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Potential biological agents for control of mosquitoes

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Abstract

There are about 3,000 different kinds of mosquitoes, and together they kill millions of people every year. Most species include adult females that use a proboscis to feed on the blood of a wide variety of victims, including mammals, birds, reptiles, amphibians, fish, and even other arthropods. They don't eat the blood, however; it's used to give their eggs a protein boost. As a result of their ability to bite and consume viruses, several species are considered vectors of illnesses such as malaria, dengue, chikungunya, zika, encephalitis, yellow fever, filariasis, west nile virus, and many more. Predators, diseases, parasites, or even rivals may all play a role as biocontrol agents. Biocontrol agents come in many forms, such as fish, frogs, lizards, mosquitoes (Including cannibalistic species), dragonflies, insects, mites, copepods, helminths, planarians, bacteria, fungi, etc. This article discusses the current and historical state of various biocontrol techniques, as well as their prospective use in the fight against mosquitoes.

Keywords: Helminths, zika, mosquitoes, biocontrol agents, predators

Introduction

Mosquitoes are members of the Culicidae family of Diptera. They are the most numerous class of hemophagous insects, and they spread disease not only to humans but also to other mammals, birds, fish, and reptiles. There are approximately 3,200 distinct kinds of mosquito, and they may be found almost everywhere except Antarctica, which is permanently frozen over.

Mosquitoes are crucial to medicine target because they spread dangerous viruses that may cause serious illness in people, including malaria, dengue fever, yellow fever, Chikungunya, Zika virus, and Japanese encephalitis ^[1]. Approximately 55 percent of the population in 124 tropical and subtropical countries is at risk for these illnesses ^[2]. There are ten different anopheline species that may spread malaria in India, but only six are really crucial. *Anopheles culicifacies* is the most common vector in rural regions, whereas *Anopheles stephensi* is the most common vector in cities.

According to the World Malaria Report (2021), malaria affects 36 percent of the global population, or 0.241% of the world's population in 107 nations. The World Health Organization (1981) estimates that in South East Asia, malaria threatens the lives of 2.5 million people, or 85.7%. Of the 2.5 million cases documented in South and East Asia, about 70% are attributable to India alone ^[3]. *Culex quinquefasciatus* is the main vector for about 102 million cases of filariasis each year. Over 90% of persons who live in Lymphatic filariasis hotspots have either patent micro filariae or chronic filarial illness ^[4]. This affects over 1,100,000,000 people. Nearly 100 million instances of dengue fever, 500,000 cases of dengue hemorrhagic fever, and 24,000 fatalities occur year due to dengue infection, which is endemic in more than 100 countries ^[5, 6]. Vector mosquito management is the only proven method for preventing and controlling malaria and other vector-borne infections, according to the World Health Organization. Prior to the discovery of DDT's insecticidal qualities in 1939, there was considerable interest in biological treatments for controlling mosquito populations. Due to the dangers to human health and the environment posed by chemical insecticides, safer options must be found. At this time, eliminating breeding grounds for mosquitoes and taking precautions against being bitten by them are the most effective means of reducing the spread of these illnesses.

Chemical insecticides, botanical insecticides, and biological control agents are the mainstays of vector control techniques [7]. Several chemical pesticides have been used successfully to reduce mosquito populations during the last several decades. The overuse of chemical pesticides has been linked to the resurgence of mosquito populations because it interferes with natural biological control mechanisms. As a consequence, it prompted a search for other control strategies due to resistance development in target species, negative impacts on non-target organisms, and human health issues [8].

Most of the public health problems caused by mosquito-borne illnesses may be mitigated by the use of vector control tactics. Prior to the development of synthetic pesticides, efforts were made to reduce mosquito populations using environmental management techniques, such as the removal of standing water and the installation of screens in doors and windows [14, 9]. The World Health Organization (WHO) developed the EMVs (Environmental Management for Vector Control) in 1982 as a component of the Concept of Integrated Vector Control.

