

## EFFECT OF PLANT EXTRACTS ON MOSQUITO POPULATION

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### ABSTRACT

*Mosquito population increases exponentially that is vast trouble for many nations because they transmit diseases such as malaria, dengue, filaria, yellow fever, chikungunya, Japanese encephalitis, Lyme disease and epidemic poly-arthritis. There are about 3500 species of mosquitoes present in tropical or sub-tropical regions. World Health Organisation has declared mosquito as "Public enemy number one". Use of chemical pesticides to manage mosquito menace has proved to be hazardous for human health and environmental well-being. Many plant extracts have been found to be effective against mosquito population (larval, adult as well as eggs). The present study reviews effect of various medicinal plant extract to manage mosquito population. Different herbal plant extracts dissolved in oil or other solutions has been used against mosquito larvae effectively.*

**Keywords:** Mosquito; Medicinal Plant; Plant Extract; *Culex*; *Anopheles*; *Aedes*

### INTRODUCTION

Mosquitoes belonging to Order Diptera of Class Insecta of Arthropods are vectors of many diseases. Mosquitoes are responsible for transmission of many viral, bacterial and protozoans' diseases.<sup>[1]</sup> Population of mosquito-increases exponentially causing major trouble for many countries by spreading diseases like filaria, Japanese encephalitis, Lyme disease, malaria, dengue, yellow fever, chikungunya and epidemic poly-arthritis.<sup>[2]</sup> There are about 3500 species of mosquitoes present in tropical and sub-tropical region.<sup>[3]</sup> WHO has declared mosquito as "Public enemy number one" because they transmit most dreadful diseases.<sup>[4]</sup> Out of all mosquito species, *Aedes*, *Anopheles* and *Culex* are major vectors. *Aedes* is responsible for spreading dengue, chikungunya and yellow fever. *Anopheles* spreads mainly filariasis and malaria while *Culex* acts as vector for Japanese encephalitis and chikungunya.<sup>[5]</sup> The disease dengue, chikungunya which is caused by *Aedes* vector affects the 2.5 million people every year.<sup>[6]</sup> The breeding places of *Aedes* mosquito vectors are major cause of increasing dengue fever. This is because of greater urbanization and growth of population.<sup>[7]</sup> Malaria caused by

*Anopheles* vector increasing 200 million-450 million infections annually worldwide.<sup>[8]</sup> *Culex* is the vector of Japanese encephalitis which happens in children with malnutrition. Major cause of encephalopathy is encephalitis.<sup>[9]</sup> There are several methods available to protect us from mosquitoes biting like physical control methods such as clearing the stagnant water and chemical control methods such as use of repellents to kill mosquitoes. The usual commercially available synthetic chemical mosquito repellent comprises Deet (N, N-diethyl-3-methyl benzamide) which has depicted excellent protection against mosquitoes.<sup>[10]</sup> Physical control methods are tiresome in use and chemicals used in chemical control methods have caused major environmental and health problems. However there are natural products that have proved to be effective against mosquito adult and larval population. These natural plant repellents are the best method for protecting against the mosquito vectors because they are safe, economical and eco-friendly. Scientific researchers have searched and studied plant products like plant extracts, essential oils and powder.<sup>[11]</sup> Traditional repellent like smoke are used in the rural areas. Several

plant crude extracts and compounds from many different families and plants have been screened as potent larvicides.<sup>[12]</sup> Researchers have established the effectiveness of plant derived secondary compounds, like steroids,<sup>[13]</sup> saponin,<sup>[14]</sup> essential oil,<sup>[15]</sup> isoflavonoids,<sup>[16]</sup> alkaloids and tannins<sup>[17]</sup> against mosquito larvae. Essential oils and plant compounds are serving as alternative root of mosquito repellent agents.<sup>[18]</sup> Taking in view the upsurge in popularity of these plant based natural products against mosquito population we tried to summarise the efficacy of these products against larval and adult population of three common mosquito vectors viz. *Aedes*, *Anopheles* and *Culex* in present review.

