



Vulnerability of carp larvae to copepod predation as a function of larval age and body length

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ABSTRACT

Several marine and freshwater predatory copepods (*Labidocera*, *Pontellopsis*, *Mesocyclops* and *Acanthocyclops*) exhibit a wide dietary niche, ranging from protists, rotifers, cladocerans, copepods, to insect and fish larvae. Predation-induced fish larval mortality in natural environments and hatcheries reduces larval abundance and may limit fish recruitment. However, few studies quantified the losses incurred by copepod predation. Therefore, this study quantified vulnerability, susceptibility, and loss of two crucial fish, *Labeo rohita* and *Catla catla*, to cyclopoid predation in relation to larval age and body length under laboratory conditions. In addition, we compared susceptibility of fish larvae to the copepod (*Mesocyclops thermocyclopoides* and *Mesocyclops aspericornis*) predation in the laboratory, based on probability of survival after attack in relation to age and size of the larvae. We also evaluated the modulatory role of alternate zooplankton species as common prey of fish larvae and copepods. Both cyclopoid species were able to subdue and kill both species of fish larvae. The two cyclopoid species did not differ considerably in their predation efficiency on larvae; the larvae of *C. catla* and *L. rohita* did not differ considerably in their respective vulnerability to cyclopoid predation. In both species of fish larvae, the copepod-imposed mortality was inversely related to larval age ($R^2 \geq 0.97$) and body length ($R^2 \geq 0.93$). The effect of alternate prey on copepod-imposed mortality was species-specific to prey. The presence of *Paramecium caudatum*, *Brachionus calyciflorus* and *Ceriodaphnia cornuta* resulted in decreased mortality of fish larvae by the copepod; however, the larvae were more susceptible to copepod predation in the presence of either *Hexarthra mira* or *Daphnia similoides* as alternate prey. The copepod-induced mortality of carp larvae was recorded, even in multispecies prey environment; however the mortality was considerably less than that in control. Overall, the maximal reduction (40–100%) in copepod-imposed larval mortality was achieved by the Rotifera *B. calyciflorus*, followed by the Cladocera *C. cornuta* and ciliate *P. caudatum*. The modulatory effects of the presence of alternate prey were more pronounced after fish larvae reached 4 days post hatch (dph). The susceptibility of carp larvae to copepod predation differed ontogenetically with maximal susceptibility between 0 and 4 dph. Threshold level for copepod-imposed mortality was estimated with bilinear model, which ranged from 5.2 to 8.4 dph and 6.8 to 7.8 mm body length. Our results provide insight and valuable inputs for enhancing the efficiency of nursery rearing.

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1. Introduction

A substantial body of literature indicates that mass scale mortality is a problem during the early life stages of a number of marine (Turner et al., 1985) and fresh water fish (Jana and Jana, 2003) in the wild, and in several nursery ponds (Letcher et al., 1996; Pillay and Kutty, 2005; Rao, 2003). High larval mortality is generally

attributed to nutritional deficiency of diets (Ayyappan and Jena, 2003; Ghosh et al., 2004), water quality (Rice et al., 1987), disease, and predation (Frimpong and Lochmann, 2005; Ludwig, 1993; Smith and Kernehan, 1981). By using an empirical model incorporating both intrinsic factors (foraging efficiency, starvation resistance, and the growth index) and the extrinsic factors (concurrent presence of prey and predator in the environment), Letcher et al. (1996) concluded that the highest variance in the larval survival rates can be attributed to the growth index as the intrinsic factor and predation as the extrinsic factor of the fish larvae.

