ORIGINAL RESEARCH ARTICLE

Evaluation of CNS Depressant Activities of Ethanolic Extract of *Ficus bengalensis* (Moraceae) Leaves

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ABSTRACT
Phytochemical analysis of the dried leaves of *Ficus bengalensis* (Moraceae) indicated the presence of flavonoids, alkaloids, triterpenoids, tannins, saponins and steroids. The pharmacological interest of these compounds, coupled with the use of this plant in traditional medicine prompted the authors to check for its possible CNS depressant activities in animal models.

METHODS: In CNS depressant test; hole cross, open field, beam walking  and thiopeptal sodium-induced sedative test was studied.

RESULTS: In CNS-depressant tests; hole cross, open field, beam walking and thiopental sodium-induced sedative test in mice it significantly (p<0.005, p<0.001) decreased the locomotor activity in mice.

CONCLUSION: The obtained results provide a support for the use of this plant in traditional medicine and its further investigation.

KeyWords: CNS-Depressant, *Ficus bengalensis* (Moraceae), locomotor activity, Phytochemical analysis.

INTRODUCTION
Some medical plants have been used for a wide variety of purposes such as food preservation, pharmaceutical, alternative medicine, and natural therapies for many thousands of years. It is generally considered that compounds produced naturally, rather than synthetically, will be biodegraded more easily and therefore being more environmentally acceptable. Thus, natural antioxidants, antibacterial, cytotoxic, antiviral, fungicidal agents and nutrients have gained popularity in recent years, and their use and positive image among consumers are spreading. Herbalism is a traditional medicinal or folk medicine practice based on the use of plants and plant extracts [1]. Many plants synthesize substances that are useful to the maintenance of health in humans and other animals. These include aromatic substances, most of which are phenols or their oxygen-substituted derivatives such as tannins. Many of the herbs and spices used by humans to season food yield useful medicinal compounds [2, 3]. Herbal therapy is used to treat a large variety of ailment and symptoms, e.g., inflammation, fever and pain; however, there are no adequate experimental evidences about their effectiveness [4, 5, 6]. Some species of *Ficus* like *Ficus racemosa*, have been reported to possess astringent, anodyne properties and is helpful in bronchitis [7]. Many plants of this genus are used in medicine for the treatment of inflammatory bowel disease, skin diseases, enlargement of liver and spleen, dysentery, diarrhoea, diabetes, leprosy, lung complaints, leucorrhoea, heart diseases, cough, asthma, piles, ulcers, gonorrhoea, rheumatism and lumbago [8, 9, 10]. Plants belonging to the genus *Ficus* have been shown to contain, a coumarins, anthocynins, alkaloids, terpenoids, sterols and flavanoids [11]. The group of flavonoid is famous for its anti-inflammatory, antiallergic, antithromtic, vasoprotective and protection of gastric mucosa. These properties have been attributed to influence of flavonoids on production of prostaglandins and their antioxidant effects [12]. Therefore, the present study was designed to investigate the CNS

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Depressant activities of the leaves of *Ficus bengalensis* in order to examine the pharmacological basis of the use of the plant in folk medicine.

**MATERIALS AND METHODS:**

**Plant material**
The leaves of *Ficus bengalensis* (Moraceae) were collected from Rangamati, Bangladesh in November 2009, and were taxonomically identified by experts at the Bangladesh National Herbarium (accession number: 50327. Then it was dried in at room temperature in a dry tidy room for 15 days.

**Extraction**
The dried plant material were finely powdered and extracted with 98% ethanol in a glass container keeping it at normal room temperature and repetitive stirring per day. Then the extract was filtered and the chlorophyll was removed. Then the solvent was evaporated at room temperature and the dried crude extract was used for investigation. Percent yield of the extract was 19.4%.

**Animals**
Young Swiss albino mice (20-30 g) of either sex were obtained from the animal house of the International Centre for Diarrhoeal Disease and Research, Bangladesh (ICDDR, B). The animals were housed under standard laboratory conditions (relative humidity 55-65%, room temperature 23±2 °C and 12 h light/dark cycle). The animals were fed with standard diet and water *ad libitum*. The University Animal Research Ethical Committee approved the experimental protocol.

**Drugs**
Diazepam (Square Pharmaceuticals Ltd, Bangladesh).

**Phytochemical group tests**
The ethanol extract was screened by using standard methods [13, 14]. The extract was screened for the presence of alkaloids, saponins, gums, glycosides, glycosides, flavonoids, steroids or tannins, carbohydrates. The reagents were first tested by using standard drugs of corresponding groups available in market. The resulting data is summarized in the (Table 1).

