Comparative growth and morphometric assessment between cultures of wild and hatchery-produced mud crabs

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ABSTRACT

This paper reports the comparative growth, nutritional performance, and morphometric variation between wild and hatchery-reared juvenile mud crabs (Scylla olivacea) in earthen ponds. The crabs were fed daily with boiled tilapia paste at a feeding rate of 5–8% body weight for the first two weeks, followed by feeding with chopped eviscerated tilapia until termination of the experiment. Selected phenotypic trains, including carapace width (CW), carapace length (CL) and abdominal width (AW), were measured weekly. The protein content of the muscle (21.13%), gill (13.51%) and egg (43.28%) were significantly higher in the hatchery-sourced compared to wild female crabs (muscle = 19.15%; gill = 10.09%; egg = 38.15%). Likewise, the hatchery sourced crabs exhibited higher lipid content in the muscle (2.45–2.51%) and eggs (7.51%) compared to the wild counterparts (muscle = 1.45–1.47%; egg = 6.15%). These findings suggested a superior nutritional quality of the hatchery-reared compared to the wild-sourced crabs. Although some selected phenotypic traits did not vary among the wild and hatchery-reared crabs (p < 0.05), their survival rates varied significantly depending on the stocking density (p < 0.05). Overall, the findings suggest that the growth characteristics of the hatchery produced and wild-sourced crabs were similar, which will help to remove the misconception among the crab farmers about the hatchery seeds and promote diversification of the crab production system for long-term sustainability.

1. Introduction

The mud crab (Scylla olivacea) is widely distributed in Southeast Asia, Indo-Pacific, and Southeastern and Eastern Africa. They mainly inhabit the estuarine environment adjacent to the mangrove forests (Jirapunpit et al., 2009; Kellison et al., 2000; Sarower et al., 2017). Recently, S. olivacea has experienced increasing demand due to its resilience in varied environments and excellent taste, forming a delicacy across vast populations (Tamsil, 2019; Rahman et al., 2017). Its emerging resilience in varied environments and excellent taste, forming a delicacy across vast populations (Rahman et al., 2017).

Crab fattening and production of soft-shelled crabs are the main culture systems in the production of wild-sourced crab juveniles (Begum et al., 2009; Ferdoushi and Xiang-Guo, 2010). However, due to the lack of reliable commercial hatcheries, most farmers (for example, in Bangladesh) depend upon the capture and domestication of wild juveniles for the stock establishment. Therefore, the reliance on wild sources of juvenile crabs forms a significant constraint for the future expansion of aquaculture. In addition, this could lead to uncontrolled harvesting of wild juvenile crabs, leading to a critical reduction in the population, thus affecting their sustainability (Rahman et al., 2017).

The need for sustainable crab farming has recently shifted the attention to hatchery mud crab production. However, most crab farmers still believe that wild-sourced seeds are better than hatchery seeds, which is another challenge for expanding crab hatcheries, for instance, in Bangladesh. Various scientific studies (Hamasaki et al., 2011; Azra and Ikhwanuddin, 2016) have been instituted to understand the appropriate technique for producing hatchery crabs. However, to date, information is still limited on the performance of hatchery-produced S. olivacea in major aquaculture producing areas. Indeed, previous studies focused on feeding regimes (Furuta, 1998), rearing techniques (Stoner and Davis, 1994) and hatchery substrata (Kellison et al., 2000) or genetic changes (Berejikian, 1995).
S. olivacea species from Bangladesh wild variety has recently been characterized and confirmed using nuclear and mitochondrial DNA markers (Sarower et al., 2017). Moreover, their peak breeding season was also reported (Ali et al., 2020). To this end, it is crucial to investigate the performance of hatchery-produced juvenile crabs compared to wild-sourced types, which could add more confidence among the aquaculture producers (Turan et al., 2006).

Therefore, the study aimed to develop proper management and conservation strategies for S. olivacea by determining comparative growth performance, nutrition quality and survivability of hatchery-produced and wild juvenile mud crab. The study was implemented through morphometric and meristic variation analysis as previously reported by Mamuris et al. (1998) and Hockaday et al. (2000). Furthermore, truss network systems constructed with landmark points were used for stock identification.

