

NOVEL TECHNOLOGIES FOR MOSQUITO CONTROL

Abstract

Mosquito act as a vector of several deadly diseases causing illnesses and continues to pose a huge hazard to international public fitness and socio-financial development. Mosquito-borne sicknesses are infections caused due to microorganisms like virus, protozoa or other parasites which might be transmitted to humans through mosquito bites. Each year, nearly 700 million people suffer from mosquito borne illnesses leading to the loss of life of nearly 725,000 people globally. As we are all aware that the order diptera is the most structurally developed order with insects gaining resistance development against pesticides. Further, these insects can quickly adapt to diverse environments and have excessive reproduction rates. Hence, there's an urgency to develop innovative management techniques for this notorious vector. The chapter discusses some of the novel techniques of mosquito control like biocontrol techniques, genetic amendment strategies along with CRISPR/Cas9 technology, wolbachia strategies, sterile insect method, use of deterrents and repellents and many others. Overall the chapter highlights the importance of using numerous processes for mosquito control. The challenges and limitations confronted inside the quest for effective mosquito management are discussed in conjunction with ethical concerns associated with genetically changed mosquitoes.

Keywords: Advanced techniques, Biocontrol, CRISPR/Cas9, Genetic modification, Mosquito control

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I. INTRODUCTION

Mosquitoes are recognized as a significant threat to global human and veterinary health due to their diverse species and ability to transmit dangerous diseases. With over 3500 unique species of mosquitoes found in diverse ecosystems and capable of feeding on a variety of host species, their adaptability and abundance contribute to their success as disease vectors [1]. The danger of mosquitoes arises from several factors. Firstly, female mosquitoes require a blood meal to broaden their eggs, and they have developed specialised mouthparts to puncture human pores and skin and reap this critical nutrient. Secondly, some mosquito species show off a desire for human beings as their favored hosts, making them much more likely to transmit sicknesses at once to human populations. Finally, mosquitoes can deliver several viruses and parasites without experiencing damage themselves, allowing them to spread these pathogens effectively [2]. Mosquitoes play a vital position in transmitting illnesses like malaria, yellow fever, dengue, encephalitis, Chikungunya, West Nile virus, and Zika. Their coevolution with ailment marketers, reservoir hosts, and human communities has made them fantastically powerful vectors of debilitating pathogens affecting both humans and animals. The impact of mosquito-borne illnesses is very high, with nearly seven-hundred million individuals are affected every year, leading to greater than one million deaths globally [3]. To combat mosquito-borne diseases several techniques are adopted. One approach involves targeting the disease agent directly to prevent its transmission to humans. This method often includes the use of chemical or microbiological ovicides, larvicides, and pupicides, sometimes in conjunction with synthetic insecticides. However, the use of insecticides requires careful regulation due to the widespread development of resistance and potential environmental and health concerns [4, 5]. In recent years, researchers have explored innovative strategies focusing on limiting the population of host-seeking female mosquitoes. These novel approaches include the use of attractive toxic sugar baits, mass-trapping techniques, auto-dissemination of hormone mimetics, push-pull strategies, and the release of *Wolbachia*-infected or irradiated sterile males/genetically modified individuals into the field [2,6]. Attractive toxic sugar baits involve luring mosquitoes with sugar-based solutions containing insecticides, reducing their numbers by targeting females. Mass-trapping techniques aim to capture a large number of mosquitoes at once, leading to population reduction. Auto-dissemination of hormone mimetics uses infected mosquitoes to deliver control agents to their breeding sites, hindering mosquito reproduction. Push-pull strategies combine repellent barriers and attractive stimuli to divert mosquitoes away from targeted areas. Lastly, the release of *Wolbachia*infected or irradiated sterile males or genetically modified mosquitoes disrupts breeding and reduces population sizes [2]. Implementing these new mosquito control strategies requires careful evaluation of their effectiveness, ecological impact, and cost-efficiency. By adopting integrated vector management strategies and combining multiple approaches, we can enhance mosquito control efforts and combat the devastating impact of mosquito-borne diseases on a global scale. Some of the newly advanced techniques adopted for mosquito population suppression are discussed in this chapter.

II. BIOLOGICAL CONTROL

Biological control of mosquito larvae proves to be the most economical and effortlessly applicable method among various mosquito control measures. The vulnerable stages of mosquitoes are targeted by an array of aquatic organisms, forming a natural

predator-prey relationship. These mosquito young instars become the sustenance for a diverse range of creatures, such as fish, amphibians, copepods, odonate young instars, water bugs, and even larvae of different mosquito species. By harnessing the prowess of these natural predators the population of mosquitoes can be effectively curbed without relying heavily on costly and potentially harmful chemical interventions. Emphasizing and encouraging biological control measures can lead towards a more sustainable and environmentally friendly approach to managing mosquito-related issues.

