Effects of Stocking Density and Water Exchange Rates on Growth Performance of Tiger Shrimp, Penaeus Semisulcatus Cultured in Earthen Ponds

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Abstract
The trial was conducted in earthen pond with average initial weight of 4.5 ± 0.4 mg/PLs of green tiger shrimp, to examine the effect of three stocking density and two water exchange rate on growth performances, production traits and feed composition. Eighteen earthen ponds (2200 m²) were stocked with either, 5, 15 and 25 PLs/m³, and received either 10 or 20% of water exchange rate. The results revealed that, mean final weight (g/PLs), gain in weight (g/PLs), gain in weight %, SGR (%/day), feed conversion ratio, protein productive value (PPV), protein efficiency ratio (PER), fat gain and energy utilization were significantly (p<0.01) the best at the lowest stocking density. While, total production exhibited significantly the opposite trend. There significant differences (P<0.05) were found between water exchange rate in term of mean final weight (g/PLs), gain in weight (g/PLs), gain in weight %, SGR (%/day), feed conversion ratio, PPV, PER, fat gain and energy utilization.

From the above results and the economic information of these study it can be concluded that, stocking density of 15 PLs/m² of green tiger shrimp and 20% water exchange rate exhibited the highest net profit and would seem to be the most desirable density and water exchange rate in the system studied.

Keywords: Green tiger shrimp; Stocking density; Water exchange rate

Introduction

Shrimp farming is one of the most profitable and fast-growing segment of the aquaculture industry (Tacon, 2002) [1]. The world production of shrimp increased 250 folds during the last 35 years from 10,000 ton in 1970 to 2, 461, 000 tones in 2005 (FAO, 2002) [2]. The green tiger shrimp, Penaeus semisulcatus used in this experiment, which resembles giant tiger shrimp, P. monodon in geographical distribution in the world. It is tolerant to high salinity and temperature, with their minimum first sexual maturity 6 month age size at about 130 mm length or 6 months age [3]. It is easy to propagate in captivity with or without lateral eyestalk ablation [4]. Stocking density is a major factor that affects fish growth under farmed conditions [5,6]. Stocking density and therefore, the volume of water per fish is a significantly factor in determining production in earthen ponds. Increasing stocking density results in stress, which leads to enhanced energy requirements causing reduced growth and food utilization [7].

Consequently, identifying the optimum stocking density for a species may be a critical factor affecting growth and feed intake when shrimp cultured in earthen ponds. The level of water exchange rate appeared to influence size in shrimp. Shrimp received more food grow faster, matured at larger size, whereas shrimp that received less water exchange rate grow slowly, and matured at smaller size [6]. Little information is available concerning the effects of stocking density and water exchange rate under the earthen ponds rearing system condition. The major objective were to determine the combined effects of stocking density and water exchange rate on growth performance, food utilization, and finally the economic feasibility of green tiger shrimp as species suitable for earthen ponds culture in Egypt.

Materials and Methods

Experimental system and management

The experiment was conducted in Shrimp and Fish International Company (SAFICO) in Sharm El Sheikh Resort, South Sinai, Egypt. This farm is located in a protected area. Water source and the discharged water were monitored twice a weekly. Three stocking densities (5, 15 and 25 Post larvae of shrimp (PLs/m²) and two water exchange (WER) (10 and 20% daily) were tested. Therefore, we have six treatments in three replicates. Shrimp PLs were produced in the hatchery of the same farm using pond reared brood-stocks. The experiment was conducted in eighteen earthen ponds each of 2200 m² water area (1.3 m water depth) with one paddlewheel (1hp) per pound. The inlet water is coming from the Red Sea (40-41 ppt salinity).

