



## Comparison of reproductive performance and offspring quality of giant freshwater prawn (*Macrobrachium rosenbergii*) broodstock from different regions

Dinh The Nhan<sup>a,b</sup>, Mathieu Wille<sup>a</sup>, Le Thanh Hung<sup>b</sup>, Patrick Sorgeloos<sup>a,\*</sup>

<sup>a</sup> Laboratory of Aquaculture & Artemia Reference Center, Ghent University, Rozier 44, 9000 Gent, Belgium

<sup>b</sup> Faculty of Fisheries, Nong Lam University, HCM City, Vietnam

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### ABSTRACT

An experiment was conducted to compare the reproductive performance and offspring quality of *Macrobrachium rosenbergii* broodstock from four different sources: (1) Vietnam wild; (2) Vietnam pond-cultured; (3) Hawaii pond-cultured and (4) China pond-cultured *M. rosenbergii* females were individually followed for 180 days in three 1200-l fresh water recirculation systems and fed a commercial diet. Ovarian development, moulting and spawning events were checked daily. In addition a number of egg and larval quality parameters were determined. The breeding frequency, fecundity, egg laying success rate, egg dimensions and egg hatchability were not significantly different between animals from the four different sources. However, there were significant differences in terms of offspring quality between the different broodstock sources. Individual dry weight, larval development rate, time to reach the postlarval stage, postlarval survival and tolerance to ammonia toxicity were all better in the offspring originating from China pond-reared and Vietnam pond-reared broodstock sources compared to those originating from Vietnam wild and Hawaii pond-reared sources. Moreover, offspring quality from Chinese and Vietnamese pond-reared broodstock proved more stable in terms of ammonia tolerance over three consecutive reproduction cycles. In general, the pond-reared broodstock from China and from Vietnam resulted in better offspring quality than the Hawaii pond-reared and Vietnam wild broodstock. These results indicate that broodstock sourcing deserves proper attention in hatchery operations of *M. rosenbergii*. It furthermore proves that domesticated (pond-reared) animals are not necessarily inferior as breeders compared to wild-sourced animals. The results may also point out the potential to selectively breed stocks with improved characteristics adapted to the local culture environment.

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### 1. Introduction

The giant freshwater prawn *Macrobrachium rosenbergii* (de Man) is the largest species in the genus and is the most favored species for farming. Broodstock of *M. rosenbergii* was introduced from Malaysia to the Anuenue Fisheries Research Centre in Hawaii-USA in 1965 (Hedgecock et al., 1979). Later, broodstocks from Hawaii and SE Asia were introduced into many regions where *M. rosenbergii* was not indigenous, including North and South America, Africa, Europe and parts of Asia to initiate culture industries (New, 2000a). Freshwater prawns are suitable candidates for inclusion in polyculture systems, and in integrated aquaculture–agriculture. The relatively long larval rearing period (18–35 days) and low larval survival rates are however disadvantages to commercial culture of this species (Maclean and Brown 1991).

In Vietnam, the giant freshwater prawn is becoming an increasingly important aquaculture species, as its culture, especially in rice fields, is considered to have the potential to raise income among

impoverished farmers. The aquaculture production of *M. rosenbergii* reached over 10,000 tons per year in 2002, having increased from about 3000 tons in the 1990s (Phuong et al., 2006). The lack of a stable seed supply has been an important obstacle to the further expansion and development of *M. rosenbergii* culture (Phuong et al., 2006). Farmers have traditionally depended on wild sources to obtain seed for aquaculture but are now faced with dwindling resources and a shortage of natural spawners (Wilder et al., 1999). Poor performance (in terms of survival and metamorphosis rates) of larvae from wild captured parent stock remains a bottleneck (Thang, 1995; Amrit and Yen, 2003). At present about 52% of the hatcheries use wild broodstock, the rest relies on farmed broodstock (Phuong et al., 2006).

