Effect of Different Stocking Densities on Growth Performance and Survival of Tire Track Eel (*Mastacembelus favus*) in Recirculating Aquaculture System (RAS)

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Abstract: ***A study was conducted to evaluate the effect of different stocking densities on the growth performance of Tire track eels (Mastacembelus favus) in RAS to advise the farmers to adopt this technology of eel culture. This study was divided into two phases: nursing and grow - out phase. In the nursing phase, M. favus eels with the initial weight of 0.005 g were stocked at the densities of 500, 1000, 1500, and 2000 eel/m^3 in the culture tanks of RAS. Feeding was done with Moina and Tubifex twice a day up to satiation and nylon substrates were placed for the hiding purpose of the eel fry. The highest survival was in the stocking density of 1000 eel/m^3 (71.44%) and the lowest was in density of 2000 eel/m^3 (56.44%). Statistical analysis by One - way ANOVA showed that there is a significant difference in the weight gain, length gain, DWG, DLG, and SGR among the treatments ($P < 0.05$). The stocking density of 1000 eel/m^3 showed the highest growth. Similarly, for grow - out phase, the tire track eels were stocked at 100, 200, 300, and 400 eel/m^3 with an initial body weight of 4.43±1.232 g. Feeding was done 2 times (8 AM and 5 PM) per day manually at satiation level for all treatments using commercial pellet feed (Micro 80) containing 42% CP. The growth parameters were not significantly different in all treatments ($P > 0.05$). FCR resulted lowest with the value of 0.94±0.12 in the highest stocking density treatment (400 eel/m^3). The best suitable stocking density was found to be 1000 eel/m^3 for the nursing and 400 eel/m^3 for the grow - out of M. favus in RAS. Based on these findings, it is clear that stocking density highly influences the growth performance and the survival of M. favus in RAS.***

Keywords: *Mastacembelus favus*, Tire Track eel, Stocking density, RAS, Growth performance, Survival rate

1. Introduction

Aquaculture plays a major role in aquatic food production, which helps in maintaining the per capita consumption in the world. It is the most possible and feasible approach to provide aquatic products for ever - increasing demand in the market [1]. The production increase is followed by ecological concerns related to environmental issues such as untreated wastewater disposal, sediment accumulation, and also the quality and safety of the products. The major obstacles towards conventional aquaculture are lack of space for expansion, high competition for new sites, and limited water availability. RAS is one successful solution for fish rearing to address this challenge of space and water scarcity [2].

Recirculating aquaculture systems (RAS) have been designed to overcome pollution issues and the stocking capacity limitations of traditional land - based aquaculture facilities [3]. Modern aquaculture has become an important component of recirculating aquaculture. The rapid growth of RASs, which appear to predominate concerning the traditional fish pond aquaculture "flow - through" systems, [4] was prompted by the need to position the production units near the markets i. e. near the areas with high population density [5]. Recirculating aquaculture systems are mechanically sophisticated but biologically complex culture systems. Fish must be reared intensively for the production to be cost - effective. The main purpose of developing the RAS system is intensive fish farming when water availability is restricted. Compared to the conventional aquaculture system, RAS uses less than 1% of the land area and provides environmentally safe waste management treatment [1]. This principle of fish culture can be implemented for the farming of fish, shrimps, clams, etc. [6]. The culture of African Catfish (*C. gariepinus*) in Europe and America has been introduced as an intensive culture of fin fishes in the Recirculating Aquaculture System (RAS), a processing technique that reuses fish rearing water more than once, thus reducing space and water requirements for fish culture [7].

Adoption of RAS comes with the issues of the high cost of establishment, limited knowledge on its operation, the expertise on the working principle of RAS, and the inappropriate design for certain species. Along with the modernization of aquaculture practices, RAS also has been enhanced with a variety of techniques such as the application of computer vision technology [8]. Intensification in aquaculture raises the concern of water quality. So, to aid the RAS technology development of different equipment like foam separator (protein skimmer), biofilter, thermal regulation system, UV - sterilizer lamp, ozonizer, and denitrification chamber [4].

In aquaculture, stocking density is the term that is normally used to define the initial number or biomass of fish per unit volume of tank or pond capacity or per unit volume in unit time of water inflow through the holding environment [9]. The pivotal factor which affects the fish growth performance in aquaculture is stocking density. Especially, in recirculating aquaculture systems, the main goal is aimed at high productivity with higher stocking densities in confined environments [10].