Shrimp meal, and soybean meal were added at 113 kg/ha as an organic fertilizer to improve natural productivity of the experimental ponds during the preparation period only (three weeks before stocking post-larval shrimp). The inorganic fertilizers were added weekly at 30 kg/ha of urea, 1.4 l/ha of phosphoric acid and 16.7 kg/ha of sodium silicate. The quantity of fertilizers was divided into 2-3 doses per week for the first three months. Then, the quantity was reduced to the half doses during the last three months. The nitrogen: phosphorus (N: P205). The concentration of silicate was 5 ppm. PLs with average weight of 4.5 mg were stocked in shrimp ponds simultaneously over a period of two weeks at night after pond preparation. Shrimps were fed on starter diet containing 52% protein during first 104 days, Table 1 and finishing diet containing 42% protein until the end of the experiment (205 days).

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Bi-weekly samples of shrimp (60 PLs at least) were caught by cast net to estimate the periodical growth parameters, total biomass of shrimp and to adjust feeding quantity. At the termination of the experiment, representative samples of shrimp during the harvest were taken. Growth parameters, production and feed utilization were calculated as follows: SGR (% day⁻¹)=100 (Ln final weight-Ln initial weight)/days. Gain in weight (g/PLs)=mean final body weight - mean initial body weight; Feed conversion ratio (FCR)=total dry feed fed (g)/ total wet weight gain (g). Net profit was determined by the difference between the sale price of the shrimp after harvest and the costs of larvae and food [12].

**Water quality parameters**

Water temperature and dissolved oxygen were measured every other day using YSI model 56 oxygen meters (Yellow Springs Instrument Company, Yellow Springs, OH, USA). Salinity was measured daily using refractometer. Un-ionized ammonia and nitrite were measured once weekly using a DREL 2000 spectrophotometer (Hach Co., Loveland, CO). pH was monitored twice weekly using an electronic pH meter (pH pen. Fisher Scientific, Cincinnati, OH). During the 10 months feeding trial, the average water quality parameters (mean ± SD) were water temperature, 26.8 ± 0.2°C; dissolved oxygen, 6.2 ± 0.5 mg/L⁻¹; Un-ionized ammonia 0.06 ± 0.03 mg/L⁻¹; nitrite, 0.05 ± 0.03 mg/L⁻¹; pH, 8.6 ± 0.16 and salinity 45.5 ± 0.2%.

**Statistical analysis**

Data were analyzed by two-way analysis of variance using the SAS General Linear Models procedure [13]. Significance between stocking density, between water exchange rate, and their interaction was determined using Duncan’s multiple range tests (Duncan 1955) [14]. Treatments effects were considered significant at (P<0.05). All percentage and ratio data were transformed to arcsin values prior to analysis (Zar, 1984) [15].

**Results**

**Water quality parameters**

Changes of water temperature, dissolved oxygen, pH, nitrite and un-ionized ammonia were shown in Table 2. It was indicated that changes in dissolved oxygen and un-ionized ammonia due to

(a) As PLs stocking density increases, DO2 decreases and ammonia increases.

| Ingredient % | Diets       | Starter | Finishing |
|--------------|-------------|---------|-----------|
| Fish meal (72% C.P.) | 51.2 | 40.0    |
| Wheat flour  | 20.3 | 22.3    |
| Fish meal (72% C.P.) | 16.0 | 25.2    |
| Soybean meal (44%, C.P.) | 5.0  | 5.0     |
| Yeast        | 1.2  | 1.2     |
| Dun fat*     | 2.5  | 2.5     |
| Cod liver oil| 0.5  | 0.5     |
| Gelatin      | 1.5  | 1.5     |
| Soy lecithin | 1.0  | 1.0     |
| Amino-vita sol* | 0.3 | 0.3     |
| Chemical composition % (wet bases) | 9.5 | 07.4    |
| Moisture     | 51.95 | 42.87   |
| Crude protein| 9.10 | 09.12   |
| Nitrogen free extract (NFE) | 8.39 | 19.31   |
| Crude fiber  | 0.53 | 00.38   |
| Ash          | 20.53| 20.92   |
| P/E Ratio (mg protein/kcal energy) | 116.00 | 88.11  |

* Dun fat is a marine powdered lipid.
** Amino vita sol is a product composed of amino acids, vitamins and minerals.

