Vol. 11/2 (2011) 177-181

JOURNAL OF NATURAL REMEDIES

Smoke Toxic Effect of *Vitex Negundo (L)* for The Control of Malarial Vector, *Anopheles Stephensi* Liston.

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Abstract

Malaria occupies the centre stage of vector-borne diseases in the world. Synthetic insecticides remain as the first line of defence in public health protection against insect disease vector. In view of growing concern about safety of chemical based repelents, interest in reviewed in smokes extracted from plants as repellent for mosquitoes. Smoke toxicity from the botanicals (Various parts of *vitex negundo* are highly effective, safe and ecologically acceptable will give widespread relief from some of the more serious diseases. In the present study it was cleared that the toxicity of smoke from the leaves *of Vitex negundo* against *A.stephensi* were highly effective in controlling their population.

Key words : Smoke toxicity, *Anopheles stephensi*, *Vitex negundo*.

1. Introduction

Mosquito of the genus *Anopheles* transmit the four parasites *(plasmodium flaciparum p.vivax ,P. malaria and P.ovale)* that cause human malaria which breed in the *Anopheles* intestine. The parasites kill red blood cells and cripple the liver. Today 500 million people are exposed to endemic malaria and it is estimated to cause half a million deaths annually, one million of which are children. The deadly diseases carried by these insects and the annoyance they cause is likely to have encouraged the discovery of methods of personal protection.(Moore *et al* .,2002)

Mosquitoes are well known for the public health importance. Though chemical insecticides are widely used in their control, the development of resistance among the mosquitoes and the harmful effects of these chemicals upon the environment have caused concern among the

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scientists and that has created a need to find easily degradable alternative insecticides. Botanical insecticides are highly effective, safe and ecologically acceptable. The common trend of the past two decades towards reducing reliance on synthetic insecticides for control of insect pest in agriculture, forestry and human health has renewed world interest towards botanicals.

Plants contain many chemicals, which are important in their defense against insects.

Vitex negundo L. (Verbenaceae) showed a promising pesticidal activity against insect and it is used predominantly for its pest properties. Infusion or suspensions of *Vitex* species have been used as natural insecticides (Hebbalkar *et al.*, 1992). *Vitex negundo* leaves, which contain terpinines, terpeneol and sesquiterpene alcohols. Almost all the parts are employed, but the leaves and roots are important as drugs. Analgeric and anti-inflammatory actions of *V.negundo* seeds and fruits have been reveiewed thoroughly and dried leaves powder of *V.negundo* showed anti-arthritic activity in rats (Telang *et al.*, 1999). In view of the above fact, an attempt has been made to evaluate effect of *Vitex negundo* on the smoke toxic effect of the malarial vector, *Anopheles stephensi.*

2. Materials and Methods

2.1 Collection of Plant Materials

Vitex negundo L. (Verbenaceae) was collected from Maruthamalai hills, Western ghats, Southern India, Coimbatore. The plants were authentified at BSI (Botanical Survey of India) and the specimens were deposited at Zoology Department, Vivekanandha College of Arts & Science, Namakkal.

2.2 Smoke Toxicity Test

Vitex negundo parts (leaves, stem and roots) were used for smoke toxicity assay. The mosquito coils were prepared by following method of Saini *et al.* (1986) with minor modifications by using 4 grams from the powder sample plant powdered sample considered as active ingredient two grams of saw-dust as binding material and two grams of coconut shell charcoal powder as burning material. All the three were thoroughly mixed with distilled water to form a semisolid paste. Mosquito coils (0.6 cm thickness) were prepared manually from the semisolid paste and were dried in shade. The control coils were prepared without plant ingredient.

The experiments were conducted in glass chamber measuring 140x120x60 cm. A window measuring 60x30 cm was situated at mid bottom of one side of the chamber. Three or four days old blood starved hundred adult female mosquitoes, fed with sucrose solution, were released into the chamber. A belly shaven Rabbit was kept tied inside the cage in immobilized condition. The experimental chamber was tightly closed. The experiment was repeated five times on five separate days including control using mosquitoes of same age groups. The data were pooled and average values were subsequently used for calculations. Control was maintained in two sets. One set was run with coil lacking the active ingredient of plant powder (control 1) another one is Mortein coil which is used for calculated in terms of percentage of unfed mosquitoes due to treatment.

$$\text{Percentage Reduction} = \frac{\text{Number of unfed mosquitoes in treatment} - \text{Number of unfed mosquito in control}}{\text{Number of mosquitoes treated}} \times 100$$

The alive mosquitoes fed with blood meal were reared in a mosquito cage, measuring 30 x 30 x 15 cm. The top and bottom of the cage were fit with glass and all other sides were covered with muslin cloth. Water soaked rasins and 5% sucrose solution soaked in cotton balls were kept as food inside the cage. Water containing powdered yeast and dog biscuits were also kept inside the cage in a glass bowl to collect eggs. The eggs from the cage were collected daily till all the mosquitoes died. 50 to 100 eggs were allowed to hatch in each plastic tray measuring 30 x 25 x 6 cm, containing about 2.5 litres of unchlorinated tap water. The larvae hatched from the eggs were fed with a mixture of dog biscuits and yeast powder in the ratio of 2:1 and water in the tray was changed daily. The number of larvae hatched was counted at second instar stage. The reduction in the population from the smoke treated mosquitoes was calculated using the formula.

