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# Advances in Vector Mosquito Control Technologies, with Particular Reference to Herbal Products

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## Abstract

Despite several decades of control effort, vector-borne diseases (VBDs) are still regarded as a major public health problem in the tropical and subtropical regions of the world. They are more common in the developing as well as resource-poor countries and inflict enormous burden in terms of morbidity and mortality. Control of vectors is the primary available intervention for some of the most devastating VBDs, particularly those lacking vaccines such as malaria, dengue, chikungunya, trypanosomiasis, filariasis, leishmaniasis and Chagas disease. Since long decades vector control programme based on chemical agents has been in place but their effectiveness was soon undermined due largely to the prowess of development of resistance amongst most vector species of these diseases. Thus, in turn, there is a growing need for searching of alternative novel interventions and exploration of next-generation vector control strategies, including repellents, attractants, traps, pneumatic/tracheal explosions in larval populations, biocides integrated with nanoparticles, genetically modified vectors (transgenics), paratransgenics, etc. Plant-based repellents have been used for generations in traditional practice as a personal protection measure against host-seeking mosquitoes. Knowledge on traditional repellent plants obtained through ethnobotanical studies is a valuable resource for the development of new natural products. Recently, commercial repellent products containing plant-based ingredients have gained increasing popularity amongst consumers, as these are commonly perceived as 'safe' in comparison to long-established synthetic repellents although this is sometimes a misconception. There is a need for further meticulously evaluating repellent compounds and develop new products that offer high repellency as well as good consumer safety. Such innovative technologies must also

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fit into the concept of integrated vector control management (IVM) which derives today a much greater meaning now by bringing within its fold all the available technologies than ever before, for an effective vector control.

## 1.1 Introduction

Arthropods, or jointed legs, are invertebrate animals belonging to phylum Arthropoda and include insects, spiders, centipedes, shrimp and crayfish which make the group largest amongst animals, accounting for approximately 80 % of extant species (anywhere close to 30 million) estimated to be present in the world. Insects are one of the most abundant groups of arthropods, with phenomenally diverse behaviour and adaptation. Even though an astoundingly large number of insects are beneficial as pollinator or producer of economically important products, some of them are nefariously known as either dreaded pests or transmitters of deadly or debilitating diseases of both public health and veteri-

nary importance. These arthropod vectors (insects, ticks, mites, etc.) transmit protozoa, helminths, bacteria and viruses, spirochaetes, fungi, etc. These vectors and the diseases transmitted by them inflict heavy losses to humans and their livestock, both directly by biting and sucking blood and indirectly by transmitting vector-borne diseases (Table 1.1). In the early days of vector control, inorganic compounds were used, but later they were largely replaced by organic chemical compounds due to their wider and quicker effects, besides large production. Repellents have always been man's fascination in keeping the mosquito at bay and the world has by now seen highly effective repellents such as DEET, DEPA, etc., albeit the list of effective candidate compounds is very long!

**Table 1.1** Some important vector groups and the diseases transmitted by them (WHO 2014a)

Vector		Diseases
Mosquitoes	<i>Aedes aegypti</i>	Dengue, yellow fever, chikungunya, Zika virus
	<i>Aedes albopictus</i>	Chikungunya, dengue, West Nile virus
	<i>Culex quinquefasciatus</i>	Lymphatic filariasis
	<i>Culex</i> species	Japanese encephalitis
	<i>Anopheles</i>	Malaria, lymphatic filariasis (in Africa)
	<i>Haemagogus</i>	Yellow fever
Flies	Sandflies	Leishmaniasis, sandfly fever (phlebotomus fever)
Bugs	Triatomine bugs	Chagas disease (American trypanosomiasis)
Ticks		Crimean-Congo haemorrhagic fever, tick-borne encephalitis, typhus, Lyme disease, relapsing fever (borreliosis), rickettsial diseases (spotted fever and Q fever), tularaemia
Fleas		Plague, murine typhus, rickettsiosis
Cyclops		Dracunculiasis (guinea worm disease)
Flies	Flies (various species)	Human African trypanosomiasis, onchocerciasis (river blindness)
Aquatic snails		Schistosomiasis (bilharziasis)

