A study on larval rearing of the local freshwater shrimp, *Macrobrachium dolatum*, in Laos: a potential target species for aquaculture

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Abstract

Freshwater shrimp are a valuable aquatic product, in terms of their high economic value, for the people living along the rivers of Laos. However, no freshwater shrimp farming system has thus far been established in Laos. With the aim of developing such technique in Laos, the present study found four local species, *Macrobrachium dolatum*, *M. eriocheirum*, *M. amplinus*, and *M. dienbienphuense*, inhabiting Luang Prabang province. *M. dolatum* was selected for further study because of its relatively greater body size and fecundity, which are desirable features for aquacultural purposes. Hatched larvae were obtained from wild broodstock reared in an experimental tank. Next, a larval rearing trial was conducted on *M. dolatum*, using freshwater and brown pond water, and survival rates were observed for 4 weeks. The survival rate at 4 weeks of rearing was 1.8% and 20.5% when the larvae were reared in freshwater and brown pond water (collected from a fish-rearing pond), respectively. Further studies, such as using salt water as rearing water, are in progress to elucidate preferable rearing conditions for the larvae of *M. dolatum*.

Introduction

Freshwater shrimp are particularly important and in high demand in Southeast Asian countries as a source of protein and income. A larvae production system has been established for the giant freshwater prawn, *Macrobrachium rosenbergii*, and this species has now become the most commercially farmed freshwater prawn in the world (New1990; New 2002). Although the Laos government has a policy of supporting the culture of indigenous aquatic animals, sustainable aquaculture of local shrimp has yet to be established in Laos. Since Laos is the only inland country in Southeast Asia, landlocked fluvial shrimp species are only available in this country (Hanamura et al. 2011). And since the ecological and biological features of amphidromous (e.g., *Caridina leucosticta*) and landlocked (e.g., *Caridina denticulata ishigakiensis*) shrimp species differ greatly

(Shokita 1976; Leberer and Cai 2003; Yatsuya et al. 2013), the most advanced knowledge and techniques for freshwater shrimp culture, such as those developed for *M. rosenbergii*, which has an amphidromous life cycle, cannot be applied directly to the landlocked fluvial shrimp inhabiting Laos. Especially in the larval stage, different biological characteristics, such as salinity tolerance, are exhibited even among the same species, such as *M. nipponense* (Ogasawara et al. 1979). The present study, then, was conducted with the aim of developing a larval rearing technique specifically suitable for landlocked local shrimp in Laos.

Materials and methods

Animal collection and species identification

Local freshwater shrimp were collected from the Mekong River, or purchased in the local market in Luang Prabang province, Laos, in October 2016. For species identification, the collected shrimp were morphologically examined according to the criteria in Cai et al. (2004) and Hanamura et al. (2011). Among the various species examined, *M. dolatum* (Fig. 1) was selected for further study because of its relatively greater body size and fecundity, which are desirable characteristics for aquacultural purposes.

Collection and rearing of M. dolatum

M. dolatum was regularly (roughly each month) collected from the Mekong River in Xanghai village (20°00'12" N, 102°13'50" E), Pak Ou district, Luang Prabang province (Fig. 2). The specimens were kept in outdoor concrete tanks (dimensions: 1 m (W) × 1 m (D) × 0.5 m (H)), with 250 L of freshwater and an aeration, circulation, and filtration system, in the Living Aquatic Resources Research Center (LARReC) in Chansavang village, Sikodtabong district, Vientiane Capital (Fig. 2), until used for the experiments. They were maintained by feeding to satiation with commercial pellets once a day.

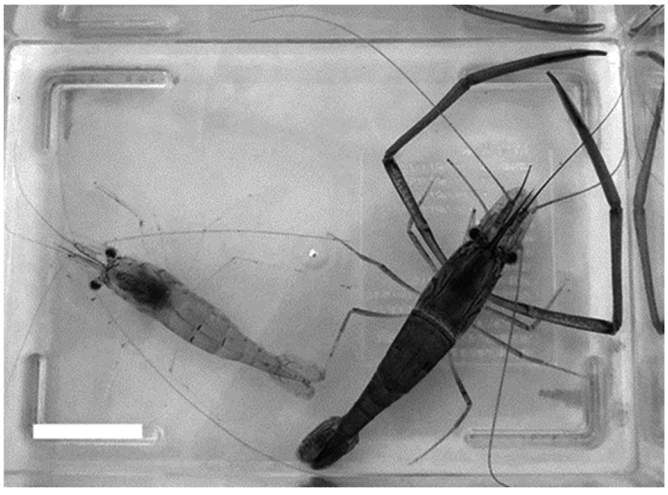

Fig. 1. Female (left) and male (right) *M. dolatum* captured from the wild (Bar: 2 cm).