- 1. Fish:** To mitigate the impact of mosquito-borne illnesses, researchers have explored natural enemies' potential to regulate mosquito populations in aquatic environments. Among these natural predators, larvivorous fish have garnered significant attention due to their effectiveness in consuming mosquito larvae. The western mosquito fish (*Gambusia affinis*) and the eastern mosquito fish (*Gambusia holbrooki*) are generally utilized for mosquito management purposes, but their aggressive nature and capability to damage native fauna boost worries [7]. Studies have shown that larvivorous fish, which includes species from the genus *Gambusia* and *Poecilia* (Poeciliidae), can substantially lessen mosquito larval populations in numerous habitats internationally [8, 9]. These small fish are introduced in numerous countries to combat mosquito-related issues effectively [10, 11]. However, introducing such species can lead to unintended consequences. For instance, they may compete for resources with native fish and amphibians, potentially leading to declines in the populations of these species [12]. The ecological cost of introducing predatory fish for mosquito control must be carefully evaluated to ensure that it does not disrupt the existing aquatic ecosystem. While the *Gambusia* species, especially *G. affinis*, have shown beneficial effects in consuming mosquito eggs, larvae, and young, their impact on mosquito control is not always consistent, resulting in unpredictable outcomes. Moreover, their presence may not deter anopheline mosquitoes, which are also vectors of diseases, from ovipositing in the vicinity [13]. This inconsistency underscores the need for a comprehensive evaluation of their effectiveness and consideration of alternative approaches to mosquito control. In certain regions, native fish species have been explored for their potential in controlling mosquito populations. For example, in the floodplain of the Gambia River, *Tilapia guineensis*, a common native floodplain fish, has been studied for its predatory capacity on mosquito larvae. Semifield trials revealed that *T. guineensis* effectively removed both culicine and anopheline larvae within a day, reducing the likelihood of finding culicine larvae in areas where these fish were present. This suggests that *T. guineensis* could be a promising candidate for mosquito control in The Gambia [13]. Another native fish species, *Epiplatys spilargyreus*, also demonstrated a significant predatory capacity in the same study, further supporting the idea of exploring indigenous fish as potential agents in mosquito control efforts. However, the effectiveness of native fish species in controlling mosquito populations can vary depending on the ecosystem and species composition, necessitating region-specific assessments. Additionally, in rice fields of the Shahjahanpur district, Uttar Pradesh, the mosquito control potential of *G. affinis* was evaluated[14]. Stocking these mosquito fish at a rate of 5 fishes per square meter resulted in a significant reduction in larval and pupal densities in experimental fields compared to control fields during a 42-day observation period. This indicates that *G. affinis* shows promise in controlling mosquito breeding in rice fields, offering a potential solution for mosquito control in specific agricultural settings.

