EVALUATION OF THREE MEDICINAL PLANTS OF BANGLADESH FOR THROMBOLYTIC POTENTIALS

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ABSTRACT
The crude methanol extracts of whole plant of Wedelia chinensis Osbeck. Merr., stem bark of Mimosa diploptricha Sauvalle. and leaves of Bauhinia purpurea Roxb. as well as their hexane, carbon tetrachloride, chloroform and aqueous soluble partitionates were subjected to screening for thrombolytic activity. The extractives of W. chinensis demonstrated varying extent of clot lysis activities within the range of 4.11 % to 44.24 %. The highest thrombolytic activity was demonstrated by the carbon tetrachloride soluble fraction (44.24±0.48 %) when compared with the standard thrombolytic drug streptokinase (66.77 %). Among the extractives of M. diploptricha and B. malabarica the carbon tetrachloride soluble fraction (19.76±0.88 % clot lysis) and the aqueous soluble fraction (25.83±0.88 %) revealed the highest thrombolytic activities, respectively.

Key words: Wedelia chinensis Osbeck. Merr., Mimosa diploptricha Sauvalle., Bauhinia purpurea Roxb. thrombolysis, streptokinase

INTRODUCTION
Wedelia chinensis Osbeck. Merr. (Synonyms: Solidago chinensis Osbeck., Verbesina calendulacea L.; Bengali name: Kesraj, Bangra, Bhimiraj, Bhimra, Mahavringaraj) commonly known as Chinese Wedelia; is a yellow-flowered perennial herb of sunflower family Asteraceae. In Bangladesh, the plant is found in Chittagong, Dhaka, Mymensingh, Patuakhali, Tangail and Nijum Deep. W. chinensis extract has been reported to attenuate androgen receptor activity and orthotopic growth of prostate cancer (Tsai et al., 2009). The essential oil of W. chinensis is capable of reducing oxidative stress due to cancer development (Manjamalai and Grace, 2012).

Mimosa diploptricha Sauvalle. (Synonyms: Mimosa invisa C. Mart., Morongia pilosa Standl.) commonly known as giant sensitive plant, is a shrubby or sprawling annual vine of Fabaceae family. The plant is native to Brazil and is extremely invasive in the Pacific, where it has been introduced on all island groups. 5-Deoxyflavones with cytotoxic activity have been isolated from M. diploptricha (Lin et al., 2011).

Bauhinia purpurea Roxb. (Synonyms: Bauhinia acida Korth., Casparea castrata Hassk. Hassk.; Bengali name: Phutki, Kanchan, Karmai) is an erect low bushy tree of Caesalpiniaeeae family. The plant is available in evergreen and deciduous forests of Sylhet in Bangladesh. Seven flavonols, including 6, 8-di-C-methyl kaempferol 3-methyl ether, kaempferol, afzelin, quercetin, isoquercitrin, quercitrin, and hyperoside were isolated from the
methanol extract of leaves (Kaewamatawong et al., 2008). The stem bark has been found to possess significant antioxidant activity (Krishnaswamy et al., 2013). Racemosol and demethylracemosol, together with their possible biogenetic precursors, preracemosol A and preracemosol B, were isolated from the roots of B. malabarica (Kittakoopa et al., 2000).

As part of our ongoing investigations on medicinal plants of Bangladesh (Sharmin et al., 2015, 2014, 2013 and 2012; Sarker et al., 2014), the crude methanol extracts of whole plant of W. chinensis, stem bark of M. diplotricha and leaves of B. malabarica growing in Bangladesh, as well as their organic and aqueous soluble fractions were studied for thrombolytic activity for the first time and we, here in, report the results of our preliminary investigations.

MATERIALS AND METHODS

Collection of plant materials and extraction:
The whole plant of W. chinensis, stem bark of M. diplotricha and leaves of B. malabarica were collected in March 2012 from Dhaka and voucher specimens for these collections have been deposited in Salar Khan Herbarium, Department of Botany, University of Dhaka.

The collected plant materials were cleaned, sun dried and pulverized. The powdered materials (500 g each) of the collected plants were separately soaked in 2.0 liters of methanol at room temperature for 7 days. The extracts were then filtered through fresh cotton bed and finally with Whatman filter paper number 1 and concentrated with a rotary evaporator at reduced temperature and pressure. An aliquot (5 g) of each of the concentrated methanol extract was fractionated by the modified Kupchan partition protocol (VanWagenen et al., 1993) and the resultant partitionates were evaporated to dryness with rotary evaporator to yield hexane (HXSF), carbon tetrachloride (CTCSF), chloroform (CSF) and aqueous (AQSF) soluble materials (Table 1). The residues were then stored in a refrigerator until further use.

