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(REVIEW ARTICLE)

A review on biological mosquito control measures-past, present and future

Aneha. K, Hemaladkshmi Padmanaban, Bhagyashree Bora, Manikandan Sivaprakasam, Kakhuangailiu Gangmei, Jibi Lukose, Abhisubesh Vijayakumar, Sahadiya Mandodan, Mathivanan Ashokkumar, Vijayalakshmi Krishnamoorthy and Poopathi Subbiah *

Department of Health Research, Unit of Microbiology and Immunology, *ICMR-Vector Control Research Centre, Ministry of Health and Family Welfare, Pondicherry- 605006, India.*

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Abstract

Over 40% of the world's population is under danger from mosquito transmitted diseases, which are a growing global health challenge. Since 2000, malaria control has made significant progress, but of late it has slowed down. Aedes-borne arbovirus risk is also rising quickly due to the remarkable expansion of the dengue and chikungunya viruses, yellow fever outbreaks, and the Zika virus pandemic. To counteract this issues people used many vector control measures. Synthetic pesticides play a significant role in mosquito larval and adult control strategies, although their deployment is hampered by the development of resistance. As an alternative, various biological measures are being used including bacterial bio pesticides. They are extremely effective at controlling larvae due to their lack of resistance development and environmental friendliness. In this present review various biological control measures used for mosquito control are discussed.

Keywords: Mosquito borne diseases; Vector control; Chemical insecticides; Resistance; Biopesticides; Sterile insect technology

1. Introduction

Mosquito has major public health importance as it is vectoring various diseases to mankind. Mosquito borne diseases are spreading rapidly and are responsible for the morbidity and mortality worldwide with a disproportionate effect on children and adolescents [1, 2, 3]. Malaria, filariasis, dengue, chikungunya, Japanese encephalitis (JE), Zika virus fever are the important diseases transmitted by mosquitoes causing millions of fatalities throughout the world every year [4,5,6].

Malaria causes global disease burden and the causative agent is the protozoal parasite of plasmodium species. Malaria is transmitted mainly by *Anopheles* mosquitoes. The most recent WHO report estimate that there would be 241 million new cases and 627 000 deaths worldwide from malaria in 2020

Dengue fever is an important arboviral infection which is transmitted by *Aedes* mosquitoes majorly *Aedes aegypti,* but also *Aedes albopictus,* which are widespread in tropical and subtropical areas. [7]. In India, dengue fever is the second-most dangerous disease spread by mosquitoes. Mostly all cases of dengue fever occur in urban, rural, and semi-urban regions. There is no specific treatment for dengue & severe dengue. According to the most recent WHO report, the dengue virus has infected 390 million people worldwide, with 96 million new cases.

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* Corresponding author: Poopathi Subbiah
Department of Health Research, Unit of Microbiology and Immunology, Vector Control Research Centre, Indian Council of Medical Research, , Ministry of Health and Family Welfare, Puducherry- 605006, India.

The primary cause of viral encephalitis in Asia is the Japanese encephalitis virus (flavivirus). Culex mosquitoes are responsible for spreading the Japanese encephalitis virus from one animal to another. Over 68000 cases of Japanese encephalitis occur annually, with 13600 to 20400 deaths. Children are particularly affected by Japanese encephalitis. After childhood infection, most adults in endemic regions develop natural immunity, but anyone of any age might be infected [8].

Aedes mosquitoes are transmitting other important viral disease called Chikungunya. The causative organism for this disease is Chikungunya virus (CHIKV) [9]. By 2019, Asia and America were the regions most affected by Chikungunya in the past years. India experienced 62,000 instances, while Pakistan reported 8,387 cases of a chronic outbreak [4]. Lymphatic filariasis is an important mosquito borne disease, commonly known as elephantiasis*. Brugia malayi, Brugia timori, and Wuchereria bancrofti* are major the nematode parasites responsible for filariasis. It is spread by a variety of mosquito species, including the *Culex* mosquito, which is more frequently found in urban and semi-urban areas, the *Anopheles* mosquito, which is mostly found in rural areas, and the *Aedes* mosquito, which is mostly found in endemic Pacific islands [10,11]. The disease affects 1.5 million persons in India, including 1.2 million filariasis sufferers and 2 million carriers of microfilaria [12].