2. Materials and methods

2.1. Ethical approval

The study was performed under the ethical guideline and regulations of ‘Biosafety and Ethics Committee’, Fisheries and Marine Resources Technology (FMRT) Discipline, Khulna University, Bangladesh. The Ref. no. is: KRAEC-2020/04/08: 28/04/2020.

2.2. Production of hatchery-produced crablets

Wild female brood crabs weighing 250–377 g and carapace length of 10–15 cm were collected using bamboo traps and long ropes from the downstream of tidal rivers adjacent to Sundarbans in Munshigonj, Satkhira (22.27° N 89.20°E, 22.26°4’ N 89.19°E, 22.27° N 89.19°E), Bangladesh. The crabs were immediately transported to a local crab hatchery, Nowabeki Gonomukhi Hatchery, and disinfected with 100 μL/L of formalin solution (Sigma Chemicals, St. Louis, MO, USA) for two hours. After that, the crabs were transferred to rearing tanks filled with water at optimum quality parameters (salinity: 25–27 ppt, pH: 7.8–8.1, ammonia: 0.001, alkalinity: 151 mg/L) (Ahmmed et al., 2017).

Before transferring the crabs, the tanks were disinfected using 12.5% sodium hypochlorite (Sigma Chemicals, St. Louis, MO, USA). Next, the brood crabs were fed twice per day using a mixture of chopped mussel, shrimp and Artemia biomass at a feeding rate of 3% body weight. Following a week of growth, the eyestalks of the berried crabs were ablated and disinfected using formalin (100 μL/L). After that, each crab was transferred into individual breeding tanks (250 L) and allowed 6–7 weeks of growth before the spawning. Finally, the newly hatched larvae were nurtured for four weeks to become the crablets.

2.3. Collection of the wild crablets

In late summer, wild juvenile crabs were captured from the same locality where the mature female brood had been captured. The crabs were harvested in their first instar stage (weight: 24.09 ± 4.0 g; carapace length: 2.77 ± 0.50 cm), and all were collected simultaneously. The crabs were then graded similarly to the hatchery-produced juveniles and placed in dry perforated plastic containers until their release to the experimental ponds.

2.4. Acclimatization and determination of survival rate

The harvested crablets were transferred into cages (9 m² nylon nets) at stocking densities of 500, 1000, 1500 crabs/cage. The caged crabs were acclimatized for five hours in pond water (without feeding) before placing them in six experimental ponds (38–40 m²). The crablets were fed daily in the first two weeks using a cooked tilapia paste at a rate of 5% body weight. Next, the survival rates were determined by counting the live crabs in the hatchery- and wild-sourced experimental ponds. Finally, the surviving crablets were fed using chopped eviscerated tilapia from the third week onwards. They were reared for three weeks then released from their cages into the experimental ponds. The water quality in all ponds was maintained constant during the culture period, and the experiment was conducted in triplicate using independently sourced crablets.

2.5. Stocking in pond

After three weeks of acclimatization and nurturing in the cages, 600 hatchery-produced and wild-caught crablets (25–30 g each) were released into the experimental ponds. This was followed by rearing the crablets for 11 weeks in earthen ponds fenced with nylon nets to contain the crablets and prevent the entry of predators. Chopped eviscerated tilapia were fed daily at a rate of 6–8% body weight.

2.6. Water quality parameters

Water turbidity measurements were performed using a Secchi disk method, which involved determining the highest depth to which the disk could be seen (Bowers et al., 2020). The temperature and salinity levels were measured using a regular mercury thermometer and refractometer, respectively (Ahmmed et al., 2017). The pH, dissolved oxygen (DO) and ammonia were measured using a chemical test kit (HI-3823 Combination test kit for Aquaculture, Hanna Instruments Ltd, Leighton Buzzard, UK). The water in the ponds was not replenished during the experiment to mimic the practices in the existing cultures.

