Central Nervous System (CNS) Depressant Activity of *Trema cannabina* Linn

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**Abstract:** The methanolic extract of leaves of *Trema cannabina* Linn. were subjected to neuropharmacological experiments on the central nervous system (CNS) to observe depressant effect. The extract prolonged the pentobarbital induced sleeping time. It also caused a significant reduction in exploratory behavior pattern in mice at the dose of 250 and 500 mg/kg of body weight.

**Key words:** *Trema cannabina* • Methanolic extract • CNS depressant

**INTRODUCTION**

*T. cannabina* belongs to the Cannabaceae family. The plant is distributed in almost all districts of Bangladesh and is used in traditional medicine by the rural people and possesses various interesting pharmacological activities [1]. The root of the plant is used in the treatment of diarrhoea, asthma and passing of blood in urine; the bark is used as poultice in muscular pain; the roots, barks and leaves are used in epilepsy [1,2]. In African folk medicine, it is used in many diseases including dysentery, hypertension, etc [3] Fruit, leaves, bark, stems, twigs and seeds are also used in traditional medicine. The leaves are used to treat coughs and sore throats and the bark is used to make cough syrups. Other reported uses include remedies for bronchitis, gonorrhoea, malaria, yellow fever, toothaches and intestinal worms [1,4].

The root of the plant is used in the treatment of diarrhoea, asthma and passing of blood in urine; the bark is used as poultice in muscular pain; the roots, barks and leaves are used in epilepsy [1,2] and to reduce inflammation [5]. The extract of leaves of *T. cannabina* showed significant analgesic and antidiarrhoeal activities on mice [1]. The methanol extract of *T. cannabina* showed strong antioxidant activity [6]. It is evident from the existing information that this plant may possess some important biological activities, which were not studied and reported before. The main objective of this study was to evaluate the neuropharmacological activities of the methanolic extract of leaves of *Trema cannabina*.

**MATERIALS AND METHODS**

**Experimental Animals:** White albino mice (*Swiss-webstar* strain, 20-25 gm body weight) bred in the animal house of the Department of Pharmacy, Jahangirnagar University, Bangladesh was used for the experiments. The animals were provided with standard laboratory food and tap water and maintained at natural day night cycle. All the experiments were conducted on an isolated and noiseless condition. The test animals were divided into three groups consisting of seven mice in each group. Group I was the control group and group II and III were the experimental groups. The experimental groups were administered with the methanolic extract of leaves of *T. cannabina* (prepared by distilled water and tween-80) at the doses of 250 and 500 mg/kg body weight intra-peritoneally (I.P.), while the animals of group I (control) were supplied with distilled water containing 0.1% tween-80.

**Plant and Material Extraction:** Leaves of *T. cannabina* were collected in early of 2007 from Khulna region of Bangladesh and was identified by the experts of Bangladesh National Herbarium, Mirpur, Dhaka (Accession no. 31,285) and a voucher specimen was kept.

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for future reference. The dried leaves of *T. cannabina* were ground into a fine powder with the help of a suitable grinder (Capacitor start motor, Walu motor factory, China). About 400 g of powered material was extracted by soxhlet apparatus with 90% methanol at 55°C temperature. The extract thus obtained was concentrated by using a rotary evaporator (Bibby RE200, Sterilin Ltd., UK) to get a viscous mass. The viscous mass was then kept at room temperature under a ceiling fan to get a dried extract (about 16% yields). The extract thus obtained was used for pharmacological screening.

**Pentobarbital Induced Hypnosis:** Pentobarbital induced sleeping time test was carried out by the method of Williamson *et al.* [7]. The time for onset of sleep was the time taken for the loss of righting reflex whereas total sleeping time represents the time between the loss and regain of righting reflex. The onset of sleep and total sleeping time were recorded for both control as well as for treated groups.

**Test for Exploratory Behavior in Mice:** This experiment was performed by (i) Open field test, (ii) Hole cross test and (iii) Hole board test. The observations were made on 0 min before injection and 30, 60, 120 and 240 min after injections (i. p.) of the test samples and control.

**Open Field Test:** The number of squares traversed by the animal was recorded for 2 min in this experiment [8].

**Hole Cross Test:** Spontaneous movement of the animals through the hole from one chamber to the other was counted for 2 min in this test [9].

**Hole Board Test:** In this experiment each animal was placed carefully in the center of the board and the number of holes passed, head dipping and the number fecal boluses excreted recorded for 2 min [10]. The observations were made on 0, 30, 60, 90, 120, 180 and 240 min after i.p. injection (at the doses of 250 and 500 mg/kg body weight) of the test drugs.

**Statistical Analysis:** Student's *t*-test was used to determine a significant difference between the control group and experimental groups.

## RESULTS

**Pentobarbital Induced Hypnosis:** The effect of the methanolic extract of *T. cannabina* on pentobarbital induced hypnosis in mice are shown in Table 1. The total sleeping time was about 58 min and 93 min at the doses of 250 and 500 mg/kg respectively where as in control group it was about 31 min. But it had no significant effect on the time of onset of sleep.

**Exploratory Behavior Effects**

**Open Field Test:** It was observed that the methanolic extract of *T. cannabina* caused a significant decrease in the open field score from 30 min to 120 min (*P* < 0.05 at the dose of 250 mg/kg and *P* < 0.001 at the dose of 500 mg/kg). This decrease was more at dose 500 mg/kg than at 250 mg/kg (Table 2).