The goal of environmental management operations for vector control is to prevent or reduce vector propagation and vector-pathogen contact by modifying environmental factors or their interactions with humans [10, 11]. DDT, the first persistent pesticide, was introduced to the arsenal for controlling vectors after WWII [9]. Pesticides, such as dieldrin, pyrethrin, and others, were created for use both inside and outdoors. Bed netting with built-in insecticides were available later [9]. The use of synthetic chemical pesticides for vector control is on the decline because to their high prices, the evolution of resistance in many target populations, and the perception of dangers to the environment and human health. There is still a need for chemical pesticides in vector control programs, but the problems they cause and the slow pace at which new types are developed have piqued interest in non-chemical methods for some time. When faced with such a situation, it was determined that biological control tactics were the most effective course of action for reducing vector populations.

The World Health Organization (WHO) defines "integrated control" as "the combined use of biotic, physical, and chemical measures to supplement natural enemies and other factors regulating populations." Biological control is a strategy for managing pests that makes use of living creatures and requires the participation of humans.

Larviciding and adulticiding are two forms of biocontrol. Predators, viruses, parasites, and competitors are all examples of biocontrol agents. Importation, in which a natural enemy of the pest is introduced to control them, Augmentation, in which a large population of natural enemies is introduced for quick control, and Conservation, in which natural enemies are maintained by regular reestablishment, are the usual three strategies used in biological control. Insecticides used extensively in the past have been outlawed by environmental protection organizations, yet chemical control is still widely utilized. Insect resistance to insecticides, fewer adverse effects, and lower costs explain why biocontrol methods are more popular than chemical agents today. Attacks on non-target species are another way in which biological management may have unintended consequences for biodiversity [12].

Fungi in biological control of mosquitoes

For many years, scientists have been looking for mosquito-specific diseases that might be employed in mosquito control efforts. The fungi that have been demonstrated to be effective

as mosquito larvicides have been studied both in the lab and in the field. Fungi are being examined for use in the microbial control of mosquito larvae, and there are several species under consideration. Mosquito larvae may be vulnerable to the biocidal effects of some fungi, including those belonging to the genera *Coelomomyces*, *Lagenidium*, *Metarhizium*, *Culicinomyces*, *Entomophthora*, etc. [13].

However, the method of action of these fungi has been documented by researchers, and it shows that the cuticle and abdomen of mosquito larvae are the primary targets [14]. The spores of some fungi stick to the cuticle of mosquito larvae at first. After that, the spore is formed, and it penetrates the cuticle to reach the hemocoel, where growth and development take place (2009). Saprophytic feeding begins, fungi reappear, and the larva eventually perishes. Both of them are potential entry points for the fungus that kills the larvae (Seye *et al.*, 2009/15). When dealing with fungal formulations, these are the most common areas of infection [16]. Fungi kill larvae by attaching to and growing within the perispiracular valves of the siphon tube, preventing the larvae from breathing [14].

Lagenidium, *Coelomomyces*, *Entomophthora*, *Culicinomyces*, *Beauveria*, and *Metarhizium* species get the most of the attention.

Planarians in biological control of mosquitoes

Planarian species *Dugesia bengalensis* may be used to effectively eradicate mosquito populations by feeding on mosquito larvae. Producing a big number of mosquito predators is a useful strategy for biologically regulating mosquito growth and checking its population. Planarians, the free-living helminths, are crucial in this regard.

Stagnant water containing other microbes (34,) and ponds and lakes are ideal for cultivating these species. These creatures are carnivorous by nature reproduce asexually and can survive in temperatures as low as 32 degrees Celsius.

The term feeding mode can be defined as how aquatic invertebrates acquire their food in nature. We use the term habit to refer to how individuals maintain their location or existence (i.e. planktonic, clingers, etc) or move (i.e. swimmers, divers, etc) (34)

Mosquito larvae, along with other aquatic invertebrates, have evolved morphological adaptations that allow behavioral flexibility for feeding on diverse resources (35, 36). Thus, several investigations have described different feeding modes and modifications of the larval mouth parts and their relationship to different modes (37, 38).