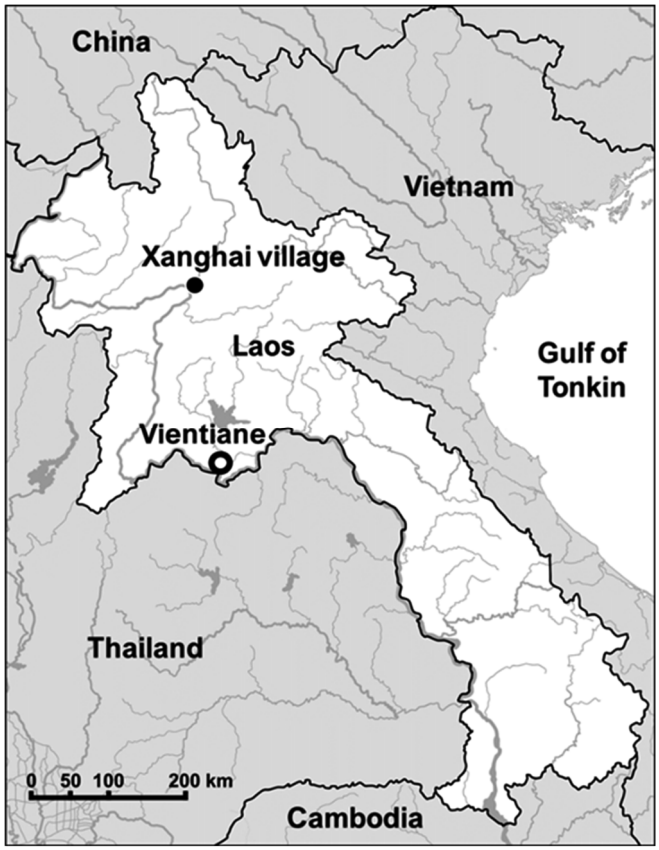

Fig. 2. Laos; solid circle: Xanghai village, collection site for *M. dolatum*; open circle: Vientiane Capital, location of the Living Aquatic Resources Research Center (LARReC, 18°01'05.3" N,102°31'07.2" E).

Obtaining and rearing the M. dolatum larvae

Some of the *M. dolatum* specimens gathered in the abovementioned collection site were found to be carrying eggs. In time, *M. dolatum* larvae were successfully hatched in the experimental tank in LARReC. After hatching, the larvae from one mother were separated into two groups of 10 to 20 larvae, with one group reared in freshwater, and one in brown pond water, each in a 1-L plastic box. The freshwater was prepared by aerating tap water for at least 24 hours to eliminate the chlorine, and the brown pond water was collected from a LARReC fish-rearing pond and used immediately. The water temperature ranged from roughly 23 to 27 °C in this study. Both groups were fed to satiation with commercial powder feed (Gold prawn, Higashimaru; manually grounded before use) once a day, and the larval survival rates in each group were examined for four weeks after hatching. Six replicates, each using larvae from a different mother, were run.

Results

Species identification and basic biological criteria

The morphological examination in the present study revealed that four species, *M. dolatum* (Fig. 1), *M. eriocheirum*, *M. amplinus*, and *M. dienbienphuense*, inhabited Luang Prabang province. The maximum carapace length (CL) of *M. dolatum* was 18.0 mm, and its clutch size ranged from 135 to 502 eggs, both larger than those of the other species: *M. eriocheirum* (15.7 mm and 31-125 eggs), *M. amplinus* (17.8 mm and 22-68 eggs), and *M. dienbienphuense* (17.7 mm and 96 eggs) (Table 1). As aforementioned, *M. dolatum*'s relatively greater body size and fecundity, which are particularly important biological features for aquacultural purposes, were the reason why this species was selected for the present study and used in the following experiments.

Table 1. Maximum carapace length (CL) and clutch size of the four shrimp species identified in the present study.

Species	Maximum CL (mm)	Clutch size (eggs)	
M. dolatum	18.0	135-502	(n = 8)
M. eriocheirum	15.7	31-125	(n = 5)
M. amplinus	17.8	22-68	(n = 5)
M. dienphuensis	17.7	96	(n = 1)

Larval rearing of M. dolatum

The water temperature ranged from roughly 23 to 27 °C in the present study. Based on the results of the six replicates, the average survival rate of the group reared in freshwater was 29.5, 1.8, 1.8, and 1.8% at 1, 2, 3, and 4 weeks after hatching, respectively (Fig. 3); and the average survival rate of the group reared in brown pond water was 85.7, 75.9, 58.0, and 20.5% at 1, 2, 3, and 4 weeks after hatching, respectively (Fig. 3). The total larval length at hatching was around 3 mm, and reached a maximum of 11.2 mm at 4 weeks of rearing.