2.7. Comparative growth performance

The growth differences between the hatchery- and wild-sourced crablets were determined by weekly sampling of fifteen crabs from each pond. First, the crabs were weighed to obtain the body weight (BW), then sized for carapace length (CL) (Gopurenko et al., 1999), carapace width (CW) and abdominal width (AW) (Stoner and Davis, 1994; Ut et al., 2007). Additionally, photographs were taken and analyzed for precise recording of CL, CW and AW using ImageJ software (Figure 1). Crabs were then harvested after 11 weeks and used for the determination of the proximate composition.

2.8. Determination of proximate composition

The proximate composition, including moisture, protein, lipids Ahmmed et al. (2020) and ash contents, was determined as described by AOAC (2003) and Ahmmed et al. (2021). Briefly, 36 hatchery and wild crabs of different sizes, including three males and three females, were randomly selected from each pond, euthanized on ice for two hours, then washed in chilled water. Each crab was weighed then dissected to separate the muscle, gills and ovary. The separate body parts were packaged in airtight bags and stored at -80 °C until further analysis.

2.9. Morphometric analysis

The harvested crabs were imaged on both the dorsal and ventral sides using a camera. After that, the images were analyzed using Image J (software v1.50) to obtain measurements of some selected phenotypic traits (e.g., CL, CW, AL and AW). Five different landmark distances of the dorsal view and three different landmark distances of the ventral view were also recorded (Figure 2). A “Truss network system” (Turan et al., 2004), constructed using the software, was used to obtain the measurements. The data were recorded in a spreadsheets sheet and used for further analysis.

2.10. Statistical analysis

At first, size-dependent variation was eliminated using the allometric formula described in Eq. (1) (Elliott et al., 1995). The elimination was
performed because of the strong correlation of the measured parameters and CW, which suggested that the differences were related to the body shape and not the relative sizes of the crab.

\[ M_{adj} = M \left( \frac{L_s}{L_o} \right)^b \]  

where \( M \) is the original measurement, \( M_{adj} \) is the size-adjusted measurement, \( L_o \) is the CW of the crab, and \( L_s \) is the overall mean of the CW from all crabs. The parameter \( b \) was estimated from the slope of the regression of \( \log M \) against \( \log L_o \), for each measured parameter. The data was analyzed using a two-way analysis of variance (ANOVA) on SPSS™ (version 23), and the differences between the means were separated using post hoc comparison (\( p < 0.05 \)).

3. Results

3.1. Water-quality parameters of crablets ponds

Inferior water quality is considered one of the most critical limiting factors in the production of aquatic animals. Furthermore, water quality parameters can affect the crablets growth rates and potentially lead to a higher mortality rate. Therefore, multiple water quality parameters were determined weekly during the culture period, as summarized in Figure 3. The temperature and salinity of the ponds ranged from 27-31 °C and 4–10 ppt, respectively. The pH (8.0–9.0) shows that the ponds were slightly alkaline with a DO of 4.5 mg/L to 5.8 mg/L. The results indicated that there were no significant differences in the water quality parameters in the current experiment.

3.2. Survivability of the crablets

The survival rate (%) of the crablets determined in the net cages before the release into the open earthen ponds are shown in Table 1. Despite the high mortality of the crablets in the first two weeks, the survival rates remained constant throughout the remaining period. Additionally, cages with a high stocking density (S1, 1500/9m²) were found to yield lower survival rates compared to moderate (S2, 1000/9m²) and low (S3, 500/9m²) stocking densities. Overall, the stocking density of 500/9m² (S3) was the most suitable for hatchery-reared and wild-caught crablets. However, there were no significant variations (\( p > 0.05 \)) in the survival rates among the crablets obtained from various sources.

3.3. Comparative growth performance

The growth performance of mud crabs from both the hatchery and wild sources was assessed based on BW, CW, CL, and AW (Gopurenko and Hughes, 2002). Weekly measurements obtained until the termination of the experiment at the 11th week are summarized in Figure 4. The weight growth curve depicted a steady increase in the crabs body weight, ranging from 30 g to 155 g and 27 g–135 g for hatchery-produced and wild crabs, respectively. However, there were no significant variations (\( p > 0.05 \)) in the growth performance and diagonal increases in body weight between the hatchery-reared and wild crablets. Both sexes of the hatchery-produced and wild crabs showed increases in the carapace width with rearing time, analogous to increased body weight. However, no significant differences were observed in the growth rate between the hatchery-produced and wild crabs and between the sexes. Other phenotypes measured to determine the growth rate in mud crabs were the carapace length and abdominal width, as shown in Figure 4. Both genders had similar growth patterns, as demonstrated by the carapace width. However, there were no significant differences in growth among the treatments, i.e., crab production system (hatchery vs wild) or gender.