Beetle in Biological Control of Mosquitoes

There are four phases in a mosquito's life cycle: egg, larva, pupa, and adult. Due to the fact that the first three stages occur only in water, they were able to aid in the mosquito control effort by preying on aquatic insects. Most aquatic insect predators are members of the insect groups Coleoptera, Diptera, Hemiptera, and Odonata, and they feast on mosquito larvae. Some of these predators are generalists that feed on a wide variety of prey (Polyphagous), while others have more limited diets (Oligophagous) or even no prey at all (Monophagous) and are thus classified as specialist predators. Polyphagous organisms are the norm for mosquito larvae (50). Depending on the structure of their mouths, these predators use a variety of techniques to predate their prey. Chewing mouthparts allow several predators in the order Odonata to consume their prey. The bodily fluid (hemolymph) of their

victim is sucked by predators like beetle larvae and Hemiptera, who have sucking mouth parts. The larvae and pupae of mosquitoes are preyed upon by a wide variety of aquatic

insects. This includes many species of Coleoptera (especially the Dytiscidae), Hemiptera (especially notonectids), and Odonata (51).

Table 1: List of fungal species reported against mosquito.

S. No.	Name of fungi	Mosquito species	Reference
1.	<i>Beauveria bassiana</i>	<i>Culex sp.</i> <i>Aedes sp. Anopheles Sp.</i>	Clark <i>et al.</i> , 1968 ^[17]
2.	<i>Beauveria tenella</i>	<i>Aedes aegypti Aedes dorsalis</i>	Pinnock <i>et al.</i> , 1973 ^[18]
3.	<i>Crypticola clavulifera</i>	<i>Aedes aegypti</i>	Frances <i>et al.</i> , 1989 ^[19]
4.	<i>Entomophthorales</i>	<i>An. stephensi Culex pipiens Culex spp</i>	Kramer, 1982 ^[20] ; Roberts.1974 ^[21] , Roberts and Strand, 1977 ^[22]
S. No.	Name of fungi	Mosquito species	Reference
1.	<i>Beauveria bassiana</i>	<i>Culex sp.</i> <i>Aedes sp. Anopheles Sp.</i>	Clark <i>et al.</i> , 1968 ^[17]
2.	<i>Beauveria tenella</i>	<i>Aedes aegypti Aedes dorsalis</i>	Pinnock <i>et al.</i> , 1973 ^[18]
3.	<i>Crypticola clavulifera</i>	<i>Aedes aegypti</i>	Frances <i>et al.</i> , 1989 ^[19]
4.	<i>Entomophthorales</i>	<i>An. stephensi Culex pipiens Culex spp</i>	Kramer, 1982 ^[20] ; Roberts.1974 ^[21] , Roberts and Strand, 1977 ^[22]
5.	<i>Fusarium culmorum</i>	<i>Culex pipiens</i>	Badran and Aly, 1995 ^[23]
6.	<i>Leptolegnia chapmanii</i>	<i>Aedes aegypti Culex quinquefasciatus Anopheles albimanus Anopheles quadrimaculatus</i>	McInnis and Zattau, 1982; ^[24] Lord and Fukuda, 1990; ^[25] McInnis and Zattau, 1982 ^[24]
7.	<i>Pythium carolinianum</i>	<i>Aedes albopictus Culex quinquefasciatus</i>	Su <i>et al.</i> , 2001 ^[26]
8.	<i>Smittium morbosum</i>	<i>Anopheles hilli Aedes albifasciatus</i>	Sweeney, 1981d ^[27] Garcia <i>et al.</i> , 1994 ^[28]
9	<i>Tolypocladium cylindrosporum</i>	<i>Aedes aegypti</i> <i>Aedes vexans</i> <i>Culiseta inornata</i> <i>Culex tarsalis Ochlerotus triseriatus</i>	Goettel, 1988 ^[29] Goettel, 1987 ^[30] , Goettel 1987 Soares, 1982 ^[31] Nadeau and Biosvert, 1994 ^[32]
10.	<i>Verticillium Lecanii</i>	<i>Ochlerotatus triseriatus</i>	Ballard and Knapp, 1984 ^[33]