### EFFECT OF MEDICINAL PLANT EXTRACTS ON *Aedes* POPULATION

*Aedes aegypti* and *Aedes albopictus* is the major vector for dengue and chikungunya diseases. In January and August of 2010, high dengue alert was there across India as well as in Malaysia, referable to the total deaths management because larvae were found in specific areas and more than 103 people were critically affected.<sup>[19]</sup> Chikungunya and Dengue Fever (DF) / Dengue Hemorrhagic Fever (DHF) were described from different part of India.<sup>[20, 21]</sup> There are several methods for holding *Aedes* mosquito vector population under check. Components of plant may be possible substitute for chemical pesticide and several plants were found to have an effect on growth, behaviour and development of mosquito vector.<sup>[5]</sup> *Azadirachta siamensis* (Thai neem) shows the better efficacy against the *Aedes aegypti* larvae. It is found in Thailand and may be different species than the *Azadirachta indica* (Indian neem).<sup>[22]</sup> To measure the efficacy of *A. siamensis* oil emulsion and an alginate bead of *A. siamensis* oil against the IV<sup>th</sup> instar larvae of *Aedes aegypti* were tested. Oil emulsion demonstrated greater larvicidal activity than the alginate bead oil preparation at 12 to 60 hours post-exposure ( $p$ -value  $< 0.01$ ). Neem oil shows 100% mortality at 48 hours and alginate bead oil formulation shows 98% mortality at 84 hours and shows 100% mortality at 96 hours against the fourth instar larvae of *Ae. aegypti*. Different parts of *A. indica* (Neem) bark, leaf, root and stem also demonstrated a good effect on mosquito vector

*Ae. aegypti* larvae. The secondary compounds of Neem have demonstrated insecticidal properties.<sup>[23]</sup> Different parts of Neem such as bark, leaf, root and seed have been extracted with different solvent viz. acetone, chloroform and ethanol. 1 mL of DMSO (*dimethyl sulphoxide*) was applied to solubilise extracts in water. Data were measured and LC<sub>50</sub> and LC<sub>90</sub> values were studied. The crude extracts were deviated and the LC<sub>50</sub> and LC<sub>90</sub> value limits from 50 to 837.5 ppm and 94 to 950 ppm. Experiment demonstrated that leaf acetone extract and root chloroform extract were more toxic and cause 100% mortality in 24 hours at concentration of 1000 ppm. Other extracts demonstrated 100% mortality at 48 hours. The fruits of *Sapindusemarginatus* and *Holothuria atra* also used against the mosquito vector *Ae. aegypti*. *Sapindusemarginatus* is a medicinal plant which is widely circulated in the dry-zone of Sri Lanka. *H. atra* is having medicinal values.<sup>[24]</sup> The crude extracts of *S. emarginatus* (a medicinal plant) and the skin of *H. atra* have been used against *Ae. aegypti*. Bioassay investigation was expressed with crude extracts of fruit *S. emarginatus* that were extracted with the solvent ethanol disclosed LC<sub>50</sub> and LC<sub>90</sub> values of 92.9 and 152.6 ppm. The skin extracts of *H. atra* were extracted with the solvent methanol disclosed LC<sub>50</sub> and LC<sub>90</sub> values of 68.82 and 180.76 ppm. Both the extracts were positive because the presence of Saponin. The investigation on plant *Pongamiapinnata* and *A. indica* seed oil against vector was carried out.<sup>[25]</sup> Combination of seed oil repellent also used on mosquito *Ae. aegypti*. The repellent were developed into 3 group's seed oils with petroleum ether, combination of seed oil, seed oil with carrier oil (Coconut oil, Olive oil and Mustard oil). Their combination ratios were 1:1, 2:1 1:2 at the concentration of 1 and 5%. Results demonstrated that 5% formulation combination of oils *P. pinnata* and *A. Indica* at the ratio of 1:1, the protection time was 300 and biting percentage rate of 6. At 5% concentration of *P. pinnata* and *A. indica* seed oil in mustard oil base provided 86.36% and 85% protection time of 230 and 240 minute respectively. The study supports that *P. pinnata* and *A. indica* having a good repellent potential. Different medicinal plant species aqueous extracts were used for the prevention of mosquito

*Ae.albopictus*. The plant species and the part used were *Ocimumgratissimum* Whole plant, *Adhatodavasica* Leaf, *Averrhoabilimbi* Fruit, *Aeglemarmelos* Leaf, *Careyaarborea* Fruit, *Commelinadiffusa* Leaf, *Lantana camara* Flower, *Phyllanthusemblica* Dry seed, *Scorparisdulis* Whole part, *Terminaliachebula* Dry seed. The effect of respective plant extracts on mosquito larvae displayed to 96 hrs. Out of 10 plants *O.gratissimum* (whole plant) and *T.chebula* (Dry seed) found 100% mortality. *P.emblica* (Dry seed), *L.camara*(Flower) and *A.marmelos* (Leaf) expressed 95, 90 and 55% larvicidal properties. Leaf extracts of *Adhatodavasica*, *Commelinadiffusa* fruit extracts of *Averrhoabilimbi*, *Careyaarborea* and whole plant extracts of *Scorparisdulis* exhibited no larvicidal efficacy.<sup>[26]</sup>

