INVITRO ANTIOXIDANT POTENTIAL OF METHANOLIC EXTRACT OF WHOLE PLANT OF EUPHORBIA HIRTA L. (EUPHORBIACEAE)

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ABSTRACT

Euphorbia hirta L. of the family Euphorbiaceae has been considered as an important herb and is used in the treatment of various diseases like bronchitis, skin diseases kidney stone etc. The objective of present investigation was to evaluate the antioxidant potential of Euphorbia hirta L. Antioxidant potential was determined by DPPH free radical scavenging activity using ascorbic acid as standard. The result showed that methanolic extract of whole plant of Euphorbia hirta L. possess antioxidant property (IC50 33.64 µg/ml) compared to that of the standard ascorbic acid (IC50 12.93 µg/ml).

KEYWORDS: Euphorbia hirta L., Antioxidant, Ascorbic acid, DPPH.

INTRODUCTION

Euphorbia hirta L. is a small annual herb belonging to the family Euphorbiaceae. The stem of the plant is slender, reddish in colour & produce white juice on cutting.[1] The leaves of this plant is opposite, elliptical, lanceolate having a faintly toothed margin and are green in colour.[2] The flowers are green, unisexual, small, crowded together in dense cyme. The male flowers are sessile, linear bracteoles, fringed, lack perianth, and possesses one stamen,
whereas the female flowers have short pedicel, the perianth is rimmed, with superior ovary.\textsuperscript{[3]} Fruits are yellow in colour which contains three brown, four-sided, angular, wrinkled seeds,\textsuperscript{[1]} \textit{Euphorbia hirta} L. is used in the treatment of many disease including bronchitis, skin diseases, cough, hay asthma, bowel disease, worm infestation, kidney stones, bronchial disease, to decrease lactation; as sedative, anxiolytic, analgesic, antipyretic, and as anti inflammatory agent.\textsuperscript{[4]}

**MATERIAL AND METHOD**

**Collection of Plant Material**

The Indigenous plant \textit{Euphorbia hirta} L. were collected from different locations of Bhopal (M.P.) region. The plants were acknowledged by a senior Botanist Dr. Tayaaf Safi Principal Gandhi P.R. College Bhopal.

**Preparation of Extract**

Plant material was washed with water and then allowed to dry in shade for about 3 to 4 weeks. Dried plant materials were grinded by using the electronic grinder. The powder of the whole plants of \textit{Euphorbia hirta} L. was extracted according to (Harborne and Baxter., 1995).\textsuperscript{[5]} The dried plants sample was powered and filed into the soxhlet using petroleum ether and methanol respectively. Almost all the chlorophyll and lipid was deposited on the side of the flask and removed carefully. The extracts were stored in refrigerator till any further use.

**Antioxidant Activity**

**DPPH free Radical Scavenging Assay\textsuperscript{[6,7,8]}**

**Principle:** The scavenging reaction between (DPPH·) and antioxidant (H-A) can be written as:

\[
\text{(DPPH·) + (H-A)} \rightarrow \text{DPPH-H + (A)}
\]

(Purple) \hspace{2cm} (Yellow)

**Preparation of Standard Ascorbic acid solution**

Various solution of the ascorbic acid was prepared in 90% methanol to obtain different concentration (10-100µg/ml). 200µM solution of DPPH in methanol was prepared and 1.5ml of this solution was added to 1.5ml of methanolic ascorbic acid solution to different
concentration and incubated for 30 min (at room temperature) in dark. After 30 minutes, the absorbance of each solution of ascorbic acid was taken against methanol (as blank) at 517nm.

**Preparation of Test solution**

Various solution of plant extract was prepared in 90% methanol to obtain different concentrations (10-100µg/ml). 200µM solution of DPPH in methanol was prepared and 1.5ml of this solution was added to 1.5ml of methanolic extract solution of different concentration and incubated for 30 min (at room temperature) in dark. After 30 minutes, the absorbance of each solution of ascorbic acid was taken against methanol (as blank) at 517nm.

