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# LABORATORY EVALUATION OF TWO NATIVE FISHES FROM TROPICAL NORTH QUEENSLAND AS BIOLOGICAL CONTROL AGENTS OF SUBTERRANEAN *AEDES AEGYPTI*

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ABSTRACT. The ability of 2 freshwater fishes, eastern rainbow fish *Melanotaenia splendida splendida* and fly-specked hardyhead *Craterocephalus stercusmuscarum stercusmuscarum,* native to North Queensland to prey on immature *Aedes aegypti* was evaluated under laboratory conditions. The predation efficiency of the 2 species was compared to the exotic guppy, *Poecilia reticulata,* which is commonly used as a biological control agent of mosquito larvae. Of the 3 fish species tested, *M. s. splendida* was shown to be the most promising agent for the biological control of *Ae. aegypti* that breed in wells. *Melanotaenia s. splendida* consumed significantly greater numbers of immature *Ae. aegypti* than *P. reticulata,* irrespective of developmental stage or light conditions. Unlike *C. s. stercusmuscarum, M. s. splendida* could be handled, transported, and kept in captivity for extended periods with negligible mortality. However, *M. s. splendida* was also an efficient predator of *Litoria caerulea* tadpoles, a species of native frog found in wells during the dry season. This result may limit the usefulness of *M. s. splendida* as a biological control agent of well-breeding *Ae. aegypti* and suggests that predacious copepods, *Mesocyclops* spp., are more suitable. However, the use of *M. s. splendida* as a mosquito control agent in containers that are unlikely to support frog populations (e.g., aquaculture tanks and drinking troughs) should be given serious consideration.

**KEY WORDS** Native fishes, biological control, *Aedes aegypti,* frog tadpoles, *Mesocyclops*

## INTRODUCTION

Dengue is an important arboviral disease affecting North Queensland (Kay et al. 1984). Because no vaccine is available, the only practical method of controlling dengue is the elimination of its major vector, *Aedes aegypti* (L.). Larval source reduction has proved to be the only effective management approach for *Aedes* populations (Service 1992). In the past, chemicals such as temephos have supplemented source reduction programs. Now, because of environmental concerns and mosquito resistance to pesticides, biological control is playing an increasingly important role in integrated management strategies.

Domestic wells are important breeding sites for north Queensland populations of *Ae. aegypti* and may serve as dry season refuges for mosquitoes (Russell et al. 1996). In 1994, the predacious freshwater copepod *Mesocyclops aspericornis* (Daday) was successfully introduced to control larval *Ae. aegypti* populations in wells at Charters Towers (Russell et al. 1996). However, because *M. aspericornis* is only effective against 1st and 2nd instars, there is a need for a biocontrol agent that rapidly eliminates the more advanced stages of immature *Ae. aegypti.*

Mosquito problems in small water bodies have been quickly alleviated by the ad hoc addition of larvivorous fish (Homski et al. 1994). Specifically, *Poecilia reticulata* (Peters) has proven effective against well-breeding *Aedes* spp. (Lardeux 1992, Rajnikant et al. 1993). Unfortunately, the worldwide dissemination of *P. reticulata* (and *Gambusia* spp.) has resulted in the displacement of native fish species, which in many cases have proven superior larvivores (Lloyd 1986). Consequently, the World Health Organization has discouraged the widespread use of exotic species while advocating research into the use of native fishes (WHO 1982). Apart from mosquito breeding, wells provide refuge for a variety of native fauna during prolonged dry seasons. For example, tadpoles of the protected native frog *Litoria caerulea* (White) have been found in wells in north Queensland.

Accordingly, the primary aim of this study was to evaluate the efficiency with which 2 native north Queensland freshwater fishes, *Melanotaenia splendida splendida* (Peters) and *Craterocephalus stercusmuscarum stercusmuscarum* (Günther), prey on *Ae. aegypti* and their environmental compatibility with respect to predation of nontarget species.

## MATERIALS AND METHODS

*General:* The methods used in this paper were a modified version of those detailed by Homski et al. (1994) for the laboratory evaluation of fish as biocontrol agents. *Melanotaenia s. splendida* (mean total length 6.20 cm ± 0.82 SE) were collected from freshwater aquaculture tanks at James Cook University, Townsville. *Craterocephalus s. stercusmuscarum* (5.00 ± 0.16 cm) were collected from the Ross River, Townsville. *Poecilia reticulata* (1.56 ± 0.04 cm) were collected from animal drinking troughs at Charters Towers.

