

## LARVICIDAL EFFICACY OF PLANT OILS AGAINST THE FILARIAL VECTOR, CULEX QUINQUEFASCIATUS (L.) (DIPTERA: CULICIDAE)

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### ABSTRACT

The bioactivity of ten plant oils, Basil (*Ocimum basilicum*), Catnip (*Nepeta cataria*), Clary Sage (*Salvia sclarea*), Cornmint (*Mentha arvensis*), Lavender (*Lavandula angustifolia*), Marjoram (*Thymus mastichina*), Patchouli (*Pogostemon cablin*), Rosemary (*Rosmarinus officinalis*), Spearmint (*Mentha spicata*), Thyme (*Thymus vulgaris*), were tested at 100, 200, 300, 400, and 500 ppm concentrations against the fourth instar larvae of *Culex quinquefasciatus*. Larval mortality was observed after 24 hours. The data obtained from the present investigation clearly revealed that the oils have significant influence on the experimental larvae and the results were discussed.

**KEYWORDS:** The bioactivity *Culex quinquefasciatus*.

### INTRODUCTION

Mosquitoes transmit deadly diseases like malaria, filariasis, yellow fever, dengue fever, Japanese encephalitis; they contribute significantly to poverty and social debility in tropical and subtropical countries (Jang *et al.*, 2002). It is reported that every year, at least 600 million people suffer from one of these infections (WHO 1996). The most important and nuisance causing mosquitoes belong to the genera *Anopheles*, *Culex*, *Aedes*, *Mansonia*, *Haemagogus*, *Sabethes* and *Psorophora*. In India the various species of *Anopheles*, *Culex*, *Aedes* and *Mansonia* are vectors of severe diseases. Insecticides such as Organochlorides, Organophosphate, Carbamates and Pyrethroids have been used for vector control. This method of control has proved to be ineffective and undesirable because of development of insect resistance and environmental pollution. Now, there is considerable interest in

developing natural products as alternative to synthetic pesticides to control invertebrate pests of medical and economic importance. The use of plant essential oil for the pest and disease management has recently been reviewed (Beier 1998).

Essential oils are natural volatile substances found in a variety of plants. When isolated from plants, essential oils are not usually extracted as chemically pure substances, but consist of mixtures of many compounds. Essential oils were the first preservatives used by man, originally in their natural state within plant tissues and then as oils obtained by water distillation. Essential oils composed by isoprenoid compounds, mainly mono- and sesquiterpenes are the carriers of the smell found in the aromatic plants (Franzios *et al.*, 1997). Commercially, essential oils are used in four primary ways: as pharmaceuticals, as flavor enhancers in many food products, as odorants in fragrances, and as insecticides.

The search for alternative pesticides and control measures that pose no risk or posing minimal risk to human health and the environment is of great interest from the preventive medicine point of view (WHO 1999). Pyrethrin-based mosquito liquid formulations are widely used in many countries, especially in the house hold of rural population. Interest in botanical pesticides revived during recent years, because of the deleterious effects of synthetic insecticides, including lack of selectivity, impact on the environment and the emergence and spread of pest resistance. The naturally occurring pesticides appear to have a promising role in the development of future commercial pesticides for safety of the environment and public health (Bowers, 1992). *Cx. quinquefasciatus*, a domestic mosquito mainly found in urban areas, is a vector of human filariasis in India. My present investigation demonstrates that the efficacy of effective nine essential oils in killing the larval *Cx. quinquefasciatus*, under laboratory conditions.

## MATERIALS AND METHODS

### Essential oils

Essential oils were obtained from Tegraj & Co (P) Ltd, India (commercial producers of plant essential oils and aromatic substances) were used in this study.

### Test organism

The larval stages of *Cx. quinquefasciatus* culture was raised at  $28\pm 2^{\circ}\text{C}$ , 65-70% RH with a photo period of  $11\pm 0.5\text{h}$ . The reared larvae were fed on dog biscuits and yeast powder in the ratio of 3:1 in plastic tray (24x35x5cm). Adults were fed with 10% sucrose solution. In

addition to sucrose feeding, the female mosquitoes were also fed with live rat blood every four days as to produce a satisfactory number of eggs and then larvae for the testing. The fourth instar larvae were used for the bioassays.

### Bioassay Procedure

Larvicidal activity was determined by following the standard procedure (WHO, 1982). Effective oils were tested at five different concentrations viz, 100, 200, 300, 400 and 500 ppm. Twenty numbers of fourth instar larvae of *Cx. quinquefasciatus* were kept in a 500 ml glass beaker containing 249 ml of dechlorinated water and 1.0 ml of desired plant essential oil concentrations. four replicates for each concentration were maintained. In control 1.0 ml of Tween80 (0.01%) was added. Mortality of larvae was recorded after 24 hrs of treatment. While recording the percentage mortalities for each concentration, the moribund and dead larvae in five replicates were combined. It has been described that dead larvae are those that cannot be induced to move when they are probed with a needle in the siphon or the cervical region; moribund larvae are those incapable of rising to the surface. The lethal concentration (LC50) and LC90 values and 95% of lower and upper confidence limits were calculated by using probit analysis (Finney, 1971). Mortality was corrected by using Abbott formula (Abbott, 1925).

$$\text{Abbotts Percent Corrected Mortality} = \frac{\% \text{ MT} - \% \text{ MC}}{100 - \% \text{ MC}} \times 100$$

Where,

% CM = % Corrected Mortality;

% MT = % Mortality in treated;

% MC = % Mortality in control.

