Updates on the Seed Production of Mud Crab

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Abstract

Widespread interest in mud crab species is increasing because these are highly prized both in domestic and export markets. Among the three mud crab species commonly found in the Philippines, *Scylla serrata, S. olivacea,* and *S. tranquebarica, S. serrata* is preferred by farmers because it is larger and less aggressive than the other species. Likewise, *S. serrata* is the most widely distributed species in the Indo-west Pacific region.

Hatchery-produced seedstock are presently used by some crab farmers in their grow-out operations. In the hatchery phase, feeding mud crab larvae with shrimp formulated diets and natural food was found to reduce the occurrence of molt death syndrome, one of the major problems in seed production. Larvae given 25% formulated diet (FD) + 75% natural food (NF; rotifers and *Artemia*) and 50% FD + 50% NF showed better performance than those larvae fed 100% FD, 100% NF and 75% FD + 25% NF indicating that usage of natural food, especially the expensive *Artemia*, can be reduced. Since the early crab instar (C) produced in the hatchery need to be grown further before stocking in grow-out ponds, two phases of nursery culture have been developed. C_{1-2} are grown to 1.5-2.0 cm carapace width (CW) size in the first phase and further grown to 3.0-4.0 cm CW in the second phase. Nursery rearing is done in net cages installed in ponds for easy retrieval. A combination of mussel or trash fish and formulated diet is used as feed.

Domestication of the mud crab *S. serrata* as a prerequisite to selective breeding has been done at SEAFDEC/AQD. Likewise, defining criteria for the determination of quality of newly hatched zoeae for stocking in the hatchery was initiated. Newly hatched zoeae were subjected to starvation and stress test using formalin. Starvation failed to elicit responses that were significantly different between the good and poor quality larvae hence it is not suitable for larval quality evaluation. Based on three-year data, the formalin stress test gave mean cumulative mortalities of 2.38±0.32, 8.24±0.88, 20±1.58 in good quality larvae, and 43.74±2.39 while 22.93±4.19, 63.68±7.17, 84.29±3.88 and 97.65±1.06 for poor quality larvae at 0 (control), 20, 30 and 40 ppm formalin, respectively. As formalin level increased, cumulative larval mortality also increased regardless of the quality of the larvae. Formalin stress test proved to be a reliable method to determine whether a batch of newly hatched zoeae was of good or poor quality.

Keywords: *Scylla* spp., mud crab, quality seed stocks, domestication

Introduction

Mud crab is highly prized in both domestic and export markets, thus widespread interest in its culture is increasing. Among the three mud crab species commonly found in the Philippines, *Scylla serrata, S. olivacea,* and *S. tranquebarica, S. serrata* is the most widely distributed in the western and central Indo-Pacific regions. This species is preferred for farming because it is larger and less aggressive than *S. olivacea,* and *S. tranquebarica*.

Total mud crab production in the Philippines was 14,438 t in 2010 (valued at US$86,521,000) and increased to 16,360 t in 2012 (valued at US$114,236,000) (FAO ISS, 2014). At present, the Philippines is one of the leading producers of market size mud crab from aquaculture. The major source of seedstock for farming in many countries is from the wild. The overexploitation of mud crabs and habitat losses have resulted in both reduced landings and mean capture size. The depletion of wild stocks highlights the need to develop alternative sources of seedstock like hatcheries. In Viet Nam and the Philippines, crablets are also sourced from the hatchery. In the Philippines, to stem the wild harvest, the provincial and municipal government along with the Bureau of Fisheries and Aquatic Resources have introduced ordinances that prohibit the gathering and selling of crablets (≤ 3 cm) outside the municipality of origin. This resulted to increased acceptability of hatchery-reared crabs by crab growers.

Domestication

The life cycle of the three mud crab species, *S serrata, S. tranquebarica* and *S. olivacea* has been completed but the focus on the seed production is on *S. serrata*. Domestication of the *S. serrata* as a prerequisite to selective breeding has been initially done at SEAFDEC/AQD (Quinitio *et al.*, 2011). The criteria for the evaluation of the quality of various stages were first established for selective breeding. Stress and challenge tests were used to evaluate the quality of the zoeae and juveniles, respectively. The stress test for the zoeae is now being employed in the larval quality assessment in the hatchery (see section on **Hatchery phase**, this paper).