**Amino acid**

| Ingredient | Starter | Finishing |
|------------|---------|-----------|
| Phenylalanine 950 mg | Tyrosine 700 mg | Valine 850 mg |
| Lysine 2,000 mg | Glycine 700 mg | Histidine 500 mg |
| Leucine 3,000 mg | Isoleucine 900 mg | Threonine 1,500 mg |
| Methionine 1,000 mg | Systeine 1,000 mg | Arginine 1,500 mg |
| Pantothonate 4,000 mg | Calcium 3,000 mg | Folic acid 300 mg |
| Vitamin C 4,000 mg | Lysine 2,000 mg | Vitamin B1 2,000 mg |
| Vitamin B6 1,200 mg | Vitamin K3 100 mg | Vitamin A 12,000,000 IU |
| Vitamin D3 3,200,000 IU | Vitamin E 3,200 mg | Vitamin B12 2,000 mg |

| Classification | Temperature | DO₂ | pH | NH₃ | Salinity (ppt) |
|----------------|-------------|-----|----|-----|---------------|
| SD             | N.S.        | 26.7 ± 0.4 | 6.7 ± 0.1 | 0.04 ± 0.001 | N.S.          |
| 5              | N.S.        | 26.9 ± 0.4 | 6.0 ± 0.1 | 0.06 ± 0.001 | 45.5 ± 0.2   |
| 15             | N.S.        | 26.8 ± 0.4 | 4.8 ± 0.2 | 0.08 ± 0.002 | 45.4 ± 0.3   |
| 25             | N.S.        | 26.8 ± 0.4 | 4.8 ± 0.2 | 0.08 ± 0.002 | 45.4 ± 0.3   |
| WER            | N.S.        | 27.0 ± 0.4 | 5.4 ± 0.2 | 0.05 ± 0.003 | 46.2 ± 0.1   |
| 10%            | N.S.        | 26.8 ± 0.4 | 5.4 ± 0.2 | 0.05 ± 0.003 | 44.7 ± 0.1   |
| 20%            | N.S.        | 27.0 ± 0.4 | 5.4 ± 0.2 | 0.05 ± 0.003 | 46.2 ± 0.1   |
| SDxWER         | N.S.        | N.S.      | N.S. | N.S. | N.S.          |

Means (+SE) in the same column having the same superscripts are not significantly different (p<0.05).

Significant level: N.S.=P>0.05, *=P≤0.05, ** P≤0.01. Water exchange rate=WER 10D2O=Dissolved oxygen, 2NH₃=Unionized Ammoni
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Figure 1: Effect of three stocking densities (5, 15, and 25 pcs/m²) and two water exchange rates (WER) (10% and 20%) on growth of marine shrimp (Penaeus semisulcatus) cultured in earthen grow-out ponds.

Growth performance

The changes in mean body weight (g/PLs) of tiger shrimp stocked at three densities and two-water exchange rate 10% or 20% during the period of the experiment (10 months) are shown in Figure 1. At the beginning of the experiment, mean weight were not significantly different among the densities at two-water exchange rate (P>0.05). The overall averages ± SD of tiger shrimp 4.5 ± 0.3 mg/PLs for weight. At the end of the experiment, the mean weight (Figure 1) ranged between 22.4±1.2 g at the lowest stocking density (5 PLs/m²) and 15.50±1.0 g at highest stocking density (25 PLs/m²). The mean weight was significantly (P<0.05) smaller at high densities than low densities Table 3. The overall data of total production, final individual weight, gain in weight, and specific growth rate (SGR) for tiger shrimp reared at three stocking density and two water exchange rate for a period of 10 months in earthen pond are presented in Table 3. It can be concluded from this table, mean final weight, gain in weight, gain in weight %, and SGR were significantly (P<0.05) the best at the lowest stocking density.

While, total production exhibited significantly the opposite trend. Harvests and production estimates increased with increasing stocking density.

At the end of the experiment, total production was 1045.1 ± 35.8 kg/ha at 5 PLs/m² stocking density and 2073.3 ± 49.6 kg/ha at 15 PLs/m² stocking density. Production was opposite to the individual weight during the growth period, that the individual weight decreased with the increasing stocking density (Figure 2 and Table 3), while total production exhibited significantly the opposite trend. Harvests and production estimates increased with increasing stocking density.