Table 2: Feeding Mode of Different Species

Species name	Habitat	Predominant Feeding Mode	References
Most Anopheles spp	Lentic: lake edges, swamps and marshes, shallow permanent ponds; lotic: flowing streams depositional areas	Collecting-filtering	[39, 40]
<i>Culex spp., Culiseta spp., Orthopodomyia spp., Uranotaenia spp., Aedes spp. (in part)</i>	Lentic: ponded streams, lake edges, swamps and marshes, shallow permanent ponds, in part) termittent ephemeral puddles, natural (phytotelmata) and artificial containers, subterranean habitats	Collecting-filtering	[41, 42]
<i>Coquillettidia spp. Mansonia spp.</i>	Lentic: swamps and marshes, shallow permanent ponds (on vascular hydrophytes)	Collecting-filtering	[43, 44]
<i>Aedes spp. (inpart), Psorophora spp., Haemagogus spp., Wyeomyia spp.</i>	Lentic: swamps and marshes, shallow temporary pools, intermittent ephemeral puddles, natural containers (phytotelmata), subterranean habitats	Collecting- gathering	[45, 46]
<i>Aedes atropalpus</i>	Lentic: intermittent ephemeral puddles (rock holes and pools), artificial containers (tires)	Scraping	[47]
<i>Aedes triseriatus, Aedes aegypti, Culex bitaeniorhynchus group, Culiseta in-ornata, Culiseta longiareolata, Tripteroides spp.</i>	Lentic: ponded streams, marshes, natural and Artificial containers (e.g. tree holes, tires, water jars)	Shredding	[48, 49]

Table 3: List of Beetles as Mosquito Control

Name	Mosquito Spp	Reference
<i>Acilius sulcatus</i>	<i>Cx. quinquefasciatus</i>	Chandra <i>et al.</i> , 2008b ^[52]
<i>Lestes congener</i>	<i>Culiseta incidens</i>	Lee, 1967 ^[53]
<i>Toxorhynchites splendens</i>	<i>Cx. quinquefasciatus</i>	Aditya <i>et al.</i> , 2006 ^[54]
<i>Ischnura forcipata</i>	<i>Cx. quinquefasciatus</i>	Mandal <i>et al.</i> , 2008 ^[55]
<i>Orthemis ferruginea</i>	<i>Ae. aegypti</i>	Sebastian <i>et al.</i> , 1980 ^[56]
<i>Notonecta sellat</i>	<i>Cx. Papiens</i>	Fischer <i>et al.</i> , 2012 ^[57]
<i>Ranatra elongata</i>	<i>Cx. quinquefasciatus</i>	saha <i>et al.</i> , 2020 ^[58]
<i>Mesocyclops thermocyclopoides</i>	<i>Ae. Aegypti</i>	soto <i>et al.</i> , 1999 ^[59]
<i>Mesocyclops australiensis</i>	<i>Aedes sp</i>	Brownetal.,1991 ^[60]
<i>Diplonychus annulatus</i>	<i>Cx. quinquefasciatus</i>	Saha <i>et al.</i> , 2007 ^[61]

Fish in Biological Control of Mosquitoes

Biological control, which includes a wide range of organisms that help to control mosquito populations naturally through predation, parasitism, and competition, is increasingly the focus of malaria control programs in both developed and developing countries, particularly in urban and peri urban areas. The employment of larvivorous fish in a mosquito control program was likewise determined to be the most effective method. The introduction or manipulation of organisms is known as biological control, and it is used to reduce the prevalence of vector-borne diseases. The use of larvivorous fish as a biological mosquito control agent dates

back to the early 1900s (62) and is widespread around the globe. Fish, amphibians, copepods, odonates, water bugs, and even the larvae of different mosquito species are among the numerous aquatic creatures that feast on mosquito larvae and pupae.

To combat mosquito populations, numerous nations have released larvivorous fish species as *Gambusia*, *Poecilia*, *Carassius*, *Aplocheilus*, etc. Several species of copepods have been documented as predators of mosquito larvae, including *Cyclops vernalis*, *Megacyclops formosanus*, *Mesocyclops guangxiensis*, *Mesocyclops longisetus*, and *Mesocyclops thermocyclopoides* [63].