The culture of Asian swamp eel (*Monopterus albus*) has shown better growth in the aquaponics recirculating aquaculture system. Producing 1kg of *M. albus* requires 17.70±1.31 m^3 water in 65 culture days in a recirculating aquaculture system [11]. A higher stocking density of 15.0 kg/m^3 (180 ind. /m^2) yielded higher production of swamp eel. *Anguilla marmorata* reared in 1.0 m^3 culture tanks of recirculating aquaculture systems at the stocking densities of 12 kg/m^3 (70 fish/m^3), 20 kg/m^3 (117 fish/m^3), and 28 kg/m^3 (165 fish/m^3), showed a limited increase in growth with the increasing stocking density. Also, the stress level was higher with the increased stocking density [12]. The present study aims to determine the effects of stocking density on the growth performance and survival rate of *Mastacembelus favus* in Recirculating Aquaculture System (RAS) at two stages.

2. Materials and Methods

2.1 Experiment location

This study was conducted in the wet laboratory of the Department of Freshwater Aquaculture in College of Aquaculture and Fisheries, Can Tho University, Vietnam. The experiment was carried out in an indoor facility for 105 days.

2.2 Experimental fish

Tire track eel (*Mastacembelus favus*) were brought and stocked in 150 L tanks and were treated with iodine (2 ml/m^3) for the removal of all the possible ectoparasite. They were acclimatized for 5 days to inspect the signs of any kind of pest or disease. After monitoring the water quality parameters, they were finally transferred to the specialized tanks for recirculating the aquaculture system.

2.3 Experimental Design and Operation

This research is mainly comprised of two experiments. Each treatment had three replicates and separate RAS units. One RAS was designed with a 150 L - circular culture tank, 100 L - biofilter tanks, 100 L - solid collection tanks, and 50 L - mechanical filter tanks. All the culture tanks were fitted with air stone diffusers for oxygen supply and a pump was used to circulate the water throughout the systems. Plastic biomedia (80 L) with a total surface area of 750 m^2/m^3 were used as biofiltration media in all the biofilter tanks.

The nursing phase comprised of 4 treatments and 3 replicates. The eels of 0.005 g (about 0.8 cm in length) were stocked with different densities (500, 1000, 1500, and 2000 ind. /m^3). Nylon substrates were used in the culture tanks. After rearing the eels for about 1.5 months the experiment was stopped. *M. favus* fry was fed with Moina and Tubifex. The system was stopped when feeding to maintain the live food in the culture tanks only. The fry was fed two times a day up to satiation that is until they stopped eating or searching for food. Strong aeration was provided to maintain the dissolved oxygen as the amount of water recirculated was less. Nylon substrates were placed in the culture tanks to provide the hiding place for the eel fry. Monitoring was done every day to check the mortality of the fry.

Similarly, the grow - out phase comprised of 4 treatments and 4 replicates. Then, the eels with an initial body weight of 4.43±1.232 g and initial body length of 12.01±1.117 cm were cultured with different stocking densities (100, 200, 300, and 400 eel/m^3). This experiment was conducted for about 2 months. For the grow - out phase, plastic substrates were used in the culture tanks for the shelter. Feeding was done 2 times (8 AM and 5 PM) per day for all treatments using commercial pellet feed containing 42% CP (Micro 80). Feeding was done manually at satiation level. The total amount of feed given to the eel was measured at the end of the experiment to calculate the Food Conversion Ratio (FCR).

2.4 Water quality monitoring

Water quality parameters like pH, dissolved oxygen (mg/L), and temperature (°C) were measured daily by using the, pH meter, DO meter, and thermometer. The parameters were measured every day at 8: 00 AM and 2: 00 PM. The water samples were collected from the culture tanks and biofilter tanks. Water samples were taken every three days to measure the ammonia, nitrite, and nitrate levels using the Sera test kits. The flow rate of water was adjusted and recorded about 1.5 L per minute.

2.5 Sampling

The growth rate of tire track eel was periodically measured every 30 days. The length and weight were measured and recorded by random sampling of ten eels from each rearing tank by using a small scoop net. From the date of stocking, fish was sampled. After the completion of the experiment, the total number of eels in all the tanks was counted and recorded to calculate the survival rate of the eels. Similarly, all the eels in each tank were batch weighed for the determination of average individual weight and biomass per system. The data obtained was used to determine the effect of different stocking densities on the eel growth performance and survival.

2.6 Data Analysis

All the data was calculated into mean and standard deviation by using Microsoft Excel. The difference in LG, WG, DLG, DWG, and SGR of fish and water quality parameters among the treatments was statistically analyzed with the help of One - way ANOVA and DUNCAN tests using SPSS to identify variations between treatments with a significance level of P <0.05. Data were expressed as mean±standard deviation (SD).