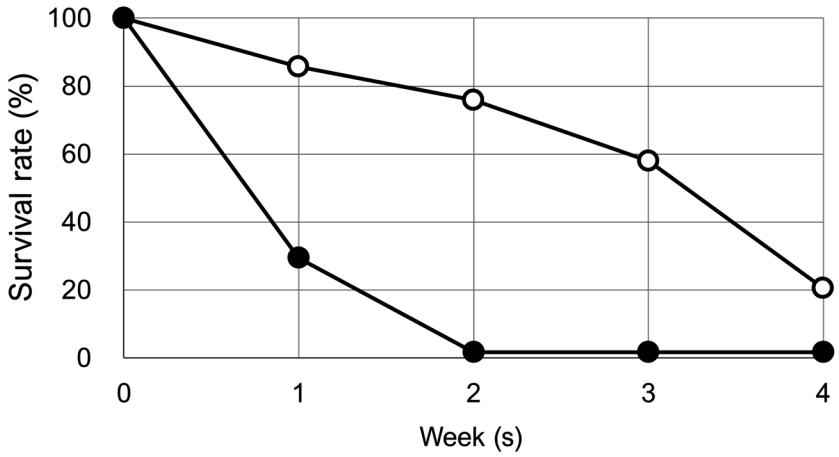

Fig. 3. The survival rate of *M. dolatum* larvae reared in freshwater (solid circles) and brown pond water (open circles). Each point indicates the weighted average survival rate of the larvae from six different mothers ("Week 0" indicates the onset of the experiment).

Discussion

Overall, the survival rate of *M. dolatum* larvae obtained in this study, in the case of both freshwater and brown pond water, was not good enough for larvae production in aquacultural practice. That said, the survival rate (at 4 weeks of rearing) in brown pond water (20.5%) was greater than in freshwater (1.8%), perhaps because the brown pond water may include edible components such as *Moina* and rotifers, which may contribute, as feed, to improved survival for the larvae. In larvae culture of the giant freshwater prawn *M. rosenbergii*, the most popular freshwater shrimp for aquacultural production, live planktonic feed such as *Artemianauplii* is widely used in hatcheries (New 1990), and *Moina* or rotifers can be used as supplemental or substitutional feed (Lovett and Felder 1988; Alam et al. 1993). Microalgae contained in the brown pond water might also play a role in improving survival, similar to that of the edible components above, by providing less stressful culture conditions for the larvae, as reported in detail by Lober and Zeng (2009) in the case of larval rearing of *M. rosenbergii*. The maximum total larval length at 4 weeks of rearing was 3.7 times that at hatching; however, it is unclear whether this growth rate is good or not, since there is, at present, no reported data on *M. dolatum* larval growth. In other *Macrobrachium* species, the growth and/or development of larvae can be improved by modifying the feed, salinity, and/or water temperature (Anger and Hayd, 2010; Lal et al., 2012; Chand et al. 2015). Our previous study showed that the salinity of rearing water was a critical factor for the survival of the larvae of *M. yui* (Okutsu et al. 2018), a species reported as landlocked fluvial shrimp found locally in Laos (Kounthongbang et al. 2015). In that study, the cumulative mortality rates of *M. yui* larvae at the end of the experimental period (from hatching to settling to the bottom, 3-4 weeks) were 58.9. 14.8, 8.3, and 35.4% when reared in freshwater, 1.7 ppt, 3.5 ppt, and 10.5 ppt artificial seawater, respectively. The study clearly showed that salt water was a better environment for the survival of *M. yui* larvae. However, in general, it is known that the

larvae of landlocked fluvial shrimp species (e.g., *Caridina mccullochi*, *Macrobrachium lanchesteri*, *Caridina formosae*) do not require salt water, but can be reared and develop normally in freshwater (Benzie 1982; Wong 1994; Shy et al. 2001; Lai and Shy 2009). In contrast, the larvae of amphidromous shrimp need to be reared in salt water (Hunte 1979a, b; Hayashi and Hamano 1984; Bauer and Delahoussaye 2008; Boudour-Boucheker et al. 2016). Though the reasons for the anomalous results in the *M. yui* study are unclear, a possible reason is that the streams in the habitat of *M. yui* flow through karst formations that contain electrolytes derived from an ancient sea, and the larvae of *M. yui* may utilize these (detailed discussion in Okutsu et al. 2018). The *M. dolatum* larvae investigated in the present study may also require saline water for their survival and development, since *M. dolatum* originates from the same region as *M. yui*. Further studies are underway to explore favorable feed and salinity conditions for survival and growth in *M. dolatum* larval production.

Acknowledgements

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