3.4. Proximate composition of mud crabs and its feed

The proximate composition presented in Table 2 shows significant variations in the nutrient profile of the organs. Generally, crab eggs had
the highest lipid (6.2–7.5%) and protein (38–43%) contents compared to the gills (lipids = 0.2–0.4%, protein = 9–13%) and muscle (lipids = 1.4–2.5%, protein = 18–21%). Due to the difficulty in the dissection of the male gonad (testes), determination of the proximate composition in reproductive organs was restricted to the females only. Hatchery-reared crabs exhibited a high protein content in all examined organs (Table 2). Moreover, female crabs had a higher protein content in all the organs than their male counterparts. The results of the study also indicated significant variations (p < 0.05) in the lipid content across the organs (Table 2). Typically, the eggs had the highest lipid content, followed by the muscles and gills. However, the muscles and eggs of the hatchery-sourced crablets had higher lipid content than their wild counterparts. Regardless of the crab source, the ash content varied from 1.09% to 7.75% and 1.11%–6.68% in males and females, respectively. The highest (7.75 ± 0.62%) and the lowest (1.11 ± 0.11%) ash content values were recorded in the gills and eggs of the hatchery-reared males and females, respectively. Overall, the gills had a higher ash content than the other body parts regardless of gender and source.

### 3.5. Morphometric analysis

The CL, CW, AL, and AW along with five different landmark distances of dst A (1-2), dst B (2-3), dst C (3-4), dst D (1-4), dst E (1-3) and dst F (2-4) of dorsal view, and three different landmark distances of dst G (1-2), dst H (2-3), dst I (3-1) of ventral view, were measured with the help of the “Truss network system”. A comparison of the carapace and abdominal measurements between the hatchery- and wild-reared stocks is shown in Figure 5. Furthermore, the distance from landmark to landmark of the ventral side of the two stocks are presented in Table 3. There were no significant differences in any of the analyzed morphometric traits between the hatchery- and wild-reared stocks of the mud crabs.

### 4. Discussions

#### 4.1. Growth and nutritional assessment

The present study compares the growth, survival rate and proximate composition of wild- and hatchery-reared crablets. The study results showed that the crablets growth performance (growth rate, survival, and proximate composition) was similar irrespective of the culture system.
The findings agreed with earlier studies reporting equal growth performance of *S. paramamosain* in both the hatchery and wild culture systems (Ut et al., 2007). Additionally, the protein content of both the hatchery (18.78%) and wild-sourced male crabs (18.95%) was similar to the protein (17.28%) in the muscle of male crabs of *Scylla diviacea* as reported by Yusof et al. (2019). However, the current study observed higher lipid contents (hatchery male = 2.51%; wild male = 1.45%; hatchery female = 2.45%; wild female = 1.47%) compared to the values (wild male = 0.57%; wild female = 0.20%) reported by Yusof et al. (2019). The variation in lipid content might be attributed to the diversity of geographical locations, seasons, and food availability (Bledsoe et al., 2003).

The protein and lipid contents of the hatchery-sourced crabs in the present study were higher than their wild-sourced counterparts (Table 2). Earlier, it was suggested that the wild-sourced crab might struggle during domestication and take longer to adapt to the feeds, which could be the possible reason for this variation (Quinitio et al., 2011). Copeman et al. (2012) also reported a higher lipid content in the hatchery-reared red king crab (*Paralithodes camtschaticus*) (131.1 ± 13.4 μg/mg) than the wild-sourced juveniles (118.6 ± 38.1 μg/mg). In addition, the gills exhibited a higher ash content compared to other organs examined in the current study. The higher ash content in the gills agreed with a previous study by Sarower et al. (2013), where double the amount (5.0–6.3%) was reported in the gill of *Scylla serrata* than in the muscle (2.0–2.4%) and significantly lower in the eggs (1.0–1.5%). This variation was not surprising because the gills generally have higher mineral content than the muscles and eggs (Ayanda et al., 2019).