The growth parameters were calculated by using the following formulae:
Weight Gain = Final weight - Initial weight
Length Gain (cm): Final length - Initial length
Daily Weight Gain (DWG): (Final weight - Initial weight) / Culture Days
Daily Length Gain (cm/day) = (Final length - Initial length) / Culture Days
Specific Growth Rate (%) = (ln Final weight - ln Initial weight) /Culture Days×100
Survival Rate (%) = (No. of eel harvested/No. of eel stocked) ×100
Total initial biomass (kg) = Total initial number × Individual weight
Total final biomass (kg) = Total final number × Individual weight

3. Results and Discussion

3.1 Water quality parameters

In the nursing phase, the effect of different stocking densities on the growth performance of *M. favus fry* was observed in recirculating aquaculture system. The temperature ranged from 26°C - 29°C, pH from 7 - 8, and dissolved oxygen from 5 - 6 mg/L. In the study done by [13] in the recirculating aquaculture system, the pH ranged from 6.2 - 7.5, temperature 25 - 27ºC, dissolved oxygen from 6 - 7 mg/L, ammonia 0.001 - 0.002 mg/L, nitrite 0.06 - 0.13 mg/L, and nitrate from 15 - 93 mg/L. *M. pancalus* tolerates a pH between 6 and 8 but pH 7 is suitable for better survival [14]. The study also revealed that larval rearing of this species is very sensitive to the water quality. The dissolved oxygen level reached the maximum value of 7.16mg/L and the minimum value of 3.74mg/L while rearing.

In the grow - out phase, the influence of the stocking densities on the water quality parameters in the culture tanks and biofilter tanks was observed. The temperature ranged from 28.0 - 29.7ºC, pH 7.4 - 8.2, dissolved oxygen 5.5 - 5.7 mg/L which did not drop down below 5 mg/L because of the recirculating aquaculture system. Also, kH in the culture tanks was recorded to know the buffering capacity of the water. Low kH indicates the less buffering capacity of the water which means pH fluctuates easily but in this study kH ranged from 75 to 85 mg/L. The concentration of NO_3, NO_2, and NH_3 increased with the increase in the stocking density. The highest nitrite level recorded was 0.47±0.036 mg/L in the high stocking density treatment and the lowest was 0 mg/L in 100 and 200 eel/m^3 densities. Similarly, the highest levels of nitrate and ammonia were observed in the high stocking density treatments (20.3±3.59 mg/L and 0.005±0.0016 mg/L). Nitrite is considered to have a toxic impact on the fish which in this experiment was below 0.5 mg/L in all the culture tanks. Stocking density also affects the nitrate and nitrite levels in the rearing of eels in recirculating systems. The nitrate levels ranged from 1.43±1.36 mg/L to 2.36±1.43 mg/L and from 3.61±3.20 mg/L to 6.31±3.77 mg/L in lower to higher stocking density (100 and 260 inds. /m^2) respectively [11].

Stocking density is the key element for the deterioration of the water quality in the culture system which means higher the stocking density higher the production of waste like feces and uneaten feed [15].

3.2 Survival Rate

Stocking density showed great influence on the survival of eel during nursing. The highest survival was 71.57% which was obtained in treatment with stocking density 1000 eel/m^3 and the lowest was 56.41% in treatment 2000 eel/m^3. The treatment with stocking density 1000 eel/m^3 was significantly different as compared to other stocking densities ($P < 0.05$) (Figure 1). A similar, result can be observed in the previous study where the survival rate was highest in low stocking density than in the high stocking density which was 90.0% in 200 fish/m^2 and 83.3% in 600 fish/m^2 [16]. Similarly, the survival rate declined from 59 - 61% as the density increased where stocking densities 100 fry/m^2 and 200 fry/m^2 attained significantly higher survival rates ($P < 0.05$) [17]. The survival rate of *M. favus* reared for 45 days was found to be 60% [18].

