Antinociceptive Activity of *Mimosa pudica* Linn

M. KARTHIKEYAN and M. K. DEEPA

*For author affiliations, see end of text.*

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ABSTRACT

*Mimosa pudica* Linn (Mimosoideae) is a plant used in traditional medicine for various disorders. The aim of this work was to evaluate the acute toxicity and antinociceptive activity of the aqueous extract of *Mimosa pudica* in animal models. In the acute toxicity study, a single dose of aqueous extract of 2000 mg kg\(^{-1}\) body weight p.o. was administered. For 48 h, animals showed no clinical signs and mortality. In the acetic acid-induced writhing model, the extract at a dose of 200 & 400 mg kg\(^{-1}\) body weight showed significant (\(p<0.001\)) inhibition of writhing response of 46.24 and 56.0 % respectively. In the hot plate test, the extract produced a significant (\(p<0.001\)) increase in the latency in a dose-related manner. This study established the analgesic properties of *Mimosa pudica* Linn.

**Keywords:** *Mimosa pudica* Linn, Whole plant, Aqueous extract, Analgesic activity, Writhing test, Hot plate test

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Plant still remains a major source for drug discovery in development of synthetic molecules. The use of traditional plant extract in the treatment of various diseases has been flourished. *Mimosa pudica* Linn belonging to the family Mimosoideae is commonly known as sensitive plant. The plant has very sensitive leaves which fold on touching and has reddish roots. The plant is being used as the drug source in ayurvedic system of medicine in Kerala and in Siddha medicine. The plant is widely distributed through tropical and subtropical parts of India, common in waste place where the climate is moist and warm. The useful parts of this plant are roots, leaves and flower heads. The whole plant is used medicinally in ayurvedic folk medicine and its photochemical studies revealed the presence of minosine, orientin, isoorientin, β- sterol, D-pinitol, norepinepherine, crocetin, tannins and turgorins. *Mimosa pudica* is an important plant which is used for various ailments in ayurvedic system of medicine [1-7]. In previous studies, it was found that the decoction of *Mimosa pudica* leaves has anticonvulsant properties [8]. Both the ethanolic and aqueous extracts of the leaves of *Mimosa pudica* possess hyperglycemic and antidepressant activities in mice and rats respectively [9, 10]. There is no scientific report on the analgesic activity of *Mimosa pudica*, therefore the present study was undertaken to examine the possible antinociceptive activity of the aqueous extract of *Mimosa pudica* using acetic acid-induced visceral nociceptive responses and thermal-induced nociception (chemical and thermal models) in mice.

**MATERIALS AND METHODS**

**Study centre**

The present study was carried out during December 2006 to June 2007 at Postgraduate Research Laboratory, Department of Pharmacology, SRM College of Pharmacy, SRM University, SRM Nagar, Kattankulathur, Kancheepuram, Tamil Nadu, India.

**Plant material**

*Mimosa pudica* Linn (Mimosoideae) was collected from tropical areas in Kancheepuram district, Tamil Nadu. It was identified by Prof.P.Jayaraman, Director, Plant Anatomy Research Center (PARC), Pharmacognosy Institute, West tambaram, Chennai, Tamil nadu, India. A voucher specimen (No: PARC/07/SRM/33) was deposited in the herbarium of the institute.

**Preparation of extract**

The plant material was washed well with water, dried under shade and powdered to a fine grade by using laboratory scale mill. A batch of 100 gm of the whole plant powder was suspended in one liter of the distilled water and the mixture was boiled for 30 minutes. The...
obtained decoction was centrifuged and filtered by using filter paper. The filtrate was evaporated in vacuum to give a residue. The yield of the product was approximately 4.2 % (w/w) of the whole plant of *Mimosa pudica* Linn. The final product was stored in a vacuum desiccator at room temperature until analysis.

**Animals**

Male Swiss albino mice weighing 20-30 gm were procured from the inbred stock of the King’s Institute, Guindy, Chennai, India. They were housed in well-ventilated polypropylene cages with a 12h light/12h dark cycle, received a standard pellet diet (Hindustan lever limited, Bangalore, India.) and water *ad libitum*. The mice were acclimatized to laboratory condition for 7days. Animals were kept under fasting for overnight, and allowed free access to water before commencement of experiment. The study was done with approval from the Institutional Animal Ethical Committee (IAEC) of Committee for the purpose of control and supervision of experiments on animal (CPCSEA).

