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(RESEARCH ARTICLE)

Studies on the biocontrol of mosquitoes using *Oreochromis mossambicus*

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Abstract

Mosquitoes cause nuisance to human beings by biting and transmitting vector-borne diseases. Though several control strategies are available for their control, biocontrol holds much promise. Among biocontrol agents, larvivorous fishes are gaining importance. Hence the present study has been undertaken to test the efficiency of *O. mossambicus* in the larval control of *Culex quinquefasciatus.* Experiments were designed with different ecological conditions like prey density, volume of water, container shape, presence of vegetation, predator density and time of the day to test the biocontrol potential of the fish. The fish was able to control the third instar larve of *C. quinquefasciatus* and hence it can be used in mosquito control programmes.

Keywords: *Oreochromis mossambicus*; *Culex quinquefasciatus*; Biocontrol; Mosquitoes; Predatory activity; Hydrilla

1. Introduction

Mosquitoes may be nature's most effective bioterrorists because they transmit some of the world's most life threatening and debilitating parasitic and viral diseases including Malaria (*Anopheles*), filariasis (*Culex, Mansonia* and some *Anopheles* sp.) and Dengue fever (principally *Aedes aegypti*). Alarmingly, these diseases are on the rise in many tropical and sub-tropical areas [1]. These vector-borne diseases are increasing due to the risk of transmission fuelled by developmental activities, demographic changes and introduction of new products. Owing to rapid human movement/migration, diseases are spreading at a much faster rate and the problem is no more of local but global concern [2].

The management of mosquitoes require adequate knowledge about the species diversity, density, distribution pattern and biting behaviour for evolving an appropriate management strategy and to implement the same for the effective control of population and in turn to reduce the menace and the incidence of the diseases in the endemic and epidemic areas [3]. Since the vector and non-vector mosquitoes are increasing in large numbers, it is necessary to use an effective tool and all available management techniques to bring about an effective degree of control in a cost-effective way. In addition to this, the problem of the management of mosquitoes in different regions is not uniform and the variation is mainly due to dynamic and differential nature of the mosquito population [4].

Different control strategies such as use of chemicals, biological agents, repellents, ecomanagement and biocides can be employed to control the mosquito vectors. As biological mosquito control agents, larvivorous fish are being used extensively all over the world since the early 1900s. *Gambusia affinis* has high larvivorous potential and hence it was introduced from its native Texas (USA) to the Hawaiian Islands in 1905. In 1921 it was introduced in Spain, then in Italy during 1920s and later to sixty other countries. A major portion of the National Health budget is spent on the control of mosquito-borne diseases. Various types of control methods have been developed and adopted for vector control but with little success. The task is yet to be addressed expeditiously. Drastic reduction in mosquito/vector population and mosquito-borne diseases can be achieved by judicious use of various available control strategies developed along with

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the involvement of community for defeating man's enemy, the mosquito. Fishes have been used for controlling mosquito larvae and many species have proved effective [5-11]. The present study has been carried out to find the effectiveness of *Oreochromis mossambicus* (Tilapia) in controlling the larvae of *Culex quinquefasciatus*.

2. Materials and methods

Healthy *O. mossambicus* fingerlings were collected from private fish farm, Madurai, Tamil Nadu, India. The fingerlings were acclimated to laboratory conditions for ten days in a well aerated and illuminated tank and maintained under room temperature till the commencement of the experiment. Fishes of uniform weight were chosen for the study. Fishes were fed on fish feed before the experiment, but the fishes were subjected to starvation for one day before the commencement of the experiments.

Mosquito larvae were collected from open drainages at Usilampatty, Madurai district, Tamil Nadu, India and the species was identified as *C. quinquefasciatus*. From the collections, third instar stage was selected for the experiment.

2.1. Prey density

In the first set up of experiments, 500 ml of water was taken separately in five different transparent containers. In containers, 100, 150, 200, 250 and 300 larvae were introduced for one hour experiment. Then one *O. mossambicus* fingerling was introduced in each container and the time was noted. After one hour the fishes were removed and the remaining larvae were counted. From this, the number of larvae consumed was determined. The experiments were conducted in triplicate. Similar experiments were conducted for a period of 24 hours but the numbers of larvae introduced in the containers were 150, 200, 250, 300 and 350.

2.2. Effect of volume of water

In the second set of experiments 400, 500, 600, 700 and 800 ml of water was taken separately in five different transparent containers. One hundred *Culex* larvae were introduced in each container for one hour experiment. Then one *O. mossambicus* fingerling was allowed in each container and the time was noted. After one hour, the fishes were removed, the remaining larvae were counted and the number of larvae consumed was determined. The experiments were conducted in triplicate to find out the influence of water volume. The same experiment was carried out for 24 hours with the prey density of 200.