Aedes mosquitoes are primary vectors of the Zika virus (ZIKV), a flaviviridae-related arbovirus [13]. On July 8, 2021, a resident of Kerala state, south-west India was found to have the Zika virus (ZIKV) **infection.** This is the first incidence of Zika virus illness in Kerala. Belsar, a village in the Purandar Taluka administrative division of the Pune district, was the site of Maharashtra's first laboratory-confirmed Zika infection where 3600 persons were affected [10].

Vector control strategies are the primary methods for lowering the public health burden of the vast majority of diseases spread by mosquitoes. These strategies which focused on environmental management before the pesticides were synthesized, concentrating on eliminating mosquito breeding sites and improving homes with screens to stop mosquitoes from entering through doors and windows [14]. A subset of the Concept of Integrated Vector Control, the EMVs (Environmental Management for Vector Control) were devised by the World Health Organization (WHO) in 1982. Environmental management operations for vector control include the planning, organizing, carrying out, and monitoring of measures to alter environmental elements or their interactions with people in order to avoid or limit vector propagation and vector-pathogen contact [15, 16]. The first residual pesticide, DDT, was added to the vector control toolkit after the Second World War [14]. For both indoor and outdoor use, chemical pesticides like dieldrin, pyrethrin, and other substances were developed. Insecticides were later incorporated to bed nets [14]. Because of their high costs, emergence of resistance in many target populations, and the perception of hazards to the environment and public health, the use of synthetic chemical insecticides for vector control is on the wane. Chemical insecticides will still play a significant role in vector control programmes, but the issues they have raised and the scarcity in development of new varieties have long sparked interest in alternative control strategies. Considering these scenario of vector control, biological control measures were found to be the best alternative.

2. Biological control measures

Biological pest control is becoming more popular nowadays as a promising strategy for reducing mosquito vector populations. Numerous mosquito-killing biocontrol methods were examined for effectiveness, environmental impact, and safety to non-targets organisms. Researchers have looked into the possibilities of bacteria, nematodes, viruses, fungi, protozoa, fish, and invertebrate predators as vector control agents [16].

The target population is sought to be lowered to "acceptable" level through biological control without endangering the ecology. Biological control methods for mosquitoes should balance human mosquito protection with biodiversity preservation while minimizing toxicological and Eco toxicological effects. When predators like dragonflies were introduced in the late 1800s, it was the first time that the use of beneficial animals for mosquito control was acknowledged [17]. However, there could be a number of problems brought on by mass reproduction and the effective introduction of predators like hydra, flatworms, predatory insects, or crabs. Biological control of insect agents has previously succeeded in replacing chemical insecticides.

3. Use of biological mosquito control agents

3.1.1. Insects

For a long time, people have recognized the value of dragonflies (Odonata) as mosquito predators. Mosquitoes are consumed by both nymphs and adults [18, 19]. Dragonfly nymphs are only found in permanent bodies of water because they take so long to mature. In feeding studies, anisopteran nymphs were demonstrated to be voracious feeders.

Nymphs were once thought to eat up to 100 mosquito larvae every day [18]. *Coenagrion puella* is an example of a zygopteran nymph that is a less successful predator (eating 10 third-instar larvae each day on average).

Mosquitoes are consumed by the majority of water bugs (Hydrocorisa and Amphicorisa). Mosquito breeding grounds commonly contain *Sigara striata, Corixa punctate, and Cymatia coleoptrat.* Because of their primarily omnivorous feeding habits, their importance as mosquito predators is fairly negligible [19]. *Naucoridae*, *Nepidae*, *Notonectidae*, *Pleidae*, *Gerridae* and *Hydrometridae* are the major examples of mosquito predatory hymenopterans [18,20,21, 22,23,24,25].

Numerous water beetles are good aquatic predators due to their abundance and voracity. These predators' capacity is increased by the fact that they may survive and breed in high numbers in a range of mosquito breeding environments. Among the water beetles, dytiscids are the most important predators. Their tiny larvae, on the other hand, can swallow more than 100 *Aedes vexans* fourth instar larvae per day. *Hydrophilus caraboides, Colymbetes fuscus, Guignotus pusillus* are the major examples [18,26].