**Hole Cross Test:** In hole cross test, it was observed that there was a significant (*P* < 0.001) decrease in the number

### Table 1: Effect of methanolic extract of *T. cannabina* on pentobarbital induced hypnosis

<table>
<thead>
<tr>
<th>Animal group</th>
<th>Treatment</th>
<th>Time of onset of sleep</th>
<th>Total sleeping time (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I (Control)</td>
<td>0.1% Tween 80 solution</td>
<td>7.42±0.143</td>
<td>31.34±2.04</td>
</tr>
<tr>
<td>II (Test group-1)</td>
<td>Me. Extract 250 mg/kg</td>
<td>7.13±0.0598*</td>
<td>58.07±2.32**</td>
</tr>
<tr>
<td>III (Test group-11)</td>
<td>Me. Extract 500 mg/kg</td>
<td>7.29±0.1222*</td>
<td>93.16±7.13**</td>
</tr>
</tbody>
</table>

* *P* < 0.05; **P* < 0.001 vs. control, Student's *t*-test; Me = Methanol

### Table 2: Effect of *T. cannabina* on open field test

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg)</th>
<th>At 0 min</th>
<th>At 30 min</th>
<th>At 60 min</th>
<th>At 120 min</th>
<th>At 240 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td></td>
<td>146.1±1.41</td>
<td>92.0±1.71</td>
<td>76.0±1.62</td>
<td>34.67±3.68</td>
<td>13.67±1.76</td>
</tr>
<tr>
<td>Me. Extract 250</td>
<td>123.3±3.13*</td>
<td>59.17±5.21*</td>
<td>49.8±2.13*</td>
<td>20.3±1.42</td>
<td>11.0±0.98</td>
<td></td>
</tr>
<tr>
<td>Me. Extract 500</td>
<td>103.3±1.59*</td>
<td>31.17±4.51*</td>
<td>31.8±3.59*</td>
<td>12.3±1.29</td>
<td>8.2±1.22</td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.E.M. Me., methanolic. * *P* < 0.001, * *P* < 0.05, * *P* < 0.02 vs. control, Student's *t*-test.
Table 3: Effect of *T. cannabina* on hole cross test

<table>
<thead>
<tr>
<th>Treatment Dose</th>
<th>At 0 min</th>
<th>At 30 min</th>
<th>At 60 min</th>
<th>At 120 min</th>
<th>At 240 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>8.50±0.43</td>
<td>6.83±0.477</td>
<td>6.17±0.48</td>
<td>4.67±0.80</td>
<td>3.67±0.56</td>
</tr>
<tr>
<td>Me. Extract 250 mg/kg</td>
<td>7.50±0.51&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.17±0.37&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.33±0.29&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.17±0.23&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1.83±0.19&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Me. Extract 500 mg/kg</td>
<td>7.33±0.67&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.50±0.43&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.67±0.33&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.67±0.33&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1.00±0.26&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.E.M. Me., methanolic. <sup>a</sup>P<0.001; <sup>b</sup>P<0.05; <sup>c</sup>P<0.02; <sup>d</sup>P<0.2 vs. control. Student's t-test

Table 4: Effect of *T. cannabina* on hole board test (head dipping)

<table>
<thead>
<tr>
<th>Treatment Dose</th>
<th>At 0 min</th>
<th>At 30 min</th>
<th>At 60 min</th>
<th>At 120 min</th>
<th>At 240 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>09.33±0.76</td>
<td>09.0±1.37</td>
<td>12.33±2.25</td>
<td>16.33±2.95</td>
<td>18.0±2.50</td>
</tr>
<tr>
<td>Me. Extract 250 mg/kg</td>
<td>11.0±0.98&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.33±1.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>06.17±1.13&lt;sup&gt;c&lt;/sup&gt;</td>
<td>08.33±1.06&lt;sup&gt;d&lt;/sup&gt;</td>
<td>10.67±1.68&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Me. Extract 500 mg/kg</td>
<td>10.33±0.96&lt;sup&gt;a&lt;/sup&gt;</td>
<td>04.83±1.33&lt;sup&gt;b&lt;/sup&gt;</td>
<td>03.83±1.08&lt;sup&gt;c&lt;/sup&gt;</td>
<td>07.33±1.64&lt;sup&gt;d&lt;/sup&gt;</td>
<td>08.33±2.25&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.E.M. Me., methanolic. <sup>a</sup>P<0.001; <sup>b</sup>P<0.05; <sup>c</sup>P<0.02 vs. control. Student's t-test

of hole crossed from one chamber to another chamber by mice from 30 min to 240 min at doses of 250 mg/kg and 500 mg/kg (Table 3).

**Hole Board Test:** It was observed that there was a significant (P<0.001 and P<0.05) decrease in head dip responses in mice from 30 min to 240 min at the doses of 250 mg/kg and 500 mg/kg of body weight (Table 4).

**DISCUSSIONS**

The experimental findings from the study showed that the methanolic extract of *T. cannabina* significantly potentiated the pentobarbital induced sleeping time in mice at the dose of 250 and 500 mg/kg of body weight which suggests its central depressant activity [11], thus suggesting the probable tranquilizing action [12]. Moreover the extract produced a significant decrease in the exploratory behavior pattern in mice, as shown by the results of the head dip in the hole board test, the open field scores in the ‘open field test’ and the number of hole crossed from one chamber to other in the ‘hole cross test’. These results, therefore further support the central sedative properties of the extract. The overall results tend to predict the central nervous system depressant action of the extract.

**REFERENCES**