

RHYOTHEMIS VARIEGATA LINNAEUS (ODONATA: LIBELLULIDAE): A POTENT BIOCONTROL AGENT AGAINST COMMON MOSQUITO LARVAE IN THRISSUR DISTRICT, KERALA

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ABSTRACT -Predatory efficiency of nymphal stages of dragonfly *Rhyothemis variegata* on the larval instars of *Aedes*, *Anopheles* and *Culex* mosquitoes were evaluated under simulated conditions. *R. variegata* nymphs were collected and were reared by providing mosquito larvae as their food. The observations were conducted between 10.00 AM to 6.00 PM. *R. variegata* showed preference for *Aedes* larvae than *Anopheles* and *Culex* probably because both *R. variegata* and *Aedes* larvae shared a common habitat; stagnant fresh water pools. *R. variegata* mostly consumed third instar larvae of *Aedes* (70.6%) followed by second instar (38.6%) larvae. The maximum consumption rate in *Anopheles* (34.6%) and *Culex* (28.6%) were significantly less when compared to *Aedes*. Percentage consumption was higher for the third instar larvae in all selected mosquitoes ($F < 0.01$). The experiments on prey choice among the four larval stages suggested that prey size affected predatory efficiency. Mortality was observed in the fourth instar larvae of all mosquito species due to the injury caused by *R. variegata* during prey capture. The maximum predation rate was recorded during the initial period of observation between 10.00 AM to 12.00 Noon and gradually decreased.

Index Terms- *R. variegata*, *Aedes*, *Anopheles*, *Culex*, predatory efficiency, biological control.

I. INTRODUCTION

Several species of mosquitoes are public health nuisance worldwide. There are about 2500 of species mosquitoes on the planet, of which 300 are well known disease carriers. Approximately seventy million people per annum catch diseases from mosquito bites worldwide. There are several vector control measures exist viz., chemical control, pheromonal control, environmental control and biological control. Among these methods, most reliable method is biological control method, because all other method involves either an environmental modifications or manipulations. Biological control measures are simple, sustainable and will not create any resistance on vector species. Introduction of predators which can breed in the environment may provide a continuous control over vector species. Vector – borne disease control strategies were emphasized on eliminating preimaginal stages are more effective as compared to the adult control measures (Kumar and Hwang, 2006). Mosquitoes are the vectors of several diseases like dengue fever, yellow fever, chikungunya etc. Recently *Zika* virus has spread throughout the tropical and subtropical continents using *Aedes* mosquitoes as their vectors. In order to control these mosquito borne diseases, we have to control mosquitoes properly. Even in smaller mosquito larval habitats like tree holes the larvae of damselfly and dragonfly exhibit predation on mosquito larvae. In Indian context odonata larvae of *Mesogomphus lineatus* (Mathavan, 1976), *Sympetrum drum* (Chatterjee *et al.*, 2007), *Ceriagrion coromandelianum*, *Brachydiplax chalybea* (Saha *et al.*, 2009) has been recorded as predators of mosquito larvae. The controlling methods should not cause any environmental impact. Odonata is recognized as an effective and important predator of mosquito all over the world (Corbet, 1980; Antony *et al.*, 1990). The mass rearing of odonata also have a conservatory status. Odonata communities have become an increasingly important tool in the studies on the ecological evaluation of aquatic systems (Schmidt, 1985). Wetlands are the common source of mosquitoes and the present study will enable to understand the utility of *R. variegata* larvae in mosquito regulation of wetland ecosystem services (Dale and Knight, 2008).

II. MATERIALS AND METHODS

A. STUDY AREA

Rhyothemis variegata was collected from the tributary of Puzhakkal River in Thrissur district. Puzhakkal River is a west flowing river in Thrissur District, Kerala, India. It has a total length of 29 km and a total of 234 km² drainage area.

B. COLLECTION METHOD

The study was conducted during December 2014 to January 2016. Dragon fly nymphs were collected by using hand nets made up of nylon of 1mm dimension and provided a metal handle of 1meter length. Samples were brought to the laboratory in plastic buckets along with some amount of *Salvinia* and water from the source area. The captured predators were maintained in the laboratory conditions by providing mosquito larvae as their food. They were kept in a glass container of 2x4x6 (LxBxH) inches in size.

