nociceptive, anti-inflammatory and antipyretic potential of *Mikania cordata* (Burm. f.) Robinson in experimental animal model

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ARTICLE INFO

Article history:
Received 2 November 2017
Revised 10 December 2017
Accepted 15 January 2018
Available online xxxx

Keywords:
Mikania cordata
Essential oil
Chloroform extract
Ethyl acetate extract
Anti-nociceptive
Anti-inflammatory
Antipyretic activity

ABSTRACT

Mikania cordata is widely used for the treatment of cuts, wounds, and dengue fever in Bangladesh. In the present study, essential oil (12.5, 25 and 50 mg/kg) and two extracts, *viz*., chloroform and ethyl acetate extracts (200, 400, 800 mg/kg b.w.) were tested for peripheral and central anti-nociceptive activity by acetic acid-induced writhing and hot plate method, respectively. Carrageenan-induced rat paw edema assay and yeast-induced hyperthermia assay were also carried out to evaluate anti-inflammatory and antipyretic properties of oil and extracts, respectively at aforesaid doses. The essential oil (50 mg/kg), chloroform extract (800 mg/kg) and ethyl acetate extract (800 mg/kg) showed potent peripheral anti-nociceptive activity having 47.33%, 29.33% and 16.65% of writhing inhibition, respectively, comparable with standard diclofenac (52.0%). Essential oil (50 mg/kg), chloroform extract (800 mg/kg) and ethyl acetate extract (800 mg/kg) presented promising central anti-nociceptive activity as well having 95.86%, 79.18% and 42.37% elongation of reaction time, respectively, at 90 min after administration of essential oil, ethyl acetate extract and 60 min after administration of chloroform extract. In anti-inflammatory activity screening, the essential oil (50 mg/kg) produced the highest 72.80% edema inhibition at 4 h after administration of carrageenan which was comparable with that of standard phenylbutazoe (87.87%). On the other hand, chloroform extract (800 mg/kg) and ethyl acetate extract (800 mg/kg) showed up to 34.31% and 15.27% of edema inhibition, respectively, at 4 h after administration of carrageenan. In antipyretic assay, the essential oil and chloroform extract displayed a strong antipyretic effect in yeast-induced rats, whereas the ethyl acetate extract had no antipyretic activity. The present study revealed anti-nociceptive, anti-inflammatory and antipyretic potential of *M. cordata* which could be the therapeutic option against fever, inflammations as well as painful conditions and confirmed the traditional use of *M. cordata*.

1. Introduction

Mikania cordata (Burm. f.) Robinson (Asteracae) is a twining herb, characterized by its cordate leaves, capitulum inflorescence, whitish flowers, narrowly oblong cypsela type of fruits and white pappus. In Bangladesh, *M. cordata* is locally known as *Assamlata* or *Tarulata*, and is widely distributed throughout the country. This plant is used as ethnomedicine in treating cuts and wounds in Bangladesh. A decoction of leaves is administered in the treatment of indigestion, dysentery, and gastrointestinal sore (Ghani, 1998).

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.sjbs.2018.01.009
1319-562X/

The *Mandi* ethnic people in Tangail district of Bangladesh eat young fried leaves of *M. cordata* in gastric pain (Partha and Hossain, 2007). The *Garo* tribal people of Netrokona district use this plant in cuts and wounds (Rahmatullah et al., 2009a). The *Santals* tribal people residing in Thakurgaon district use the leaf of *M. cordata* for the treatment of cuts and wounds, and as remedy of dengue fever (Rahmatullah et al., 2009b). The plant is also used as leafy vegetable and applied against coughs, eye sores and gastro-intestinal disorders. In Assam (India), the local herbal practitioners (*Kabiraj*) use the leaf juice of *M. cordata* in the case of insect and scorpion sting (Sastri, 1962). *M. cordata* is also indigenously used by the tribe *Seminoles* to treat itchy skin as well as for circumcision, wounds, and tumors (Sturtevant, 1954). It is reported to have psycho-pharmacological, neuro-pharmacological, antibacterial, antifungal activities and therapeutic properties against pain, inflammation, hyperthermia, ulcer and carcinogenesis (Mosaddik and Alam, 2000). It contains different biologically active compounds e.g. mikanin, friedelin, butyryloxy kaurenic acid, benzoyloxy kaurenic acid, stigmasterol and beta-sitosterin (Ghani, 2003).

The CNS depressant activity with potential antioxidant properties are found in different plant aerial extracts (Hyun et al., 2016) and it is also common in *M. cordata* (Hasan et al., 2009). The hydroalcoholic leaf extract of this species also found to have neuropharmacological as well as CNS-depressant activity (Dey et al., 2011). Crude ethanolic extract of *M. cordata* found to have antinociceptive, cytotoxicity, and antibacterial activities in animal model (Nayeem et al., 2011). However, no study was carried out on the anti-nociceptive, anti-inflammatory and antipyretic potential of the essential oil as well as chloroform and ethyl acetate extracts of *M. cordata* leaves so far. Therefore, this study aimed at investigating the anti-nociceptive, anti-inflammatory and antipyretic effects of leaf essential oil and extracts of *M. cordata* occurring in Bangladesh.