Table 4: List of Fish as Mosquito Control

Larvivorous fishes	Mosquito species	reference
<i>Aphanius dispar</i>	<i>Culex quinquefasciatus</i> . <i>Anopheles arabiensis</i> . <i>Anopheles gambiae</i> .	Fletcher 1992 [64] Haqand Srivastava, 2013 [65] Imbahale <i>et al.</i> 2011 [66] Ataur-Rahim, 1981 [67]
<i>Gambusia affinis</i>	<i>An. subpictus</i> . <i>Cx. quinquefasciatus</i> . <i>An. subalbatus</i> . <i>Ae. aegypti</i> . <i>An. stephensi</i> . <i>An. gambiae</i> .	Chatterjee and Chandra, 1997 [68] Zvantsov, 2008 [69]
<i>Puntius tetrazona</i>	<i>Cx. Vishnui</i>	Barik <i>et al.</i> , 2018 [70]
<i>Oreochromis mossambica</i> (Tilapia)	<i>Cx. quinquefasciatus</i> . <i>An. Culicifacies</i>	Sharma and Ghosh, 1989 [71]
<i>Poecilia reticulata</i> (Guppy)	<i>An. subpictus</i> . <i>An. gambiae</i> . <i>An. subpictus</i> . <i>An. Funestus</i>	Sitaraman <i>et al.</i> , 1976 [72]
<i>Chanda nama</i>	<i>An. Culicifacies</i> , <i>An. balabocensis</i> <i>balabocensis</i> <i>An. Varuna</i>	MESV, 1988 [73]
<i>Colisa sota</i>	<i>An. annularis</i> .	[73]
<i>Colisa lalia</i>	<i>An. annularis</i>	[73]
<i>Colisa fasciatus</i> .	<i>Anopheles annularis</i>	[73]
<i>Aplocheilus panchax</i>	<i>An. Culicifacies</i> , <i>An. sundaicus</i> . <i>Cx. Quinquefasciatus</i> . <i>Cx. Vishnui</i>	[73]

There are a plethora of native and non-native larvivorous fishes that might be used as biocontrol agents, including *Aphanius dispar* (Dispar topminnow) and *Aplocheilus lineatus*. The panchax minnow (*Aplocheilus panchax*), the giant gourami (*Colisa fasciatus*), the dwarf gourami (*Colisa lalia*), and the sunset gourami (*Colisa sota*), the elongated glass perchlet (*Chanda nama*), the estuarine rice fish (*Oryzias melastigma*), the zebra danio (*Danio rerio*), the spiked paradise fish [74].

Bacteria in biological control of mosquitoes: Forty percent of insect species are naturally infected with the Wolbachia, which are endosymbiotic bacteria. Invasive and persistent

Wolbachia infections in mosquitoes have been shown to shorten mosquito longevity, affect reproductive, and impede the spread of pathogens. To curb the spread of dengue illness, *Aedes aegypti* mosquitoes infected with Wolbachia have been released in certain regions of Australia. Gram-positive, aerobic, entomopathogenic soil bacteria *Bacillus thuringiensis* serotype *israelensis* (BTI) and *Bacillus sphaericus* produce insecticidal toxins that specifically kill mosquito larvae.

Mosquito larvae absorb the bacterial strain's crystal toxin via their digestive systems. It interacts to particular receptors in the brush border membrane of the midgut and is activated from a protoxin by a protease (Silva-Filha *et al.*, 2021 75). Then the poison is taken within the cell, and eventually the cell dies.

