LABORATORY EVALUATION OF THE BIOCONTROL POTENTIAL OF *MESOCYCLOPS THERMOCYLOPOIDES* (COPEPODA: CYCLOPIDAE) AGAINST MOSQUITO LARVAE

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Abstract. Biocontrol potential of *Mesocyclops thermocyclopoides* against first instar larvae of *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus* was studied under laboratory conditions. It was found that *M. thermocyclopoides* had the highest predation efficacy against *Ae. aegypti* followed by *An. stephensi* and *Cx. quinquefasciatus*. There was a significant reduction in the predation efficacy of *M. thermocyclopoides* against *Cx. quinquefasciatus* in the presence of alternate food ($p < 0.01$). The cage simulation trial indicated that *M. themocyclopoides* has the potential to control *Ae. aegypti* breeding effectively in a container type of habitat.

INTRODUCTION

Cyclopoid copepods have long been known for their predation upon mosquito larvae (Horlbut, 1938; Bonnet and Mukaida, 1957). However their true biological control potential against mosquito larvae has been established only recently. Rivière *et al* (1987) demonstrated the successful use of *Mesocyclops aspericornis* as a practical and cost effective means for controlling the larvae of *Aedes polynesiensis* and *Ae. aegypti* in French Polynesia. Since then, there has been renewed interest in evaluation of the biocontrol potential of various species of cyclopoid copepods against mosquito larvae (Andreadis and Gere, 1992; Brown *et al*, 1991; Marten, 1989, 1990 a, b). Recently Kay (1996) in a review on use of copepods for contolling vectors of dengue and other vector borne disease, concluded that cyclopoids can provide over 99% control of *Aedes* larvae and can also be used along with *B. thuringiensis* H 14.

In India, about 18 species of cyclopoid copepods have been reported (Joshi, 1996) of which *Mesocyclops leuckarti* (Clause, 1857) and *M. hyalinus* (Rehberg, 1880) are two major vectors of Guinea worm disease. Sharma and Wattal (1981) have reported 7 species of cyclops from the Delhi region which included both the vectors of Guinea worm disease.

While carrying out a survey of breeding habitats of mosquitos in Delhi, we noticed the presence of high densities of copepods at Wazirabad ponds which were later identified as *M. thermocyclopoides* (Harda, 1931). Keeping in view the biocontrol potential of copepods against first instar larvae of mosquitos, it was thought prudent to evaluate the predation efficiency of these cyclopoid copepods.

This paper presents the results of laboratory studies on the predation efficiency of *M. thermocyclopoides* (Harda, 1931) against the larvae of three common mosquitos of urban habitats viz. *Anopheles stephensi* Liston, *Aedes aegypti* Linn and *Culex quinquefasciatus* Say.

MATERIALS AND METHODS

Mesocyclops thermocyclopoides were collected from Wazirabad pond in Delhi and mass cultured in the laboratory using water from a cement tank constructed in the campus of laboratory for undertaking laboratory trials. This water contained both protozoa and planktonic algae which are a natural source of food for *M. thermocyclopoides*. First instar *Aedes aegypti* larvae were provided as supplementary food for the copepods. The mosquito larvae of *An. stephensi*, *Culex quinquefasciatus* and *Ae. aegypti* used in the experiments, were obtained from mosquito colonies being maintained at the Malaria Research Center. For determining Biocontrol potential of *M. thermocyclopoides* a series of experiments were performed in the laboratory.

The first set of experiments was done to find out the predatory potential of copepods by offering a 'choice' of alternative food in the form of plankton (algae and protozoa) and 'no choice' (only first instar mosquito larvae). For 'choice', the experiments were performed by providing 25 larvae to one *Mesocyclops* in plastic bowls of 200 ml capacity with 100 ml water from the tank containing the plankton. The 'no choice' experiments were performed by using the plankton free water obtained by filtering the tank water through a fine nylon cloth. Four replicates were set for each species and the number of larvae consumed/killed in 24 hours was recorded.

The second set of experiments was performed by varying the number of *M. thermocyclopoides* (from 2 to 20) while the number of prey mosquito larvae was kept constant (25). These experiments were also done in 200 ml capacity bowls containing 100 ml of filtered water from the tank.