Table 1: Kupchan partitioning of W. chinensis, M. diplotricha and B. Malabarica

<table>
<thead>
<tr>
<th>Crude extract/ Fractions</th>
<th>W. chinensis (g)</th>
<th>M. diplotricha (g)</th>
<th>B. malabarica (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME</td>
<td>5.0</td>
<td>5.0</td>
<td>5.0</td>
</tr>
<tr>
<td>HXSF</td>
<td>1.0</td>
<td>1.3</td>
<td>1.0</td>
</tr>
<tr>
<td>CTCSF</td>
<td>1.5</td>
<td>0.8</td>
<td>1.0</td>
</tr>
<tr>
<td>CSF</td>
<td>1.0</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>AQSF</td>
<td>0.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
</tbody>
</table>

ME= Methanolic crude extract; HXSF= Hexane soluble fraction; CTCSF= Carbon tetrachloride soluble fraction; CSF= Chloroform soluble fraction; AQSF= Aqueous soluble fraction

Thrombolytic activity
The thrombolytic activity was evaluated by the method developed by Prasad et al. (2006) by using streptokinase as positive control.

STATISTICAL ANALYSIS
For all bioassays, three replicates of each sample were used for statistical analysis and the values are reported as mean ± SD.
Table-2 Thrombolytic activities of W. chinensis, M. diplotricha and B. malabarica extractives

<table>
<thead>
<tr>
<th>Samples/Standard</th>
<th>W. chinensis</th>
<th>M. diplotricha</th>
<th>B. malabarica</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME</td>
<td>12.53±0.54</td>
<td>8.42±0.52</td>
<td>0.55±0.17</td>
</tr>
<tr>
<td>HXSF</td>
<td>11.40±0.39</td>
<td>8.22±0.63</td>
<td>8.95±0.84</td>
</tr>
<tr>
<td>CTCSF</td>
<td>44.24±0.48</td>
<td>19.76±0.88</td>
<td>13.98±0.23</td>
</tr>
<tr>
<td>CSF</td>
<td>4.26±0.23</td>
<td>6.98±1.03</td>
<td>6.42±0.45</td>
</tr>
<tr>
<td>AQSF</td>
<td>4.11±0.68</td>
<td>6.02±0.41</td>
<td>25.83±0.88</td>
</tr>
<tr>
<td>Water</td>
<td>3.79±0.55</td>
<td>6.79±1.08</td>
<td>66.77±1.08</td>
</tr>
<tr>
<td>Streptokinase</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

ME = Methanol crude extract; HXSF = Hexane soluble fraction; CTCSF = Carbon tetrachloride soluble fraction; CSF = Chloroform soluble fraction; AQSF = Aqueous soluble fraction.

RESULTS AND DISCUSSION

The crude methanol extracts of whole plant of W. chinensis, stem bark of M. diplotricha and leaves of B. malabarica as well as their hexane, carbon tetrachloride, chloroform and aqueous soluble partitionates were subjected to screenings for thrombolytic potentials.

In order to identify the drugs with the ability to promote lysis of blood clot from natural resources, the extractives of W. chinensis, M. diplotricha and B. malabarica were assessed for thrombolytic activity. Addition of 100 μl streptokinase, a positive control (30,000 I.U.) to the clots of human blood and subsequent incubation for 90 minutes at 37°C showed 66.77 % lysis of clot. On the other hand, distilled water, treated as negative control, revealed a negligible lysis of clot (3.79 %). Among the extractives of W. chinensis and M. diplotricha, the carbon tetrachloride soluble fractions of both the extracts exhibited 44.24±0.48 % and 19.76±0.88 % clot lysis, respectively. On the other hand, B. malabarica extractives showed mild to moderate thrombolytic activity and the highest thrombolytic activity was demonstrated by the aqueous soluble fraction (25.83±0.88 %) (Table 2).

CONCLUSION

The objective of the study was to evaluate the thrombolytic potential of crude methanol extracts of whole plant of W. chinensis, stem bark of M. diplotricha and leaves of B. malabarica as well as their hexane, carbon tetrachloride, chloroform and aqueous soluble partitionates. It is clearly evident from the above findings that the carbon tetrachloride soluble fraction of W. chinensis bark exhibited significant thrombolytic activity but the M. diplotricha and B. malabarica extractives demonstrated mild to moderate thrombolytic activity. Therefore, these plants are good candidates for further systematic, chemical and biological studies to isolate the active principles.

REFERENCES


