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Mosquito vector management knowledge, attitude, practices and future of user & environment friendly new generation botanical Mosquitocide formulations: A review

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Abstract

Every step taken to control the mosquitoes has a cumulative effect and contributes immensely to control mosquito borne diseases. The eggs developing within the female mosquito need human blood for nourishment and so the female mosquito bites humans. Protection of humans against mosquito bite by using bed nets and mosquito repellent. By preventing water logging and destroying unwanted water collections sources of egg laying can be denied and breeding of mosquitoes can be prevented. Most of insecticides have effects on the human beings as well as the environment and other life forms. The real benefits of botanical insecticides can be best realized in developing countries, where people may not be able to afford synthetic insecticides, and the traditional use of plants and plant derivatives for protection of mosquitoes is long established. Recently, attention has been paid to use user & environment friendly new generation botanical mosquitocide formulations for protection against mosquitoes.

Keywords: Mosquito, vector, management, botanical mosquitocide, formulations

1. Introduction

Mosquitoes are among the most disturbing blood sucking insects afflicting human beings. Several mosquito species belonging to genera *Anopheles*, *Culex* and *Aedes* are vectors for the pathogens of various diseases like Dengue fever, Malaria, Yellow fever, Japanese Encephalitis and several other infections. Mosquitoes alone transmit diseases to more than 700 million people and over one million deaths are reported annually across the globe. Malaria which is caused by *Plasmodium* parasites transmitted through the bites of female *Anopheles* mosquitoes continues to impart a major disease burden on infants and young children in endemic regions [1]. The *Aedes aegypti* mosquito which spreads Dengue fever is responsible for more than 100 million infections worldwide every year, leading to thousands of deaths and more than 2.5 billion people or over 40% of the world's population are now at risk of Dengue.

Having a mosquito infestation in close proximity to you and your family is an extremely dangerous situation and can be fatal. The risk of mosquito-borne diseases is especially high in humid areas. In the more arid regions of India, mosquito infestation commonly peak during and after the annual monsoon rainfalls. In the more tropic and humid regions, the peak of mosquito activity normally falls into the summers, when high temperatures fasten the reproductive cycle of mosquitoes. Therefore, the control of mosquitoes is an important public health concern around the world.

The use of synthetic adulticides for mosquito control is not advisable owing to environmental hazards and prohibitive cost. Personal protection measures such as using mosquito nets or synthetic repellents are not a practical solution in view of the socio-economic background of the community and poor sustainability. The other approach is to eradicate immatures of mosquitoes or to make the habitats unsuitable for mosquito breeding through environmental modification/ manipulation. The latter may be considered as an environment friendly approach but cannot be applicable to all the habitats in all situations. Though such source reduction is a permanent solution for the prevention of mosquito breeding, it needs an engineering solution, which involves huge expenditure.

Therefore, botanical insecticidal operation considered to be a user & environment friendly alternative measure of choice has received adequate attention in recent times to keep people safe from mosquitoes.

2. Types of mosquito problem

There are thousands of different species of mosquito, but the most common mosquito problems in India are caused by-

2.1 Anopheles Mosquito

The development from egg to adult takes only between 6-10 days. The female lay between 60-150 eggs after a blood meal. *Anopheles* mosquitoes commonly bite at night and rest indoors and outdoors during the day. *Anopheles* mosquitoes are transmitters of Malaria.



Fig. 1: *Anopheles* Mosquito

2.2 Culex Mosquito:

Culex mosquitoes are transmitters of Japanese B Encephalitis, a very dangerous type of brain fever. They bite at night and rest before and after blood meals. *Culex* breeds in polluted stagnant water, and is a major pest problem in urban areas and metropolises of India, where it breeds in drains.



Fig. 2: *Culex* Mosquito

2.3 Aedes Mosquito

Unlike above mentioned species, *Aedes* mosquitoes are active during the day and they breed in clean water in man-made containers, such as flower vases, water accumulation in tyres and cans. In India, *Aedes* mosquitoes are responsible for Chikungunya and Dengue. Their development from egg to mosquito is quite rapid (6-8 days).



Fig. 3: *Aedes* Mosquito

3. Vector surveillance

3.1 Larval surveys:

For larval surveys, the basic sampling unit is the house or premise, which is systematically searched for water holding containers. Containers are examined for the presence of mosquito larvae and pupae. Depending on the objective of the survey, the search may be terminated as soon as larvae are found, or it may be continued until all containers have been examined. The collection of specimens for laboratory examination is necessary to confirm the species.