Despite four decades of domestication (Ling and Merican, 1961; New, 2000a,b), little information is published concerning the effects of many generations of domestication on cultured stock (e.g. inbreeding level). Amrit and Yen (2003) compared performance of larvae originating from Thai and Vietnamese *Macrobrachium* breeders. Larvae from Thai breeders were found to have higher survival and develop more uniformly as compared to larvae from Vietnamese breeders. As Thai prawns were pond-reared and Vietnamese prawns were wild sourced, this study could however not separate the effect of

\* Corresponding author. Tel.: +32 926 43753; fax: +32 92644193.  
E-mail address: [Patrick.Sorgeloos@UGent.be](mailto:Patrick.Sorgeloos@UGent.be) (P. Sorgeloos).

geographical origin from the effect of wild versus captive source. There is still a controversy whether it is better to use wild or pond-reared breeders and local or imported prawn breeders. Wild breeders are generally considered better, but quality may vary depending on capture techniques and transport conditions. Moreover, breeders from different geographical origin might have different characteristics in terms of reproduction and offspring quality.

Evaluating reproductive characteristics and offspring quality of different prawn strains could also be considered as a first step in the development of selective breeding programs. Thanh et al. (2009) rightly noted that in this respect, very little efforts have been made on crustacean species.

To date, there are five national hatcheries in Vietnam for the production of *M. rosenbergii*. Hatchery output nevertheless is still insufficient to meet demands both in terms of both quantity and quality. Therefore, large numbers of *M. rosenbergii* postlarvae (PL) are imported from China to supply the farmers. Ongrown specimens from this Chinese source are used as pond-reared breeders in hatcheries. Early 2005 the Fishery Department of An Giang province imported *M. rosenbergii* PL from Hawaii which are believed to have better quality in terms of reproductive and growout performance.

In the current study, an experiment was conducted to compare the reproductive performance and offspring quality of *M. rosenbergii* broodstock from four different sources: Vietnam wild; Vietnam pond-reared; Hawaii pond-reared and China pond-reared with the objective to determine which broodstock source is most suited for seed production under conditions prevailing in South Vietnam. This knowledge may add to the development of improved hatchery protocols and seed quality of *M. rosenbergii* culture and serve as a starting point to set up a selective breeding programme.

## 2. Materials and methods

### 2.1. Broodstock sources

Adults of the giant freshwater prawn *M. rosenbergii* were selected from four different sources: (1) Vietnam wild breeders (VW) were captured in the Cua Dai river, Ben Tre province, belonging to the lower section of the Mekong river system; (2) Vietnam pond-reared breeders (VP) were collected from growout ponds which had been stocked with postlarvae originating from wild breeders from Cu Chi, Ho Chi Minh peri-urban; (3) Hawaii pond-reared breeders (HP), introduced from Malaysia into Hawaii in 1965, but kept for many generations in captivity since, were introduced to Vietnam in early 2005 as postlarvae for on-farm trials in the Mekong Delta and collected as broodstock at the end of a culture trial from a local farm in An Giang province; and (4) China pond-reared breeders (CP) were imported from China in April 2005 as postlarvae by Nguyen Thi Suong (prawn hatchery owner in Thanh Phu district, Ben Tre province) and were grown out in culture ponds in Binh Dai district, Ben Tre province, South Vietnam and collected as broodstock at the end of the grow-out culture period. The individual weight and length of the animals were not significantly different between the sources at the beginning of the experiment. Numbers of initial breeders for each source ( $n$ ) and average weight (g) were: VW ( $n = 67$ ),  $37.4 \pm 7.9$  g; VP ( $n = 86$ ),  $31.8 \pm 4.7$  g; HP ( $n = 54$ ),  $34.9 \pm 5.1$  g and CP ( $n = 75$ ),  $37.8 \pm 7.3$  g.

### 2.2. Experimental conditions and set-up

#### 2.2.1. Broodstock system

Three separate recirculation units were set up, each one containing one 1000-l holding tank ( $1.4 \times 2.4 \times 0.3$  m) and one 200-l overhead biological filter tank. The biofilter was filled with coral stone. An airlift system provided aeration and aided the water to pass through the filter media. Water was continuously pumped from the central chamber of the broodstock holding tank into the biofilter tank and then returned to the holding tank by gravity.

The holding tank was divided into twenty-four square rearing compartments ( $0.35 \times 0.35$  m) and one central pump compartment, with each compartment containing one female. There were 3 replicate units with a total of 18 females per source of broodstock.