Numerous articles have emphasized how important caddisfly larvae are as mosquito predators [27, 28]. They are important semi-permanent water body mosquito predators in marshy forests. The 2-3 cm long larvae of *Limnephilus* and *Phryganea* species have been seen on numerous occasions catching snow-melt mosquito larvae. Common mosquito larval predators include the carnivorous Culicidae and Chaoboridae families. In North America, *Toxorhynchites* species have long been researched as possible mosquito pests [29, 30, 31]. Instead of feeding on blood, the female mosquitoes consume nectors. These mosquitoes are mainly found in warm temperature. These naturally rabid and cannibalistic mosquito larvae prefer to lay their eggs in water-filled containers and prefer to devour other mosquito larvae as food. As a result, they are successful in preventing mosquitoes from creating nests inside of containers. *Aedes albopictus* and *Aedes aegypti,* which primarily breed in artificial containers, were eradicated using *Toxorhynchites* species [32, 33, 34]. The Dolichopodidae, Empididae, Ceratopogonidae, and Muscidae (*Genus: Lispe*) are dipterans that can prey on adult mosquitoes [35, 36, 37].

In French, Polynesia, Queensland, and Australia, the copepod crustacean species like macrocyclops and mesocyclops has been used as biological control of *Aedes* mosquitoes [38, 39]. They primarily eat first instar larvae. These discoveries have prompted the development of straightforward techniques for maintaining and widely dispersing copepod species before release.

3.1.2. Nematodes

The nematodes of Steinernematidae and Mermithidae families are the important insect parasites [41]. Steinernematidae are potent parasitic predators on terrestrial insects, particularly their larvae that grow in the soil. It is still controversial whether nematodes like as *Steinernema* species or *Heterorhabditis* species should be used to control *Diptera,* especially *Musca domestica.* It was discovered that mosquito larvae can only be successfully infected in the laboratory. The biological control of mosquitoes is more dependent on aquatic mermithid parasites. In several parts of the world, mermithid nematode species have been researched as biological control agents [42, 43]. *Romanomermis culicivorax* is a biological control agent, contagious between 20 °C and 32°C and a pH range of 5.4 to 7.9. hence temperature and pH are crucial factors to take into account [44].

3.1.3. Larvivorous Fish

Larvivorous fish, which eat mosquito larvae, are the main focus of the biological control of mosquitoes using vertebrate creatures [45]. The most well-known aquatic mosquito predators are *Gambusia affinis, Poecilia reticulate.* For the purpose of controlling mosquitoes, they have been introduced in more than 60 nations [46]. The most extensively used mosquito-controlling organism is *Gambusia affinis* [47]. Hackett released a study on *Gambusia affinis'* efficiency in preventing malaria in Europe as early as 1937. *Gambusia affinis* had greatly aided Turkey and Iran for decrease in malaria cases [48, 49]. The World Health Organization no longer recommends the indiscriminate use of *Gambusia affinis* for mosquito control because of its aggressive behavior toward a variety of aquatic species and its questionable contribution to the management of mosquito-borne diseases [50]. Because they can consume aquatic plants, fish are occasionally introduced as well. As predators, they can reduce mosquito numbers, but as consumers of aquatic vegetation, they can also do so by reducing the habitats where mosquito larvae can grow. *Tilapia zilli, Orechromis mossambia, and Orechromis hornor*, three subtropical cichlids, the common carp, *Cyprinus carpio*, the grass carp, *Ctenopharygodon idella, Alpocheilus panchax, and Cynolebias bellotii* are additional species that are employed to control mosquito populations. [51, 52]. *Oreochromis mossambicus* (Tilapia) is a mouth brooder found in Mozambique. Cichlids is fish that belong to the Cichlidae family and are found in East Africa. They are commonly grown in paddy fields to prevent mosquito larvae and are also utilized as a source of food. This freshwater fish can also be found in brackish water and it reproduces best at a temperature of 20 ºC [53].

3.1.4. Protozoans

Microsporidia are obligate parasites without mitochondria since it can quickly bind to host cells and utilize them as a source of energy [54]. Microsporidia include *Nosema, Amblyospora, Thelohania, Vavraia,* and *Parathelohania*. [55, 56, 57, 58]. The parasite's virulence is determined by the quality of the host [59]. *Nosema algerae* are infecting *Culex quinquefasciatus, Stegomyia aegypti, Anopheles stephensi, and Armigeres subalbbatus.*