3.2 Adult Surveys

3.2.1 Landing/biting collection

Landing/biting collections of humans is a sensitive means of detecting low level infestations, but are very labour intensive. Because adult males have low dispersal rates, their presence can be a reliable indicator of clear proximity to hidden larvae habitats. It is usually expressed in terms of landing/biting counts per man hour.

3.2.2 Resting collection

During periods of inactivity, adult mosquitoes typically rest indoors, especially in bedrooms and mostly in dark places, such as cloth closets and other sheltered sites. Resting collection requires systematic searching of these sites for adult mosquitoes with the aid of flashlight. Following a standard timed collection routine in selected rooms of each house, densities are recorded as the number of adults per house or number of adults per man hour of human efforts.

3.2.3 Oviposition traps

Ovitrap are devices used to detect the presence where the population density is low and larval surveys are largely unproductive as well as normal conditions. The ovitrap is used for surveillance in urban areas to evaluate the impact of adulticidal space spraying on adult female population.

4. Vector management

4.1 Environmental Management

The major environmental management methods used for control of immature stages of vector are-

4.1.1 Environmental modification

The best way to avoid mosquito bites is to locate and remove standing water where mosquitoes can lay eggs. Larvae are usually found on the surface of stagnant water. Remove any items located outdoors that can collect water, including buckets, old tires, bottles, wheelbarrows, and cans. Keep swimming pools circulating and chlorinated at all times. Remove debris from rain gutters regularly and keep street gutters clear to prevent water runoff from pooling.

4.1.2 Changes in human habitations

Efforts are made to reduce man-virus contact by mosquito proofing of houses with screens on doors/windows [2, 6]. Exclude mosquitoes from your home by keeping doors and windows tightly shut or add screens with insect-proof netting. Keeping fine mesh screens in good repair will maintain an effective barrier [7, 8].

4.2 Personal Protection

Protective clothing and repellents are common means of personal protection against mosquitoes and other biting insects [3]. Household insecticide products, namely, mosquito coils, pyrethrum space spray and aerosols have been used extensively for personal protection against mosquitoes. Insecticide treated mosquito nets have limited utility in dengue control, since the vector species bite during the day time. However, insecticide treated bed nets can be effectively used to protect infants and night workers while sleeping in daytime [19].

Mosquito nets act as physical barriers by blocking the vector mosquitoes. Application of pyrethroid insecticides adds a chemical barrier to the physical one, further reducing human-vector contact and increasing the protective efficacy of the mosquito nets [18]. Pyrethroid insecticides have a long residual

action and low mammalian toxicity and provide prolonged protection by their excito-repellent effect. As mosquitoes are positively attracted by the odour of the sleeper inside the net, these insecticides treated nets (ITNs) acts like a baited trap and the mosquitoes that come into contact with the ITN are, most often, killed [18]. Due to multiple effects, the ITNs have been shown to avert around 50% of malaria cases and provide at least double the protection than that provided by untreated nets [17, 18].

ITNs have been found to be the most cost-effective interventions against malaria, and long-lasting insecticidal nets LLINs were found to be significantly cheaper to use than conventionally treated nets [18]. ITNs/LLINs are particularly useful for high-risk populations that cannot be reached by residual spraying, for people in forest-fringe areas who are at risk of infection from forest stay, and for pregnant women who are highly vulnerable to malaria [18].

Conventional ITNs, treated with pyrethroids such as alpha-cypermethrin, cyfluthrin, deltamethrin, lambda-cyhalothrin or permethrin, need to be re-treated after three washes, or at least once a year to ensure continued insecticidal effect. Long-lasting insecticidal nets [LLINs] are factory-treated mosquito nets, made with netting material that has the insecticide incorporated within or bound around the fibers and the insecticide is progressively released so that the net retains the efficacy after repeated washings [5]. The LLINs are expected to retain their effective biological activity without re-treatment for at least 20 standard washes and for three years of recommended use under field conditions. Permethrin (high density polyethylene monofilament yarn blended with 2% permethrin), Deltamethrin (multifilament polyester netting treated with deltamethrin 55mgm⁻²), and alpha cypermethrin (multifilament polyester netting treated with alpha cypermethrin 200mgm⁻²) are used in LLINs [5].

4.3 Biological Control

4.3.1 Mosquito-Eating Fish

One of the safest and interesting methods in mosquito control is the use of biological agents that eat or destroy the larvae. Several species of birds, bats, fish, spiders and predatory insects also eat mosquitoes and will complement other control methods. The Eco-friendly larvivorous top water minnow or mosquito-eating fish, *Gambusia affinis* is an important control agent for immature mosquitoes. It feeds on the larvae and is most effective in ornamental ponds or other man-made bodies of water that do not connect with natural waterways. These fish can be introduced into all collections of potable water like

wells, tanks, ponds and lakes, particularly in rural and peri-urban areas and in freshwater bodies in rural areas. Never release mosquito-fish into streams, ponds or lakes as they can become invasive. Gold fish and koi will also eat mosquito larvae.