Freshwater predatory cyclopoids belonging to the genera *Mesocyclops*, *Acanthocyclops* and *Diacyclops* are generalist feeders that use a wide spectrum of prey types, ranging from ciliates, rotifers,

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cladocerans, mosquito larvae, their own nauplii, and copepodites (Kumar, 2003; Kumar and Rao, 2003; Rao and Kumar, 2002). They also use fish larvae as food (Frimpong and Lochmann, 2005; Ludwig, 1993, 1999; Smith and Kernehan, 1981). In addition, they are wasteful killers (Kumar, 2003; Kumar and Rao, 2003; Rao and Kumar, 2002) that generally attack and subdue more prey than their actual nutritional requirements (Kumar et al., 2008). Predatory cyclopoids are common in freshwater and rice field ecosystems, and are also found in live fish feed culture systems worldwide.

Anecdotal evidence suggests that, despite having high nutritional content, copepods must only be used as prey for larger fish fry because they can prey upon tiny newly-hatched larvae of common carp fry (Jhingran, 1991). However, the essentiality of live feed, mainly zooplankters, is widely accepted by fish farmers during early larval stages of fish (Hepher and Pruginin, 1990; Rao, 2003). Collection of wild zooplankters as a food source for developing larvae is a common practice in traditional hatcheries (Szlauer, 1983/1984; Szlauer and Szlauer, 1980).

Wild collected zooplankton mainly includes ciliates, rotifers, cladocerans, and copepods. Although copepods, especially cyclopoids, are comparable in size to the desirable feed of fish larvae (Szlauer, 1995; Szlauer and Szlauer, 1980), they can subdue and exploit the developing fish larvae as a food source (Kumar, 2003; Piasecki et al., 2004; Smith and Kernehan, 1981).

The foraging behavior of copepods is directly influenced by the presence of alternate prey species in the environment (Kumar and Rao, 2003; Kumar et al., 2008). Because of their favorable size and nutritional profile, copepods and rotifers are often used as the first food of fish larvae in several nursery ponds (Farkas et al., 1981; Shields et al., 1999). Among the possible causes of fish larval mortality in early stages, copepod predation may be considered as one of the causal factors of high larval mortality (Frimpong and Lochmann, 2005; Kabata, 1985; Ludwig, 1999).

Despite a number of anecdotal observations on the piscicidal behavior of copepods, quantitative information on the prey–predator relationship between copepods and carp larvae is rare. Furthermore, limited information is available on the effects of the presence of other zooplankters constituting potential prey (Frimpong and Lochmann, 2005; Valderrama et al., 2000) on copepods–carp larvae interaction.

In South Asia, the ideal breeding habitats of commercially crucial Indian major carps (e.g., *Labeo rohita* and *Catla catla*) are also inhabited by the predatory cyclopoids (such as *Mesocyclops*). These two fish are commercially crucial fish, and are indigenous to the Indian subcontinent. However, they were recently introduced to various countries worldwide for aquaculture purposes. They are of greatest economic importance in the South Asian subcontinent, widely exploited by artisanal fisheries, and are frequently consumed by people of all economic groups (Jhingran, 1991). The temporal and spatial co-occurrence of rotifers and cladocerans with the cyclopoids, especially *Mesocyclops*, during the breeding seasons of Indian major carps, and the concurrent presence of *Mesocyclops* in the live feed culture tanks and plastic pools encouraged us to estimate the copepod predation on carp larvae. Therefore, the objectives of this study were as follows: (1) enumerate the copepod imposed larval mortality in relation to age and body length of carp larvae; (2) elucidate the modulatory effects of the presence of alternate zooplankter prey in the system; and (3) test the hypothesis that the probability of survival of carp larvae after attack is higher with increasing age and body length.

2. Materials and methods

2.1. Experimental animals

All experimental animals (except fish larvae) in our study were cultured in the laboratory and maintained for at least 3 months before use in the experiments.

The two copepod species, *Mesocyclops thermocyclopoides* (Harada) and *Mesocyclops aspericornis* (Von Daday) are widespread pantropical predatory zooplankton (Aguirre et al., 2003; Dussart and Fernando, 1988). Methods of isolation and maintenance of *Mesocyclops* (Chang and Hanazato, 2003; Kumar and Rao, 1999; Kumar, 2003; Hwang et al., 2009), cladocerans (Kumar and Rao, 2003; Kumar and Hwang, 2008), rotifers (Kak and Rao, 1998; Kumar and Rao, 2001), and ciliates (Rao and Kumar, 2002) were described earlier. We selected these two copepod species because general practices of carp larval rearing and complications of copepod predation were discussed in previous studies (Fregadolli, 2003; Frimpong and Lochmann, 2005; Jana and Jana, 2003; Kabata, 1985).