## 1.2 Global Distribution of Vector-Borne Diseases and Need for Indigenously Developed Technologies

The emergence and re-emergence of vector-borne diseases are the most important challenges threatening global public health in the twenty-first century. VBDs account for more than 17 % of the estimated global burden of infectious diseases, causing more than 1 billion cases and over 1 million deaths annually (WHO 2014b). Mosquitoes and ticks account for the majority of transmissions of the most important vector-borne diseases. At present, mosquito-transmitted diseases are present in more than 125 countries worldwide, mainly in the tropical and subtropical regions, posing major risks to half the world's population. Malaria and dengue remain the most prevalent mosquito-borne infections in many parts of the world with over 50 % of world's population – more than 3.3 billion and 2.5 billion people – at risk for malaria and dengue, respectively, with severe impact on economic and social development. Added to it are other vector-borne diseases like West Nile virus in the Americas,

chikungunya and Japanese encephalitis in Asia and Oceania and Rift Valley fever in Western and Eastern Africa rapidly emerging (Gould and Solomon 2008).

India, with over 1.21 billion people, is the second most populous country in the world (75 % of South Asia region's people live in India, World Bank 2010) and the tenth largest economy (with a GDP of US\$ 1847.9 billion in 2011), but still continues to share as high as 21 % of the world's global burden of diseases in terms of increased morbidity, mortality and disability (WHO 2012; National Commission on Macroeconomics and Health 2005). Overall, out of 4.2 million disability-adjusted life years lost due to vector-borne diseases, malaria alone was responsible for an estimated 1.85 million years lost/annum in India (Table 1.2) (Kumar et al. 2007; Peters et al. 2001). Insecticides formed the main plank of control strategy in the past, and in fact they are invariably deployed even today in the wake of any disease outbreak. Owing to the environmental and animal health concerns, in addition to development of resistance in the vectors of diseases against most of these synthetic pesticides, there has been an urgent need for exploring novel

**Table 1.2** Burden of major vector-borne diseases in India (WHO 2012)

Diseases	Number of cases	Percentage of global burden	Reference
Malaria	1.07 million (2012) and 0.88 million (2013)	India contributes about 70 % of malaria in the Southeast Asian Region	Kumar et al. (2007)
Dengue	75,454 (2013)	Asia accounts for 70 %, in which India alone contributed 49 % and 34 % of the Asia and global burden, respectively	Gubler (1998), WHO (2014a)
Visceral leishmaniasis (kala-azar)	13,869 cases and 20 deaths (2013)	67 % of total visceral leishmaniasis (VL) cases reported by Indian subcontinent	WHO (2014a), Lobo et al. (2011)
Lymphatic filariasis	6 million	About 70 % of the infection worldwide contributed by India, Indonesia, Nigeria and Bangladesh	WHO (2014b)
Onchocerciasis	NA	NA	
Schistosomiasis	NA	NA	
JE	1078 JE cases and 199 deaths (2013)	NA	WHO (2014a)
Chikungunya	18,639 cases (2013)	NA	WHO (2014a)

and innovative methodologies that are eco-friendly and less expensive and can be integrated with IVM approach (Patz et al. 2005; Gubler 1998; Monath 1994; Lobo et al. 2011).