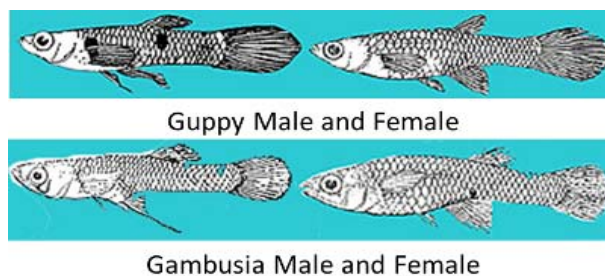


Fig.4: Mosquito-eating fish

4.3.2 Pathogens

Endotoxin-producing bacteria, *Bacillus thuringiensis* serotype H-14 (*Bt* H-14) has been found an effective mosquito control agent. Bacteria such as *Bacillus sphaericus* and *Bacillus thuringiensis varisraelensis* are also effective larvicides. However, they need to be re-introduced every 15 days and their culture may need expertise. Mermitid Nematode (*Romanomermis culicivorax*), Notonectid (Bug), Ambylospora (Protozoa), Coelomomyces (Fungus), Nuclear Polyhedrosis (Virus), and Cyclopoid copepods (Crustacean) are the other biological larvicides found to be effective.

4.4 Chemical Control

There are many mosquito-control products sold to reduce mosquito populations, including liquid, granular and powder etc. formulations (Table 1). When using these chemical products, take precautions to minimize human, pet, and environmental exposure. Use of aerosol sprays, foggers and misting systems is not recommended, due to the high probability of harm during the application from inhaling the pesticides and touching the residues they leave behind. People and pets may be exposed to pesticides through direct contact with spray droplets, contact with objects in the treatment area on which residues have landed, or inhalation of aerosolized pesticide in the air. Outdoor sprays can drift away and pose a risk to non-target wildlife such as fish, honey bees, ladybugs, and butter flies. These systems provide only temporary relief from mosquitoes, and widespread use can lead to mosquito resistance.

Table 1: List of registered larvicide/adulticide under the Insecticides Act, 1968, Government of India, (UPTO- 30.06.2016)

Insecticide Compound	Formulation	Group	Larvicide / Adulticide	Dosage	Uses
Alphamethrin	0.1 w/w (RTU)	SP	Adulticide	25-50 mg a.i./sq.m	-
Allethrin	0.5% Coil	SP	Adulticide	-	-
	0.2% Coil			-	
	4% Mat			-	
	5% Aerosol			-	
	3.6% LV			-	
Bifenthrin	0.05% Coil	SP	Adulticide	-	-
Cyfluthrin	10 % WP	SP	Adulticide	25-50 mg a.i./sq.m	100 gm of Cyfluthrin 10% WP to be diluted in 8 liters of potable water 40 gm of Cyfluthrin 10% WP to be diluted in 10% liters water
Chlorpyrifos M	40%EC	OP	Adulticide	-	-
Cyfluthrin	5% EW	SP	Adulticide	8 ml./ sq.m.	-
Cyfluthrin + Transfluthrin	0.025+0.04%	SP	Adulticide	-	-
Deltamethrin	2.5% Flow	SP	Adulticide	25 mg /sq.m	For impregnation of polyester, nylon and cotton bed