The bacteria *Vibrio harveyi* was used for the challenge tests to evaluate the disease resistance of each batch belonging to the base population (P_o) and first generation (F_1). Juvenile crabs from various families were injected with *V. harveyi* at 10^6, 10^7 and 10^8 cfu/ml, and saline solution as control. Juveniles from batch Sam2A (F_1). (3 x $10^{5.6}$ cfu/crab) had the highest resistance to *V. harveyi* followed by batch Sam2C (P_o) (2 x $10^{5.5}$ cfu/crab), Sam2D (P_o) (3 x 105.4 cfu/crab) and batch CamB (F_1) (3 x105.5). Juveniles from batch CamA (P_o) (2 x $10^{4.4}$ cfu/crab) and Sam2B (P^o) (2 x $10^{4.5}$ cfu/crab) had high mortality even at low levels of *V. harveyi*. The same pattern was observed in terms of duration to 100% cumulative mortality. The median lethal dose for *V. harveyi* was estimated as $10^{5.696}$. The stress tests in hatchery-reared juveniles, using the white spot syndrome virus, are currently being done by another colleague.

The batches that passed as good quality larvae and juveniles were further reared to broodstock size in ponds and subjected to another evaluation (e.g. growth and reproductive performance), including screening for viruses, prior to selection.

Hatchery phase

The inconsistent survival from later zoea to megalopa stage due to molt death syndrome (MDS) remains the major problem in mud crab hatchery. It has been suggested that poor nutrition, low water temperature and prophylaxis application in the zoeal stage are some of the causes of MDS.

Rotifer and *Artemia* are the most commonly utilized natural food for crab larvae. Later megalopa stage and crab instar are fed with minced fish and mussel. At present, there are several commercially available shrimp formulated diets with various levels of HUFA and other essential nutrients that can also be fed to mud crab larvae hence, reducing the use of rotifers and *Artemia*. Feeding of mud crab larvae with shrimp formulated diets and natural food was found to lessen the occurrence of MDS. Mud crab larvae given 25% shrimp formulated diet (FD) + 75% natural food (NF, rotifers and *Artemia*) and 50% FD + 50% NF showed better performance than those larvae fed 100% FD, 100% NF and 75% FD + 25% NF indicating that use of natural food, especially the expensive *Artemia*, can be reduced (Burlas, 2014) (Figure 1).

Likewise, defining criteria for the determination of quality of newly hatched zoeae for stocking in the hatchery was initiated. Newly hatched zoeae were subjected to starvation and stress test using formalin. Starvation failed to elicit responses that were significantly different between the good and poor quality larvae hence it is not suitable for larval quality evaluation. Based on three-year data, the formalin stress test gave mean cumulative mortalities of 2.38±0.32, 8.24±0.88, 20±1.58 in good quality larvae, and 43.74±2.39 while 22.93±4.19, 63.68±7.17, 84.29±3.88 and 97.65±1.06 for poor quality larvae at 0 (control), 20, 30 and 40 ppm formalin, respectively. As formalin level increased, cumulative larval mortality also increased regardless of the quality of the larvae. Formalin stress test proved to be a reliable method to determine whether a batch of newly hatched zoeae was of good or poor quality.

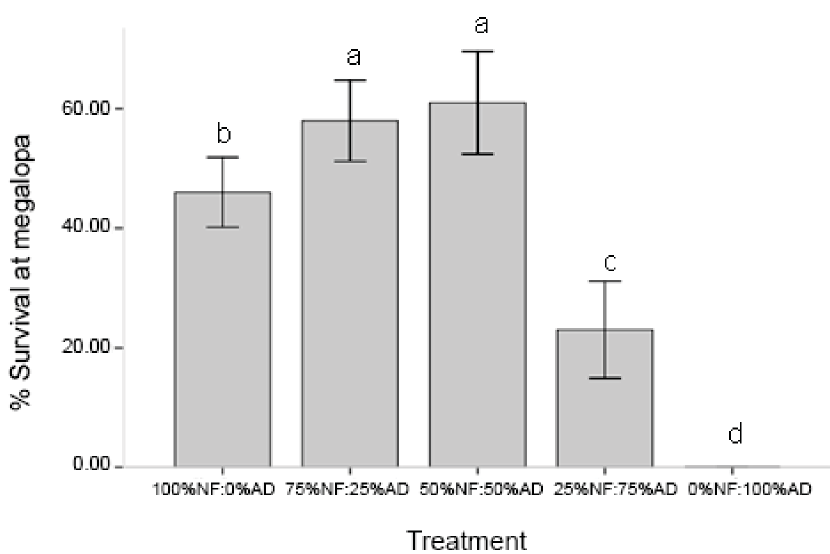

Figure 1. Survival of mud crab larvae fed various combinations of natural food and formulated diet.