The different instars of 3 different mosquitoes were reared separately under laboratory conditions. *Aedes* mosquito was obtained from rotten plantain materials. *Anopheles* was obtained by keeping fresh water in plastic container under shady places. *Culex* mosquito was obtained from rotten cabbage leaves. Identification of mosquito larvae up to the species level was not done because of the lack of taxonomic expertise.

C. EXPERIMENTAL SETUP

The consumption rate of dragon fly nymphs against different instars of 3 selected mosquito larvae was determined. For this the collected predators was kept starved for 24hrs. They were then transferred to a separate glass container of 1 liter volume filled with 750ml of source water. To this container 50 mosquito larvae of a particular developmental instar was introduced and their feeding rate was noted. Observation was done for a total of 8 hours between 10.00 AM to 6.00 PM. This was repeated 3 times for 4 developmental instars of each selected mosquitoes.

D. STATISTICAL ANALYSIS

Data collected during the experiment was computed and the mean consumption, percentage consumption of all the selected predators over all the developmental instars of *Anopheles*, *Aedes* and *Culex* mosquitoes was calculated. Percentage mortality rate of selected prey species were calculated using mathematical formulas. The significance of the results were checked by using Univariate ANOVA of Statistical Package for social Science (SPSS) software version 21.0. Two way analysis of variance was done with type of mosquitoes as first factor and instars as second factor. Significance of Interaction between type of mosquitoes and instars was done separately by using least significant difference test (LSD).

III. EXPERIMENTAL ENVIRONMENT

The study revealed that *R. variegata* mostly prefer the *Aedes* mosquitoes than *Anopheles* and *Culex* mosquitoes. *R. variegata* prefer third instar of *Aedes* mosquitoes followed by its second instar. Maximum consumption rate of *R. variegata* was observed as 70.6%, in the third instar of *Aedes* mosquitoes. The lowest percentage consumption rate of *R. variegata* was observed as 9.3% in the first instar of *Culex* mosquitoes. Results revealed that *R. variegata* less prefer *Culex* mosquitoes. (Table 1).

Table 1: Consumption rate of *R. variegata* on the larval instars of *Anopheles*, *Aedes* and *Culex* mosquitoes.

| Mosquito selected | <i>Anopheles</i> | | | | <i>Aedes</i> | | | | <i>Culex</i> | | | |
|------------------------|------------------|----|------|------|--------------|------|------|------|--------------|------|------|------|
| | I | II | III | IV | I | II | III | IV | I | II | III | IV |
| Mean consumption | 2.5 | 4 | 4.3 | 2.1 | 2.7 | 4.8 | 8.8 | 3.5 | 1.1 | 2.3 | 3.5 | 2.1 |
| Percentage consumption | 20 | 32 | 34.6 | 16.6 | 22 | 38.6 | 70.6 | 28.6 | 9.3 | 18.6 | 28.6 | 14.6 |

In the case of *R. variegata*, percentage consumption was higher in the case of third instars (Fig 1.). In the case of *Anopheles*, no significant difference was found in the percentage consumption between third instar and second instar and also between first and fourth instar larvae. However, in the case of *Aedes* and *Culex*, Percentage consumption in second instars was significantly lower than that in the third instar and significantly higher than first and fourth instar larvae. No significant difference in percentage consumption was found between second and fourth instars and first and fourth instars of *Culex* mosquitoes. Comparison of percentage consumption between *Aedes* and *Culex* in each instars revealed that there exists significant difference in the percentage consumption between species in all instars of these mosquitoes. There is no significant difference in the percentage consumption between first instars of *Aedes* and *Anopheles* mosquitoes. No significant difference in percentage consumption was found in third and fourth instars of *Anopheles* and *Culex* mosquitoes (Table 2).