$$\text{Population reduction (\%)} \frac{\text{Number of larvae hatched in control} - \text{Number of larvae hatched in treated.}}{\text{Number of larvae hatched in control}} \text{ X } 100$$

2.3 Fecundity Studies

The fecundity experiments were conducted by taking an equal number of male and female *Anopheles stephensi* which had emerged from the control and treated sets. They were netted in the cages of 30 x 30 cms dimension individually of each concentration. Three days after the blood meal eggs were collected daily from the small plastic bowls containing water kept in ovitraps in the cages. The fecundity was calculated by the number of eggs layed in ovitrap divided by number of females let to mate (the death of the adults in the experiment was also considered).

2.4 Egg Hatchability Test

The eggs were placed in the enamel tray for hatching. The percentage of hatchability was calculated as number experimented by number of eggs hatched.

$$\text{Hatchability (\%)} \frac{\text{Number of eggs hatched}}{\text{Number of eggs tested}} \text{ X } 100$$

2.5 Statistical Analysis

All data were subjected to analysis of variance and the treatment means were separated by Duncans Multiple Range test. (Duncan) 1995.

Table 1: Smoke toxic effect of *Vitex negundo* parts ensured population of *Anopheles stephensi*

***Vitex negundo* parts used**	**No.of mosquitoes tested**	**Total No. of eggs**	**Total No. of larve hatched fro the egg**	**% of reduction in population over control 1**
Leaf	50	400[b]	125[b]	92.4[a]
Root	50	853[a]	358[c]	80.8[ab]
Stem	50	984[a]	534[b]	71.3[b]
Control I*	50	1063[b]	780[a]	90.8[ab]
Control II*	50	245[b]	156[b]	00[c]

Control I* - Coil lacking the active ingredient of plant powder
Control II* - Mortein Coil
Within a column means followed by the same letter(s) are not significantly different at 5% level by DMRT

Table 2 : Smoke toxic effect of *Vitex negundo* parts against biting activity of *Anopheles stephensi*

***Vitex negundo* parts used**	**No.of mosquitoes tested**	**Fed mosquitoes**	**Unfed Mosquitoes**			**% Unfed over control 1**
			Alive	**Dead**	**Total**	
Leaf	100	21[be]	46[ab]	33[ab]	79[ab]	60[a]
Root	100	23[b]	50[d]	27[c]	77[b]	48[b]
Stem	100	26[b]	53[a]	21[d]	74[b]	45[b]
Control I*	100	71[a]	29[c]	0[c]	29[c]	0[c]
Control II*	100	16[c]	42[b]	42[a]	84[a]	0[c]

Control I* - Coil lacking the active ingredient of plant powder
Control II* - Mortein Coil
Within a column means followed by the same letter(s) are not significantly different at 5% level by DMRT

3. Results and Discussion

Smoke is a common method of repelling biting insects used throughout the world. Fresh or dried plants are frequently added to fires to enhance the repellent properties of the smoke. Other methods are hanging the plants around the house or sprinkling leaves on the floor. Mosquito coils made from dried plants and combustible material such as saw dust are also a cheap and often an effective method of repelling mosquitoes. In Java today, the same in cense used in ceremonies to honour ancestors is also used on a daily basis to repel mosquitoes (Sangat-Roemanty, 199).

In the present study also smoke emerged from *Vitex negundo* parts have considerably affected the adult mosquitoes and brought out considerable mortality and also treated individual layed minimum number of eggs (Table 1).

Vitex negundo leaves are used as a mosquito repellent in India (Parrotta, 2001). In Maharashtra state, tribal people use smoke from the leaves of this plant at night to protect them from mosquito bites. Hebbalkar *et al.*, (1992) this study was further supporting the present work. Traditionally smoke from burning dried plant leaves such as *Vitex negundo,* neem, pongamia seed kernel powder and Acorus *Calamus* rhizome powder have been used for domestic protection, in Malaysia, (Janten *et al.*, 1999) have prepared nineteen mosquito coil formulations, each containing a different plant and investigated for their knockdown and 24 hrs mortality values against *Aedes aegypti*. Mosquito coil of the leaves of *Cymbopogon mardus* and *Aloevera* and seed kernel; of *Azadirachta indica* incorporation with D-trans-allethrin significantly increased their efficiency against mosquitoes in terms of knocking down and killing effects. Therefore, use of these plant materials as organic filters in and reduced health hazards. In view of growing concern about safety of chemical based repellents, interest in reviewed in oils extracted from plants as repellent for mosquitoes. Traditional repellents not only provide protection against mosquito bites but also prevent malarial transmission. In the present study the smoke from the *Vitex negundo* also had considerable smoke repellency. Hence, these plant parts can be preferably employed for the development of mosquito coil in future.

4. Acknowledgement

The authos is thankful to **Dr. K. MURUGAN,** Reader, Department of Zoology, Bharathiyar University, Coimbatore, Tamilnadu, India. For his suggestions, unstained support and encouragement during the study period.

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