2.3. Container shape

In the next set of experiments, influence of the shape of the container on the predation was studied. Different shapes of containers such as square, round, rectangle and oval were taken with 500 ml of water. Two hundred *Culex* larvae and one *O. mossambicus* were introduced in each container and the time was noted. After one hour the fishes were removed, the remaining larvae were counted and the number of larvae consumed was determined. The experiments were conducted twice. The same experiment was conducted for 24 hours with 200 larvae.

2.4. Influence of aquatic plants

In the fourth set of experiments 500 ml of water was taken in two different transparent plastic containers. Two hundred *Culex* larvae were introduced in each container. In each container, one *O. mossambicus* was allowed. Few branches of *Hydrilla* plants were put in one container and the time was noted. After one hour the fishes were removed from the respective containers, the remaining larvae were counted and the number of larvae consumed was determined. The experiments were conducted thrice. Similar experiments were conducted for a period of 24 hours to find out the influence of *Hydrilla* plant on the predation.

2.5. Number of predators

In another set of experiments, the influence of number of predators on the predation was studied. For this, 500 ml of water was taken in three separate containers and two hundred larvae were introduced in each container. Then, one, two, three and four *O. mossambicus* fingerlings were allowed individually in each container and the time was noted. After one hour, the fishes were removed, the remaining larvae were counted and the number of larvae consumed was determined. The same experiment was conducted for 24 hours, but the number of larvae introduced in each container was kept as 300.

2.6. Influence of time of the day

In the last set of experiments, the influence of time of the day on the predation of the fish was studied. For this, 500 ml of water was taken in one container with two hundred larvae and one *O. mossambicus* fingerling. The experiment was started at 8 am and continued till 8 am the next day. After every one hour, the fish was removed, remaining larvae were counted and the number of larvae consumed was determined. Again the fish was reintroduced.

3. Results

Biocontrol of the filarial vector mosquito *C. quinquefasciatus* employing the larvivorous fish, *O. mossambicus* was studied. The efficacy in controlling *C. quinquefasciatus* was experimented in different, prey densities, volumes of water, and shapes of the containers, in the presence of *Hydrilla* plant and by increasing the predator densities.

In the case of *O. mossambicus*, maximum predation was at 200 prey density and the number of larvae consumed was 40 after one hour. At the end of 24 hrs, 174 larvae were killed with a percentage of 20 and 87 for one hour and 24 hour respectively (Figure 1). In 250 prey density, after one hour, minimum predation was observed. The number of larvae consumed was only fifteen with a percentage of six. The maximum predation was at the prey density of 150 and the predation rate decreased with the increase in prey densities. *O. mossambicus* feeding decreased with the increase in prey densities.