					net
Diflubenzuron	2% Tab	BU	Larvicide	0.5-1.0 ppm	½- 1 Tablet in 40 lit. water
	25% WP			25-50 g a.i./ha	-
Deltamethrin+Allethrin	0.05+0.04%	SP	Adulticide	12.5-25	-
Deltamethrin + D-transallethrin	2.5+2% EC	SP	Adulticide	12.5-25	-
Diazinon	25% CS	OP	Adulticide	-	-
D-transallethrin	2% MAT	SP	Adulticide	-	-
	0.1% Coil			-	
D-allethrin	21.97% Mat	SP	Adulticide	-	-
Fenthion	2% Spray	OP	Adulticide	-	-
Lambda Cyhalothrin	2.43% CS	SP	Adulticide	10mg a.i./sq.m	Impregnation of bed nets to prevent attack from mosquitoes.
	2.43% CS			20-30 mg/sq.m.	-
Malathion	2% Spray	OP	Adulticide	-	-
Metofluthrin	0.005% Coil	SP	Adulticide	-	-
Permethrin	2% w/w	SP	Adulticide	-	For control of mosquitoes both indoors and outdoors. After unpacking and before using the new bed net, keep it in and open place for 12 hours away from the sunlight
Proteamphos	1% Spray	OP	Adulticide	-	-
Propoxur+Cylin	0.75+0.025 Aero	Carbamate / SP	Adulticide	-	-
Propoxur	20%EC	Carbamate	Adulticide	200mg a.i./sq.m	-
Pirimiphos-methyl	1% Spray	OP	Adulticide	50 ml/100m ³	-
Pyrethrin+Malathion	0.05+1%	SP/OP	Adulticide	-	-
Pyrethrin	0.2% w/w	SP	Adulticide	-	-
Propoxur	1% Spray	Carbamate	Adulticide	-	-
Prallethrin	1% w/w Red Mat	SP	Adulticide	-	-
	0.04% Coil			-	
	0.8% w/w Mat			-	
	0.5% Coil			-	
	1.2% Mat			-	
	19% w/w VP			-	
2.4% w/w Liq.	-				
S-Bioallethrin	2.4% Mat	SP	Adulticide	-	-
Transfluthrin	0.88,1.6% liq. Vap.	SP	Adulticide	-	-
	1.6% liq. Vap.			-	
	20% w/w MV Gel			-	
	0.03% w/w Coil			-	
	1% EU			-	
	1.2% liq. Vap.			-	

SP= Synthetic Pyrethroids; OP= Organophosphorus; BU= Benzyl Urea.

4.4.1 Larvicide

All the larvicides, which are safe, without any odour or colour, have residual effect with low mammalian toxicity and do not pose any health hazard should be used. Temephos, an organophosphate compound meets all the above mentioned requirements and this insecticide is being used under the public health programme. The recommended dose for application of Temephos (50 EC) is 1 ppm (1 mg per liter of water). Themiphos and Fenthion are the two commonly used larvicidal agents. Themiphos is used on potable water collections and Fenthion, being more toxic, is used on non-potable water collections. Oils may be applied to the water surface, suffocating the larvae and pupae. Most oils in use today are rapidly biodegraded. An insect growth regulator such as methoprene is specific to mosquitoes and can be applied in the same way as chemical insecticides.

4.4.2 Adulticide

The following methods are recommended for the control of adult mosquitoes:

4.4.2.1 Pyrethrum spray

It may be used in indoor situations as space spray at a concentration of 0.1% - 0.2% @ 30-60 ml/1000 cu. ft.

Commercial formulation of 2% pyrethrum extract is diluted with kerosene in the ratio of one part of 2% pyrethrum extract with 19 parts of kerosene (volume/volume). Thus, one liter of 2% pyrethrum extract is diluted by kerosene into 20 liters to make 0.1% pyrethrum formulation (ready-to-spray formulation). After dilution, pyrethrum extract is sprayed with Flit pump or hand operated fogging machine fitted with micro discharge nozzle.

4.4.2.2 Malathion fogging or Ultra Low Volume (ULV) spray

In application of ULV, minimum volume of liquid insecticide formulation is applied per unit area. That is, the insecticide is broken down into small droplets of a volume median diameter (VMD) of 40-80 microns with an objective of producing a cloud of insecticide droplets that remain suspended in air for an appreciable time and driven under the influence of wind [20]. This provides maximum effectiveness against target vectors. Since no diluent is used, the technique is more cost-effective than thermal fogging but it does not generate a visible fog. Most organophosphorus insecticides in their technical form can be applied as ULV spray. Under the public health programme, ULV spray (fogging) is undertaken by

using 95% or pure technical malathion. The ground equipment mostly used for ULV spray includes portable motorized knapsack blowers and cold aerosol generators.

4.4.3 Indoor Residual Spraying (IRS)

IRS is an integral component of the global mosquito control plan and currently DDT, pyrethroids (Deltamethrin 2.5% WP, Cyfluthrin 10% WP, Alpha cypermethrin 5% WP and Lambda cyhalothrin 10% WP) or Malathion 25% are used in different parts of the world for this purpose [12]. All the interior walls and ceilings as well as the underside of furniture, back of the doors and porches of permanent human dwellings as well as Jhoom huts where people sleep during the plantation or harvesting season are sprayed. For protection during the entire transmission season, two rounds of DDT or synthetic pyrethroids or three rounds of Malathion are used [16].