2. **Copepods:** Omnivorous copepods, including *Cyclops vernalis*, *Megacyclops formosanus*, *Mesocyclops aspericornis*, *M. edax*, *M. guangxiensis*, *M. longisetus*, and *M. thermocyclopoides*, have proven their capability as effective predators of younger mosquito larvae [15, 16, 17]. In Vietnam, copepods were effectively employed for mosquito control. They particularly focused on the dengue virus vector *i.e. Ae. aegypti* [18]. Following their introduction into a village in northern Vietnam in 1993, those copepods played an essential position in removing *Ae. aegypti* from sizable surrounding regions by 2000, leading to an outstanding decline in dengue transmission. Even after reliable interventions ceased, the nearby communities in Vietnam persevered their active efforts in copepod biocontrol against *Ae. Aegypti*, reflecting the long-lasting effect of this biocontrol approach.
3. **Frogs and toads:** Ref [19] were the first to highlight the potential of anurans, particularly frogs and toads, in controlling mosquitoes. Their research revealed that mosquito eggs are actively preyed upon by tadpoles with diverse life-history characteristics. Interestingly, the mosquito species *Ae. aegypti* showed a preference for laying eggs in tadpole water, where tadpoles of *Polypedates cruciger*, *Bufo*, *Ramanella*, *Euphlyctis*, and *Hoplobatrachus* genera were found to predate on and destroy these eggs. Studies have confirmed that all tested tadpole species exhibited mosquito egg predation [19]. This highlights the potential of tadpoles in contributing to mosquito control efforts.
4. **Bti and Entomopathogenic Fungi:** *Bacillus thuringiensis* var. *Israelensis* (*Bti*) is a gram positive, spore-forming bacterium widely used as a mosquito larvicide in European countries because of its selective target on insect larvae [20]. The bacterium releases toxins and virulence factors that particularly have an effect on the larval stages of mosquitoes like *Ae. aegypti* [21] and *Ae. albopictus* [22]. Its application has been powerful in decreasing larval mosquito populations, making it a precious device in mosquito control applications. However, prolonged use of *Bti* increases worries, since many insects like diamond backmoth have developed resistance towards *Bt* also [23, 24]. Therefore, there's a need for careful management and rotation of mosquito management techniques to prevent the emergence of resistant populations. In Germany, *Bti* has been effectively used alongside *B. sphaericus* as organic manipulate marketers towards mosquitoes for over a decade. Studies have proven that *Bti* remedies overlaying over 1000 km² of mosquito breeding sites in Germany have resulted in a high-quality decrease of the mosquito population through greater than 90 per cent with none observed harmful effect on the surroundings [25]. The experimental tablet system XL-47, containing 48 per cent *Bti* technical powder, validated promising results in inhibiting pupal formation even underneath prolonged exposure to daylight [26]. However, non-stop tracking and responsible use are crucial to keep the efficacy of *Bti* and save you resistance improvement in mosquito populations. Ref [27] in their study reported that the blastospores of the fungi *Metarhizium anisopliae* (ESALQ 818 and LEF 2000) were highly virulent to adult *Ae aegypti*. Similar studies were carried out by Ref [28] using *Beauveria bassiana* for the simultaneous control of *Ae albopictus* and *Cx. pipiens* mosquito adults. . Among the 30 isolates tested, *B. bassiana* JN5R1W1 was selected as the most effective fungus for the simultaneous control of *Ae. albopictus* and *Cx. pipiens* adults.

5. Wolbachia Endosymbiotic Bacteria: *Wolbachia* are fascinating endosymbiotic bacteria that infect about 40 per cent of insect species [29]. These intracellular bacteria have the extraordinary capability to manipulate the reproduction in their arthropod hosts, and they may be predominantly transmitted vertically from mother to offspring, with occasional horizontal transmission among species. As a result, *Wolbachia* can infect a huge variety of arthropods, inducing diverse reproductive phenotypes. One of the major reproductive phenomena brought about by using *Wolbachia* is cytoplasmic incompatibility (CI) in mosquitoes. Natural *Wolbachia* infections are discovered in fundamental mosquito ailment vectors consisting of *Cx quinquefasciatus* and *Ae albopictus*. However, *Aeaegypti* does not harbor *Wolbachia* naturally but it has been introduced into it to control mosquito populations. Nonetheless, a recent report in Burkina Faso recognized a singular *Wolbachia* pressure in *Anopheles gambiae* and *Acoluzzii*, which might be fundamental malaria vectors in Sub-Saharan Africa. The usage of *Wolbachia* for mosquito-borne disease management dates back to the past due 1960s whilst it was successfully used to remove *Cx. quinquefasciatus* mosquito populations in Myanmar [30]. This strategy, known as the incompatible insect technique (IIT), involves releasing massive numbers of *Wolbachia*-inflamed male mosquitoes, which compete with wild-type males and induce sterility, thus suppressing the mosquito population. To suppress *Ae. Albopictus*, a triple *Wolbachia*-inflamed strain (wAlbA, wAlbB, and wPip inflamed) has been generated [31], and MosquitoMate, a biotech organisation, has been pioneering the usage of IIT using *Ae. Albopictus*, with ongoing releases of inflamed male mosquitoes. In case of *Ae. aegypti*, where *Wolbachia* are not found naturally, scientists have introduced *Wolbachia* into the eggs of *Ae. Aegypti*. When these male mosquitoes with *Wolbachia* mate with wild mosquitoes which don't carry *Wolbachia*, the eggs don't hatch. As a result the number of *Ae. Aegypti* decreases. Also, introduction of *Wolbachia* (wMel strain) into *Ae. Aegypti* mosquitoes reduces their capacity to transmit dengue and other arboviruses. The exact mechanisms for this are unclear, but according to some experts *Wolbachia* outcompetes the virus for resources such as lipids, or turbocharges the host's immune response. Ref [32] in their study found that *Wolbachia* deployments were associated with a 69 per cent reduction in dengue cases in Niteroi, Brazil compared to the control plot. Further, studies on *Ae. aegypti* have shown that every one *Wolbachia* strains exhibit near 100 per cent maternal transmission rates and set off excessive ranges of CI [33]. The wMel *Wolbachia* infection, delivered into *Ae. Aegypti* from *Drosophila melanogaster*, efficaciously invaded natural *Ae. Aegypti* populations in Australia, spreading unexpectedly following releases of wMel-infected adults [34]. Encouraging outcomes from those preliminary trial releases have led to subsequent releases in countries experiencing high dengue instances, including Indonesia, Vietnam, Colombia, and Brazil (www.Eliminatedengue.Com). As this *Wolbachia*-based technique gains traction, issues approximately the capability improvement of resistance to *Wolbachia*'s inhibitory outcomes were raised. Since the strategy remains in its early ranges, further research is wanted to determine the simplest *Wolbachia* strains or combinations for dengue manipulation. Ongoing collaboration between researchers, corporations like MosquitoMate, and other such mosquito control programmes can be important in refining and optimizing this innovative method.