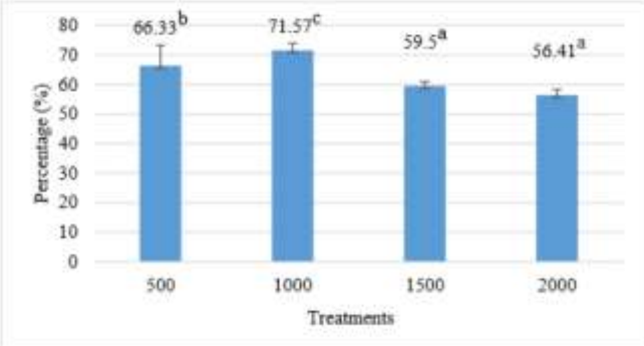

Figure 1: Survival rate (%) of eel during nursing (*Note: The value are in mean±SD. Different letters (a, b, c) among the bars indicate a significant difference, p < 0.05)*

However, during the grow - out phase, all the stocking densities resulted in a 100% survival rate. This result shows that all the stocking density did not have any influence on the survival of tire track eel during the grow - out in RAS. A similar result was observed in a study done on European eel (*Anguilla anguilla*) where the survival rate was observed to be about 100% in the recirculation aquaculture system [13]. No mortality was observed in any of the stocking densities while rearing *A. marmorata* [12]. This result indicates that marbled eels adapt greatly to the recirculating aquaculture system. A study conducted on the Asian swamp eel (*Monopterus albus*) revealed that the highest survival of the eels was found to be 86.44% in treatment with the stocking density of 180 inds. /m^2 whereas the lowest was 77.84% in 260 inds. /m^2 but there was no significant difference among them ($P < 0.05$) [11]. Rearing *Anguilla japonica* with the initial weight of 20g/tail at the stocking density of 163 tails/ tank for 104 days resulted in the survival rate of 91% where mortality was mainly caused due to power outage and water reconditioner problem [19]. The previous study conducted in *Clarias gariepinus* in the recirculating aquaculture systems with the stocking densities of 20, 000 - 31, 000 for fingerlings, 6, 000 - 9, 000 for juveniles and 150 - 300 fish/m^3 for adult fish resulted in the survival rate of 75 - 80%, 75 - 93%, and 77 - 88% respectively [7].

3.3 Growth parameters

3.3.1 Nursing phase

The growth performance of *M. favus* during the nursing with different stocking densities shows that there is a significant difference in final weight and weight gain of tire track eels (P < 0.05) (Table 1). The weight gain ranged from 1.05 g to 1.53 g where the highest was 1.53±0.07 g in the stocking density 1000 eel/m^3 and the lowest in stocking density of 2000 eel/m^3 (1.05±0.04 g). Similarly, the daily weight gain was highest in 1000 eel/m^3 (0.03±0.01 g/day). There was no significant difference in the daily weight gain of the tire track eels among the treatments (P > 0.05). Statistical analysis by One - way ANOVA showed that there is a significant difference in the length gain parameters among the treatments (P < 0.05) during the nursing of *M. favus*. The average final length was higher in stocking density of 1000 eel/m^3 ($^($8.46±0.59 cm) and lowest in 2000 eel/m^3 density (6.26±0.48 cm). In the stocking density of 1000 eel/m^3, the length gain and daily length gain were the highest (7.64±0.08 cm and 0.17±0.00 cm/day) but the lowest was in 2000 eel/m^3 (5.46±0.00 cm and 0.12±0.00 cm/day). Similarly, the results of SGR of the eels showed a significant difference among all the treatments (P < 0.05) where 1000 eel/m^3 density had the highest SGR (12.72±0.11%) and lowest in 2000 eel/m^3 density. The study conducted on *M. armatus* attained the highest daily growth (0.045g/day) in the low stocking which declined with the increase in stocking densities. The weight gain and SGR (1.09 g and 3.79%/day) were also highest in the low stocking density 200 fish/m^2 as compared to the medium and high stocking densities [16]. A similar study conducted on glass eels (*Anguilla bicolor*) shows that the stocking density (54.95, 109.89, and 164.84 fish/m^3) did not affect significantly on the final weight, weight gain, SGR, FCR, and survival however it had a significant effect on the yield of the eels [20]. In a similar study conducted in *Anguilla bicolor*, the stocking density showed a significant effect on the growth performance of the eels (P<0.05) [21]. The highest weight gain (1.43g), specific growth rate (1.62%/day), survival rate (79%), feed conversion ratio (5.17), and feed efficiency (19.52%) was obtained in the treatment with a stocking density of 3eels/L. The stocking density did not affect significantly on feed conversion ratio and feed efficiency but only on weight gain, SGR, and survival rate (P>0.05) [21].

M. favus fed with Moina and Tubifex resulted in the growth from 1.44±0.54 cm to 6.5±0.18 cm [18]. Similarly, feeding the eel fry with tubifex worms showed significantly better growth, SGR (3.01±0.11%), and survival rate (95.00±1.56%) over other feeds (P<0.05) [22].