Table 3: Effect of stocking density (SD) and water exchange rate (WER) on growth performance of marine shrimp (Penaeus semisulcatus) post-larvae cultured in earthen ponds. Culture period was 205 days.

| Classification | Average body weight (g) | ADG (mg/PLs/day) | SGR (%/Day) | TP kg/ha |
|----------------|-------------------------|------------------|-------------|---------|
| SD 5           | 26.4 ± 1.2              | 4.1 ± 0.03       | 10.3 ± 0.5  | 1260.5 ± 137 |
| 15             | 22.4 ± 1.2              | 4.2 ± 0.03       | 10.0 ± 0.5  | 1206.5 ± 137 |
| 25             | 19.9 ± 1.5              | 4.3 ± 0.03       | 9.7 ± 0.5   | 2065.5 ± 206 |
| WER 10%        | 16.9 ± 1.4              | 4.0 ± 0.04       | 9.8 ± 1.5   | 1285.0 ± 137 |
| 20%            | 19.9 ± 1.5              | 4.1 ± 0.04       | 9.7 ± 1.5   | 2065.5 ± 206 |

Means (+SE) in the same column having the same superscripts are not significantly different (P>0.05). Significant level: N.S.=P>0.05, *P<0.05, ** P<0.01.

1 Average daily gain (ADG)= (Final wt—Initial wt.) / period (days).
2 Specific growth rate (SGR)=100 × (ln Final wt—ln Initial wt.) / period (days). TP= total production
3 Protein Productive Value (PPV %)=100 × protein gained in shrimp / protein fed.
4 Energy Utilization (EU) = 100 × energy gained in shrimp / energy fed.

Energy Utilization (EU)=100×energy gained in shrimp / energy fed.

| Classification | FCR | PPV (%) | PER | Fat Gain (g) | Energy utilization (%) |
|----------------|-----|---------|-----|--------------|-----------------------|
| SD 5           | **  | **      | **  | **           | **                    |
| 15             | **  | **      | **  | **           | **                    |
| 25             | **  | **      | **  | **           | **                    |
| WER 10%        | **  | **      | **  | **           | **                    |
| 20%            | **  | **      | **  | **           | **                    |

Table 4: Effect of stocking density (SD) and water exchange rate (WER) on feed and nutrient utilization efficiency of marine shrimp (Penaeus semisulcatus) post-larvae cultured in earthen grow-out ponds.

(b) Higher water exchange rate (WER) led to higher DO2 and pH and lower ammonia.

(c) There were a significant interaction between PLs stocking density (SD) and WER for DO2 and ammonia.

Growth performance

The changes in mean body weight (g/PLs) of tiger shrimp stocked in earthen pond are presented in Table 3. It can be concluded from this table, mean final weight, gain in weight, gain in weight %, and SGR were significantly (P<0.05) the best at the lowest stocking density.

While, total production exhibited significantly the opposite trend. Harvests and production estimates increased with increasing stocking density.

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| Classification | FCR | PPV (%) | PER | Fat Gain (g) | Energy utilization (%) |
|----------------|-----|---------|-----|--------------|-----------------------|
| SD 5           | **  | **      | **  | **           | **                    |
| 15             | **  | **      | **  | **           | **                    |
| 25             | **  | **      | **  | **           | **                    |
| WER 10%        | **  | **      | **  | **           | **                    |
| 20%            | **  | **      | **  | **           | **                    |

Means (+S.E.) in the same column having the same superscripts are not significantly different (P>0.05). Significant level: N.S.=P>0.05, *P<0.05, ** P<0.01.

