

SHORT COMMUNICATION

## Botanicals as Mosquito Larvicides

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### ABSTRACT

Methanol extracts of 19 indigenous plants were evaluated as mosquito larvicide. Among these, pericarp of *Zanthoxylum limonella* was found to have the most promising larvicidal properties against *Aedes(s) albopictus* and *Culex quinquefasciatus* with LC<sub>90</sub> values at 0.47 ppm and 0.73 ppm, respectively. The extract of *Piper nigrum* was also found very effective (LC<sub>90</sub>) on the larvae of both the species at 6.8 ppm and 8.4 ppm, respectively. The extracts of the remaining plant parts showed LC<sub>90</sub> values at above 100 ppm concentration. Extract of *Calotropis gigantea* was found to be the least effective (LC<sub>90</sub> values at 962.8 ppm and 1091.8 ppm) against the larvae of both the species. However, plant extracts were found more effective against *Aedes(s) albopictus* larvae than against *Culex quinquefasciatus* larvae.

**Keywords:** Mosquito larvicide, plant extract, larvicidal activity, *Zanthoxylum limonella*, *Piper nigrum*, *Calotropis gigantea*, *Aedes(s) albopictus*, *Culex quinquefasciatus*

### 1. INTRODUCTION

The geo-climatic condition of north-eastern region of India supports fast proliferation and growth of natural fauna and flora. Many of the herbs and shrubs are found to have promising medicinal properties, mosquito larvicidal and mosquito repellent properties. Owing to the fact that application of synthetic larvicide has envenomed the surroundings as well as non-target organisms, natural products of plant origin with insecticidal properties have been tried as an indigenous method for the control of a variety of insect pests and vectors in the recent past<sup>1-5</sup>. Phytotoxicological activity of *Tagetes erecta* was demonstrated by Sharma and Saxena<sup>6</sup> against second and third instar larvae of *Anopheles stephansi*. Azadiractin, the active ingredient of *Azadiracta indica* has long been recognised for its mosquito larvicidal capability. Long before the advent of synthetic insecticides, plants and their derivatives

were being used to kill the pests of agriculture, veterinary and public health importance. Insecticidal activity of plant-derived compounds have been evaluated and few of these exploited commercially<sup>7</sup>. Laboratory and field trials of plant extracts and purified chemicals showed mosquito larvicidal activity. Larvicidal activity of certain extracts of plant parts such as *Acorus calamus*, *Adhatoda vesica*, *Croton tiglium*, *Mentha arvensis*, *Ocimum basilicum*, and *Vitex negundo* against *Culex* larvae was reported by Deshmukh<sup>8</sup>. Also, the alcoholic extract of *Artemisia annua* was reported to be more toxic than the hexane extract and acetone extract of *Artemisia vulgaris* against *Culex* larvae. Similarly, the extract of *Cuscuta reflexa* was found more effective than the extract of *Cannabis sativa*, against *Culex* larvae<sup>9</sup>. According to Sharma and Shrivastava<sup>10</sup>, the extract of *Cuscuta reflexa* was the most toxic to *Culex* larvae, followed by extracts of *Artemisia annua*, *Carica papaya*,

and *Lantana indica*. Latha<sup>11</sup> reported *Piper longum* and *Zingiber wightianum* extracts at 80 mg/l causing complete mortality on *Culex quinquefasciatus* and 60 mg/l for *Culex sitiens*. Fruit and seed extracts of *Abrus precatorious* were found toxic to adult mosquitoes<sup>12</sup> and crude extract of *Withospermum arvense* were found toxic to *Aedes aegypti* larvae<sup>13</sup>. Several such reports related to the toxic properties of plant products against mosquito larvae like peel oils of three citrus fruits against *Culex pipiens*<sup>14</sup> and larvicidal properties of leaf extracts of *Solanum nigrum*<sup>15</sup> are available.

Insecticides of plant origin have been extensively used on agricultural pests, and to a vary limited extent, against insects vectors of public health importance, which deserve careful and thorough screening. The use of plant extracts for insect control has several appealing features, as these are generally more biodegradable, less hazardous, and rich storehouse of chemicals of diverse biological activity. Moreover, herbal sources give a lead for discovering new insecticides. Therefore, biologically active plant materials have attracted considerable interest in mosquito control programmes in the recent times. The present study deals with the screening of locally available herbs and shrubs for mosquito larvicidal properties.