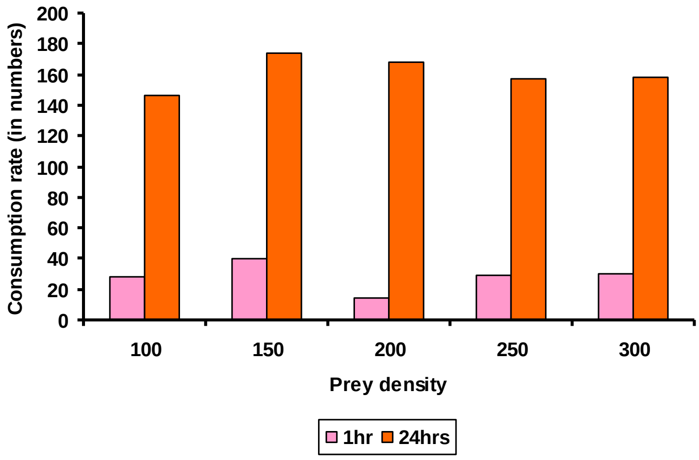

Figure 1 Effect of prey density on the predation of *Oreochromis mossambicus*

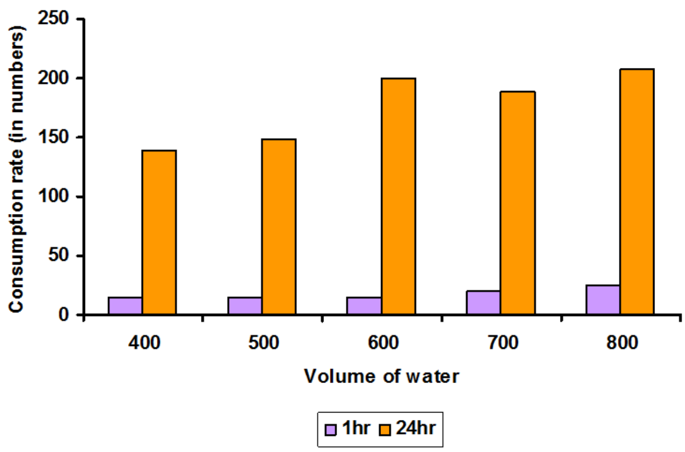

Figure 2 Effect of volume of water on the predation of *Oreochromis mossambicus*

With reference to volume of water, the predation of *O. mossambicus* was maximum at 800 ml and it consumed 208 larvae with the percentage of 83 within 24 hours. Minimum predation was found at 400ml of water as only 139 larvae were killed by *O. mossambicus* with an average of 56 (Figure 2). The predation of *O. mossambicus* was conducive at 600 to 800ml and predation rate was at peak.

The predatory activity of *O. mossambicus* with reference to different shapes of container is shown in Figure 3. It was the maximum in rectangular container with a percentage of 73 and the number of larvae consumed was 182 out of 250. Minimum predation activity was found in round shape with a percentage of 46 in one day. Rectangular shape favoured the predatory activity of *O. mossambicus*.

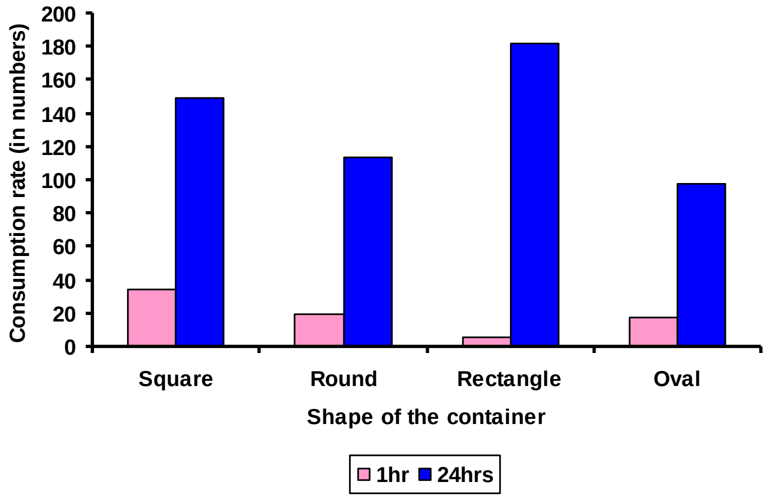

Figure 3 Effect of shape of container on the predation of *Oreochromis mossambicus*

O. mossambicus showed maximum predation in the absence of *Hydrilla*. In one day, it consumed 190 larvae with a percentage of 76 (Figure 4). *O. mossmbicus* showed that in the absence of aquatic plants effective control of mosquito larvae can be done. This shows that they feed on mosquito larvae to a certain extent when chance permits them.

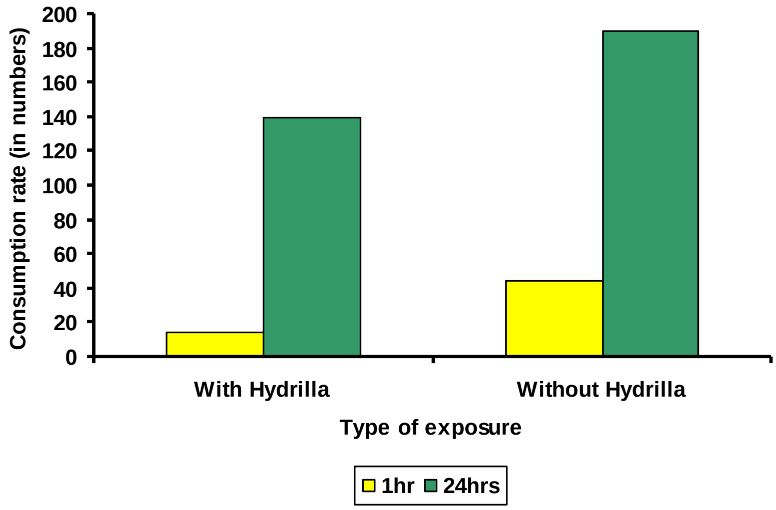

Figure 4 Influence of *Hydrilla* plant on the predation of *Oreochromis mossambicus*

With reference to predator densities, after one hour, the predation was the maximum with two predators which consumed 65 larvae. But at the end of 24 hours, the maximum predation was observed in the group of four *O. mossambicus* with a percentage of 61 (Figure 5). *O. mossambicus* showed that feeding increased with the increase in predator densities.