**Drugs and Chemicals**

The following drugs and chemicals were used for the study: Acetyl Salicylic Acid (USV limited, Mumbai.), Pentazocine (Ranbaxy Laboratories Ltd, New Delhi.), Acetic acid (Ranbaxy Fine Chemicals, New Delhi.), Sodium chloride (Ranbaxy Fine Chemicals, New Delhi).

**Acute toxicity studies**

Acute toxicity studies were carried out using acute toxic class-limit test dose guidelines 425 of Organization for Economic and Cultural Development (OECD). Acute toxicity of the plant extract was carried out using groups of three Swiss albino mice by administering a dose of 2000 mg kg\(^{-1}\) body weight per o.s.( p.o.), while control group received normal saline. The toxicological effects were assessed on the basis of mortality and behavioral changes during 48 h [11].

**Analgesic activity**

**Writhing test**

The peripheral analgesic activity was determined by acetic acid-induced writhing. Animals were injected intraperitoneally(i.p.) with 0.6 % acetic acid [11-14], 10 ml kg\(^{-1}\) body weight and pretreated with *Mimosa pudica* extract at 200 & 400 mg kg\(^{-1}\) body weight, p.o. Positive control group were received acetyl salicylic acid intramuscular (i.m.) 200 mg kg\(^{-1}\)body weight, 30 minutes prior to the peritoneal irritation. Control group received 10 ml kg\(^{-1}\) body weight of 0.9 % Sodium chloride solution, i.p. The resulting writhings were observed and counted for 60 minutes after acetic acid injection.

**Hot plate test**

The method of Woolfe and McDonald [15] was used. The paw of the mice is very sensitive to heat at temperature which is not damaging the skin. The response in the form of jumping, withdrawal of the paw or the licking of the paws was defined as hot plate latency [13, 14]. The animals were placed on Eddy’s hot plate kept at a temperature of 55±1°C. Those that showed a reaction time below 15 sec were placed again on the hot plate and the latency was recorded at 30, 60 and 120 minutes after administration of the test compounds. The test was terminated at 30 sec in the absence of a response. Control group received normal saline (10 ml /kg p.o.), Pentazocine was used as positive control (10 mg/kg). The test group received *Mimosa pudica* aqueous extract at the dose of 200 and 400 mg/kg p.o. The latency was recorded of the above timings. Average reaction time and percentage variation were calculated using the following ratio [16].

\[
\text{Percentage of protection} = \frac{(\text{Drug latency} - \text{Base line latency})}{\text{Base line latency}} \times 100
\]

**Statistical analysis**

The statistical analysis of all the results was carried out using one-way ANOVA followed by Dennett’s multiple comparison using graph pad instat 3 software and all the results obtained in the study were compared with the control group. The \(p\) values <0.05 were considered statistically significant.

**RESULTS**

**Acute toxicity**

The animals showed no clinical signs without any mortality recorded. About 2000 mg kg\(^{-1}\) body weight may be assumed.

**Analgesic activity**

**Writhing test**

As shown in Table 1, our results showed that the number of acetic acid-induced writhing was significantly reduced by aqueous extract administered orally at 200 & 400 mg kg\(^{-1}\) body weight in dose-related manner with 46.24 and 56.0 % of inhibition respectively. The results were statistically significance.
Antinociceptive Activity of *Mimosa pudica* Linn

Table 2. Effect of aqueous extract of *Mimosa pudica* Linn on the latency of mice exposed to hot plate

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Dose (mg kg⁻¹ Body weight)</th>
<th>Basal 30 min</th>
<th>60 min</th>
<th>120 min</th>
<th>Percentage protection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>-</td>
<td>3.20 ± 0.032</td>
<td>2.57 ± 0.047</td>
<td>3.07 ± 0.042</td>
<td>3.04 ± 0.050</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>200</td>
<td>3.48 ± 0.094*</td>
<td>5.25 ± 0.055**</td>
<td>4.42 ± 0.090**</td>
<td>3.50 ± 0.112***</td>
</tr>
<tr>
<td></td>
<td>400</td>
<td>3.69 ± 0.061**</td>
<td>6.46 ± 0.157**</td>
<td>4.87 ± 0.182**</td>
<td>3.341 ± 0.102N</td>
</tr>
<tr>
<td>Pentazocine</td>
<td>10</td>
<td>3.90 ± 0.056**</td>
<td>7.38 ± 0.176**</td>
<td>6.51 ± 0.129**</td>
<td>5.67 ± 0.074**</td>
</tr>
</tbody>
</table>

*Values are expressed in Mean ± SEM (n = 6); Difference between groups were statistically analyzed by one-way ANOVA; *p*<0.05, **p*<0.001, ***p*<0.001, Dunnett test as compared to control which received normal saline.