Although genetic variability was expected to introduce variations in the growth performance of hatchery- and wild-reared crabs, the present study did not observe any significant change. These insignificant differences were also reported in a previous study among the hatchery- and wild-reared crablets during the entire culture period (Ayanda et al., 2019). In addition, it had been anticipated that the hatchery system could lower the ability of the crabs to compete for food and resist cannibalism but less cannibalistic characteristics were observed in the current study, owing to the similar age group of the crablets (Sainte-Marie and Lafrance, 2002). The optimal rearing conditions (salinity = 25–27 ppt; DO = 7–10 ppm, pH = 7.8–8.1; NH₃ = 0.001 ppm; temperature = 27–30 °C) in the

**Figure 4.** Plots of growth parameters against growth time. (A) body weight; (B) carapace width, (C) carapace length; and (D) abdominal width of male and female crabs. Values are mean ± SD of three individual replicates, and error bars represent the standard deviation.

### Table 2. Proximate composition (g/100 g wet tissue) of different organs from hatchery and wild and sourced crabs.

| Organ  | Source  | Sex  | Moisture (%) | Protein (%) | Lipids (%) | Ash (%) |
|--------|---------|------|--------------|-------------|------------|---------|
| Muscle | Hatchery| Male | 74.75 ± 2.5 | 18.78 ± 1.0  | 2.51 ± 0.1** | 2.4 ± 0.2** |
|        |         | Female | 73.95 ± 3.5 | 21.13 ± 0.9** | 2.45 ± 0.2** | 1.83 ± 0.3** |
|        | Wild    | Male | 74.45 ± 2.8 | 18.95 ± 0.75 | 1.45 ± 0.25/b | 3.20 ± 0.15/y |
|        |         | Female | 75.42 ± 1.8 | 19.15 ± 1.5b  | 1.47 ± 0.18b  | 3.47 ± 0.15x |
| Gill   | Hatchery| Male | 78.95 ± 3.0 | 11.83 ± 0.8** | 0.48 ± 0.05  | 7.65 ± 0.25** |
|        |         | Female | 79.28 ± 3.5 | 13.51 ± 0.9** | 0.49 ± 0.06  | 6.23 ± 0.25** |
|        | Wild    | Male | 84.25 ± 3.2 | 9.43 ± 1.0**  | 0.27 ± 0.09  | 5.6 ± 0.15/y |
|        |         | Female | 82.66 ± 2.0 | 10.09 ± 1.5b  | 0.39 ± 0.03** | 6.77 ± 0.30 |
| Egg    | Hatchery| Male | N/A         | N/A         | N/A        | N/A        |
|        |         | Female | 46.85 ± 2.5 | 43.28 ± 1.54  | 7.51 ± 0.2*  | 1.09 ± 0.5  |
|        | Wild    | Male | N/A         | N/A         | N/A        | N/A        |
|        |         | Female | 52.49 ± 2.0 | 38.15 ± 1.2b  | 6.15 ± 0.4b  | 1.25 ± 0.30 |

Data are presented as mean ± SD (n = 3 from independent samples and replications). Different letter (a, b) and (x, y) in the same column indicates significant differences (P < 0.05) within the same organ depending on the source and sex, respectively.
The survival rates of the crabs determined in the acclimatization stage agree with the findings of a previous study (Khan et al., 2017). In general, cannibalistic activities of the crabs are expected to lower their survival rate (Trino and Rodriguez, 2002). However, compared to the survival rate of 21.88% reported in a previous study (Karim and Tahya, 2019), higher survival rates (55–57%) were recorded in the current study. This finding implies that it was viable to proceed with the pond culture.