### EFFECT OF MEDICINAL PLANT EXTRACTS ON ANOPHELES POPULATION<sup>1</sup>

A single most important disease inflicted by mosquito vector is malaria<sup>[27,28]</sup> and it is a major cause of infant mortality rate.<sup>[29, 30]</sup> The biggest challenge in battle against this pandemic is resistance of the parasite to anti-malarial drugs and their toxicity to human health and high treatment cost. Recent WHO calculates are that there are 300-500 million cases per year.<sup>[31, 32]</sup> *Anophelesgambiae* are very notorious vector of the parasite.<sup>[29, 30]</sup> There is no vaccine to control infection caused by mosquito vector *An.gambiae*. To avoid this problem vector control is a best alternative. A large number of plant derivatives have established to be effective against mosquitoes.<sup>[33]</sup> A recent researcher has concentrated on plant product alternative. There are several varieties of plants present all over the world that can be used against malarial vector *Anopheles*. *Clausenaanisata* is a plant which demonstrates bioactivity against various diseases and inhibits the larvicidal activity. *C.anisata* belongs to family Rutaceae and originates in tropical region. The extracts of plant *C. anisata* were extracted with the solvent Hexane, Ethyl acetate, Chloroform, Acetone and Methanol and oils were received by hydro-distillation in qualified Clevenger-type of newly whole plant. The oils of *C.anisata* demonstrate markedly increased activity against III<sup>rd</sup> instar larvae of

*An.gambiae*. In this study the plant *C.anisata* crude ethyl acetate gave higher LC<sub>50</sub> value of 2095.46 mg L<sup>-1</sup> and LC<sub>99</sub> value of 4438.75 mg L<sup>-1</sup>. The *C.anisata* oils gave LC<sub>50</sub> value of 76.96 mg L<sup>-1</sup> and LC<sub>99</sub> value of 256.80 mg L<sup>-1</sup>. These essential oils (volatile) and crude extracts (Non-volatile) compounds of plant *C.anisata* are helpful in malaria and mosquito management programmes. Different types of oils extracted from plants act as potent repellent for mosquitoes.<sup>[34]</sup> The repellent action of plant *Cymbopogan martini martini* *Stapfvarsofia* oil against mosquito vector *Anopheles sunndaicus* was studied. Pure oil of *varsofia* was received from Plant Genetic Resource Division, Indian Agriculture Research Institute, New Delhi. *C.martini* oil supplies 98.7 % protection in indoor and outdoor protection gives 96.52% against the vector *An.sunndaicus*. The plant extracts of *Artemisia parviflora* are very effective against vector *Anopheles stephensi*. This plant prevents malaria in tribal village. The extracts of plant are extracted with the methanol solvent. The mortality rate of larvae and pupae was recorded at LC<sub>50</sub> and LC<sub>90</sub> values. The *A.parviflora* plant is biologically active which may be due to the presence of dynamic compounds like  $\alpha$ -Caryophyllene, Camphor, germacrene D, Artemisia ketone, 1-8 Cineole, D-Copaene and Sabinyl acetate. These active compounds are poisonous against the mosquito. The LC<sub>50</sub> measure for first instar larvae is 45.61 and is modified in the fourth instar larvae as 59.60. Eucalyptus based repellent (PMD) *p*-menthane-3, 8 diol was evaluated in the field comparison with Deet (N, N-diethyl-3-methyl benzamide). According to Trigg and Hill (1996)<sup>[35]</sup> this compound is nearly as effective as Deet against *An.gambiae* Gills. But in their study field comparison of PMD against two malaria mosquito vectors, *An.funestus* and *An.gambiae* was investigated. Three formulation of PMD (50% AI), a pump spray, a stick, and a gel were assessed. These repellent were applied to the legs and feet at doses chosen in this experiment and they gave complete security from biting for between 6 and 7.75 hour and it depends on their formulation type. Neem products are also very helpful for controlling Malaria vector, *An.stephensi*. Larvicidal, Repellent, and Smoke