(p<0.001) and similar to that of standard drug ASA with significant (p<0.001) inhibition at dose of 200 mg kg⁻¹ body weight.

**Hot plate test**

In hot plate test (Table 2), the *Mimosa pudica* aqueous extract at doses of 200 and 400 mg/kg showed a significant increase in the latency time in a dose-dependent manner. The result was found to be statistically significant (p<0.01) as compared with control and is similar to that of pentazocine-treated group at a dose of 10 mg/kg.

**DISCUSSION**

The results showed that the *Mimosa pudica* aqueous extract administered orally to mice produced significant antinociceptive action when assessed, using acetic acid-induced writhing and thermal-induced nociception tests in mice. In the acute toxicity study, the LD₅₀ of the aqueous extract is high because there was no death recorded, even at 10 times of the effective dose. This indicates that, the extract has high margin of safety.

The acetic acid-induced writhing test in mice is regarded as a model of inflammation pain, and it is used as screening tool for evaluation of analgesic or anti-inflammatory agents. Intraperitoneal injection of acetic acid produces pain through activation of chemosensitive nociceptors [17]. It has been suggested that acetic acid acts by releasing endogenous inflammatory mediators or irritation of the visceral surface, which leads to the liberation of histamine, kinins, prostanoids, serotonin and substance P. It is a sensitive procedure to evaluate peripherally- and centrally- acting analgesics [18-22]. The nociceptive activity of acetic acid may be due to cytokine release, such as TNF-α, interleukin-1β and interleukin-8, by resident peritoneal macrophages and mast cells [23]. The intraperitoneal injection of acetic acid induced an increase in the concentration of glutamate and aspartate in the cerebrospinal fluid [24]. We have reported that the *Mimosa pudica* aqueous extract inhibited, in a dose-dependent manner, the nociception induced by acetic acid, when compared with the well-known NSAID, aspirin.

In this work, we measured the nociceptive reactivity to thermal stimuli in mice using the hot plate test, which is sensitive acute pain test for detecting opiate analgesia as well as several types of hyperalgesia reactions from spinal origin. The hot plate test could be a simple and sensitive procedure to evaluate analgesics and hyperalgesics reactions in mice. This method is considered to be selective for opioid-like compounds in animals [25]. The results indicate that the oral administration of aqueous extract of *Mimosa pudica* significantly attenuated the hot plate thermal stimulation. Hot plate is normally used to study the central analgesics effects of drugs. Therefore, it is probable that *Mimosa pudica* could be producing its effects centrally. These shows the extract increased the stress tolerances capacity of the animals by possible involvement in higher centre. Although the underlying mechanism is unknown, the observed activity can be attributed to the overall effects of the plant constituents or the components having similar structure to NSAIDs or opioids.

In conclusion, the results of the present work clearly demonstrated the antinociceptive activity of *Mimosa pudica* aqueous extract. It also could be concluded that the aqueous extract has both peripheral and central analgesic properties the crude extract has been reported to have interaction with opioid receptors [26]. The antinociceptive effect of this extract may be a result of inhibition or reduction of proinflammatory mediators [27]. The analgesic activity of *Mimosa pudica* aqueous extract can be due to the presence of sterols. Analgesic activities of some sterols have already been shown on the models of pain induced by acetic acid and formalin [28, 29]. Detection of this class of compounds from *Mimosa pudica* may justify their antinociceptive activity. Work is in progress to isolate, characterize and find the mechanism of action of the active compounds in aqueous extract responsible for both peripheral and central analgesic activity.

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CURRENT AUTHOR ADDRESSES

M. Karthikeyan, Department of Pharmacology, Malik Deenar College of Pharmacy, Seetangoli, Bela post, Kerala state, India.

M. K. Deepa, Department of Pharmaceutics, JDT Islam College of Pharmacy, Marikkunnu, Jamthi hill, Malabar, Kerala state, India.