In the third set of experiment while the number of predators (*M. thermocyclopoides*) was kept constant (50), the number of mosquito larvae varied form 25 to 500 and the water volume was increased to 500 ml in plastic bowls of one liter capacity.

In addition to aforesaid experiments, a trial was made in the laboratory cages to simulate natural condition by placing a container for the breeding of *Ae. aegypti* to find out the impact of *M. thermocyclopoides* on the control of cyclic breeding of *Ae. aegypti*. For this purpose two cloth cages, each having 25 female and 25 male adult mosquitos were taken. An enamel tray containing 2 liters of water was kept in each cage for oviposition. In the experimental cage 200 gravid cyclopids were released in the tray. The female mosquitos were allowed to lay eggs in the tray and the larvae hatching out of the eggs served as prey for the cyclopids in the experimental cage. The larvae which escaped from being preyed upon and developed as adults were fed on rabbits placed inside the cages, as and when needed. Fresh water was added in the tray to maintain the same water level. Dog biscuit and yeast tablets were also added as larval food in both the trays. The density of adult and immature mosquitos in both the cages were recorded at weekly intervals. These observations were continued up to 8 weeks.

RESULTS AND DISCUSSION

In 'no choice'experiments mesocyclops consumed greater number of mosquito larvae as compared to 'choice' experiments (Table 1). The difference was highly significant with *Cx. quinquefasciatus* larvae ($p < 0.01$) but with *An. stephensi* and *Ae. aegypti* the difference was not significant. Maximum predation was recorded against *Ae. aegypti* followed by *An. stephensi* and *Cx. quinquefasciatus* larvae.

Table 2 shows the effect of predator (*Mesocyclops*) density on predation of mosquito larvae. The percent predation upon larvae increased gradually with increased predator density. The copepod density required to produce over 90% predation in 24 hours against larvae of *Ae. aegypti, An. stephensi* and *Cx. quinquefasciatus* was found to be 10, 20 and > 20 copepods/100 ml of water respectively (Fig 1).

Table 1

Predation efficacy of *M. thermocyclopoides* against first instar mosquito larvae in the presence (choice) or absence (no choice) of alternate food (other planktons).

Species	Percent predation (Means ± SE) in 24 hour		Student's *t*-test
	no choice experiments	'Choice' experiments	
Cx. quinquefasciatus	19.0 ± 2.9	9.0 ± 1.2	3.7*
An. stephensi	23.0 ± 4.8	16.0 ± 4.2	1.27**
Ae. aegypti	32.0 ± 8.2	30.0 ± 8.4	0.18**

There were four replicates for each test
* Significant $p < 0.01$
** Not significant

Table 2

Effect of predator density on predation efficacy of *M. thermocyclopoides* against first instar mosquito larvae.

No. of Mesocyclops	Percent predation (Means ± SE in 24 hours)		
	An. stephensi	*Cx. quinquefasciatus*	*Ae. aegypti*
2	34.0 ± 10.7	34.0 ± 19.3	56.0 ± 6
5	57.0 ± 14.3	44.0 ± 17.6	80.0 ± 7.1
10	78.0 ± 8.9	71.0 ± 10.9	95.0 ± 0.6
20	99.0 ± 1.2	88.0 ± 6.5	100.0 ±

Note: The experiment was done in bowls of 200 ml capacity containing 100 ml water with varying predator density.

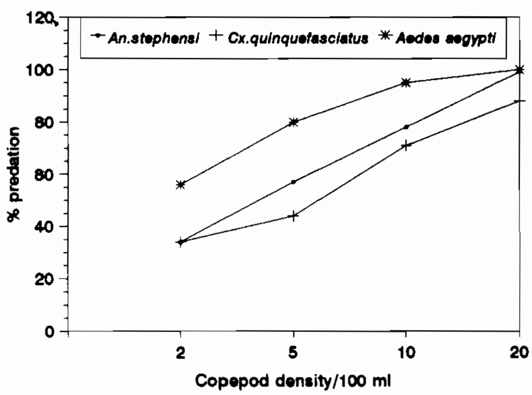

Fig 1–Effect of varying density of *M. thermocyclopoides* on the predation efficacy against first instar mosquito larve.