toxicity effect of neem products is well demonstrated. Each component of neem tree has insecticidal properties.<sup>[36, 37, 38]</sup> Neem seeds comprising 99 biologically active components like azadirachtin, nimbin, nimbolides and nimbidin. Some these derived products have ovicidal activity, antifeedancy, fecundity suppression following insect growth regulation and repellency against vector insect.<sup>[39, 40]</sup> Six neemlemonoids (purity >99%), such as azadirachtin, salannin, gedunin, deacetylgedunin, 17-hydroxyazadiradione and deacetylnimbin can be chemically extracted. Larvicidal bio-assays were carried in the laboratory with neemlemonoids at the lethal concentration of LC<sub>50</sub> and LC<sub>90</sub>. Repellency test was acted by human volunteers by using neem oil. Smoke toxicity was function on adult *Anopheles* female mosquito in laboratory by using neem kernels. The LC<sub>50</sub> and LC<sub>90</sub> values of *Azadirachtin* treatment at 0.05, 1.0 and 1.5 ppm concentration were 0.299% and 1.061%. After the neem oil treatment at concentration 0.05, 1.0 and 1.5 ppm LC<sub>50</sub> and LC<sub>90</sub> values were 0.503 and 1.324 respectively. *Salannin* was 0.438% and 1.420%, *Gedunin* was 0.558% and 1.568%, *Deacetylgedunin* was 0.461% and 1.384, *17-Hydroxyazadiradione* was 0.387% and 1.553%, *Deacetylnimbin* was 0.594% and 1.542% respectively. The repellent activity of neem products was tested at different concentrations (0.2 %, 0.4 %, and 0.6 %). The products of neem oils are *Azadirachtin*, *Salannin*, *17-Hydroxyazadiradione*, *Deacetylgedunin*, *Deacetylnimbin* and *Gedunin*. Neem oil demonstrated higher repellency (<300 minutes at 0.6 concentration) complied by *Gedunin* which expressed less activity (<200 minutes at 0.6 concentration). For Smoke repellency test leaves and pods were applied. When smoke emerged from leaves pods had possible powerful effect and percentage of repellency on leaf was 59% and pod was 53% and commercial oil shows positive control of 65% repellency.<sup>[41]</sup>

### **EFFECT OF MEDICINAL PLANT EXTRACTS ON CULEX POPULATION**

Out of various mosquito species belonging to order Diptera, family Culicidae three important vector species are *Culex pipiens*, *Culex quinquefasciatus*, *Culex tritaeniorhynchus*. These three

spread *Filariasis*, *Japanese encephalitis* and *Dengue haemorrhagic fever*. There are several varieties of medicinal plants which can be used as repellents for *Culex* species like *Salvia officinalis*,<sup>[42]</sup> *Caesalpinia pulcherrima*,<sup>[43]</sup> *Tephrosia purpurea*,<sup>[44]</sup> *Menthapulegium*,<sup>[45]</sup> *Oputiadillenii*,<sup>[46]</sup> *Centella asiatica*,<sup>[47]</sup> *Eucalyptus*, *Azadirachtaindica*,<sup>[48]</sup> *Andrographis paniculata*,<sup>[49]</sup> *Hyptis suaveolens*<sup>[50]</sup> and *Calotropis gigantea*.<sup>[51]</sup> The screening of plant *S. officinalis* found the phytochemicals like sterols, tannins, flavonoids, gallic tannins, catechic tannins, mucilages and glycosides components. The plant *S. officinalis* extracts has been extracted with Ethanol solution. The ethanolic solution was found to be very effective with a LC<sub>50</sub> value of 287 ppm and 487 ppm for LC<sub>90</sub> against *C. pipiens* larvae. Larval mortality of *C. pipiens* was assessed at 600 ppm. It gives 100% mortality at the concentration of 0.5. The testing of *C. pulcherrima* obtained phytochemicals from the whole parts and extracted by different types of solvents like methanol, aqueous, benzene, chloroform and acetone. The testing of this plant against the mosquito vectors like *C. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus* gave fruitful results. The highest larvicidal activity was detected in benzene extract with LC<sub>50</sub> and LC<sub>90</sub> values were 150.47, 135.24 and 119.27 ppm and 282.57, 261.55 and 243.37 ppm respectively.<sup>[43]</sup> The testing of *T. purpurea* plant has been utilized in popular medicine to cover many diseases. This plant is having activities like anticancer, antioxidant, antimicrobial, anti-plasmodial, larvicidal. The extracts of plant and stem of *T. purpurea* were extracted with methanol solvent and tested against the Filarial vector *C. quinquefasciatus*. The efficacy of stem extract against III<sup>rd</sup> instar larvae appeared less effective with LC<sub>50</sub> 2348 ppm than leaves extract with LC<sub>50</sub> 58.3 ppm. Leaves extracts demonstrated a greatest reduction effect on adult emergence, fecundity and fertility. Moreover, morphological deformities on pupal and adult intermediate were detected after treatment with leaves extract.<sup>[52]</sup> *M. pulegium* (Lamiaceae) are herbaceous plants and perennial. These plants are aromatic and are used in traditional medicine, confectionery, culinary preparations, cosmetics and perfumery.<sup>[53]</sup> After the screening of this plant the phytochemicals were