## 2. MATERIALS AND METHODS

Plant materials collected from the foothill forests of Sonitpur district, Assam, were segregated as leaf, twig, flower, tuber, root, etc and were air dried in a shady place to retain their active ingredients intact. Dried materials were powdered in a table model grinder for extraction and each of the powdered plant material (500 g) was soaked in methanol in a airtight wide mouth bottle and kept for 7 days. After that, the cold extracts from the bottles along with methanol were filtered and kept in petridishes for drying at room temperature. The average yield of crude extract (500 g) of powder was 3.5 g. Dried extracts were used for larvicidal bioassay as per WHO standards. Stock solutions were prepared by dissolving plant extract (1 g) in water (1 l) to make its strength 1000 ppm. Teepol (1 ppm) was also added as emulsifying agent. Different concentrations

were prepared by adding required doses of stock solution in beakers containing water (250 ml).

Laboratory-reared *Aedes albopictus* and *Culex quinquefasciatus* were used for larvicidal bioassay under laboratory conditions ( $27 \pm 2$  °C and  $75 \pm 5$  % RH). Twenty five mosquito larvae of third or early fourth instar were released in each beaker for 24 h with a concurrent control for every set of experiment. Three replicates were kept for each concentration. No food was added in the beaker as per WHO norms. Mortality was recorded after 24 h of treatment and mortality corrected by Abbot's formula. Data were analysed by probit analysis<sup>16</sup>.

$$\text{Abbott's formula} = \frac{\text{Percentage of test mortality} - \text{Percentage of control mortality}}{100 - \text{percentage of control mortality}} \times 100$$

## 3. RESULTS AND DISCUSSION

Out of 19 plants, pericarp extract of *Zenthoxylum limonella* was found to possess the most effective larvicidal activity against *Aedes(s) albopictus* and *Culex quinquefasciatus* with LC<sub>90</sub> values at 0.47 ppm and 0.73 ppm, respectively, followed by seed extract of *Piper nigrum*, *Piper longum*, and leaf extract of *Spilanthes acmella* (Table 1). Among the others, peel extract of *Citrus aurantifolia*, leaf extract of *Mentha arvensis*, and flower extract of *Tagetas patula* were also found to have larvicidal properties. Leaf extracts, of *Lippia nodiflora*, *Chrysanthemum cinerarifolium*, *Leucus linifolia*, *Lantana camara*, fruit extract of *Capsicum annum*, root extract of *Moringa oleifera*, and rhizome extract of *Zingiber officinalis* also showed larvicidal (LC<sub>90</sub>) activity against both the species with a higher dose.

As compared to the other herbal extracts, concentration of *Zenthoxulum limonella* extract for LC<sub>90</sub> value was very low for both the species. Results of *Piper nigrum* were also encouraging considering other plant extracts tested against *Aedes albopictus* and *Culex quinquefasciatus*. LC<sub>50</sub> values of *Zenthoxulum limonella* showed at 0.01 ppm and 0.02 ppm and for *Piper nigrum*, at 0.56 ppm and 0.65 ppm accordingly. It may be the plant alkaloid that have toxic effect on mosquito larvae. According to Saxena<sup>5</sup>, *et al.* plant alkaloids resulted in a significant loss in fecundity