Table 1: Growth parameters of M. favus during nursing

Parameters	Treatments (eel/m^3)			
	500	1000	1500	2000
Initial weight (g)	0.005	0.005	0.005	0.005
Initial length (cm)	0.812	0.814	0.804	0.801
Final weight (g)	1.34±0.29^b	1.53±0.31^c	1.07±0.25^a	1.05±0.27^a
Final length (cm)	7.29±1.70^c	8.46±0.59^d	6.77±0.64^b	6.26±0.48^a
Weight gain (g)	1.33±0.04^b	1.53±0.07^c	1.07±0.07^a	1.05±0.04^a
Length gain (cm)	6.48±0.14^c	7.64±0.08^d	5.96±0.07^b	5.46±0.00^a
DWG (g/day)	0.03±0.00^{bc}	0.03±0.01^c	0.02±0.01^{ab}	0.02±0.00^a
DLG (cm/day)	0.15±0.01^c	0.17±0.00^d	0.13±0.00^b	0.12±0.00^a
SGR (%)	12.42±0.07^b	12.72±0.11^c	11.93±0.14^a	11.88±0.09^a

Note: The value shown in the table are in mean±SD. Means with different superscript letters (a, b, c) in the same row indicate a significant difference (p<0.05)

3.3.2 Grow - out phase

In the grow - out phase, all the growth performance parameters such as initial weight, initial length, final weight, final length, weight gain, length gain, daily weight gain (DWG), daily length gain (DLG), specific growth rate (SGR), and survival rate (SR) were not influenced by the stocking density (P < 0.05) (Table 2). The highest weight gain was observed in the stocking density of 400 eel/ m^3 and the lowest was in 200 eel/m^3. The highest stocking density 400 eel/m^3 showed the highest daily weight gain and specific growth rate with the values 0.07±0.01 g/day and 1.06±0.19% respectively. All the growth parameters were observed to be lower in the treatments with the stocking density of 200 eel/m^3.

The average final length was higher in stocking density of 300 eel/m^3 with the value of 15.13±0.43 cm and lowest in 200 eel/m^{3}density (6.26±0.48 cm). In the stocking density of 300 eel/m^3, the length gain was the highest (3.03±0.30 cm) whereas daily length gain was similar in both stocking densities of 300 and 400 eel/m^3. Similarly, the results of SGR of the eels showed that there is no significant difference among all the treatments (P > 0.05) where 400 eel/m^3 density had the highest SGR (1.06±0.19%). During the rearing of marbled eel in the recirculating aquaculture system, the stocking density affected significantly on the final individual weight, growth efficiency, and feed intake. However, the SGR, feed conversion ratio, and protein efficiency rate showed no difference between different stocking densities [12]. In a study done on the swamp eel, the stocking density did not show any significant difference in the daily weight gain and SGR where the highest was in treatment with stocking density 180 inds. /m^2 which was 0.70±0.15 g/day and 1.79 ±0.60%/day respectively. The highest weight gain attained by the swamp eels reared with a stocking density of 180 inds. /m^2 was 41.71±9.05g and the lowest was 26.40±4.02g at the density of 100 inds. /m^2 in the culture period of 65 days [11].

Table 2: Growth parameters of *M. favus* during grow - out

Parameters	Treatments (eel/m^3)			
	100	200	300	400
Initial weight (g)	4.47±0.16^{a}	4.34±0.14^{a}	4.82±0.31^{a}	4.60±0.36^{a}
Initial Length (cm)	12.16±0.19^{a}	11.92±0.14^{a}	12.21±0.25^{a}	12.26±0.36^{a}
Final weight (g)	8.11±1.63^{a}	7.58±1.08^{a}	8.06±0.24^{a}	8.67±0.84^{a}
Final length (cm)	14.44±1.01^{a}	14.04±0.50^{a}	15.24±0.06^{a}	15.13±0.43^{a}
Weight gain (g)	3.64±1.65^{a}	3.24±1.07^{a}	3.25±0.27^{a}	4.07±0.83^{a}
Length gain (cm)	2.28±1.02^{a}	2.12±0.50^{a}	3.03±0.30^{a}	2.87±0.39^{a}
DWG (g/day)	0.06±0.03^{a}	0.05±0.02^{a}	0.05±0.00^{a}	0.07±0.01^{a}
DLG (cm/day)	0.04±0.02^{a}	0.04±0.01^{a}	0.05±0.00^{a}	0.05±0.01^{a}
SGR (%)	0.97±0.33^{a}	0.92±0.23^{a}	0.86±0.09^{a}	1.06±0.19^{a}