### 1.3 Vector Control Strategies

For many years, much of the medical research community has been focusing on the development of vaccines or drugs against mosquito-borne diseases, and there has been substantial success achieved as well in this direction (e.g. JE, KFD, etc.). Still there are many other serious and deadly infections, such as dengue, chikungunya and malaria, which have so far been eluding the discovery of an effective vaccine. In cases of certain diseases, such as lymphatic filariasis, leishmaniasis and dengue, it has been proven beyond doubt that an effective control of vector could bring about an appreciable depletion in the disease cases as well. The 'vector population suppression' involves various methods such as use of insecticides, pathogens, predators, lure and kill trapping, environment management, etc., while 'vector population replacement' involves vector's genetic manipulation so that vector should either reproduce its nonviable generations or becomes unfit for reproduction or for disease transmission.

#### 1.3.1 Biochemical Strategies

Like in other countries, in India too, insecticides played a key role in effecting vector control under a national disease control programme such as those of National Malaria, Filariasis and Leishmaniasis Eradication/Control Programmes. The Directorate of National Vector Borne Disease Control programme (NVBDCP, earlier known as NMEP) is the nodal department in preventing and control of vector-borne illnesses in India. Since mosquito is the prime arthropod vector in transmitting the vector-borne infection in India, all the vector control strategies are mainly targeted on mosquito control. Currently, India is using insecticides for public health purposes,

namely, the organochlorines (DDT), organophosphates (malathion) and certain groups of synthetic pyrethroid (deltamethrin, cyfluthrin, alpha-cypermethrin, lambda-cyhalothrin, etc.) for indoor residual spraying (IRS), fogging and aerial spraying (ultra-low volume spraying). Since organophosphates are highly toxic to mammals and have a short residual life, it is not used for IRS, whereas pyrethroids are widely used for IRS and are the only insecticide currently used to impregnate bed nets.

Larvicidal treatment of water with temephos and Bti is approved by the WHO. The organophosphate temephos (Abate) has been most widely used against the container-breeding malaria vector *Anopheles stephensi* and the dengue/chikungunya vector *Aedes aegypti*. Destroying the larvae, either chemically or biologically, while in the aquatic environment is an effective vector control method. Recently, insect growth regulators (IGR), such as diflubenzuron and methoprene, have been implemented as larvicide which are target specific but may be toxic to immature stages of other aquatic insects. Although insecticides have been effective in bringing disease under control in the initial stage of application, the current disease burden indicates that the strategies deployed are no longer effective due to the development of resistance and that the widespread and long term application is not cost effective and logistically difficult especially in developing and underdeveloped countries. In recent decades, resistance to newer classes of insecticides has also been reported quickly after usage because the formulations used for vector control were originally used for agricultural purposes. Multiple and cross-resistance to insecticides in major mosquito vectors is reported against organochlorines, organophosphates, carbamates and pyrethroid insecticides (Hemingway and Ranson 2000).

The modes of insecticide resistance mechanisms in mosquitoes are through (i) metabolism-based resistance (altered activities of enzyme groups, which inhibit the insecticides from reaching their potential target sites), (ii) behaviour resistance (changing their customary behaviour in a way to exposure to chemical), (iii) target

site resistance (changes in the target sites and proteins that insecticides bind to) and (iv) penetration resistance (occurs when the cuticle (outer layer of insects) absorbs the insecticides molecule's much more slowly than the susceptible insects) (Hemingway and Ranson 2000). The development of resistance over various biochemicals makes a shift to insecticide combinations and also searching for new chemical formulations. This has compelled to look for eco-friendly alternative control methods so that the use of insecticides can be minimized. Several environment-friendly methods involving use of insectivorous fishes, biopesticides, pheromones, sterilized males, refractory mosquitoes, endosymbiont, midgut symbionts, etc. are being developed with varying degrees of success. Apart from these, a whole array of effective repellents, sticky traps, lethal traps, etc. is also proposed.

### 1.3.2 Biological Control Agents

Biocontrol is the use of natural enemies for control of vector population. Biological agents, such as parasites, pathogens and predators, can be used to target various life stages of the mosquito. Bacteria, fungi, viruses, fish, predaceous insects such as dragonflies and notonectids and copepods have been employed to decrease the mosquito larvae populations. These agents are inexpensive to implement, and safe for humans and nontarget organisms. They therefore provide a potentially environmentally friendly option. Fishes feeding on mosquito larvae include the world famous *Gambusia affinis*, some cyprinids (carps and minnows) and killifish which have been used for many decades in malaria endemic regions.