**Determination of CNS depressant activity**
To determine the CNS depressant activity; hole cross test, open field test, beam walking test and thiopental sodium-induced sedative tests were performed.

**Hole cross test**
The animals were divided into four groups (control, positive control and test I and test II group) with 5 mice in each group. A steel partition was fixed in the middle of a cage having a size of $30 \times 20 \times 14$ cm. A hole of 3 cm diameter was made at a height of 7.5 cm in the centre of the cage. The number of passage of a mouse through the hole from one chamber to the other was counted for a period of 5 min started at 0, 30, 60, 90 and 120th min after oral administration of the extract. Diazepam was used in positive control as reference standard at the dose of 1 mg/kg body weight [15]. The resulting data is summarized in the (Table 2).

**Open field test**
The animals were divided into control and test groups containing 5 mice each. The test group received the extract at the doses of 250 and 500 mg/kg body weight orally whereas the control group received vehicle (1% Tween 80 in water). In the board an open field of half square meter was divided into a series of squares each alternatively colored black and white. The apparatus had a wall of 40 cm height. The number of squares passed anyway by the animals was counted for 3 min started at 0, 30, 60, 90 and 120th min after oral administration of the test drugs [16]. The resulting data is summarized in the (Table 2).

**Beam walking test**
Mice were selected randomly and then well trained to walk from a start platform along a ruler (100 cm long, 3 cm wide) elevated 30 cm above the bench by metal supports to a goal box. The successful mice were divided into four groups where n=5. The mice were treated intraperitoneally with the extract at the doses of 250 and 500 mg/kg, Vehicle (1% Tween 80 in water) or diazepam (1 mg/kg body weight). Thirty minutes after the treatment; each mouse was placed on the beam at one end and allowed to walk to the goal box. Mice that fell were returned to the position they fell from, with a maximum time of 60 sec allowed on beam. The number of foot slips (one or both hind limb slipped from the beam) was recorded. The number of foot slip depicts the insufficiency in motor coordination [17]. The resulting data is summarized in the (Table 3).

**Thiopental sodium-induced sedative test:**
Four groups of mice were selected where n=5 and designated as Control group, Positive control group, Test group I and Test group II. Control group animals received thiopental sodium (i.p);
Positive control group animals received thiopental sodium(i.p) plus diazepam(i.p) and group I and II were treated with thiopental sodium(i.p) plus (p.o) ethanol extract of *Ficus bengalensis* respectively. All groups of mice (n=5) were injected with thiopental sodium (10mg/kg i.p) 15min after administration of either diazepam (1mg/kg) or the extract (250 and 500mg/kg) and the time interval between losing and regaining of righting reflex was measured as sleeping time. The resulting data is summarized in the table 5.

### Table 1. Result of phytochemical screening of the ethanol extract of *Ficus bengalensis* (Moraceae).

<table>
<thead>
<tr>
<th>Compound</th>
<th>Alkaloids</th>
<th>Glycosides</th>
<th>Steroids</th>
<th>Gums</th>
<th>Flavonoids</th>
<th>Saponins</th>
<th>Reducing sugars</th>
<th>Tannins</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observation</td>
<td>+ve</td>
<td>-ve</td>
<td>+ve</td>
<td>-ve</td>
<td>+ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
</tbody>
</table>

Key:  +ve = Presence  -ve = Absence

### Determination of CNS depressant activity

#### Hole cross test
There is negligible variation in number of hole crossed from one chamber to another by mice of the control group from 30 to 120min. The groups Test I and Test II treated with the extract at the doses of 250mg/kg and 500mg/kg showed significant decrease of movement from its initial value at 0 to 120min which was comparable with that of the group treated with diazepam (Table 2). The result was statistically significant (p < 0.001).