4.4.4 Mosquito Repellents

Mosquito repellent is a substance applied to skin, clothing or other surfaces which discourages mosquitoes from landing or climbing on that surface. Usually, mosquito repellents work by masking human scent or by using a scent which mosquitoes naturally avoid [4]. Carbon dioxide and lactic acid present in sweat in warm-blooded animals act as an attractive substance for mosquitoes. The perception of the odour is through chemo-receptors which are present in the antennae of mosquitoes. The repellents block the lactic acid receptors and destroy upwind flight. Therefore, mosquitoes lose its contact with the host. The advantage of skin repellents is their relative cheapness. Most of the insect repellent cream being produced by different manufacturers are chemical based and may cause skin problems to human being on prolonged use. DEET spray is still the most widely used mosquito repellent. It has generally been regarded as safe. However, toxic effects have been recorded, including encephalopathy in children, urticaria syndrome, anaphylaxis, hypotension and decreased heart rate [1]. Several other compounds have been evaluated for repellent activity, but none has had the commercial success of DEET.

5. User & environment friendly botanical formulations

5.1 Spreading Oil Formulations

The surface-spreading formulations are essential oil-based formulations which can be applied by dropping on water surface, and after application, the formulation spreads to the whole water surface within a few seconds (Fig. 5). Basically these formulations are stable dispersion of water-insoluble liquid or solid in oil, and if it is a solid dispersion, the dispersed particles are wet, grinded into a fine particle suspension and stabilized with the help of wetting and dispersing agents. When these formulations are applied on the water surface, the active ingredient maintains a smooth networking film on the water surface. The films formed by these formulations are not continuous films but having holes which maintain the required oxygen level within the water for other aquatic organisms present. The larvae of *Anopheles* mosquitoes feed on the water surface and the active ingredient produce mortality after ingestion. Spreading oil formulations of essential oil may directly be dropped on the surface of water sources like lake, pond, etc., and the formulations will be nontoxic to non-target organisms, fish and other aquatic species. The application does not require any spray, and good control of mosquito larvae may be obtained just by dropping it on the water surface. The formulations designed in the laboratory have the characteristics of maintaining the networking film throughout the water surface, and under

natural conditions, the film remains in networking condition even after the winds and water waves.

Advantages

- Botanical based thus safe for non-target organism
- Easy to pour and measure
- May give enhanced biological activity
- Non flammable
- Low cost of formulation



Fig 5: Spreading Oil Formulation

5.2. Floating Tablets

The floating tablets are slow release tablets which after application in water bodies floats on the surface of water due to low specific gravity and specific inert ingredients (Fig. 6). The floating tablets offer a simple and practical approach to achieve increased surface-residence time for the dosage form and sustained active ingredients' release. It can reduce the frequency of dosing required for mosquito larval control and decrease variation in larvicidal concentration. Floating tablet formulations of botanical may directly be applied on surface of water sources like lake, pond, etc.; the formulations will be non-toxic to non-target organisms, fishes and other aquatic species. The application does not require any spray, and good control of mosquito larvae may be obtained just by dropping it on the water surface. The release of active ingredient from the formulations may be designed based upon the type and infestation of the target species.

Advantages

- Dust free
- Easy packaging
- Easy to handle and apply
- Long shelf life
- Adjustable release rates



Fig 6: Floating Tablets

5.3 Nanoemulsion

Botanical-based Nanoemulsion may be a good alternative of synthetic pesticides for mosquito control spray application (Fig. 7). Due to finer droplet size, total surface area is increased many folds so total coverage of target site also increases as results dose rate can be reduced to some extent.

Advantages

- Low dose rate
- No skin and eye irritation
- Less or no solvent
- Minimal toxicity
- No flammability

A number of essential and botanical oil-based Nanoemulsions have been prepared with insecticidal activity for mosquito. These Nano emulsions may be a good alternative to any kinetically stable emulsion or suspension used for spray application.



Fig. 7: Nanoemulsion

5.4 Nanogel

Nanogels are typical formulations mainly of the size range of 100 nm, by varying solvent quality and branching the volume fraction, one can alter variably to maintain a three dimensional structure. Recently in our laboratory, we have developed a nanogel formulation for long lasting impregnation of this insecticide in the dresses which can protect personals from mosquito bite whenever they are deployed/posted to work in forest areas. This type of nanogel formulation may have good future in seed dressing/coating because of its lower particle size, large surface area and greater adhesive properties.

6. Conclusion

The real benefits of botanical insecticides can be best realized in developing countries, where people may not be able to afford synthetic insecticides, and the traditional use of plants and plant derivatives for protection of mosquitoes is long established. Even where synthetic insecticides are affordable to people (e.g.- through government subsidies), limited literacy and a lack of protective equipment result in thousands of accidental poisonings annually. Recently, attention has been paid to traditional plants used in India for protection against mosquitoes.

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