### 1.3.3 Preventive Strategies

Preventative measures to combat against VBDs include the personal prophylactics like bed nets, repellents and attractants, insecticide-treated nets (ITNs) or long-lasting insecticide nets (LLINs)

or insecticide-treated wall linings, environmental management and source reduction measures and the careful design of human settlements (covering the windows and doors of human dwellings with thin muslin).

#### 1.3.3.1 Insecticide-Treated Materials (ITMs), including Long-Lasting Insecticide Nets (LLIN)

It consists of insecticide-treated nets (ITNs) or long-lasting insecticide nets (LLINs), window curtains, sheet cover and wall hangings which have increased in demand in recent decades. The effectiveness of untreated nets could be improved through the use of chemicals, which either killed or repelled insect vectors (Lines and Addington 2001) The ITNs were not very successful because they require reimpregnation every 6 months which was quite laborious and became ineffective after a few washes. This led to the development of LLINs wherein the chemicals were bonded to the fibre of the net (Masum et al. 2010). Insecticide-treated wall linings (ITWLs) or pyrethroid-impregnated indoor linings have been proposed as a safer alternative to indoor residual spraying. Window curtains, screens, doorway or wardrobe curtains, etc. all appear to have promising results in different settings.

#### 1.3.3.2 Repellents

Repellents are made based on chemical products with an offensive smell or taste to mosquitoes. Plant-derived compounds with repellent properties are most likely chemicals that are produced in defence against insects that pose a threat to the plant itself. These chemicals can be grouped into different categories, based on the functional groups present. They include nitrogen-containing compounds, terpenoids, phenolics, proteinase inhibitors and growth regulators. These compounds are generally produced to fight off a broad spectrum of insects including mosquitoes. Plants with better repellent properties fall into distinct families, with the Poaceae family (*Citronella* based especially *Cymbopogon* spp.) being the pre-eminent one. Species of Lamiaceae, Fabaceae and Asteraceae also show promising

results. Prior to the extensive use of synthetically produced repellents, aromatic/essential oils were commonly used. The military was a significant consumer of these oils, creams containing citronella, camphor and paraffin. Numerous essential oil-producing plants from the Lamiaceae, Poaceae, Rutaceae and Myrtaceae families have very well-known repellent activity. Synthetic products that have been used as repellents include indalone, dimethylphthalate 2-ethyl-1, 3-hexane diol (Rutgers 612) and N,N-diethyl-m-toluamide (DEET). DEET is by far the most effective and widely used repellent. The Indian counterpart of the DEET is N,N-diethyl phenyl acetamide (DEPA) which is now commercially available in different formulations and has been claimed to have the protection rate of about 8 h, against not only the mosquito pests and vectors but also other hematophagous arthropods as well as ticks, mites and even leeches.

### 1.3.3.3 Insect Traps

The ovitrap or oviposition trap was mostly used and invented for the surveillance of *Aedes* vectors and then modified to render it lethal to adults or larvae of *Ae. aegypti* (Chan 1972; Lok et al. 1977). In principle, ovitraps could kill adult mosquitoes if the ovistrip was treated with insecticide (Zeichner and Perich 1999) or destroy progeny by using fine nylon netting for trapping the larvae (Lok et al. 1977). Lethal ovitraps (which incorporate an insecticide on the oviposition substrate) with deltamethrin-treated ovistrips killed 89 % of *Ae. aegypti* adults and produced more than 99 % larval mortality during 1-month field trials in Brazil (Perich et al. 2003). The autocidal ovitraps (which allow oviposition but prevent adult emergence) and sticky ovitraps (which trap the mosquito when it lands) have been used on a limited basis. The autocidal ovitrap was used in Singapore for the control of *Aedes* vectors in urban areas with a high density of *Aedes* (Lok et al. 1977). The advantages of lethal ovitraps include their simplicity, their specificity for and effectiveness against container breeders like *Ae. aegypti* and its potential for integration with other chemical or biological control methodologies.