Note: The value shown in the table are in mean±SD. Means with different superscript letters (a, b, c) in the same column indicate a significant difference (p<0.05)

Feed consumption and FCR during grow - out

Feed consumption was higher in the highest stocking density treatment 400 eel/m^3 which was significantly different as compared to the other stocking densities (100, 200, and 300 eel/m^3) (P<0.05). It indicates that the higher the stocking density the higher the feed consumption. Similarly, FCR was lowest (0.94±0.12) in the stocking density of 400 eel /m^3 whereas the highest was (1.34±0.37) in the stocking density of 100 eel/m^3. FCR tends to increase as the stocking density decreases. Similarly, feeding behavior was observed to be competitive based on the density of the eels which appeared to be similar to *Monopterusalbus* [11]. Stocking densityalso affects greatly the FCR which is shown by [11], where *M. albus* reared at 180 inds. /m^2 obtained the lowest FCR (3.55±0.06) as compared to other stocking density treatments (100 and 260 inds. /m^2). The previous study was done by [19] on the performance of Japanese eel in an intensive closed recirculating system resulted in the total feed consumption of 11.6 kg, an increase in weight was 7.7 kg, and feed conversion ratio was 67%.

A slight increase in the stocking density has a smaller impact on eel performance, but a greater increase in the stocking density results in significant stress on the fish [12]. Lower stocking density might not alter the growth performance of *A. marmorata*, but higher stocking density significantly affects the growth of the eels [12]. Higher stocking density of swamp eel (*Monopterus albus*) compared to lower stocking densities yields relatively higher production of eel also results in a higheraccumulation rate of nutrients concentration in the system [11].

Biomass production in grow - out

M. favus reared at the higher stocking density of 400 eel/m^3 resulted in the highest biomass production (3.73±0.49 kg/m^3) which was significantly different as compared to other stocking densities (P < 0.05) (Figure 2). The result of this study indicated that with the increase in stocking density the eel biomass also increased. Also, it shows that different stocking densities have a great influence on the production of the eels. A similar result was observed in the study conducted by [11], where the increase in stocking density (260 inds. /m^2) resulted in the higher production of the swamp eel which was 58.06±13.25 kg/m^3 which declined with the decrease in the stocking density with 53.57±8.35 kg/m^3 in 180 inds. /m^2 and 21.76±2.26 kg/m^3 in 100 inds. /m^2.

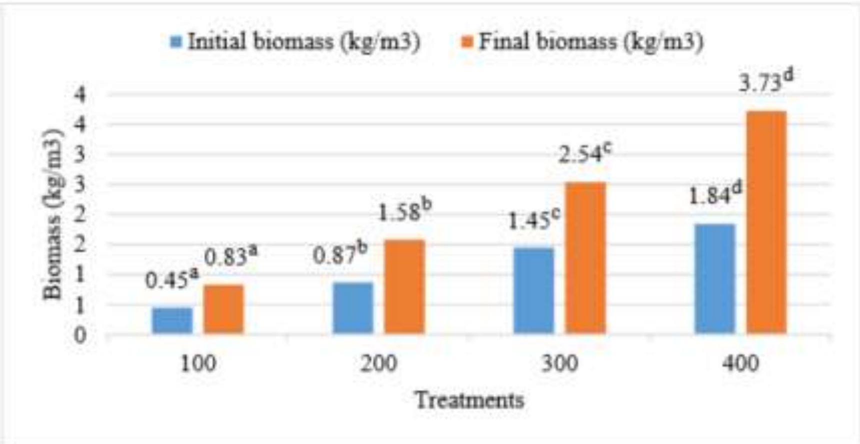

Figure 2: Effect of stocking density on the biomass of *M. favus* during grow - out (*Note: The value shown in the figure are in mean. Different letters (a, b, c) among the bars indicate a significant difference, p < 0.05)*

Variation in weight of eels during grow - out

The size of eels during the experiment was not uniform (Figure 3). The stocking density of 300 eel/m^3 had the highest and 100 eel/m^3 had the lowest variation in weight of eels. The total biomass obtained during harvest was significantly higher where fry was reared at the stocking densities 200 - 300 fry/m^2 (P<0.05). Fry of uniform sizes was achieved from the tanks stocked with the densities 100 and 200 fry/m^2 [17].