2. Materials and methods

2.1. Plant material

The aerial parts of *Mikania cordata* were plucked from Muksudpur of Gopalganj district, Bangladesh in the months of June and July 2010. The plant material was collected and identified by Prof. Dr. M. Oliur Rahman of the University of Dhaka, Bangladesh. The voucher specimen has been preserved in Bangladesh National Herbarium (DACB – Voucher No. 37894).

2.2. Extraction of essential oil

After drying of the fresh leaves at room temperature, those were converted into powdered form using pestle and mortar, and the essential oil was extracted by steam distillation for 3 h. The yielding percentage was 0.65 (w/w) and the density at 25 °C was 0.98 g/ml. In this study, one thousand and nine hundred gram (1900 g) of the powder was hydrodistilled and 12.35 g of the essential oil was yielded. The oils was preserved in refrigerator in a lightproof bottle. In order to determine the relative density of the oil 10 ml capacity bottle was employed.

2.3. Preparation of plant organic extracts

Fifty grams of powdered form of the leaves was used separately for chloroform and ethyl acetate solvent extraction at room temperature for 7 days. After evaporation of solvents using vacuum rotary evaporator at 50 °C, 6.2 g of chloroform extract and 7.4 g ethyl acetate extracts were obtained. The extracts were suspended in normal saline using 5.0% Tween 80 before administration.

2.4. Experimental animal

The animal (Swiss albino mice of 25–30 g and Wister albino rats of 150–200 g) were collected from ICDDR, B (International Centre for Diarrheal Diseases and Research, Bangladesh). Standard polypropylene cages were used to keep the animal at temperature 25 ± 1 °C where humidity remains 60–70% with normal day/night cycle (12 h each). Standard pellets as basal diet (ICDDR, B formulated) and water *ad libitum* were supplied to the animals for one week. After fasting for overnight, the animal were weighted prior to the experiment. Guidelines of the National Institute of Health for the Care and Use of Laboratory Animals (NIH Publication 80-23, revised in 1996) were strictly followed in carrying out the experiments.

2.5. Drugs and chemicals

Diclofenac sodium manufactured by ACI Pharmaceutical Ltd., Dhaka and morphine manufactured by Gonoshasthaya Pharmaceuticals Ltd, Dhaka, Bangladesh were purchased for the experiments. Acetic acid (Merck, Germany) and carrageenan (BDH, UK.) were used in this study. For other chemicals, only the analytical grade chemicals (Sigma and Merck) were employed in the experiments.

2.6. Anti-nociceptive activity study

2.6.1. Peripheral anti-nociceptive activity assay

Acetic acid writhing inhibition method as developed by Koster et al. (1959) was used to identify peripheral anti-nociceptive activity of the plant extracts and essential oil of *M. cordata*. Three different groups of mice were tested for each type of the test sample and each group included 5 randomly selected mice. Mice of a group were received 5% Tween 80 (10 ml/kg), while three different groups of mice received essential oil of 12.5, 25.0 and 50.0 mg/kg, other three different groups received chloroform extracts of 200, 400, 800 mg/kg and another three different groups received ethyl acetate extracts of 200, 400, 800 mg/kg. Thirty minutes later, the mice were received 3% acetic acid to induce pain. After 5 min of acetic acid administration, writhing count was started and continued over a period of 20 min. The writhing of the group received tested specimens and diclofenac sodium were compared to the writhing response of the control group and considered as index of analgesia. The percentage of inhibition was calculated as per following equation:

$$\%\ Inhibition = \frac{Mean\ wrihing\ count(control\ group - treated\ group) \times 100}{Mean\ writhing\ count\ of\ control\ group}$$

2.6.2. Central anti-nociceptive activity assay

For hot plate experiment, the method of Ojewole (2006) with slight modification was followed. The mice which showed responses to the hot plate (Columbus, USA) thermal stimulation after 4 s were selected for the test. In this case the hot plate temperature was kept at 55 ± 1.0 °C and the time period taken to show the response by hind paw licking or jumping was documented in seconds. The test samples (12.5, 25.0 and 50.0 mg/kg of oil, and 200, 400 and 800 mg/kg of extracts) were given orally to mice. Each of mouse under tests was also considered as the own control. Morphine (10 mg/kg) was employed as positive control, and administrated subcutaneously. The primary response time (control latency) was determined by placing the mice on the hot analgesiometer and the time (in seconds) taken to respond was recorded. The average time of the three consecutive experiment at 1 h interval was recorded as control latency. On the other hand, every group

Please cite this article in press as: Siddiqui, S.A., et al. Evaluation of anti-nociceptive, anti-inflammatory and antipyretic potential of *Mikania cordata* (Burm. f.) Robinson in experimental animal model. Saudi Journal of Biological Sciences (2018), https://doi.org/10.1016/j.sjbs.2018.01.009