Water quality parameters are essential factors determining the growth of aquatic animals. Therefore, water quality parameters were recorded weekly until the end of the experiment. The temperature (27–31 °C) and salinity (4–10 ppt) in all treatment ponds were in the optimum range (Ruscoe et al., 2004). Despite the slight variation in temperature (decrease from 31 °C to 27 °C) and salinity due to torrential downpours during the second and fifth weeks, the growth of the mud crabs was not affected. Other water quality parameters (DO, NH₃, alkalinity) also remained suitable for crab culture (Deb et al., 2017; Rahman et al., 2017; Yu et al., 2016). Overall, the water quality parameters in all experimental ponds did not fluctuate significantly during the culture period, thus not affect the growth and survival rates of the crabs.

4.2. Morphometric analysis

In our study, we did not find significant morphological variations between the wild- and hatchery-produced crabs. Nonetheless and despite the lack of significant differences in the current study, significant morphological variations among different wild fish stocks have been reported (Mahfuj et al., 2019; Ahammad et al., 2018; Gain et al., 2017; Chaklader et al., 2016; Hussain et al., 2010; Turan et al., 2004; Swain et al., 1991). However, our study expected similar results between the wild- and hatchery-produced crablets due to their sourcing from the exact geographical location. Another reason for the insignificant variation might be the high adapting capability of the mud crabs (Ali et al., 2020). Unlike fish, mud crabs are very hardy and can tolerate a wide range of variations in environmental factors without any morphological changes (Rahman et al., 2020). Fish, being more vulnerable to environmental fluctuation, change their morphometrics very quickly, as documented in previous investigations (Swain et al., 1991, Wimberger, 1992; Turan et al., 2004). The variations in phenotypic traits can be caused by habitat, feeding strategies and geographical locations (Swain et al., 1991, Wimberger, 1992). In our study, wild and hatchery-produced crablets were grown in the same water quality, habitat, and similar feeding regimes. Hence there were minimal morphological changes due to fluctuations in environmental conditions. Truss network measurements were used to determine phenotypic variations (Turan et al., 2004), which was an advantage in stock structure analysis, primarily because the time was insufficient for remarkable genetic differentiation among the stocks. In addition, the Truss network could help detect the existing genetic variation when genetic markers fail.

5. Conclusions

This study compared the survivability, growth performance and morphological differences between the hatchery- and wild-reared crablets of Scylla olivacea. The findings of the study revealed that the performance of the hatchery-reared crablets was improved compared to their wild counterparts. Therefore, this study may be beneficial in dealing with misconceptions by the crab farmers that the hatchery-produced crablets are not good in terms of growth, survivability, and robustness. Furthermore, the study could diversify the crab sources and promote hatchery establishment among the aquaculture communities.

Declarations

Author contribution statement

Md. Golam Sarower; Ghausiatur Reza Banu: Conceived and designed the experiments.
Md. Mahmud-Al-Hasan: Performed the experiments; Wrote the paper.
Md. Shohanur Rahman; Md. Mehedi Hasan: Performed the experiments.
Mirja Kaizer Ahmmed ; Muhammad Yousuf Ali; Stephen G. Giteru: Analyzed and interpreted the data; Wrote the paper.

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Data availability statement

Data will be made available on request.

Table 3. Distance from landmark to landmark of the dorsal and ventral side of the two stocks of the hatchery-reared and wild mud crab (S. olivacea).

| Landmark distance | Hatchery (mm) | Wild (mm) |
|------------------|--------------|-----------|
| **Dorsal side**  |              |           |
| Dist A (1-2)     | 43.35 ± 2.7  | 44.30 ± 3.5 |
| Dist B (2-3)     | 45.39 ± 3.9  | 44.38 ± 4.1 |
| Dist C (3-4)     | 97.53 ± 5.5  | 93.57 ± 6.3 |
| Dist D (4-1)     | 45.95 ± 3.7  | 44.29 ± 4.1 |
| Dist E (1-3)     | 78.47 ± 3.1  | 78.60 ± 3.1 |
| Dist F (2-4)     | 78.70 ± 3.9  | 79.09 ± 2.8 |
| **Ventral side** |              |           |
| Dist G (1-2)     | 53.15 ± 2.8  | 54.23 ± 2.9 |
| Dist H (2-3)     | 47.06 ± 3.7  | 46.97 ± 2.5 |
| Dist I (3-1)     | 53.28 ± 3.6  | 54.14 ± 3.4 |
Declaration of interests statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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