Table 2. Results of TWO WAY ANOVA for *R. variegata* showing percentage of consumption in each instars of different types of mosquitoes

| Instars | Percentage of consumption (mean ± SE) | Significance in comparison between species | | | | |
|---------|---------------------------------------|--|----------------------------|------------------|-------|-------|
| | | <i>Aedes</i> (1) | <i>Anopheles</i> (2) | <i>Culex</i> (3) | 1 & 2 | 1 & 3 |
| 1 | 22.00 ± 2.00 ^d | 20 ± 4.16 ^b | 9.33 ± 1.33 ^c | NS | S | S |
| 2 | 38.67 ± 1.33 ^b | 32.00 ± 2.00 ^a | 18.67 ± 1.33 ^b | S | S | S |
| 3 | 70.67 ± 1.76 ^a | 34.67 ± 1.33 ^a | 28.67 ± 1.76 ^a | S | S | NS |
| 4 | 28.67 ± 1.76 ^c | 16.67 ± 2.4 ^b | 14.67 ± 2.67 ^{bc} | S | S | NS |

S – Significant; NS – non significant
Means having same letter as superscript are homogeneous within a column

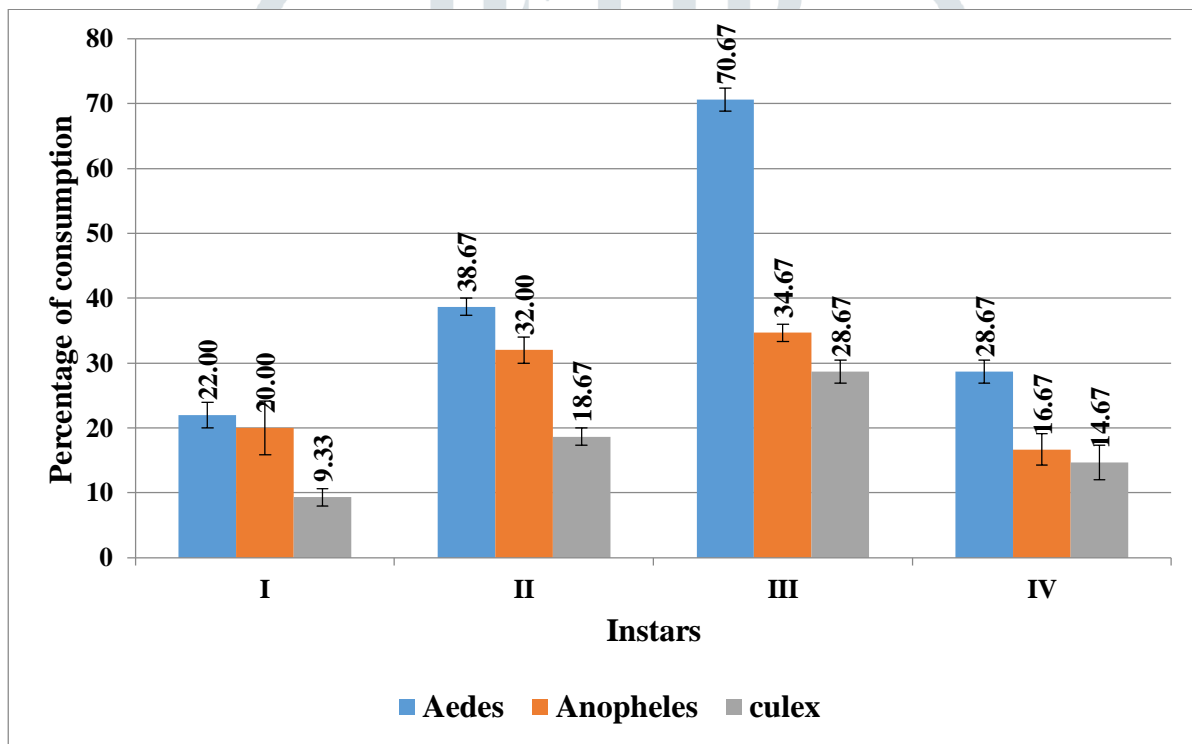


Fig. 1. predatory efficiency of *Rhyothemis variegata*

Mortality rate was observed only in the fourth instar larvae of selected mosquitoes. In the case of *R. variegata*, *Aedes* larvae showed higher mortality (9.3) followed by *Anopheles* (6.3) and *Culex* (3.3). This may be due to the injury caused by the predator on the larvae at the time of prey catching and the subsequent loss of haemolymph leading to death (Table 3).