### Table 2: Effect of the ethanol extract of *Ficus bengalensis* aerial part on hole cross test in mice

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose (mg/kg body weight)</th>
<th>Movements on open field before and after drug administration (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0 Min</td>
</tr>
<tr>
<td>Control</td>
<td>1% Tween in water (p.o.)</td>
<td>15.4 ± 2.2</td>
</tr>
<tr>
<td>Diazepam</td>
<td>1 (i.p.)</td>
<td>18.3 ± 2.7</td>
</tr>
<tr>
<td>Test I</td>
<td>250 (p.o.)</td>
<td>17.2 ± 1.9</td>
</tr>
<tr>
<td>Test II</td>
<td>500 (p.o.)</td>
<td>18.9 ± 2.3**</td>
</tr>
</tbody>
</table>

(n = 5); ** p < 0.001

### Open field test

In the open field test, test I and test II groups treated with the extract at the doses of 250 and 500mg/kg body weight showed significant (p<0.001) dose dependent decrease of movement from its initial value at 0 to 120min (Table 3).

### Table 3: Effect of the ethanol extract of *Ficus bengalensis* on open field test in mice

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose (mg/kg body weight)</th>
<th>Movements on open field before and after drug administration (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0 Min</td>
</tr>
<tr>
<td>Control</td>
<td>1% Tween in water (p.o.)</td>
<td>126.2 ± 6.5</td>
</tr>
<tr>
<td>Diazepam</td>
<td>1 (i.p.)</td>
<td>78.8 ± 5.1</td>
</tr>
<tr>
<td>Test I</td>
<td>250 (p.o.)</td>
<td>89.4 ± 4.7</td>
</tr>
<tr>
<td>Test II</td>
<td>500 (p.o.)</td>
<td>89.8 ± 4.8</td>
</tr>
</tbody>
</table>

(n = 5); ** p < 0.001

### Beam walking test

In the beam walking test, test II group treated with the extract significantly (p < 0.05, P <0.001) induce motor coordination deficit in mice at the dose of 500mg/kg. But the extract at the dose of 250mg/kg test I group did not significantly induce motor coordination deficit in mice (Table 4).

### Table 4: Effect of the ethanol extract of *Ficus bengalensis* on Beam walking test in mice

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose (mg/kg body weight)</th>
<th>Mean time (min) to complete the task (mean ± SEM)</th>
<th>Mean number of foot slips (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1% Tween in water (p.o.)</td>
<td>8.7 ± 1.2</td>
<td>5.3 ± 0.4</td>
</tr>
<tr>
<td>Diazepam</td>
<td>1 (i.p.)</td>
<td>48.6 ± 4.9**</td>
<td>7.1 ± 1.3**</td>
</tr>
<tr>
<td>Test I</td>
<td>250 (p.o.)</td>
<td>11.6 ± 2.7**</td>
<td>6.2 ± 0.7*</td>
</tr>
</tbody>
</table>
Thiopental sodium-induced sedative test
In thiopental sodium induced sleeping time test, test I and test II groups treated with the extract both the doses 250 and 500mg/kg showed significant (p<0.05, p<0.001) decrease in onset of action and increased the duration of sleep (Table 5) which was comparable with the control group. The results from the CNS-depressant tests indicated that it significantly decreased the locomotor activity as shown by the results of the open field and hole cross tests. The locomotor activity is a measure of the level of excitability of the CNS [19]. And decrease of this activity may be closely related to sedation resulting from depression of the central nervous system [20]. In case of the thiopental sodium induced sedative test both the doses of the extract produced a significant increase in the hypnotic effect in a dose dependent manner, thus suggesting a profound sedative activity. This method is a very sensitive way to detect agents with CNS depressant activity [21].The sedative effect recorded here may be related to an interaction with benzodiazepines and related compounds that bind to receptors in the CNS and have already been identified in certain plant extracts.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose (mg/kg body weight)</th>
<th>Onset of sleep in min. (mean ± SEM)</th>
<th>Duration of sleep in min. (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10</td>
<td>13.2 ± 2.312</td>
<td>67.6 ± 4.325</td>
</tr>
<tr>
<td>Positive Control</td>
<td>10 + 5</td>
<td>9.4 ± 1.312*</td>
<td>102.8 ± 5.648**</td>
</tr>
<tr>
<td>Group-I</td>
<td>10 + 250</td>
<td>12.5 ± 2.513</td>
<td>79.3 ± 5.214**</td>
</tr>
<tr>
<td>Group-II</td>
<td>10 + 500</td>
<td>10.2 ± 1.268*</td>
<td>95.8 ± 7.362**</td>
</tr>
</tbody>
</table>

CONCLUSION
In conclusion, we can confirm that the ethanol extracts of Ficus bengalensis (Moraceae) are endowed with CNS depressant activities. Further investigations are necessary for pharmacological and toxicological characterization.

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REFERENCES