3.1.5. Entomopathogenic Fungi

The fungus Coelomyces was the first entomopathogenic fungus to be utilized in the control *of Anopheles gambiae* larvae [60]. When consumed by mosquito larvae, this entomopathogenic fungus changes the larvae's physiology, reducing mosquito blood-feeding capacity, such as *Beauveria bassiana* in *Stegomyia aegypti* [61]. Other fungus species, such as *Lagenidium giganteum*, are also utilized as biocontrol agents in California to control the vectors of West Nile virus and Western Equine Encephalitis [62]. Couch initially described *Lagenidium giganteum* in 1935, when it was paired with a copepod and proved effective in suppressing Anopheles and Culex in North America [63]. It was later discovered to be effective in controlling dengue and filariasis vectors in cement tanks [64, 65]. The fungal species *Leptolegnia caudate* was efficient in suppressing the *Anopheline* population [66]. Mosquito larvae were found to be particularly susceptible to fungus infection in *Stegomyia aegypti and Culex pipiens*, but not in *Anopheles gambiae*. Also, other essential fungi used in the control of vectors are *Metazhizium anisopliae* and *Metarhizium brunneum*. These fungi are highly effective against various insects and exist in the soil and natural environment. The fungi infect mosquito larvae either by producing conidia or blastospores [67]. Since multiple distinct toxins produced during fungal infection are fatal to mosquitoes, and entomopathogenic fungi are largely targeted at adult mosquitoes, the selection pressure for resistance is immediate killing of insects [68]. Therefore, it is anticipated that fungal resistance will develop considerably more slowly than pesticide resistance [69]. Further investigation is required to ascertain the infectiousness, persistence, and viability of fungal spores in mosquito habitat populations because there is so little literature describing how fungus affects mosquito populations in 2010 [69]. To enable widespread application of fungus spores to wild mosquito populations, the best techniques must be found [70].

3.1.6. Bacteria

The discovery of the gram-positive, endospore-forming soil bacterium *Bacillus thuringiensis spp. israelensis (Bti)* in Israel's Negev desert marked the start of a new chapter in the fight against mosquitoes [71]. The development of the powerful *Bacillus sphaericus* strain also having the significant role in the mosquito control [72, 73]. *Bacillus cereus* [74] *Bacillus alvei, Bacillus brevis*, *Bacillus amyloliquefaciens*, *Bacillus circulans, Bacillus subtilis*, *Brevibacill leterosporous*, *Pseudomonas fluorescens*, *Pseudomonas aeruginosa* and *Clostridium bifermentans*, are other bacteria used to control mosquito vectors [75].

Bacillus thuringiensis subsp. israelensis (Bti) and Bacillus sphaericus (Bs)

Bti is a spore-producing, gram-positive rod-shaped entomopathogenic bacteria. They are widely dispersed throughout the natural environment, including in water, soil, plant leaves, stored grains, bug corpses, and desert bird excrement, among other things [76]. The parasporal inclusion and crystals formed after bacterial sporulation have mosquitocidal properties. The inclusions' Cry 4B (135 kDa), Cry 4A (125 kDa), Cry 11A (68 kDa), and CytlAa (28 kDa) proteins all create the toxins that have the mosquitocidal effect. They are assembling into an almost crystal-like shape. Along with the toxic proteins already discussed, Cryl0A and Cyt2Ba are also accountable for the ability to kill mosquitoes.

Bacillus sphaericus produces two-component toxins Bin B (51 kDa) and Bin A (42 kDa), which are responsible for their toxic effects on mosquitoes. They bind to epithelial cells of the gastrointestinal tract of mosquito larvae and specific midgut receptors on the gastrointestinal tract. The first *Bs* strain to be identified to be hazardous to larvae was in 1965. Since then, more than 300 strains have been found and isolated globally.*Bs* has been used to control the common house mosquitoes *Culex quinquefasciatus, Culex pipiens* and *Anopheles* larvae. Advantages of this mosquitocidal strain include high specificity, environmental safety, strong potency, and long-lasting activity. There are currently a number of possible *B. sphaericus 2362*-based biopesticides available. The US and Europe both sell VectoLex and Spherimos. B. Biocide-S, a biopesticide based on S. 1593, is also sold in India. Similarly, the People's Republic of China has access to *B. sphaericus*-C3-41[77].