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Table 1. Mean Percentage (±SE) of different stages of immature *Aedes aegypti* consumed by *Melanotaenia splendida splendida* and *Poecilia reticulata* during day and night.

| Fish species | Immature stage | | | Day | Night |
|---|---|---|---|---|---|
| | I–II | III–IV | Pupa | | |
| *M. s. splendida* | 99.5 (2.6) | 100 (2.6) | 100 (2.6) | 99.7 (2.1) | 99.9 (2.1) |
| *P. reticulata* | 89.6 (3.2) | 66.9 (3.2) | 45.7 (3.2) | 76.9 (2.6) | 57.8 (2.6) |

In the laboratory, fish were maintained in containers of aerated water equipped with biological filtration at 25 ± 2°C. The fish were fed with Wardley's tropical food flakes® (Wardley Corporation, Seaucus, NJ) and acclimated for 14 days prior to experimentation.

The trials were carried out in white 2.5-liter plastic jars containing 2 liters of filtered well water to exclude any other predators. Individual fish were placed in each jar and were held without food for 24-h prior to each experiment to standardize hunger level. Predation trials were conducted over a period of 2 h, after which time the fish were removed from the jars and the remaining larvae (alive or dead) were counted. Each trial was replicated 5 times in parallel (i.e., $n = 15$ for a trial with 3 treatments), and 5 negative control jars containing *Ae. aegypti* were used for each experiment.

*Preliminary comparison of predation efficiency:* In each trial, each species was tested against 25, 50, and 100 4th-stage *Ae. aegypti* obtained from the Queensland Institute of Medical Research (QIMR) colony.

*Diel periodicity and prey size on predation efficiency:* Because *C. s. stercusmuscarum* did not survive well after 10 min of road transport in a 100-liter container, only *M. s. splendida* and *P. reticulata* were used in this experiment. In each trial, the fish were tested against 100 immature mosquitoes of each developmental stage. The immature *Ae. aegypti* were tested as 3 groups; 1st and 2nd instar, 3rd and 4th instar, and pupae. The trials were

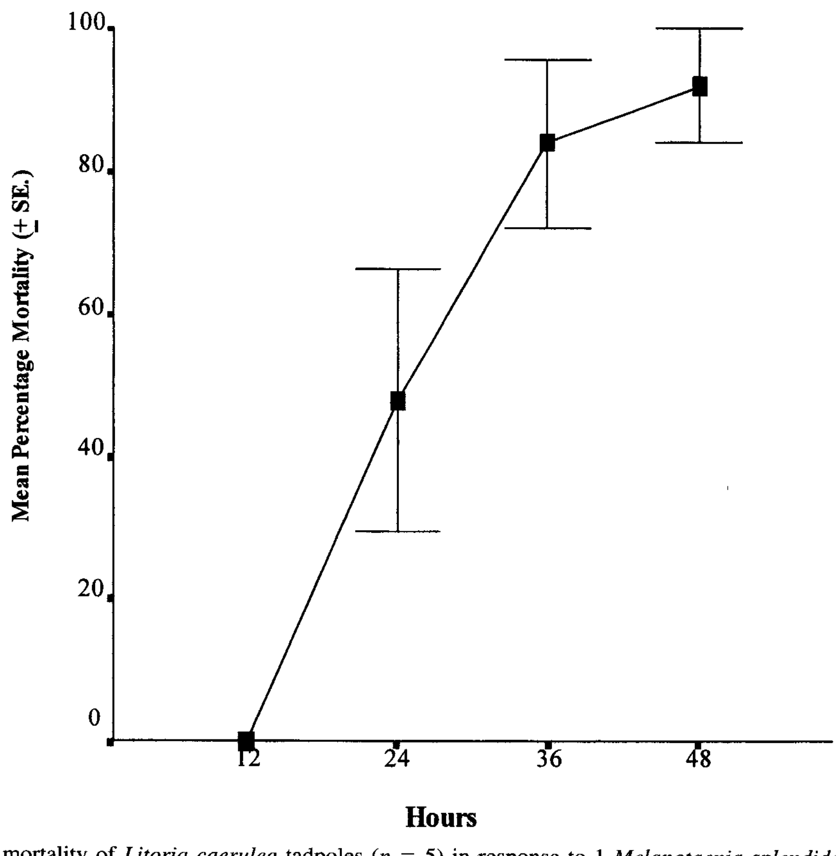


Fig. 1. Percent mortality of *Litoria caerulea* tadpoles ($n = 5$) in response to 1 *Melanotaenia splendida splendida* in 12-liter tanks. Points for *L. caerulea* mortality are shown as mean ± SE of 5 replicates.