Table 3. Percentage mortality of the selected mosquito larvae.

| Predator selected | Percentage mortality rate | | |
|---------------------|---|---------------------------------------|---------------------------------------|
| | <i>Anopheles</i> - 4 th instar | <i>Aedes</i> - 4 th instar | <i>Culex</i> – 4 th instar |
| <i>R. variegata</i> | 6.6 | 9.3 | 3.3 |

Table 4. Predation rate of *R. variegata* on the larval instars of *Anopheles*, *Aedes* and *Culex* mosquitoes during the observation period

| Type of Mosquito | Instar | Observation Period | | | |
|------------------|--------|--------------------|---------|--------|--------|
| | | 10 -12 AM | 12-2 PM | 2-4 PM | 4-6 PM |
| <i>Aedes</i> | 1 | 5.6 | 2.6 | 2.7 | 0.3 |
| | 2 | 11.3 | 4.6 | 2.3 | 1 |
| | 3 | 13.3 | 12.3 | 6 | 3.6 |
| | 4 | 4.6 | 4 | 3.3 | 2.3 |
| <i>Anopheles</i> | 1 | 4.3 | 2.6 | 1.6 | 1.3 |
| | 2 | 5 | 5.3 | 3.6 | 2 |
| | 3 | 7.3 | 6 | 2.6 | 1.3 |
| | 4 | 4.6 | 2.3 | 1 | 0.3 |
| <i>Culex</i> | 1 | 2 | 1.3 | 0.6 | 0.6 |
| | 2 | 4 | 2.6 | 1.6 | 1 |
| | 3 | 6.6 | 4 | 2.6 | 1 |
| | 4 | 3.3 | 2.3 | 1.3 | 0.3 |

Both *R. variegata* nymphs and *Aedes* larvae generally live in fresh water bodies and hence both prey and predator share a common habitat. These results are consistent with the results of Saha et al., (2012). Nymphs of *R. variegata* and *Aedes* mosquito larvae are bottom feeders which might have resulted in the higher predation of *Aedes* larvae by *R. variegata* and these results are in line with the observations of Venkatesh and Tyagi (2013). Maximum predation rate was observed during the initial period of observation. For all the larval instars and the feeding reduced subsequently as the time passed. (Table 4).

The results from the experiments on prey choice among the four larval stages suggested that prey size affected predatory efficiency (Table 3). First instar larvae spent more time near the water surface than did older larvae and this behaviour keeps them away from the predators which are mostly bottom dwelling. This might have also contributed to the lesser consumption rate in the first instar larvae of all mosquitoes. On the other hand all other larval stages have larger body size which attracted the attention of the predator. Minakawa et al., (2007) reported that prey size doesn't affect the predatory capacity. However in the present study all the three predators preferred third and second instar larvae. In the case of fourth instar larvae intense mortality was reported that might have influenced their predation. Least preference was observed for the smaller first instar larvae. In short *R. variegata* can be suggested pretty much as good predators of *Aedes* larvae, however cannot be suggested for the biocontrol of *Culex* larvae as a result of both of them inhabit completely different habitats in natural conditions.

IV. CONCLUSION

The present study disclosed that *R. variegata* could be a sensible predator of larval mosquitoes. *R. variegata* nymphs largely prefer *Aedes* larvae than *Anopheles* and *Culex* larvae, because both nymphs of *R. variegata* and *Aedes* share a common habitat such as stagnant fresh water pools for their oviposition and development. Hence *R. variegata* species can be recommended for the effective management of *Aedes* mosquitoes. For all the larval instars, predator showed most feeding throughout initial period and the feeding reduced afterwards as the time passed. Fatal rate was ascertained solely within the fourth instar larvae of selected mosquitoes. This might result to the injury caused by the predator on the larvae at the time of prey catching and also the resultant loss of haemolymph leading to death. Applied mathematics disclosed that there exists vital distinction within the predation rate between types of mosquito larvae and *R. variegata*. The results obtained here are square measure supported on laboratory experiments and therefore further studied are suggested to determine the biocontrol potential of these species in natural condition.

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