Larvicidal activity of essential oils from Indian medicinal plants against Aedes aegypti L.

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ABSTRACT

Volatile oils from Curcuma longa leaf, seeds of Leucas aspera and Allium cepa were evaluated for static larvicidal bioassay test against Aedes aegypti larvae, in searching for biocontrol agents for mosquito. The resulted mean of LC50 values placed the plants volatile oils as good larvicidal agents in the order of Curcuma longa (3.942 µl) > Leucas aspera (4.159 µl) > Allium cepa (5.909 µl).

Key words: Curcuma longa; Leaf, Leucas aspera & Allium cepa; Seeds, Larvicidal Activity

INTRODUCTION

The mosquito, Aedes aegypti (Diptera) is a vector for the etiological agent arbovirus, which is responsible for dengue fever and dengue hemorrhagic fever, and it is reported that, two-fifths of the world population are now at risk for catching the dengue. Over the last five decades many approaches were done in public health programme for control of these mosquito born diseases in the interruption of disease transmission by either killing, by using repellants or causing larval mortality in the large scale at breeding centers of the vectors (WHO 1981). Conventional use of synthetic insecticides for the mosquito control has created multifarious problems. In an attempt to overcome the problem, great emphasis has been recently placed on the research and development of vector control using eco-friendly bioresources based on plant products as alternatives of synthetic chemical insecticides.

Thus the present work has been focused on three different plants volatile oil namely, Curcuma longa leaf, Leucas aspera seed and Allium cepa seed for evaluating the larvicidal activity. The plant Curcuma longa (Zingiberaeae) is a perennial herb, short stem measures up to 1 meter high, distributed through out tropical and subtropical regions of the world and widely cultivated in India and China. It was used as spice, flavouring agent, and also used in medicines of anorexia, diabetic wounds, rheumatism, swellings and sinusitis [1]. The plant Leucas aspera (Labiatae) is a much branch, diffuse annual herb with 30-60cms. high, found in all over Indian cultivated fields as weed. The plant is used as antipyretic, external application for psoriasis, chronic skin eruption, painful swellings and also used as a medicines for cough and cold [2a, 2b]. The plant Allium cepa (Liliaceae) is a flavouring vegetable in various types of foods. These are used in medicines for fever dropsy chronic bronchitis, colic and scurvy [3].

MATERIALS AND METHODS

The Plant volatile oil: Three different plant materials of distinct character in odour namely leaf of Curcuma longa, Seeds of Leucas aspera and Allium cepa were collected from the surrounding fields of S.K. University, Anantapur, India. The voucher specimens were deposited in S.K.U herbarium. The collected plant materials were subjected to hydro-distillation in Clevenger apparatus for 5h. in order to obtain volatile oil [4]. The oil thus obtained, was stored in brown bottles under refrigeration for subsequent use in the assay. The mosquito larvae: Aedes aegypti larvae were procured from Vector Control Board, Government of Andhra Pradesh and also collected from various places of clean stagnant water, Anantapur, India. The larvae were identified and authenticated by entomologist. For the growth of the larvae, the laboratory was maintained with netted tubes at 25-30°C temperature and 80-90% relative humidity under the photoperiod 14:10 (light:dark) and glucose granules or dog biscuits were given as feed to larvae, in the animal house, S.K. University, Anantapur, India [5].

Statistic larvicidal bioassay: To evaluate the 50% lethal concentration of different plants volatile oils against Aedes aegypti larvae, standard WHO larval susceptibility test procedure was followed (WHO 1981), and from the results of mortality data, 50% toxicity was assessed by the application of Finney probit statistical method [6] and confirmatory test by Dragstedt-Beheren’s equation. The assay was carried out by maintaining a series of log concentration of volatile oil ranging from 0.301 to 1.000 µl by dissolving the plants volatile in distilled water and 25 early fourth instar larvae of Aedes aegypti were exposed for each of such oil concentration. Four replicates were run for each concentration with final total of 100 larvae tested for each oil concentration. Simultaneously control test at the rate of 25 larvae was carried out with the required quantity of distilled water. After the exposure of larvae for 24h, their mortality and partial mortality were registered and accordingly the average values were ascertainment from the replicates of each oil concentration [7].

Calculation of Regression: The effect of different plants volatile oils on the mortality of fourth instar Aedes aegypti larvae after 24hrs. exposure, were corrected for natural response by the
Table 1: Larval mortality percentage of 4th instar larvae of Aedes aegypti exposed for 24 hours to different concentrations of Plants volatile oils. 