**Table 1. Mosquito larvicidal efficacy of some plant extracts**

Plant species (with common names)	Part used	<i>Culex quinquefasciatus</i>			<i>Aedes(s) albopictus</i>		
		LC <sub>50</sub> ppm	LC <sub>90</sub> ppm	Regression equation	LC <sub>50</sub> ppm	LC <sub>90</sub> ppm	Regression equation
<i>Zanthoxylum limonella</i> (Bezaroni)	Pericarp	0.02	0.73	$Y = 0.775x + 4.062$	0.01	0.47	$Y = 0.749x + 4.279$
<i>Pipernigrum</i> (jaluk)	Seed	0.65	8.42	$Y = 1.0983x + 4.758$	0.56	6.80	$Y = 1.1225x + 4.1220$
<i>Spilanthes acmella</i>	Leaf	57.40	108.30	$Y = 4.1627x + 2.3501$	54.10	113.90	$Y = 3.8499x + 1.6784$
<i>Piper longum</i> (Pipoli)	Seed	70.10	113.90	$Y = 5.669x + 5.4992$	54.50	92.10	$Y = 5.4766x + 4.5187$
<i>Citrus aurantifolia</i>	Peel	96.40	128.40	$Y = 10.201x + 15.274$	79.50	116.70	$Y = 7.6293x + 9.5318$
<i>Mentha arvensis</i> (Pudina)	Leaf	98.90	193.90	$Y = 4.3440x + 3.6756$	90.20	111.90	$Y = 4.3528x + 3.5360$
<i>Tagetespatula</i> (Nargee)	Flower	118.10	261.070	$Y = 3.5126x + 2.3014$	84.79	185.10	$Y = 3.5780x + 1.9266$
<i>Lippia nodiflora</i> (Nemu sak)	Leaf	199.00	301.60	$Y = 6.9745x + 11.0557$	190.10	267.50	$Y = 7.9629x - 13.147$
<i>Chrysanthemum</i> <i>Cinerariifolium</i> (Indrimalati)	Leaf	219.00	398.60	$Y = 4.7213x - 6.0653$	227.00	323.90	$Y = 4.4089x - 5.4081$
<i>Capsicum annuum</i> (Jalakia)	Fruit	240.70	314.70	$Y = 10.1775x - 19.261$	208.20	320.80	$Y = 6.1373x - 9.2791$
<i>Leucas linifolia</i> (Doron)	Leaf	250.00	315.00	$Y = 12.8550x - 25.874$	233.20	305.50	$Y = 10.8577 - 20.766$
<i>Lantana camara</i> (Guti phul)	Leaf	273.50	374.70	$Y = 9.2798x - 17.6632$	268.30	364.20	$Y = 9.1651x - 17.304$
<i>Ocimum sanctum</i> (Tulshi)	Leaf	288.20	528.30	$Y = 4.5162x - 6.2776$	176.90	271.00	$Y = 7.1467x - 11.067$
<i>Moringa oleifera</i> (Sajina)	Root	317.90	486.60	$Y = 6.7917x - 12.0074$	283.40	498.20	$Y = 4.9387x - 7.1333$
<i>Hibiscus rosa sinensis</i> (Joba)	Leaf	389.50	613.30	$Y = 6.3069x - 11.3451$	294.90	501.90	$Y = 5.3382x - 8.1973$
<i>Zingiber officinalis</i> (Ada)	Rhizome	390.10	628.30	$Y = 5.9706x - 10.4801$	521.10	524.20	$Y = 5.8004x - 9.5389$
<i>Curcuma amada</i> (Amda)	Rhizome	395.10	606.90	$Y = 6.4218x - 11.7044$	481.20	783.10	$Y = 5.7232x - 10.377$
<i>Adnentera pavonia</i> (Lalchandani)	Seed	474.90	834.90	$Y = 5.0250x - 8.4563$	420.10	743.70	$Y = 4.7796x - 7.5467$
<i>Calotropis gigantea</i> (Akan)	Leaf	628.90	1091.80	$Y = 5.1569x - 9.4424$	467.00	962.80	$Y = 3.8702x - 5.3486$

and fertility in the adult species of mosquitoes. Sharma and Saxena<sup>6</sup> also found that the petroleum ether extract of *Tagetes erectes* had toxic effect on larvae of *Anopheles stephensi* and on its significant growth index. In the present study, post-treatment data analysis on growth and fertility could not be carried out. Mwangi and Mukiyama<sup>17</sup> observed that one fraction of *Melia volkensii* fruit kernel extract

had growth inhibition activity at low concentration, whereas two other fractions had acute toxic effects on the mosquito larvae. As per Pushpalatha and Muthukrishnan<sup>18</sup>, leaf extracts of *Vitex negundo*, *Nerium oleander*, and seed of extract of *Syzygium jambolanum* at very low concentration had larvicidal activity against *Culex quinquefasciatus* and *Anopheles stephensi*, and also extended the duration of larval

instars pupation. Mudrigal,<sup>13</sup> *et al.* reported that methanol extract of *Lithospermum arvense* were toxic to *Aedes aegypti* larvae. Fractionation of *Zenthoxylum limonella* extract may give more active compound for larval control of mosquitoes. It is evident from the present study that herbal extracts might have promising larvicidal efficacy.

Plants are rich source of bioactive organic chemicals and offer an advantage over synthetic pesticides as these are less toxic, less prone to development of resistance, and easily biodegradable. Screening and identification of effective compounds available in north-eastern region of India will certainly bring more success towards the control of mosquitoes.

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