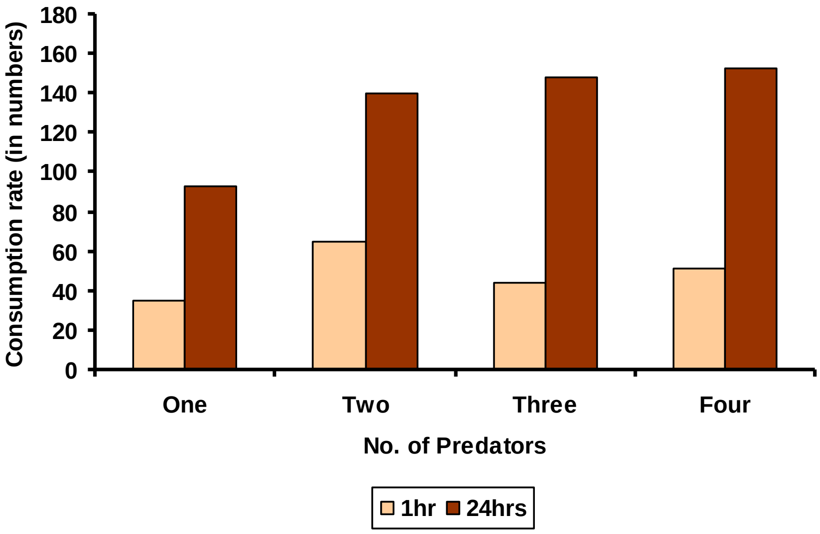

Figure 5 Effect of number of predators on the predation of *Oreochromis mossambicus*

Figure 6 indicates the predatory pattern of *O. mossambicus* for 24 hours continuously exposed to *C. quinquefasciatus* larvae. During first six hours (8-9, 9-10, 10-11, 11-12, 12-13, 13-14) the predation rate was more. After 12 hours, the predation level gradually reduced where *O. mossambicus* consumed only three larvae. Total predation at the end of 24 hours was 219 larvae out of 250 with a percentage of 88. The *O. mossambicus* was feeding on third instar larval forms voraciously throughout the day.

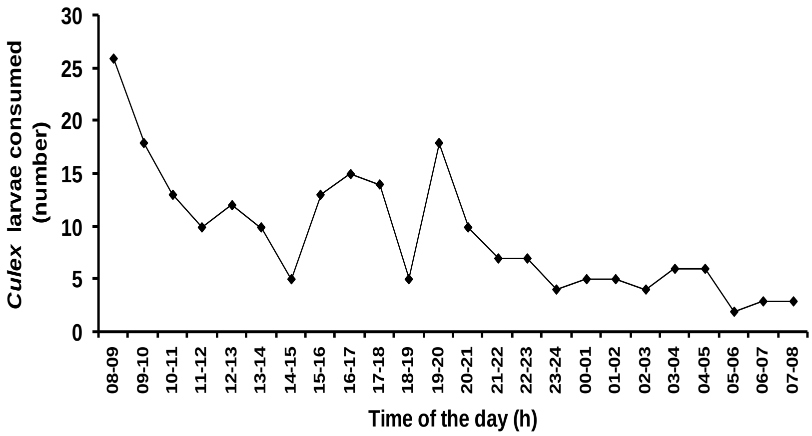

Figure 6 Predatory pattern of the fish, *Oreochromis mossambicus*

4. Discussion

Insects are the most important invertebrate group which serve as beneficial to human beings and other animals on one hand and on the other hand they also act as vectors of several of the world's most devastating diseases. The disease vectors such as tsetse flies, ticks, mosquitoes and sand flies and the parasites they carry, become more and more resistant to chemical spraying and drug therapy. Mosquitoes transmit diseases to more than 700,000,000 people each year. No single method will provide adequate control of mosquitoes and so combination of biological, chemical and environmental management and repellents is needed at present to attack mosquitoes. Biological control is expected to play an increasing role in vector management strategies of the future [12].

During the DDT era, control of mosquitoes and mosquito vectors of different mosquito-borne diseases were undertaken mainly by environmental management, pyrethrum space spraying, use of Paris green, and oiling with petro products. In the fifties attention was directed to eradicate mosquitoes using synthetic insecticides until insecticide resistance began to assume prominence [13]. The importance of climate and weather is obvious since mosquitoes are cold blooded animals and their activity cycles as well as virus replication in their tissues are influenced by temperature. Temperature and relative humidity of the season favour dengue virus propagation in the mosquito and is one of the contributing factors to the occurrence of DHF outbreaks [14, 15]. The predation of *O. mossambicus* was maximum in the prey density of 150. Feeding rate decreased with the increase in prey density. Predation of *Poecilia reticulata* was conducive at 600ml to 880 ml.