Table 5: List of Bacterial Strain as a Mosquito Larvicide

Bacterial strain	Effective against mosquito species mosquito	References
<i>Bacillus thuringiensis</i>	<i>Anopheles</i> sp.	Balakrishnan <i>et al.</i> , 2015 [76]
<i>Bacillus sphaericus</i>	<i>Culex</i> and <i>Anopheles</i>	Balakrishnan <i>et al.</i> , 2015 [76]
<i>Bacillus alvei</i>	<i>Culex fatigans</i> , <i>Anopheles stephensi</i> , <i>Aedes aegypti</i> .	Balakrishnan <i>et al.</i> , 2015 [76]
<i>Escherichia coli</i>	<i>Culex</i> mosquito	Jenkins, 1964 [77]
<i>Bacillus cereus</i>	<i>Anopheles</i> sp. and <i>Culex</i> sp.	Balakrishnan <i>et al.</i> , 2015 [76]
<i>Pseudomonas fluorescens</i>	<i>Anopheles stephensi</i> , <i>Cx. quinquefasciatus</i> , <i>Ae. aegypti</i>	Jenkins, 1964 [77]
<i>Aneurinibacillus aneurinilyticus</i>	<i>Anopheles subpictus</i> , <i>Cx. quinquefasciatus</i> , <i>Aedes aegypti</i>	Das <i>et al.</i> , 2016 [78]
<i>Bacillus sphaericus</i>	<i>Anopheles subpictus</i> , <i>Cx. quinquefasciatus</i> , <i>Armigeres subalbatus</i>	Das <i>et al.</i> , 2017 [78]
<i>Saccharopolyspora spinosa</i>	<i>Ae. albopictus</i>	Liu <i>et al.</i> , 2004a [79]
<i>Bacillus circulans</i>	<i>Cx. quinquefasciatus</i> and <i>Anopheles gambiae</i> and <i>Aedes aegypti</i> .	Darriet and Hougard, 2002 [80]

Only one family of obligatory insect parasites, the Mermithidae, has been discovered among the five orders of Nematoda's fourteen families. *Romanomermis iyengari* (Mermithidae) is one of several species of entomopathogenic nematodes which parasitize and kill mosquito larvae^[81]. Nematodes are classified as parasitic insects that are either obligate or facultative in their feeding style. There are five orders under the Phylum nematode, and 14 obligative parasitic families under those orders. Out of those, the only family found in the mosquito natural habitat is Mermithidae. The Mermithidae family is only one of nine nematode families with parasitic, lethal, or growth-altering species. Nematodes often infect hosts by breaking through the cuticle while insects are feeding on them, or by entering the body via the anus or spiracles^[82]. Allantonematidae, Diplogasteridae, Rhabditidae, Sphaerulariidae, Heterorhabditidae, Neotylenchidae, Steinernematidae, and Tetradonematidae are some of the families that belong to this phylum. The most prevalent nematode species employed to eliminate mosquito larvae are mermithids. Although insects are the primary hosts for Mermithids (they feed as obligate parasites), these creatures will also prey on spiders, crustaceans, mollusks, leeches, earthworms, and other arthropods. They might be unique to a single insect species or to a whole family, and their infection ultimately proves fatal for the host. Much attention has been paid to this family because its members pose little to no ecological risk, are not threatened by competitive beneficial organisms, and can live for extended periods of time in the control site of interest under the right inoculation conditions^[83]. Despite the fact that Mermithids have been found in over 60 different mosquito species, they have not received nearly as much attention. Because of their host specificity, ability to kill the host at a certain development stage, ease of handling, effectiveness as parasites, rapid reproductive rate, and efficient swimming, nematodes are a potentially useful biocontrol agent^[84]. The French Polynesian experience demonstrates that very basic approaches may be incorporated and deployed successfully and affordably despite the ecological obstacles of controlling the *Aedes* vectors of dengue viruses and *Wuchereria bancrofti* in so many dispersed islands.

Anurans in biological control of mosquitoes

With the frog population on the decrease and the possibility of a rise in mosquito density, the question of whether or not to utilize frogs to suppress mosquito larvae is being debated in India.

Since 1972, it has been illegal in India to kill frogs. The restriction on killing frogs has received support from those concerned about the export of frog legs. There's a widespread belief that as amphibian populations drop, mosquito populations rise^[85].

All of a frog's existence is spent in or near water, or else hidden away behind some kind of damp cover like leaves, pebbles, or logs. Most tadpole species consume a wide variety of foods, including bacteria, algae, protozoa, insect larvae, shrimp, eggs, and other amphibian offspring.

Adult frogs of almost all species are carnivores, eating things like annelids, gastropods, and arthropods like mosquitoes. A tiny number of them could even hunt vertebrates like fish, frogs, and small animals. Research indicates that 50 frogs may effectively control insect populations throughout an acre of rice paddy land. Insect populations, such as those of mosquitoes, may therefore be controlled by frogs^[86].