### Statistical analysis

Calculation of LC50 and statistical analysis LC50 values (the concentration at which 50% of the larvae were mortality) were calculated by probit analysis using software Statistical tools.

## RESULT AND DISCUSSION

The percent mortality values of 4th instar larvae treated with different concentration (ranging from 100 to 500 ppm) of the ten essential oils after 24 hrs treatment are represented Table 1 and Figure1 for *C. quinquefasciatus*. The LC50 and LC90 values between the concentration of essential oils and 24 hrs percent mortality of 4th instar larvae of *Cx. quinquefasciatus* are

represented in Table 2 respectively. Table 1 show the larvicidal activity of effective ten essential oils against the fourth instar *Cx. quinquefasciatus* larvae. All the ten oils showed concentration dependent larval mortality. Catnip oil was the most effective treatment. It shows maximum activity against fourth instar larvae of *Cx. quinquefasciatus*. The minimum Larvicidal activity was recorded in cornmint oil.

Table 2 and Figure 2 show the lethal concentration of effective oils against *Cx. quinquefasciatus* larvae. The lowest LC50 of 252.60 ppm was recorded in catnip oil. The LC90 for catnip oil was recorded as 480.51ppm. The highest LC50 and LC90 values of 426.80 and 689.29 ppm were recorded in cornmint oil. Our data clearly illustrate that selected effective oils were more potent larvicidal on *Cx. quinquefasciatus*. Our data also show a dose-dependent effect on mortality, with increasing concentrations of essential oil and its essential oils increasing mortality of the larvae.

The findings of the present investigation indicate that larvicidal properties of ten essential oils on *Cx. quinquefasciatus* reported in the present study, confirm their potential for control of the mosquito populations. These properties can be exploited to protect the environment of the user or the user from vectors of disease.

The larvicidal results are also comparable with earlier reports. The LC50 value of Clove oil towards *Cx. quinquefasciatus* has been observed to be 29.5 ppm as compared to LC50 37.7 of *Cymbopogon citratus* essential oil against *Cx. quinquefasciatus* (Pushpanathan *et al.*, 2006). More than 2000 plant species have been reported to possess chemicals with pest control properties (Ahmed *et al.*, 1984) and among them about 344 species of plants have been known to possess some degree of activity against the mosquitoes (Sukumar *et al.*, 1991).

Natural products of plant origin with insecticidal properties have been tried in the recent past for control of variety of insect pests and vectors (Wink, 1993). Though several plants from different families have been reported for insecticidal activity only a few botanicals like neem based insecticides have moved from the laboratory to field use, which might be due to the light and heat stability of neem compounds compared to synthetic insecticides (Green *et al.*, 1991). They found that the oil exhibited larvicidal activity and its LC50 against second, third and fourth instars *Cx. quinquefasciatus* larvae was 144.54, 165.70 and 184.18 ppm respectively. Carvalho *et al.* (2003) reported the larvicidal activity of the essential oil from *Lippia sidoides* Cham, against *Aedes aegypti* Linn. Thymol, an alkylated phenol derivative

and one of the major components of *Lippia sidoides* essential oil, was identified as the active principle responsible for the larvicidal action, which caused 100% larval mortality at the lowest concentration (0.017%).

Ai-Dakhi and Morsy (1999) have reported the larvicidal action of oils from the peel of lemon (*Citrus limon*), grape fruit (citrus paradise) and novel orange (*Citrus sinensis*) against fourth instar larvae of *Culex pipiens*. Choochote *et al.* (2007) stated that the efficacy of volatile oils can be improved by formulating them with vanillin. Sosan *et al.* (2001) reported larvicidal activities of essential oils of *Ocimum gratissium*, *Cymbopogon citratus* and *Ageratum conyzoides* against *Ae. aegypti* and achieved 100% mortality at 120, 200 and 300ppm concentrations respectively. Rahuman and Venkatesan (2008) reported that the petroleum ether extract of *Citrullus colocynthis*, methanol extracts of *Cannabis indica*, *Cannabis sativus*, *Momordica charantia* and acetone extract of *Trichosanthes anguina* against the larvae of *A. aegypti* (LC<sub>50</sub>=74.57, 309.46, 492.73, 199.14, and 554.20ppm) and against *Cx. quinquefasciatus* (LC<sub>50</sub>=88.24, 377.69, 623.80, 207.61, and 842.34 ppm), respectively.