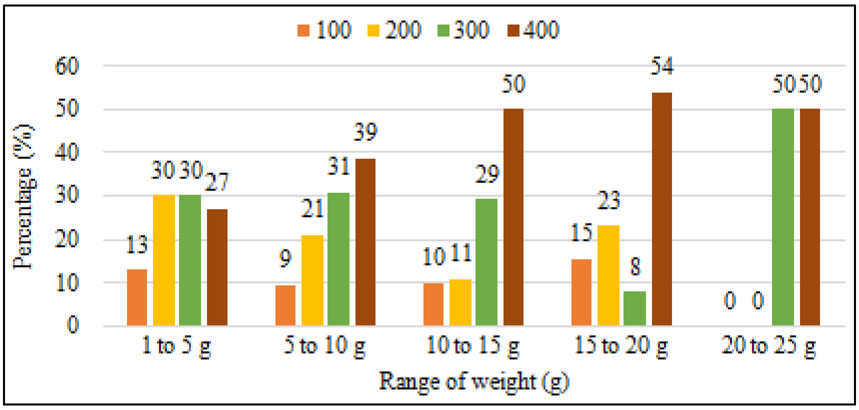

Figure 3: Variation in weight of eels in different stocking densities

4. Conclusion and Recommendation

Based on the findings of this study, it showed that during the nursing of *Mastacembelus favus* in RAS stocking density greatly impacts the growth performance and survival rate. The highest survival and weight gain were obtained in stocking density 1000 eel/m^3. It can be concluded that the stocking density of 1000 eel/m^3 is the best suitable for the nursing of *M. favus* in RAS. In contrast to the nursing, the grow - out of *M. favus* in there circulating aquaculture system showed that the stocking density did not have any significant difference in the growth parameters. All the growth parameters were highest in the stocking density of 400 eel/m^3. Similarly, the survival rate was found to be 100% in all the stocking densities. The feed conversion ratio was the lowest in the stocking density of 400 eel/m^3. The optimum stocking density for the grow - out was found at 400 eel/m^3 in RAS. This study provides an encouraging method as to how the culture of *M. favus* in the recirculating aquaculture system with the controlled environmental conditions.

Further research should be conducted on the evaluation of the effects of stocking density on the growth performance and survival of *Mastacembelus favus* during both nursing and grow - out in the recirculating aquaculture systems. However, only a little research is conducted on *M. favus* in RAS due to which it is quite difficult to adopt this technique commercially. Through this experiment, it is quite clear that more studies on feeding are also required to know the protein requirement of the Tire track eel.

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References

[1] Timmons, M. B. and Ebeling, J. M. (2007). Recirculating Aquaculture. NRAC Publ. No.01 - 007. Cayuga Aqua Ventures, Ithaca, NY.975
[2] Zhang, S. Y., Li, G., Wu, H. B., Liu, X. G., Yao, Y. H., Tao, L. and Liu, H. (2011). An integrated recirculating aquaculture system (RAS) for land - based fish farming: The effects on water quality and fish production. *Aquacultural Engineering*.45 (3): 93 - 102.
[3] Bartelme, R. P., McLellan, S. L. and Newton, R. J. (2017). Freshwater recirculating aquaculture system operations drive biofilter bacterial community shifts around a stable nitrifying consortium of ammonia - oxidizing archaea and comammox *Nitrospira*. *Frontiers in Microbiology*.8: 101.
[4] Mordenti, O., Casalini, A., Mandelli, M. and Di Biase, A. (2014). A closed recirculating aquaculture system for artificial seed production of the European eel (*Anguilla anguilla*): Technology development for spontaneous spawning and egg incubation. *Aquacultural Engineering*, Elsevier B. V.58: 88 - 94.
[5] Barbu, M., Ceanga, E. and Caraman, S. (2016). Water Quality Modeling and Control in Recirculating Aquaculture Systems. *Urban Agric*, 2: 64
[6] Bregnballe, J. (2015). A Guide to Recirculation Aquaculture. An introduction to the new environmentally friendly and highly productive closed fish farming systems, Food and Agriculture Organization of the United Nations (FAO), EUROFISH International Organisation.
[7] Akinwole, A. O. and Faturoti, E. O. (2007). Biological performance of African Catfish (*Clarias gariepinus)* cultured in the recirculating system in Ibadan. *Aquacultural Engineering*.36 (1): 18–23.
[8] Zhou, C., Yang, X., Zhang, B., Lin, K., Xu, D., Guo, Q., and Sun, C. (2017). An adaptive image enhancement method for a recirculating aquaculture system. *Scientific Reports*. Springer US.7 (1): 1 - 11.
[9] Ellis, T., Scott, S., Bromage, N., North, B., and Porter, M. (2001). What is stocking density?. *Trout News*.35 - 37.
[10] Espinal, C. A. and Matulić, D. (2019). Recirculating aquaculture technologies. *Aquaponics food production systems*, Springer, Cham.35 - 76,
[11] Nhan, H. T., Tai, N. T., Liem, P. T., Ut, V. N. and Ako, H. (2019). Effects of different stocking densities on growth performance of Asian swamp eel *Monopterus albus*, water quality and plant growth of watercress *Nasturtium officinale* in an aquaponic recirculating system. *Aquaculture*. Elsevier.503 (December 2018): 96 - 104.
[12] Tan, C., Sun, D., Tan, H., Liu, W., Luo, G. and Wei, X. (2018). Effects of stocking density on growth, body composition, digestive enzyme levels, and blood