<table>
<thead>
<tr>
<th>Plants Volatile oils Parameter in Log Concentration (m/l)</th>
<th>Curcuma longa Leaf Volatile oil % of larvace mortality % of larvace mortality</th>
<th>Leucas aspera seed Volatile oil % of larvace mortality % of larvace mortality</th>
<th>Allium cepa seed Volatile oil % of larvace mortality % of larvace mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.00</td>
<td>00</td>
<td>00</td>
<td>00</td>
</tr>
<tr>
<td>0.301</td>
<td>00</td>
<td>00</td>
<td>00</td>
</tr>
<tr>
<td>0.602</td>
<td>52</td>
<td>5.05</td>
<td>48</td>
</tr>
<tr>
<td>0.778</td>
<td>56</td>
<td>5.15</td>
<td>52</td>
</tr>
<tr>
<td>0.903</td>
<td>76</td>
<td>5.71</td>
<td>80</td>
</tr>
<tr>
<td>1.000</td>
<td>96</td>
<td>6.75</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 2: LC_{50} values (t/l) and their 95% fiducial (upper and lower) limits, regression equation and chi-square (\chi^2) values of three plant volatile oils against Aedes aegypti larvae. 

<table>
<thead>
<tr>
<th>Plants Volatile oils</th>
<th>LC_{50}±SEM</th>
<th>L.F.L</th>
<th>U.F.L</th>
<th>Regression Equation</th>
<th>Calculated \chi^2 value</th>
<th>Table \chi^2 value at (0.05)/(n-2)/df</th>
</tr>
</thead>
<tbody>
<tr>
<td>Curcuma longa Leaf Volatile oil</td>
<td>3.942 0.03</td>
<td>3.912</td>
<td>3.972</td>
<td>Y = 2.24x + 0.96</td>
<td>5.17</td>
<td>11.07</td>
</tr>
<tr>
<td>Leucas aspera seed Volatile oil</td>
<td>4.159 0.11</td>
<td>4.049</td>
<td>4.269</td>
<td>y = 2.56x – 1.29</td>
<td>4.78</td>
<td>11.07</td>
</tr>
<tr>
<td>Allium cepa seed Volatile oil</td>
<td>5.909 0.06</td>
<td>5.849</td>
<td>5.969</td>
<td>Y = 2.71x – 2.55</td>
<td>4.49</td>
<td>11.07</td>
</tr>
</tbody>
</table>

following Abbott’s formulae [8]. 

Abbott’s % mortality = \frac{\text{% Proportion of test mortality}}{\text{100 - % proportion of control mortality}} \times 100

Based on the log concentration and the mortality percentage values, regression equation were obtained. The homogeneity of population could also be tested by Chi-square test (\chi^2). By using median, lethal concentration (LC_{50}) values of the different plant volatile oils after 24 hrs. exposure against fourth instar larvae of Aedes aegypti and their fiducial limits (95% upper fiducial limit and lower fiducial limit) could be calculated.

RESULTS AND DISCUSSIONS

The percentage mortality data at different Log. concentrations (ranging from 0 to 1 t/l) of the three plants volatile oil namely Curcuma longa leaf and seeds of Leucas aspera and Allium cepa were shown in Table 1. The Regression equations (based on 24hrs. mortality) along with LC_{50} values and the Chi-square values for each concentration of each plant against Aedes aegypti larvae were represented in Table 2. Based on the probit analysis, the median lethal concentrations (LC_{50}) of the three plants namely Curcuma longa, Leucas aspera, Allium cepa are found to be 3.942 t/l, 4.159 t/l and 5.909 t/l, respectively. Among the three plants volatile oils, Curcuma longa exhibits good larvicidal activity at lowest concentrations (3.942 t/l) than Leucas aspera and Allium cepa. Further the homogeneity of population were also tested among these three plants and their chi-square (\chi^2) values were found to be 5.17 for Curcumalonga, 4.78 for Leucas aspera and 4.49 for Allium cepa plants volatile oils. Further the presence of biologically active compounds such as Thymol, Myrcene, Curcumene, Turmerone, Linalool, Carvacrol, Pinene, -Phellandrene, Cineole, Caryophyllene, Menthol and many Carboxylic and alcohols are found in volatile oil of these three plants which can promote larvicidal and insecticidal property [9]. Alluri et al., (2005) [10] assessed the mortality rate of 120 medicinal plants. Comparing with those plants, the present tested plants volatile oil namely Curcuma longa, Leucas aspera and Allium cepa were assessed themselves as a good biocontrol agents against Aedes aegypti larvae and hence these plants volatile oils, can be used as eco-friendly, biodegradable, low cost and indigenous ones for vector control with minimum care by individual and communities in specific situations as per present demand.

CONCLUSION

Keeping in view of larvicidal properties of volatile oil of Curcuma longa leaf and seeds of Leucas aspera and Allium cepa, the volatile oils of these plants can be used for the emergence of new biocontrol agent that can meet the challenges or threat of mosquito larvae, effectively.

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