Roman Pavela (2009) studied the larvicidal effects of essential oils from 22 aromatic plant species were tested for mortality of the mosquito larvae *Culex quinquefasciatus*. Lethal concentrations were determined for individual essential oils. Essential oils obtained from *Thymus vulgaris*, *Satureja hortensis* and *Thymus satureioides* plants showed, with LC<sub>50</sub> found lower than 50 g/ml (33, 36 and 44 g/ml, respectively). Madhu *et al.*, (2010) have reported that the efficacy of petroleum ether extract seemed to be effective with LC<sub>50</sub> and LC<sub>90</sub> values of 11.42 and 18.00ppm respectively. In contrast to that, the present result shows that the essential oils was five times more effective (Table 1 and 2). The literature revealed that only limited work is available on toxic nature of oils against *Cx. quinquefasciatus* larvae. Even though various researchers have undertaken essential oils, the identities of mosquito larvicidal compounds have not been determined.

In this regard, the present work is revealed the lethal efficiency of ten essential oils present in this paper. The results obtained in this work indicate that nine oils studied displayed activity against *Cx. quinquefasciatus* larvae, showing correlation between the experimental data and the uses reported for these plants in the literature.

**Table 1: Larvicidal efficacy of plant oils against *Cx. quinquefasciatus* larvae.**

| Plant oil  | Scientific name | Concentration (ppm) Mean larval mortality $\pm$ S.D. |               |                |                |                |
|------------|-----------------|--|---------------|----------------|----------------|----------------|
|            |                 | 100  | 200           | 300            | 400            | 500            |
| Catnip     | N. cataria      | 5.5 $\pm$ 0.5  | 9 $\pm$ 0.8   | 16.2 $\pm$ 0.5 | 17.5 $\pm$ 1.5 | 22.2 $\pm$ 0.9 |
| Thyme      | T. vulgaris     | 5.0 $\pm$ 0.5  | 8.2 $\pm$ 0.9 | 15.6 $\pm$ 0.3 | 16.2 $\pm$ 1.1 | 21.5 $\pm$ 0   |
| Basil      | O.basilicum     | 4.2 $\pm$ 0.5  | 7.6 $\pm$ 0.9 | 14 $\pm$ 0     | 15.5 $\pm$ 1.1 | 20 $\pm$ 0     |
| Patchouli  | P. cablin       | 3.2 $\pm$ 0.5  | 6.2 $\pm$ 0.5 | 10 $\pm$ 0.8   | 12.7 $\pm$ 0.9 | 17.7 $\pm$ 0.9 |
| Rosemary   | R. officinalis  | 2.7 $\pm$ 0.5  | 5.7 $\pm$ 0.5 | 9.7 $\pm$ 0.8  | 11.7 $\pm$ 1.2 | 17.5 $\pm$ 0.5 |
| Spearmint  | M. spicata      | 2.7 $\pm$ 0.5  | 4.7 $\pm$ 0.6 | 8.5 $\pm$ 0.6  | 10.7 $\pm$ 0.9 | 16 $\pm$ 1.4   |
| Lavender   | L.angustifolia  | 2.2 $\pm$ 0.5  | 4.5 $\pm$ 0.5 | 8.2 $\pm$ 1.9  | 9.4 $\pm$ 0.8  | 15.5 $\pm$ 0.8 |
| Marjoram   | T.mastichina    | 2.1 $\pm$ 0.5  | 4 $\pm$ 0     | 8.0 $\pm$ 0.5  | 8.7 $\pm$ 0.5  | 15.2 $\pm$ 0.5 |
| Clary sage | S.sclarea       | 1.2 $\pm$ 0.5  | 3.7 $\pm$ 0.7 | 7.5 $\pm$ 0.5  | 7.8 $\pm$ 0.8  | 14.5 $\pm$ 0.8 |
| Cornmint   | M. arvensis     | 1.1 $\pm$ 0.5  | 3.3 $\pm$ 0.1 | 6.5 $\pm$ 0.5  | 7.1 $\pm$ 0.8  | 14.1 $\pm$ 0.8 |
| Controle   |                 | 0.0 $\pm$ 0.0  |               |                |                |                |

**Table 2: Probit analysis of plant oils against *Cx. quinquefasciatus* larvae.**

| Plant oil  | Scientific name | LC 50  | LC90   |
|------------|-----------------|--------|--------|
| Catnip     | N. cataria      | 252.60 | 480.51 |
| Thyme      | T. vulgaris     | 270.53 | 516.40 |
| Basil      | O.basilicum     | 295.12 | 540.34 |
| Patchouli  | P. cablin       | 316.99 | 561.98 |
| Rosemary   | R. officinalis  | 338.19 | 580.37 |
| Spearmint  | M. spicata      | 345.77 | 600.81 |
| Lavender   | L.angustifolia  | 367.98 | 631.57 |
| Marjoram   | T.mastichina    | 392.66 | 657.02 |
| Clary sage | S.sclarea       | 412.14 | 673.12 |
| Cornmint   | M. arvensis     | 426.80 | 689.29 |

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