1 Average daily gain (ADG)= (Final wt—Initial wt.)/ period (days).
2 Specific growth rate (SGR)=100 × (ln Final wt—in Initial wt.)/ period (days).
3 TP: kg/ha = total production = total weight of shrimp in the pond at harvest (gm) / Total area of pond (m²) x 10000

Table 3: Effect of stocking density (SD) and water exchange rate (WER) on growth performance of marine shrimp (Penaeus semisulcatus) post-larvae cultured in earthen ponds. Culture period was 205 days.

| Classification | FCR | PPV (%) | PER | Fat Gain (g) | Energy utilization (%) |
|----------------|-----|---------|-----|--------------|-----------------------|
| SD 5           | **  | **      | **  | **           | **                    |
| 15             | **  | **      | **  | **           | **                    |
| 25             | **  | **      | **  | **           | **                    |
| WER 10%        | **  | **      | **  | **           | **                    |
| 20%            | **  | **      | **  | **           | **                    |

Presented data are an average of three readings + S.E. Means (+S.E.) in the same column having the same superscripts (capital) are not significantly different (P<0.05).

Significant level: N.S.=P>0.05, *P<0.05, ** P<0.01.

1 Feed conversion ratio (FCR).
2 Protein Productive Value (PPV %)=100 × protein gained in shrimp body (gm)/ protein fed (gm).
3 Protein Efficiency Ratio (PER)=Gain in the shrimp weight (gm)/ protein fed (gm).
4 Energy Utilization (EU)=100 × energy gained in shrimp / energy fed.

Table 4: Effect of stocking density (SD) and water exchange rate (WER) on feed and nutrient utilization efficiency of marine shrimp (Penaeus semisulcatus) post-larvae cultured in earthen grow-out ponds.

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production increased with increasing stocking density (Figure 2 and Table 5). The total production was significantly affected by stocking density (P≤0.05), and affected by water exchange rate (Table 5). The results of feed conversion ratio (FCR), protein productive value (PPV %), protein efficiency ratio (PER), gain in fat and energy utilization were significantly (P≤0.05) affected by stocking density and feeding levels and exhibited the best results at the lowest stocking density and lowest water exchange rate.

Survival rates

The data on the survival percent are presented in Table 5. The results of the analyses of variance of survival rates were showed highly significant (P<0.05) affected by stocking densities and water exchange rates. Survival rates were decreased significantly with increasing stocking density and increased with increasing water exchange rate. The lowest survival rate was at high stocking densities (25 PLs/m²) and the higher survival rate at lower stocking densities 5 PLs/m². The Economic information for tiger shrimp reared in earthen ponds for 10 months at three stocking density and two-water exchange rate are presented in Table 5. From this table the net profits were directly related to stocking density and water exchange rate. The total cost increase by increasing stocking density and water exchange rate. From the economic information it can be concluded that the highest net profit was achieved at stocking density of 15 PLs/m² at water exchange rate 20 % in earthen ponds.

Discussion

Besides genetics and environmental condition, food supply and stocking density are important factors affecting growth and maturation of wild and cultured shrimp [6,16]. As stocking density increases, competition for food and living space usually intensifies providing one of the most effective controls on animal production [17]. The effects of density may be divided into two categories: the density dependent and the density-independent. The stocking density that significantly affects the growth of shrimp was considered as the density dependent category, such as the cases found for Shrimp, Litopenaeus vannamei Boone [18]. In the present study, the similar case of negative curvilinear relationship was found between stocking density and growth weight of tiger shrimp (Figure 2). The changes in growth of shrimp are physiological response to environmental condition [19]. Water quality has complex side effect on high stocking density. Palomino et al. (2001) found that higher stocking density was accompanied by lower pH and dissolved oxygen and suggested that the resulting changes in water quality might play an important role in affecting growth and survival of shrimp [6]. Preston et al. (2002) indicated that metabolic wastes, which are directly proportional to stocking density, have been implicated in inhibiting the growth of fish to be toxic to shrimp [20].

Moreover, shrimp need oxygen for aerobic generation of energy for body maintenance, locomotion, feeding and biosynthesis. A minimum dissolved oxygen level of 3.0 ppm was recommended during cage culture of tilapia in freshwater [21]. In the present study, a minimum dissolved oxygen level was maintained above 5.8 mg/l there are significant differences among the ponds, and effect on growth was observed. The water flow system in the present study provided good water quality consistently throughout the experimental period at all ponds. There were no evidences of large physicochemical fluctuations, occurrence of disease and handling stress, and deterioration of water quality in the experimental ponds during the course of the experiment.