**Preparation of Control solution**

For control, 1.5ml of methanol was mixed with 200µM DPPH solution and incubated for 30 min at room temperature in dark. Absorbance of the control was taken after 30min against methanol (as blank) at 517 nm. The antioxidant activity of plant leaf extract and ascorbic acid were calculated by using the following formula in terms of % of inhibition:

\[
\% \text{ Inhibition} = \left[ \frac{(A_c \text{ 515 nm} - A_t \text{ 515 nm} / A_c \text{ 515 nm}) \times 100}{100} \right].
\]

Where,

\[ A_c = \text{Absorbance of control} \]
\[ A_t = \text{Absorbance of ascorbic acid/ methanoli plant extract.} \]

**RESULTS AND DISCUSSION**

Antioxidants react with DPPH, a stable free radical, which gets reduced to DPPH-H consequently, the absorbance gets decreased. The degree of discoloration indicates the scavenging potential of the antioxidant compounds or extract in terms of hydrogen donating ability.

The obtained results of the methanolic extracts of *Euphorbia hirta* L. plant are shown below. The scavenging activity of methanolic extract of whole plant of *Euphorbia hirta* L. was found (IC\(_{50}\) 33.64 µg/ml). The scavenging effect was compared to that of the standard ascorbic acid with IC\(_{50}\) value 12.93 µg/ml.
Table 1: % Inhibition data of DPPH free radical scavenging assay by ascorbic acid

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Conc.(µg/ml)</th>
<th>Absorbance (Control), Ac</th>
<th>Absorbance (Test), At</th>
<th>% Inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>2</td>
<td></td>
<td>0.51</td>
<td>27.143</td>
</tr>
<tr>
<td>2.</td>
<td>4</td>
<td></td>
<td>0.485</td>
<td>30.714</td>
</tr>
<tr>
<td>3.</td>
<td>6</td>
<td></td>
<td>0.458</td>
<td>34.571</td>
</tr>
<tr>
<td>4.</td>
<td>8</td>
<td></td>
<td>0.424</td>
<td>39.429</td>
</tr>
<tr>
<td>5.</td>
<td>10</td>
<td></td>
<td>0.395</td>
<td>43.571</td>
</tr>
<tr>
<td>6.</td>
<td>12</td>
<td></td>
<td>0.362</td>
<td>48.286</td>
</tr>
<tr>
<td>7.</td>
<td>14</td>
<td></td>
<td>0.336</td>
<td>52</td>
</tr>
<tr>
<td>8.</td>
<td>16</td>
<td></td>
<td>0.302</td>
<td>56.857</td>
</tr>
</tbody>
</table>

Fig 1: Standard curve of ascorbic acid. Graph representing regression curve of ascorbic acid by DPPH assay method

Table 2: % Inhibition of DPPH by methanolic extract of *Euphorbia hirta* L.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Conc. (µg/ml)</th>
<th>Absorbance (Control), A_c</th>
<th>Absorbance (Test), A_t</th>
<th>% Inhibition</th>
<th>IC50 (µg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>10</td>
<td>0.478</td>
<td>0.373</td>
<td>21.97</td>
<td>33.64</td>
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<tr>
<td>2.</td>
<td>20</td>
<td></td>
<td>0.332</td>
<td>30.54</td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>30</td>
<td></td>
<td>0.278</td>
<td>41.84</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>40</td>
<td></td>
<td>0.194</td>
<td>59.41</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>50</td>
<td></td>
<td>0.131</td>
<td>72.59</td>
<td></td>
</tr>
</tbody>
</table>
Fig.2: Graph represent regression curve of methanolic Extract of *Euphorbia hirta* L. by DPPH assay method

CONCLUSION

The present study was carried out to investigate the antioxidant property of methanolic extract of the plant *Euphorbia hirta* L. The scavenging activity of the plant methanolic extract through DPPH radicals was investigated using ascorbic acid as standard. The current results suggested that the *E. hirta* L. methanolic extract has potent antioxidant effects. The antioxidant property of the plant can be use in the treatment of various diseases.

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