III. THE STERILE INSECT TECHNIQUE

The Sterile Insect Technique (SIT) is a highly effective genetic suppression strategy utilized against agricultural pests, but its application to combat mosquito-borne diseases has been constrained by reduced performance of sterilized males and challenges in reducing wild population densities. SIT involves mass rearing and sterilization of male insects through irradiation or chemosterilizing agents, ensuring that when released, they produce no viable offspring when they mate with wild females. The first effective applications of SIT to mosquitoes were done in 1960s and 1970s with pilot trials against *Cx. quinquefasciatus* [35] and *An. quadrimaculatus* in Florida, USA [36], and *An. albimanus* in El Salvador, Central America [37]. The introduction of 8,400 to 18,000 male *Cx. pipiens quinquefasciatus* Say, treated with a sterilizing agent called thiotepa, successfully controlled and eradicated the native population of this mosquito species on an island near Florida within a span of 10 weeks. The sterile males exhibited remarkable capability in locating and mating with the female mosquitoes present on the island [35]. Development and improvement of the technical steps have led to international interest in using SIT against some major vector species of *Plasmodium* spp. (*An. arabiensis*) and dengue virus (*Ae. albopictus* and *Ae. aegypti*). Over the past 50 years, SIT has achieved remarkable success in eradicating pest populations, such as the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), Mexican fruit fly *Anastrepha ludens* (Loew), the New World screwworm fly, *Cochliomyia hominivorax* (Coquerel)), and the Cactus moth, *Cactoblastis cactorum* [38]. However, for mosquito control, particularly targeting *Aedes* species that transmit human diseases, the efficacy of SIT has been limited due to the suboptimal performance of sterilized males caused by the sterilization process itself. Additionally, a significant challenge lies in the initial phase of reducing wild mosquito population densities before introducing the sterile males. This hurdle has impeded the successful implementation of SIT programs and other mosquito suppression strategies aimed at eradication [39].

IV. GENETICALLY MODIFIED MOSQUITOES

In the quest for effective and environmentally-friendly methods to control mosquito populations and combat mosquito-borne diseases, innovative genetic engineering approaches have emerged. One such technique is called Release of Insects Carrying a Dominant Lethal (RIDL), which was developed by the British biotech firm Oxitec [40]. This technique involves the introduction of a self-limiting gene into mosquito populations, rendering them unable to produce viable offspring. The RIDL approach works by incorporating a lethal gene into male mosquitoes, which is only repressed in the presence of an antidote called tetracycline. Mosquitoes carrying this gene are reared in controlled facilities, ensuring they reach adulthood before being released into the wild. Once released, these genetically modified male mosquitoes mate with wild females. However, since the lethal gene is not actively repressed in the wild, their offspring die at the larval stage, effectively reducing the mosquito population. What makes RIDL particularly advantageous is its species-specificity, similar to other methods such as Incompatible Insect Technique (IIT) and Sterile Insect Technique (SIT). Additionally, this method has minimal long-lasting effects on the target mosquito population, as its primary goal is to suppress or eliminate the population in the release area. Field trials conducted in the Cayman Islands in 2009-2010 with a RIDL strain of *Ae. aegypti* (OX513A) demonstrated successful suppression of the local wild mosquito population [41]. Similar results were observed in Malaysia, where OX513A males exhibited comparable