Besides water quality, the effect of stocking density on shrimp might be dependent upon the biological characteristics. Such as, tolerance to environmental change, life stage, sex, social interaction and behavior, so that the density effect on growth and production might be explainable by their competition for territories, with similar case found for Litopenaeus setiferus [6,22]. Behavioral studies on L. setiferus indicating that growth inhibiting antagonistic behavioral patterns were generally unabated even at the highest stocking density. The stress on shrimp caused by the crowdedness may be the other explanation for the effect of stocking density. Allan and Maguire (1992) also found that the highest biomass (Harvest) was achieved at the highest stocking density for Penaeus monodon cultured in ponds [23]. Culture of Penaeus indicus in ponds showed that the highest stocking density achieved the highest biomass [5,24]. In our experiment, the highest biomass was achieved at stocking density of 15 PLs/m² at water exchange rate either 20%. It was been found that the stocking density affected the growth of tiger shrimp significantly and water exchange rate. Shrimp reared at low density grow better than those reared at high density Table 3 and the differences were highly significant. Final mean weight were inversely proportional to stocking density, which was particularly evident when average weight of shrimp reared at the lowest stocking density was significantly different from weight of shrimp reared at the higher densities. Stocking density also affected the growth of L. setiferus and P. vannamei cultured in concrete ponds at three different densities [25,26].

Shrimp reared at the highest density had the lowest final mean weight. These results may be attributed to the shrimp at low density consume maximum amount of food available and growing fast. In addition, reported that slow growth of shrimp at high density was probably due to that the individuals disturbing each other during feeding and normal activity [6]. Also, attributed the decrease in growth rate with increasing density to the reduced food consumption. The data on feed conversion ratio given in Table 4, confirm this finding, whereas the fish reared at low density and 20% water exchange rate possessed the better feed conversion (the shrimp used less feed to produce one unit of gain in body weight) than those reared at high density and 10% water exchange rate. While, final harvest and production values were directly related to stocking density, there must be some density at which mortality is severe for a variety of causes and growth rate is reduced. When this occurs, production will be reduced. This critical level was not reached in our experiment although the stocking density of 25 PLs/m² was high. One reason, for the ability of tiger shrimp to maintain...
high production levels when available oxygen present and unionized ammonia is reduced. Rearing densities of 15 PLs/m² for tiger shrimp would seem to be the most desirable in the system studied. There was a strong trend for both production and final harvest to increase with increasing stocking density. These results are in agreement with those of reported that productions of shrimp culture are generally depended on the stocking density, and water exchange rate [6].

Therefore, the results of the present study showed that stocking density was positively correlated with total production per unit area (r=0.987) and negatively correlated with individual weight gain (r=0.997). The levels of water supply appeared to influence on size at first maturity in shrimp. Shrimp that received more water exchange rate grew faster and matured at larger size, whereas shrimp that received less water exchange rate grew slowly and matured at smaller sizes. Similar observations have been reported in a number of shrimp species [23,27,28]. Water exchange rate availability influenced the total percentage of mature shrimp in the population considerably, being highest in shrimp receiving highest level of water exchange rate and lowest in shrimp receiving lowest level of water exchange rate. In addition, water exchange rate supply influenced significantly the mean final weight and food conversion ratio. In the present study, tiger shrimp received 20% water exchange rate had significantly the best mean final weight and feed conversion ratio compared with 10% water exchange rate. Allan and Maguire (1993) reported similar results for final mean weight and feed conversion ratio compared with 10% water exchange rate supply. There was a strong trend for both production and final harvest to increase with increasing stocking density.

Finally, semi-intensive culture of tiger shrimp in earthen ponds with flow and supplemental continuous aeration significantly influenced by stocking density and water exchange rate. Growth and production of shrimp were significantly related to stocking density and water exchange rate. It can be concluded that, the best desirable stocking density was 15 PLs/m² and 20% water exchange rate in the system studied which give the highest mean final weight, better feed conversion ratio, better production and economically had the highest net profit.

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