3.1.7. Resistance to Bacillus sphaericus and Bti

It was previously thought that the use of *Bacillus sphaericus*-derived microbial insecticides did not confer resistance to mosquitoes. Nonetheless, 30 years of previous studies have shown that the *Bacillus sphaericus* binary toxin is also complex in this resistance issue. The *Culex quinquefasciatus* larvae have been found to be resistant to the toxin known as Bin (*B. sphaericus*), according to laboratory and field study [78]. Cross-resistance is also inevitable in some strains. For instance, *Culex pipiens* larvae raised in laboratories have already developed a resistance *to Bacillus sphaericus*. Laboratory strains revealed that the genetic resistance against the majority of latent strains *of Bacillus sphaericus* was cross-resistant to other strains of related categories of toxin-producing organisms [79]. As a result, the use of this particular *Bs* strain in previously planned vector control programmes is no longer possible due to resistance to *B Bacillus Bacillus sphaericus* Bin toxin. Researchers have performed studies on the resistance to the binary toxin produced by *Bacillus sphaericus*. This includes receptor alterations that alter their affinity for toxins [79]. Mosquitoes resistant to *Bti* have already been documented by several authors [80]. Resistance *Bti* is a secondary burst that deals more damage. *P. interpunctella* was the site of the initial discovery of *Bt* resistance in 2000. *Aedes aegypti, Culex quinquefasciatus, Ostrinia nubilalis, Pectonophora gossypiella*, and other insect species have developed resistance to *Bt* toxins [81]. Therefore, it is imperative to find new, more potent alternatives to existing target-specific alternatives.

3.2. Releasing of Mosquitoes

3.2.1. Wolbachia Endosymbiotic Bacteria

An endosymbiotic bacterium called *Wolbachia* is spontaneously infecting many insect species [82]. After infection, it causes cytoplasmic incompatibility, a reproductive phenotype in mosquitoes (CI). *Wolbachia* infection causes non-viable offspring when uninfected females mate with infected male, however, infection causes viable offspring when infected females mate with both infected and uninfected males, allowing this maternally transmitted bacteria to penetrate host populations. *Wolbachia* naturally infecting some of the major mosquito vectors like *Culex quinquefasciatus, Aedes albopictus*, No natural infection in *Aedes aegypti*. Now that it has been transferred from its natural host species to *Aedes aegypti,* the bacterial endosymbiont *Wolbachia* can stop the transmission of the Zika viruses and dengue. *Wolbachia*-infected *Aedes aegypti* has been discovered in numerous field studies, and it is expected that it will spread from the release sites and settle in the target population [83]. The long-term success of this method will depend on how long the *Wolbachia* strain can continue to prevent virus transmission. Population replacement methods using *Wolbachia* rely on mosquito field releases similarly to genetic modification. The *Wolbachia* infection will likely disappear after releases stop if release programmes are too small to ensure that threshold prevalence is reached [84].

3.2.2. Sterile Insect Technique

A target species is mass-reared, sterilized (typically using radiation or chemosterilants), and then reintroduced into a wild population in the sterile insect method (SIT). If releases are sustained for long enough generations, a target population may be repressed or even eradicated due to the following induction of sterility in the natural population, which lowers its reproductive capacity [85]. SIT works best and is most cost-efficient when the sterile release populations are all male. The mechanical sorting techniques utilized in conventional mosquito sexing techniques rely on the size distinctions between female and male pupae. But modern genetic techniques that make use of sex-linked markers could be able to improve the precision and effectiveness of high throughput sex-sorting [14].

4. Conclusion

As mosquitoes are transmitting various diseases like Malaria, Dengue, Chikungunya, Zika virus fever etc. mosquito control has major public health importance. From the past decades people controlled mosquitoes by environment management. Later on after the invention of synthetic insecticides people used it for vector control. Its continuous usage led to the development of resistance in vectors, besides being harmful for environment and other non-target organisms. In this scenario biological control measures were found to be more effective vector control tool. The release of natural predators of mosquitoes like insects, larvivorous fishes, nematodes, crustaceans are found to be effective. Among them biological control using microbes like fungus and bacteria are also effective. *Bacillus thuringiensis israelensis* and *Bacillus sphaericus* are most important mosquitocidal bacteria used widely. The toxic crystals they produce at the time of sporulation has the mosquitocidal activity. Some studies have reported that mosquito is developing some resistance against these mosquitocidal bacteria. In these conditions, novel bacterial isolates have been effectively obtained recently, and it is advised that future researchers may find even more effective techniques for vector control programmes. The releasing of mosquitoes with genetic modification with *Wolbachia* are also a promising mosquito control strategy. One of the most promising novel approaches has been the use of *Wolbachia* endosymbiotic bacteria, which has been targeted at reducing DENV transmission. Large-scale experiments are required to evaluate whether *Wolbachia*-based techniques may be a successful form of mosquito biocontrol program. Effective mosquito population suppression may need a combination of synergistic tactics, including SIT, RIDL, and *Wolbachia*-induced IIT.

If vector control is properly applied, the majority of vector-borne diseases can be avoided completely. The various approaches that are becoming accessible, which are discussed in this theme issue, will give more alternatives for preventing mosquito-borne diseases and might improve on already effective approaches.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors agree no conflict of interest.

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