A newer approach to killing mosquitoes in a nontoxic way is to use a device that burns propane, thus generating carbon dioxide, warmth and water vapour which draws the mosquitoes towards the propane flame, where they are then sucked into a net or holder where they are collected. Some newer mosquito traps or known mosquito attractants are also available which are based on the principle of disabling its odour receptor as host-seeking female mosquitoes are guided by attractant odours released by their target. *Anopheles gambiae* is attracted to ammonia, lactic acid and other carboxylic acids naturally present in the body odour and sweat produced by warm-blooded animals. There are traps like black hole mosquito and midge trap and Jakmax insect trap which were developed based on this concept which release heat and CO<sub>2</sub> and captures mosquitoes, sandflies, midges, termites and other flies that are attracted by black light.

### 1.3.3.4 Strategies Based on Genetic Modification

Genetic modification of vector mosquitoes is one of such technologies, which may be mainly used either to suppress or replace the wild populations of a vector so as to decrease vector populations or reduce the vector's ability to transmit. These applications include the release of reared mosquitoes in the environment to introduce modified genetic traits in wild population. GM encompasses multiple approaches, which are broadly categorized into two types. The first category includes sterilized insect technique (SIT) for population suppression and the second category is the gene drive systems for population replacement or manipulation. The sterilized insect technique (SIT) includes the dominant lethal gene systems (RIDL), *Wolbachia*-mediated cytoplasmic incompatibility (CI) and classical radiation-induced male sterility, while population manipulative technologies include *Medea*-based gene drive, under-dominance gene drive, homing endonuclease, *Wolbachia*-mediated heritable biocontrol or genetically modified midgut bacteria and transposable element like *PiggyBac*. However, most of them are still in laboratory development. There are various methods either

based on population suppression or population manipulation; all these techniques are still in the initial stage of development and require different region-based field testing under independent monitoring in order to prove the result to gain the consent of society to implement. These techniques require large volume of mosquitoes to be released into the environment at different period of interval to either suppress or replace mosquito population.

## 1.4 Plant-Based (Herbal) Mosquito Repellents

Plants offer a great promise for offering molecules that can be exploited to develop effective antimosquito products (Tyagi and Shahi 2001; Tyagi 2002a, b; Tyagi and Shahi 2002; Tyagi 2003a, b; Shahi et al. 2000). Most plants contain compounds, falling under several categories, including repellents, feeding deterrents, toxins and growth regulators, which they use in preventing attack from phytophagous (plant-eating) insects. These compounds may be categorized under (i) nitrogen compounds (primarily alkaloids), (ii) terpenoids, (iii) phenolics, (iv) proteinase inhibitors and (v) growth regulators. Those volatile components released as a consequence of herbivory are now best known for being effective against mosquitoes and other biting insects (Pichersky and Gershenzon 2002). Insects detect odours when volatile odours bind to odorant receptor (OR) proteins on ciliated dendrites of specialized odour receptor neurons (ORNs) often on the antennae and maxillary palps of the insect. Some ORNs, such as OR83b, that is important in olfaction and blocked by the gold standard synthetic repellent DEET (N, N-diethyl-3-methylbenzamide), are highly conserved across insect species. Plants commonly produce volatile 'green leaf volatiles' when leaves are damaged in order to deter herbivores. However, it is most likely that many plant volatiles are deterrent or repellent because they have high vapour toxicity to insects.