A starter culture of the copepod *M. thermocyclopoides* was isolated from the zooplankton sample of Bhalaswa Lake, Delhi, whereas that of *M. aspericornis* was originally isolated from zooplankton sample of an ornamental pond in the premises of Tajmahal, Agra, India.

Mass cultures of both copepod species were originally developed from a single ovigerous female in each case. The smaller cladoceran *Ceriodaphnia cornuta* ($530 \pm 11.68 \mu\text{m}$) and rotifers (*Brachionus calyciflorus* ($197 \pm 12.4 \mu\text{m}$) and *Hexarthra mira*) (160 ± 7.2) were originally isolated from an outdoor experimental plastic pool maintained at the campus of AND College, University of Delhi; whereas the larger cladoceran *Daphnia similoides* and the ciliate *Paramecium caudatum* and *Blepharisma musculus* were isolated from the plankton samples of a local pond. Isolation of experimental animals was conducted by filtering the pond water through $53 \mu\text{m}$ mesh net. The experimental animals, isolated from the field samples, were first acclimatized to the laboratory conditions in the mixture of filtered lake water and autoclaved tap water for at least 2 weeks.

The copepods were maintained in 1000 ml beakers at a temperature of $25 \pm 1.5^\circ\text{C}$, and fed with a mixed diet of mosquito larvae, rotifers, the unicellular alga *Chlorella vulgaris*, and when possible, fish larvae. Only adult (5–8 d old) non-ovigerous females were used in the tests.

Cladoceran and rotifer cultures were maintained in the autoclaved tap water, and the green algae *C. vulgaris* at a concentration of $2.5 \times 10^6 \text{ ml}^{-1}$ was used as food. The entire medium was changed daily by filtering through a $53 \mu\text{m}$ mesh net. The ciliate *P. caudatum* and *B. musculus* were grown successfully in the mixture of hay and cabbage infusion. The media for the ciliate culture (infusion of hay and cabbage) were changed weekly by centrifugation. The algae *C. vulgaris* (dia- $5.6 \mu\text{m}$) was mass cultured in Erlenmeyer flasks (Vol. 250–1000 ml) using vitamin-enriched K-L medium (Kuhl and Lorenzen, 1963). The algae were harvested during the exponential phase of their growth.

The fertilized eggs of carp larvae were obtained from the Fisheries Department Haryana. Carp larvae were obtained by hatching the fertilized eggs in the 3 ft (dia) \times 3/4 ft (depth) aerated containers and subsequently rearing them under laboratory conditions (temperature: $25 \pm 2^\circ\text{C}$; pH: 7.2–8.2; photoperiod: 10 h light/14 h dark) in separate glass aquaria. Nearly 3/4th volume of water in each container was changed daily with dechlorinated tap water. From the second day post hatched (dph), the larvae were fed live zooplankton (including rotifers, cladocerans, and copepods) collected from outdoor plastic pools at 5 h intervals during the daytime. A detailed account of experimental animals, their body lengths, and culture conditions are provided in Table 1.

All experimental containers were stored in the laboratory, which was fitted with thermoregulator, set at a temperature of $25 \pm 1.5^\circ\text{C}$. The presence of live, dead, and mutilated larvae was recorded daily. The experiments spanned 25 days from March to April 2010. Sufficiently large numbers of fertilized eggs (of *C. catla* and *L. rohita* separately for experiment-I, and only *L. rohita* eggs for experiments-II and III) were retained for hatching prior to the initiation of each of the three experiments.

All three experiments were initiated with a fresh batch of carp larvae and copepods.

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