To date, the success of mud crab and majority of shrimp hatcheries is still dependent on the use of antibiotics for the treatment against *Vibrio* spp., which is one of the major causes of diseases in the crustacean larvae. The use of antibiotics as treatment for *Vibrio* spp. has proven to improve larval survival (Baticados *et al.*, 1990; Diggles *et al.*, 2000). However, misuse of antibiotics may cause mortalities, incomplete molting (Baticados and Paclibare, 1992), morphological deformities (Baticados and Paclibare, 1992; Pakingking *et al.*, 2002; Lye *et al.*, 2005), and slow growth (Ferreira *et al.*, 2007) in animals. It has been observed that frequency of antibiotic application can be reduced to every 5 days in good quality mud crab larvae. Upon reaching the benthic stage of megalopae, any prophylactic treatment is stopped.

Nursery phase

Nursery is the intermediate phase between hatchery and grow-out. Since the megalopa or early crab instar (C) produced in the hatchery need to be grown further before stocking in the grow-out ponds, two phases of the nursery culture were developed. C_{1-2} are grown to 1.5 – 2.0 cm carapace width size in the first phase and further grown to 3.0-4.0 cm CW in the second phase (SEAFDEC *et al.*, 2010). The second nursery phase was developed to address the growout farmers' preference for bigger-sized juveniles (Rodriguez *et al.*, 2007b).

Nursery rearing is done in net cages installed in ponds (Figure 2) for easy retrieval of stocks. For Phase 1, this previously involved megalopae for stocking (Rodriguez *et al.*, 2007a). However, C_1 is now used for stocking (Quinitio *et al.*, 2009; SEAFDEC/AQD *et al.*, 2010) in the nursery and this may be due to ease in transport at this stage. Phase 1 and 2 may be done one after the other separately or continuously using the same pond compartment. The culture period is 3-4 weeks in each phase depending on the desired size at harvest.

Figure 2. Net cages installed in ponds used for nursery rearing of *Scylla serrata*.

Phase 2 of the nursery may also be done in net lined ponds or in pens inside mangroves (SEAFDEC/AQD, 2010). The net enclosures, as in cages in ponds, prevent the escape of cultured stocks and the entry of other crab species. Stocking directly in ponds was tried but resulted in lower yields than those stocked in net cages (Rodriguez *et al.*, 2007b).

Several strategies have been tested to reduce cannibalism, one of the major problems in the nursery, and improve yield. Various shelter materials and designs are being tested in actual nursery culture. Crab instars use these shelters for hiding when they molt as they are most vulnerable to cannibalism immediately after molting.

Trimming of claws has been shown to be an effective means to reduce cannibalism (Quinitio and Estepa, 2011) and improve percentage survival of intermolt juveniles. This strategy was applied in the second phase of nursery rearing. Initial results revealed that, although trimming could be done easily with the use of nail clippers, this method did not significantly increase survival and was labor intensive.

Feed for the nursery usually consists of trashfish, mussel, or boiled chicken lungs (SEAFDEC/AQD *et al.*, 2010; Quinitio and Parado-Estepa, 2008) or commercially available formulated diets for shrimps (Shelley and Lovatelli, 2011). Several tests have recently shown that feeding a combination of minced mussel or low value fish and pelleted diet formulated for mud crab gives better survival in the nursery. Tryptophan has been shown to lessen the aggressive behavior in mud crab juveniles in the laboratory (Laranja *et al.*, 2010). Incorporation of tryptophan in the formulated diet is currently being tested in the nursery cage culture to determine if this will significantly augment profitability of nursery culture through increased survival or growth.