#### 2.2.2. Broodstock backup system

A recirculation system, similar to the one described above was arranged to maintain extra animals from the four sources for replacing any mortalities that occurred during the initial phase of the experimental period. In this way, from each source an extra 20 females and 10 males were maintained separately.

#### 2.2.3. Broodstock rearing conditions

Broodstock rearing methodology roughly followed the techniques described by Cavalli et al. (1999). The prawns were randomly selected and stocked into the three experimental units. Freshwater was continuously pumped from the central compartment of the holding tank into the biological filter and flowed back to the holding tank by gravity. Water was exchanged at a rate of approximately 20% per day after removing waste and uneaten feed by siphoning.  $\text{NH}_4\text{-N}$ ;  $\text{NO}_2\text{-N}$ , and  $\text{NO}_3\text{-N}$  levels were maintained below 0.2, 0.1, and 10.0  $\text{mg l}^{-1}$  respectively. The photoperiod was set at 12 h light at an intensity of 600 lx at the water surface. Temperature was maintained at  $28 \pm 1$  °C. Prawns were fed *ad libitum* with a commercial formulated shrimp diet twice a day (at 8.00 h and 17.00 h). The formulated diet contained 400  $\text{g kg}^{-1}$  crude protein, 77  $\text{g kg}^{-1}$  total lipids, and 108  $\text{g kg}^{-1}$  ash (Shrimp Maturation, Uni-President, Viet Nam).

#### 2.2.4. Larval rearing system

A larval rearing system was installed following the design of Cavalli et al. (2001). The set-up consisted of three separate recirculation systems. Each had 16 12-l cylindro-conical jars connected to a single recirculation system containing a settlement tank (100 l) and connected to a submerged biological filter tank (200 l) which was filled with plastic media. Airlifts provided the necessary oxygen in the biofilter. Water from the biological filter tank flowed into a reservoir tank from which it was pumped back to the larval rearing tanks. The water entered the larval tanks from the bottom at a flow rate of approximately 0.2–0.3  $\text{l min}^{-1}$ . The total volume of the recirculation system was approximately 600 l. Water was exchanged at a rate of approximately 10% per day after removing wastes and uneaten feed by siphoning.  $\text{NH}_4\text{-N}$ ;  $\text{NO}_2\text{-N}$ , and  $\text{NO}_3\text{-N}$  levels were maintained below 0.2, 0.1, and 10.0  $\text{mg l}^{-1}$  respectively. Water salinity was adjusted by diluting seawater to 12  $\text{g l}^{-1}$  with deionized water. Gentle aeration was applied in all rearing jars. A lamp system was installed, providing around 900–1000 lx at the water surface for 12 h  $\text{day}^{-1}$ . From each spawner, triplicate groups of 600 newly-hatched larvae were stocked. Larval stocking density was 50 larvae  $\text{l}^{-1}$ . Average water temperature was  $30 \pm 1$  °C. Newly-hatched *Artemia franciscana* nauplii (Great Salt Lake strain) were offered at a density of 10–15  $\text{ml}^{-1}$  from day 2 to day 7. The *Artemia* ration was split over two feedings at 7.00 h and 17.00 h. From day 8 until metamorphosis to PL, the larvae were also fed a supplemental commercial diet (Brine Shrimp Flakes, O.S.I., USA) containing 530  $\text{g kg}^{-1}$  crude protein, 90  $\text{g kg}^{-1}$  total lipid, 110  $\text{g kg}^{-1}$  ash, 90  $\text{g kg}^{-1}$  moisture, and 20  $\text{g kg}^{-1}$  fibre. The commercial diet was fed to satiation five times per day (7.00, 9.00, 11.00, 13.00 and 15.00 h) while *Artemia* nauplii were fed once a day at 17.00 h.

### 2.3. Evaluation parameters

#### 2.3.1. Reproductive parameters

Initial mean weight and total length of females and males were recorded. Ovarian development was classified according to colour, size and outline of the ovary following the description by Chang and Shih, 1995. Moulting and the duration of the intermoult period of the females were recorded. If the moulted female had developed ovaries

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