Mosquitoes may reproduce in a wide variety of watery environments, including ponds, marshes, ditches, pools, drains, and many types of containers¹⁴. There seem to be clear breeding preferences among many species. *Aedes aegypti* breeds in home, peri-domestic, and other small water collection including desert coolers^[87, 88]. *Anopheles* spp. are connected with fresh water habitats; *Culex* spp. and *Mansonia* spp. may also be found in contaminated settings; and *Culex* spp. and *Mansonia* spp. Frogs may decrease vector population and vector transmitted illness burden^[89] if placed into mosquito larval rearing environments such as ponds, puddles, tanks, etc. The natural regulation of mosquito larvae is disrupted due to the introduction of the mosquito fish, *Gambusia affinis holbrooki*, which is a predator of eggs, hatchlings, and tadpoles of both the native *Limnodynastes somatus* and the non-native *Bufo marinus* frogs. There may be competition for natural food sources from *Culiseta longiarolata* and young *Bufo viridis*, both of which rely heavily on periphyton for sustenance.^[90] *Rana tigrina* tadpoles prefer the pupal stage, whereas most other mosquito predators choose the early larval stage^[91]. *Hyla septentrionalis* tadpoles preferred feeding on mosquito larvae over other food sources, and this may have something to do with the fact that the number of *Cx. pipiens* was shown to decrease when *Hyla* sp. was present in the field^[92].

Integrated biological control of mosquitoes

Management of mosquito populations using biological means. They demonstrate that natural enemies may play a significant role in controlling mosquito populations by preying on mosquito larvae and pupae in watery habitats. Many aquatic animals, such as fish, amphibians, copepods, odonates, water bugs, and even the larvae of other mosquito species, feed on mosquito larvae and pupae. Many nations have imported larvivorous fishes for biological control of mosquitoes, such as the *Gambusia*, *Poecilia*, *Carassius*, *Aplocheilus*, etc. Several species of copepods have been documented as predators of mosquito larvae, including *Cyclops vernalis*, *Megacyclops formosanus*, *Mesocyclops guangxiensis*, *Mesocyclops longisetus*, and *Mesocyclops thermocyclopoides*. The mosquito species *Toxorhynchites*, often known as the elephant mosquito, is a member of the family *Culex* and feeds mostly on the larvae of other mosquito and other aquatic insect species.

The sterile insect technique (SIT) is a method of suppressing unwanted traits by causing chromosomal abnormalities and dominant fatal mutations in sperm in large numbers of males by the use of chemical sterilization. When these sterilized male insects are released, they cannot reproduce with wild females since they are sterile. Cytoplasmic incompatibility, chromosomal translocations, sex distortion, and gene substitution are other genetic techniques^[93].

Conclusion

Current methods of mosquito control rely significantly on the continuous use of pesticides; thus, biocontrol techniques for mosquito-borne illnesses are necessary.

Methods that are safe for humans and the environment and can be used repeatedly against several mosquito species are needed. The complete eradication of *Ae. aegypti* populations in rural Vietnam demonstrates the efficacy of mosquito predators under the right circumstances. Because of its capacity to preferentially kill mosquito larvae, the pathogenic bacteria *Bt* has been widely employed, and future control efforts may

benefit from the use of other diseases, such as entomopathogenic fungi. The use of *Wolbachia* endosymbiotic bacteria has been explored as a potential innovative strategy for lowering DENV transmission rates. Though promising, *Wolbachia*-based techniques for mosquito biocontrol still need larger-scale research to evaluate their efficacy. Synergistic approaches using several techniques, such as SIT, RIDL, and *Wolbachia*-induced IIT, may be necessary for efficient population suppression.⁽⁹⁴⁾ More research into the chemical ecology of mosquito mate seeking, swarming landmarks, and mate choice in swarming areas is necessary to develop more effective vector control techniques⁽⁹⁵⁾. Several natural items also have been studied for their potential to deter mosquitoes, including *Zanthoxylum armatum* (Rutaceae), *Azadirachta indica* (Maliaceae), and *Curcuma aromatica* (Zingiberaceae)^[96].

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