References

Baticados MCL, Lavilla-Pitogo CR, Cruz-Lacierda ER, de la Peña LD and Suñaz, NA. 1990. Studies on the chemical control of luminous bacteria *Vibrio harveyi* and *V. splendidus* isolated from diseased *Penaeus monodon* larvae and rearing water. Diseases of Aquatic Organisms 9: 133-139.

Baticados MCL and Paclibare JO. 1992. The use of chemotherapeutic agents in aquaculture in the Philippines. *In:* Shariff M, Subasinghe RP, Arthur JR (eds). Diseases in Asian Aquaculture I. pp 531-546. Fish Health Section, Asian Fisheries Society, Manila.

Burlas M. 2014. Evaluation of commercial shrimp larval diet as replacement of live feeds in mud crab larval rearing. M.Sc. thesis. University of the Philippines Visayas, Iloilo.

Diggles BK, Moss GA, Carson J and Anderson CD. 2000. Luminous vibriosis in rock lobster *Jasu verreauxi* (Decapoda: Palinuridae) phyllosoma larvae associated with infection by *Vibrio harveyi*. Diseases of Aquatic Organisms 43: 127-137.

Fisheries and Aquaculture Information and Statistics Service (FAO ISS) 03/03/2014. www.fao.org/figis/serviet/SQServiet.

Ferreira CSG, Nunes BA, Henriques-Almeida JMM and Guilhermino L. 2007. Acute toxicity of oxytetracycline and florfenicol to the microalgae *Tetraselmis chuii* and to the crustacean *Artemia parthenogenetica*. Ecotoxicology and Environmental Safety 67(3): 452-458.

Laranja Jr. JL, Quinitio ET, Catacutan MR and Coloso RM. 2010. Effects of dietary L-tryptophan on the agonistic behavior, growth and survival of juvenile mud crab *Scylla serrata*. Aquaculture 310: 84-90.

Lye CM, Bently MG, Clare AS and Sefton EM. 2005. Endocrine disruption in the shore crab *Carcinus maenas* – a biomarker for benthic marine invertebrates? Marine Ecology Progress Series 288: 221-232.

Pakingking Jr. RV, Cruz-Lacierda ER and Torres JL. 2002. Studies on the efficacy of sarafin (sarafloxacin hydrochloride) on vibrios in black tiger shrimp (*Penaeus monodon*). *In:* Lavilla-Pitogo CR and Cruz-Lacierda ER (eds). Diseases in Asian Aquaculture IV. pp 125-134. Fish Health Section, Asian Fisheries Society, Manila.

Quinitio ET, de la Cruz JJ, Eguia MRR, Parado-Estepa FD, Pates G and Lavilla-Pitogo CR. 2011. Domestication of the mud crab *Scylla serrata*. Aquaculture International 19: 237-250.

Quinitio ET and Estepa FDP. 2011. Survival and growth of mud crab, *Scylla serrata*, juveniles subjected to removal or trimming of chelipeds. Aquaculture 318: 229-234.

Quinitio ET and Parado-Estepa FD. 2008. Biology and hatchery of mud crab *Scylla* spp. 2nd ed. Aquaculture Extension Manual No. 34. SEAFDEC Aquaculture Department, Iloilo, Philippines. 44 pp.

Quinitio ET, Rodriguez EM and Parado-Estepa FD. 2009. Chapter 4.4. Nursery and grow-out of mud crab. *In:* Rural Aquaculture- A Handbook for Human Resource Development, SEAFDEC Aquaculture Department, Tigbauan, Iloilo. P. 87-95.

Rodriguez EM, Quinitio ET, Parado-Estepa FD and Millamena OM. 2007a. Culture of megalopae in brackishwater ponds. Asian Fisheries Science 14: 185-190.

Rodriguez EM, Parado-Estepa FD and Quinitio ET. 2007b. Extension of nursery culture of *Scylla serrata* juveniles in net cages and ponds. Aquaculture Research 38: 1588-1592.

SEAFDEC/AQD, ACE, MODE/SPPI, BVFMC, ACELT, BFAR and ACIAR/CATP. 2010. Mud crab nursery in ponds. Aquaculture Extension Manual No. 47, SEAFDEC Aquaculture Department, Iloilo, Philippines. 27 pp.

Shelley C and Lovatelli A. 2011.Mud crab aquaculture – A practical manual. FAO Fisheries and Aquaculture Technical Paper. No. 567. Rome, FAO. 78 pp.