In view of growing concern about safety of chemicals, interest is revived on plant products. The phytochemicals may serve as attractants [16]. A large number of plant extracts have been reported to have mosquitocidal or repellent activity against mosquito vectors [17]. In the present study, it was noticed that predator densities increased predation of mosquito larvae. 24hr continuous study indicated that *O. mossambicus* consumed 219 out of 250 third instar larvae of mosquito.

In the present study, biocontrol of mosquito employing the larvivorous fish, *O. mossambicus* was tested. Larvivorous fishes have been used for over hundred years in mosquito control and many species have proved effective. In Western Kenya, the larvivorus fish, *Oreochromis niloticus* is cultured and eaten but has not been previously tested in field for controlling mosquitoes. After *O. niloticus* introduction, mosquito densities immediately dropped in the treated ponds [18]. It has been found that the average consumption rate during 24 hrs study period was appreciable (219/250). Rectangular shape of container favored predatory activity. The larvivorous fish consumed more mosquito larvae in the absence of the aquatic plant, *Hydrilla*.

Biological control of vector mosquitoes (*Anopheles sinensis*) was demonstrated using fish predators, *Moroco oxycephalus* and *Misgurnus anguillicandatus* in the semi field rice paddy. A sustained control of 55% was achieved during the study period [19]. Bellini and coworkers [20] showed efficacy of various fish species (*Caraussius auratus, Cyprinus carpio, Gambusia affinis*) in the control of field mosquitoes in northern Italy. *O. mossambicus* was effective in controlling mosquitoes in cow dung pits, when introduced against III and IV instar larvae and pupae of *C. quinquefasciatus* and *An. culicifacies* [21]. *Oreochromis niloticus* is a native African fish possessing mosquito control properties known since 1917. Under laboratory conditions, this fish species has been shown to be larvivorus with a marked interest in mosquito larvae. The fry actively pursue mosquito immatures however when greater than 150 mm in length they prefer eating macrophytes. Therefore, larger fish eat the plant material in which mosquito immatures hide, allowing the fry to find them [22]. In the present study, *O. mossambicus* voraciously consumed mosquito larvae in the absence of aquatic plants. In the presence of vegetation, the efficacy towards mosquito larva reduced to a little extent. Yu and Lee [23] studied the biological control of the malarial vector (*Anopheles sinensis*) by the combined use of larvivorous fish (*Aplocheilus latipus*) and herbivorous hybrid fish (*Tilapia mossambica*) in the rice field of Korea. Stocking of *A. latipus* and *T. mossambica* in weed infested rice field resulted in an 80-82% reduction in the immature density of *A. sinensis*.

Unlike the chemical pesticides, the results are of unpredictable with biological agents. This calls for a better understanding of the biological interactions with the environment [24]. Awareness is a powerful weapon in the armoury of the mankind in its relentless battle against the forces of morbidity at all stages. Health education has always occupied a lower priority in Indian public policy. An activity associated with health education is the security of social support to enable the effective implementation of disease control measures. Community power and potential in combating vectors remain vastly untapped and underutilized [25]. To promote mosquito and malaria control measures at household and community levels, using primary and secondary schools are the entry point for awareness creation. Preventive strategies used to repel mosquitoes in the houses and dissemination of all the information on mosquito and malaria control through drama performed by the students will be effective [26]. Towards this end, we require a programme of biological research aimed at understanding the factors that limit the number of mosquitoes. The two main factors determining the efficacy of the fish are the suitability of fish species to the water bodies where the vectors breed and the ability of the fish to eat enough larvae of vector species to reduce the number of infectious bites. The fish should be of native, and able to thrive in breeding sites [27]. Aquatic plants influence fish predation and serve as refuge for the larvae of mosquitoes. The larvivorous potential of fish to control mosquitoes may vary due to ecological conditions [28].

5. Conclusion

O. mossambicus exhibits high efficiency in controlling the larvae of *C. quinquefasciatus*. If suitable environmental conditions like predator density and depth of water bodies are maintained, it can perform better in the biocontrol of mosquitoes.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors namely Santha, Appasamy Surendran and Antony Joseph Thatheyus state that the article has not been published in another publication and is not being submitted simultaneously to another journal.

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