In experiments with predator number constant (50/500 ml water) and prey variable (25 to 500 larvae) there was a slight reduction in the percent predation against larvae in containers with higher larval densities (Table 3).

In a cage simulation trial, in the experimental cage (with 25 male and 25 female *Ae. aegypti*, and 200 mesocyclops) there was heavy predation on first instar mosquito larvae. After one week only 7 larvae were observed in the tray as compared to 359 larvae in the untreated control. Subsequently the number of emerging (and surviving) adults started declining gradually. After 8 weeks all mosquitos (both larvae as well as adults) in the experimental cage were eliminated as a result of predation by mesocyclops while in the control, 32 adult mosquitos and 678 larvae were left (Table 4).

These results indicate the *M. thermocyclopoides* has a biocontrol potential against larvae of *Ae.*

Table 3

Effect of prey density on predation efficacy of *M. thermocyclopoides* against first instar mosquito larvae. [Predator constant; (50) prey density variable].

No. of larvae	Percent predation (Means ± SE in 24 hours)		
	An. stephensi	*Ae. aegypti*	*Cx. quinquefasciatus*
25	97.0 ± 2.20	99.0 ± 1.54	64.0 ± 6.25
100	73.25 ± 7.58	94.75 ± 3.47	58.75 ± 4.30
200	66.25 ± 5.69	80.5 ± 1.54	48.75 ± 3.25
500	63.35 ± 1.45	78.55 ± 3.5	45.7 ± 3.25

Note: The experiment was done in bowls of one liter capacity containing 500 ml water with varying prey density.

Table 4

Impact of *M. thermocyclopides* on *Ae. aegypti* in a cage simulation trial.

Time (weeks)	No. of mosquitos in cages			
	Experimental cage		Control cage	
	Adults	Immatures	Adults	Immatures
0	50	0	50	0
1	38	7	41	359
2	29	3	32	755
3	27	4	26	888
4	14	12	104	747
5	7	92	151	852
6	5	10	127	947
7	1	0	84	670
8	0	0	32	678

aegypti and also *An. stephensi* even in the presence of alternative food. However, against *Cx. quinquefasciatus*, there was significant reduction in the predation efficacy of *M. thermocyclopoides* in the presence of alternate food. Bapna and Renapurkar (1994) have also reported a similar reduction in the predation efficacy of *M. leuckarti* against *Cx. quinquefasciatus* larvae.

The predation efficacy of *M. thermocyclopoides* against larvae of *Ae. aegypti* is comparable with *M. aspericornis* and other copepod species reported elsewhere (Brown *et al*, 1991). The higher rate of predation against *Ae. aegypti* could be due to the Segregation behaviour of *Aedes* larvae. In 24 hour observation 100 copepods/litre were found sufficient to cause more than 90% predation against *Aedes aegypti* up to a density of 200 larvae/liter. At higher larval densities the percent predation was slightly reduced. However, the cage simulation trial clearly demonstrated the sustained control of *Ae. aegypti* breeding by *M. thermocyclopoides* with an initial density of 100 gravid copepods per liter of water in a container breeding habitat. The sustained control of *Ae. aegypti* breeding was achieved probably due to the rapid growth of the copepods in the presence of sufficient food in the breeding habitat. The overall results of our study indicate the biocontrol potential of *M. thermocyclopoides* against *Ae. aegypti* and *An. stephensi* in containers. It is further advantageous that *M. thermocyclopoides* is not a vector of Guinea worm disease in India (Joshi, 1996). Riviere *et al* (1987) demonstrated 91-99% control of *Ae. aegypti* and *Ae. polynesiensis* breeding in wells, tires, tree holes, drums and crab holes in French Polynesia. In the light of these observations, further studies are required on the role of *M. thermocyclopoides* against *Ae. aegypti* and *An. stephensi* breeding in containers and other small water collection such as, desert coolers and overhead tanks (not used for drinking purpose).

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