In India, a large number of examples are there where the plants, whole or part of it (in dried or

fresh and in different forms: leaf, fruit, seed, root, etc.), have been exploited for thousands of years most simply by hanging bruised plants in houses or by burning in fire after preparing a formulation with animal dung (neem leaves are the best example of natural repellent). All over the world, there is now a common knowledge of using these plant-based "natural" smelling repellents because plants are perceived as a safe and trusted means of mosquito bite prevention (Dam et al. 2000a, b).

### 1.4.1 Some of the Best Known Plants for Mosquito Repellency

#### 1.4.1.1 Eucalyptus

*Corymbia citriodora* (Myrtaceae), also known as lemon eucalyptus plant, is known for many centuries to have insect repellency properties. Lemon eucalyptus essential oil, comprising 85 % citronellal, is far more effective at repelling mosquitoes for several minutes. However one of its ingredients, para-menthane-3,8-diol, which has a lower vapour pressure than volatile monoterpenes found in most plant oils, provides very high protection from a broad range of insect vectors over several hours. Nanotechnology has of course recently opened new vistas in using effectively the eucalyptus extracts (Sugumar et al. 2014).

#### 1.4.1.2 Citronella

Best known as lemongrass, the essential oils and extracts belonging to plants in the citronella genus (Family:Poaceae) are commonly used as ingredients of plant-based mosquito repellents mainly *Cymbopogon citratus*, *C. nardus*, *C. schoenanthus*, *C. winterianus* and *C. jwarancusa*. Citronella, which mainly contains citronellal, citronellol, geraniol, citral,  $\alpha$  pinene and limonene, was used by the Indian Army to repel mosquitoes at the beginning of the twentieth century. Today, citronella is one of the most widely used natural repellents on the market, used at concentrations of 5–10 %. Citronella-based repellents only protect from host-seeking mosquitoes for about 2 h

although formulation of the repellent is very important. Recently, the use of nanotechnology, particularly allowing vanillin (5 %) with the essential oil, has allowed higher protection rate with slower release rates of oils. Encapsulated citronella oil nanoemulsion is prepared by high-pressure homogenization of 2.5 % surfactant and 100 % glycerol, to create stable droplets that increase the retention of the oil and slow down release.

#### 1.4.1.3 Neem

Neem is widely appreciated globally for its manifold applications, one of which being the repellency against pests of various kinds including hematophagous insects such as mosquitoes. Several field studies from India have shown very high efficacy of neem-based preparations. It is just unfortunate that it could not be yet exploited fully for its unmatched and wonderful repellency characteristics, albeit considered as a natural alternative to DEET.

### 1.4.2 Promising Developments in Plant-Based Repellents

Plants-based repellents have a great future, as these are considered much safer and eco-friendly than even the best known synthetic preparations. The field of plant-based repellents is moving forward as consumers demand means of protection from arthropod bites that are safe, pleasant to use and environmentally sustainable. Perhaps the most important consideration is improving the longevity of those repellents that are effective but volatile such as citronella. Several studies looked at improving formulations of plant oils to increase their longevity through development of nano-emulsions, improved formulations and fixatives, while alternate uses such as spatial activity and excito-repellency have also been investigated. *Eucalyptus*, *Citronella* and *Cymbopogon* species have shown huge potential in offering molecules that can be manipulated into highly effective repellents. New developments have also been seen in understanding the function of plant-based repellents in insects.

## 1.5 Conclusion

Major arthropod vector-borne diseases, particularly malaria, dengue, Japanese encephalitis, African trypanosomiasis, Chagas disease, schistosomiasis, filariasis, etc. are life threatening and infect billions of people throughout the world where the children and the poor are highly susceptible to infection. Even though conventional methodologies and techniques are successful in certain ecosystems and settings, mostly the vectors remain unaffected largely due to their potential to quickly develop resistance against the insecticides in vogue! There is therefore an urgent need to search for alternative products which function through different lethal mechanisms on the vector. Such new systems should innovatively integrate with the IVM since there is no panacea for the whole problem of vector control or the vector-borne disease control.

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