conducted using daylight hours of 1000 to 1400 h, during which trial containers were exposed to full sunlight, and nighttime hours of 2000 to 2400 h, during which jars were covered to reduce extraneous light (e.g., from street lights).

*Tadpole predation:* Experiments were conducted in 5, 12-liter plastic containers filled with 28 ± 2°C aged tap water. One *M. s. splendida* (8.21 ± 0.59 cm) and 5 *Litoria caerulea* tadpoles (1.52 ± 0.05 cm) were added to each container. One container, only, with 5 *L. caerulea* tadpoles served as a control. The number of tadpoles consumed by the fish were counted every 12 h for 2 days.

*Statistical methods:* The effect of diel periodicity, fish species, and stage of immature development on predation efficiency was tested using a 3-way replicated analysis of variance with all pairwise comparisons done by Tukey HSD test. (SPSS Australasia, Sydney, Australia).

## RESULTS

*Preliminary comparison of predation efficiency:* Both *M. s. splendida* and *C. s. stercusmuscarum* consumed 100% of the immature *Ae. aegypti* at all densities. *Poecilia reticulata* consumed 100% of the immature *Ae. aegypti* at densities of 25 and 50 per liter; however, they consumed significantly less, 83.8%, at 100 per liter ($F$ = 11.85, df 2, $P$ = 0.0006).

*Diel periodicity and developmental stage of immature* Ae. aegypti *on predation efficiency: Melanotaenia. s. splendida* consumed significantly greater numbers of immature *Ae. aegypti* than *P. reticulata,* irrespective of developmental stage ($F$ = 28.48, df 2, $P$ = 0.0001) or time of day ($F$ = 16.17, df 1, $P$ = 0.0001) (Table 1). The efficacy of *P. reticulata* progressively decreased with immature stage ($F$ = 35.09, df 2, $P$ = 0.0001) and was higher during the daylight hours ($F$ = 5.53, df 1, $P$ = 0.022).

*Frog tadpole predation:* Over 48 h, all 5 *L. caerulea* tadpoles were consumed by *M. s. splendida* in 4 of the 5 containers (Fig. 1). All tadpoles remained alive in the untreated control.

## DISCUSSION

This study clearly demonstrates the effectiveness of native fish from north Queensland as predators of larval *Ae. aegypti.* Although *C. s. stercusmuscarum* was an efficient predator in preliminary trials, 60 to 70% died shortly after capture and transportation (approximately 2 km), and the remaining individuals rarely survived for longer than 17 days. In contrast to the poor survival of *C. s. stercusmuscarum,* which limits its operational potential as a biocontrol agent, *M. s. splendida* was extremely robust, as was the introduced *P. reticulata.*

Native *M. s. splendida* were superior larval predators to the introduced *P. reticulata.* The larger *M. s. splendida* easily swallowed larger instars and pupae, whereas *P. reticulata* only consumed 1st- and 2nd-stage larvae to 89.6% efficacy, which fell away to 45.7–66.9% predation with older immature stages. Furthermore, *M. s. splendida* showed no diel periodicity, whereas *P. reticulata* was predominantly diurnal.

Despite this, *M. s. splendida* also proved to be a predator of *L. caerulea* tadpoles, the data indicating that *M. s. splendida* was capable of decimating populations that sometimes utilize wells. This characteristic is undesirable, especially considering the alarming decline in recent years of native frog populations (Richards et al. 1993). This evaluation suggests that although *M. s. splendida* may be suited as a biocontrol agent where native frogs are absent, *Mesocyclops* ssp. would still appear to be the preferred option for the control of *Ae. aegypti* in wells. Other native fish species should be considered as options.

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