longevity and dispersal capabilities [42]. Furthermore, recent releases of OX513A males in Brazil resulted in substantial suppression of the target wild population. Another potential method to control mosquito populations involves the manipulation of sex ratios by inducing an extreme male-biased sex ratio. This can be achieved through genetic modification that leads to preferential breakdown of the X chromosome during male meiosis. For example, in *An. gambiae*, a major malaria vector, synthetic distorter male mosquitoes have been created, producing over 95 per cent male offspring. The breakdown of the paternal X chromosome prevents its transmission to the next generation, resulting in fully fertile mosquito strains with predominantly male progeny [43]. Apart from these, In 2014, a groundbreaking technology emerged, merging gene drives with CRISPR-Cas9, allowing genes to rapidly spread through populations by increasing inheritance rates. This innovation, exemplified by Kyrou et al.'s work on the *A. gambiae* mosquito, targeted the doublesex gene, resulting in female mosquitoes developing male and female organs, rendering them infertile and unable to transmit the *P. falciparum* parasite [44]. Though promising, the introduction of such genetically modified organisms raises environmental concerns. Eradicating a species or facilitating crossbreeding could have unforeseen repercussions on ecosystem dynamics and gene dissemination into other species [45]. Careful evaluation of the potential risks and ethical considerations is essential before deploying these gene drives on a larger scale. Striking a balance between public health benefits and environmental protection is crucial to harnessing the full potential of this technology responsibly and sustainably.

V. THE “LURE AND KILL” TECHNIQUE

This novel technique, proposed for mosquito control, particularly targeting *Anopheles* species [46], exploits visual stimuli to attract mosquitoes to swarming sites. By manipulating artificial swarm markers or landmarks, it becomes possible to disrupt or enhance swarms, creating designated "kill zones" where large numbers of attracted mosquitoes can be exterminated [47]. This innovative approach shows promise in combatting mosquito-borne diseases, providing a potential breakthrough in vector control strategies.

VI. SOUND TRAPS

In the summer of 1948, an intriguing field experiment took place in Cuba, where researchers recorded and reproduced the sounds produced by single females of *An. albimanus*, a species of mosquito. These reproduced sounds were transmitted through a loudspeaker enclosed within an electrically-charged screen placed in the mosquitoes' natural swamp habitat. The aim was to attract male *An. albimanus* mosquitoes, and it proved successful, with several hundred males being lured and subsequently electrocuted by the charged screen [48]. Despite this early success, subsequent field trials with sound traps encountered limitations. One major challenge was the technical difficulty in designing a sound trap with sufficient amplification to attract mosquitoes from long distances. Furthermore, the positioning of the traps was crucial, and they showed better efficacy when placed in close proximity to swarming sites.

VII. MOSQUITO REPELLENTS/ DETERRENTS

Use of repellents against insect pest is not a new approach. Since ancient times there has been reports of using repellents for deterring insects. These repellents represent an

important tool in the fight against mosquito-borne disease. However, their use in integrated pest management approaches have been largely neglected. Before the discovery of synthetic chemicals, botanical extracts and mechanical barriers were mainly used by the people to prevent bites from arthropods. Some of the most successful plant extracts against mosquito bites were citronella, cassia, cedar, lavender, eucalyptus and neem treeoil [49, 50]. A six-month field study in Pakistan found that repellent soap containing DEET was extremely effective in reducing *P. falciparum* cases when compared to the control [51]. Numerous DEET-containing formulations are used as contact repellents and they effectively repel a wide spectrum of hematophagous insects. With respect to spatial repellents, numerous products are available. These include transfluthrin, metafluthrin, and various botanical oils or compounds (e.g., oils of citronella, peppermint, lemongrass) volatilized into a head space with the goal of repelling biting mosquitoes [52].

VIII. CONCLUSION AND FUTURE PROSPECTIVE

Innovative technologies offer promising solutions for mosquito control, addressing the burden of mosquito-borne diseases sustainably. Strategies range from gene editing and Wolbachia introduction to attract-and-kill methods and drone applications, showing potential in reducing mosquito populations and disease transmission. However, responsible implementation demands rigorous testing, safety measures, and ethical considerations. Collaboration between scientists, public health organizations, and engagement with local communities are vital for success. Developing eco-friendly, safe, and sustainable biocontrol strategies is essential to reduce reliance on insecticides. Effective mosquito predators and pathogenic bacteria like Bti can be used. Wolbachia, which targets DENV transmission, shows significant promise, but large-scale trials are crucial. Synergistic strategies like SIT, RIDL, and Wolbachia-induced IIT may be necessary for population suppression. Understanding mosquito behavior, such as mate searching and swarming, can enhance control programs. While genetically engineering organisms may be justifiable for public health, ethical concerns arise, especially if extinction risks outweigh benefits. Informed decision-making through detailed analysis of consequences is crucial for such interventions. Striking a balance between progress and responsible practices is essential to create a healthier, mosquito-safe world.

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