obtained. The percent yield of the hydro-ethanolic extract from the aerial parts (leaves, stem and roots) of *Menthapulegium* was  $8.7 \pm 0.2$ . The highest larvicidal activities was detected in hydro-ethanolic extract of the plant *M.pulegium* against vector *C.pipiens* with the value of  $LC_{50}$  0.38 (0.35-0.49) and the value of  $LC_{90}$  0.64 (0.50-0.69) mg/ml respectively.<sup>[54]</sup> Plant based insect repellent *Eucalyptus* and *A.indica* seed oil were screened against the filarial mosquito *C.quinquefasciatus*. Eucalyptus oil allows longer protection other than other plant based repellents.<sup>[55]</sup> *A.indica* seed oil gives 90.26% and 88.83% protection and *Eucalyptus* seed oil gives 93.37% and 92.04% at the concentration of 50% and 100% for 4 hour against *Culexquinquefasciatus*.<sup>[48]</sup> The extracts of *Oputiadilleni* stem (modified as cladode) extracted with acetone and checked the mortality of IV<sup>th</sup> instar larvae of *Ae.aegypti* and *C.quinquefasciatus*. The 24 hours  $LC_{50}$  values of stem extracts of *O.dilleni* were estimated 10.95 ppm for *C.quinquefasciatus* and 21.92 ppm for *Ae.aegypti*.<sup>[46]</sup> After the examining of *A.paniculata* plant leaves extract against *C.quinquefasciatus* the most effective phytochemical is andrographolide (2.39%) which is present in *A.paniculata* in highest amount. The plant extract is extracted with ethanol and highest mortality rate of 20% and 73.3% was shown in 300 ppm at 3 and 24 hours respectively. The standardized trend was detected in toxicity of acetone extract. The highest mortality rate of 50% and 83.3% was observed in 300 ppm in 3 and 24 hours respectively.<sup>[49]</sup> *H.suaveolens* was assayed for mosquito larvicidal vector *C.quinquefasciatus*. It is a medicinal herb commonly known as 'American mint'. The aerial part extracts of *H.suaveolens* were extracted with petroleum ether, chloroform and acetone. This test revealed that acetone extract demonstrated highest larvicidal activity and  $LC_{50}$  value was 485.61, petroleum ether and chloroform extract showed 493.44 and 625.97 mg/L after 24 hours. In case of 48 hours petroleum ether extract ( $LC_{50}$  298.76 mg/L) which expressed greatest larvicidal activity complied by acetone ( $LC_{50}$  344.03 mg/L) and chloroform ( $LC_{50}$  429.50 mg/L).<sup>[50]</sup> *C.gigantean* (Apocynaceae) flower extracts were evaluated against filarial vector *C.quinquefasciatus*. The extracts were extracted with

petroleum ether, chloroform and ethanol. The highest repellent efficacy was shown with ethanol flower extract which allowed 100% protection in 60 minutes at a higher concentration of 5.0 mg/cm<sup>2</sup>. Chloroform flower extracts gives 95.66% at the highest concentration of 5.0 mg/cm<sup>2</sup>. Petroleum ether flower extracts provided 100% at the highest concentration of 5.0 mg/cm<sup>2</sup>.<sup>[51]</sup> Shad and Andrew (2017)<sup>[56]</sup> studied larvicidal efficacy of ethanolic extracts of *Annona-squamosa* (Annonaceae) over the filarial vector, *C.quinquefasciatus* Say and found it to be effective.

## CONCLUSION

In comparison to synthetic chemical compounds plant based products are safer. Continuous application of chemical pesticides cause insecticide resistance in mosquito's and is harmful to non-target organisms and is dangerous for environment. Whereas plant based repellents do not pose risks of toxicity to human beings and domestic animals. In many parts of the world plant based repellents are used. Due to less effect on environment and low budget herbal plant repellents attracted great attention of community. Toxicity assay of the herbal plants need to be done before ascertaining their role in establishment. Larvicidal activities of plant extract change according to the species of plants. Plants can be substitute reference for chemical mosquito larvicides because they are having a possible source of bioactive chemicals which are eco-friendly by nature.

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