biochemical parameters of *Anguilla marmorata* in a recirculating aquaculture system. *Turkish Journal of Fisheries and Aquatic Sciences*.18 (1): 9 - 16.
[13] Mota, V. C., Limbu, P., Martins, C. I. M., Eding, E. H. and Verreth, J. A. J. (2015). The effect of nearly closed RAS on the feed intake and growth of Nile tilapia (*Oreochromis niloticus),* African catfish (*Clarias gariepinus*), and European eel (*Anguilla anguilla*). *Aquacultural Engineering*. Elsevier B. V.68: 1 - 5.
[14] Afroz, A., Islam, M. S., Hasan, M. R., Hasnahena, M. and Tuly, D. M. (2014). Larval rearing of spiny eel, *Mastacembelus pancalus* in the captivity with emphasis on their development stages. *International Journal of Fisheries and Aquatic Studies*.1 (6): 163 - 167.
[15] Myers, P., Espinosa, R., Parr, C. S., Jones, T., Hammond, G. S. and Dewey, T. A. (2021). The Animal Diversity Web (online). Accessed at https: //animaldiversity. org.
[16] Nguyễn Quang Đạt, Trần Đình Luân, Trần Anh Tuấn and Trương Tiến Hải. (2013). ẢNH HƯỞNG CỦA MẬT ĐỘ ĐẾN TĂNG TRƯỞNG VÀ TỶ LỆ SỐNG CÁ CHẠCH SÔNG (*Mastacembelus armatus*) GIAI ĐOẠN ƯƠNG TỪ HƯƠNG LÊN GIỐNG. Tạp chí Khoa học và Phát triển. TRƯỜNG ĐẠI HỌC NÔNG NGHIỆP HÀ NỘI, 948 - 953. (In English).
[17] Sahoo, S. K., Giri, S. S. and Sahu, A. K. (2004). Effect of stocking density on growth and survival of *Clarias batrachus* (Linn.) larvae and fry during hatchery rearing. *Journal of Applied Ichthyology*.20 (4): 302 - 305.
[18] Nguyễn Thành Trung, Nguyễn Tường Anh and Nguyễn Quốc Thanh. (2009). Thử nghiệm sản xuất giống cá Chạch lấu (*Mastacembelus favus*) ", Hội nghị khoa học thủy sản toàn quốc ngày 19/11/2009, Đại học Nông Lâm Tp HCM.16 - 21. (In Vietnamese).
[19] Suzuki, Y., Maruyama, T., Numata, H., Sato, H. and Asakawa, M. (2003). Performance of a closed recirculating system with foam separation, nitrification and denitrification units for intensive culture of eel: towards zero - emission. *Aquacultural Engineering*.29 (3 - 4): 165 - 182.
[20] Sukardi, P., Prayogo, N. A., Winanto, T., Siregar, A. S. and Harisam, T. (2018). Nursery I: The effect of stocking density on the performance of glass eels, *Anguilla bicolor* in the biofloc system. *In E3S Web of Conferences*, EDP Sciences.47: 02009).
[21] Muchlisin, Z. A., Indrianti, Y. S., Dewiyanti, I., Lisra, F., Yanti, R. Z., Nur, F. M. and Batubara, A. S. (2021). Effect of stocking density of the growth performance, survival rate, and feed utilization of the eels *Anguilla bicolor* (Pisces: Anguillidae) Larvae. In *IOP Conference Series: Earth and Environmental Science*, IOP Publishing.674 (1): 012086.
[22] Shifat, J., Rahman, M. M. and Mollah, M. F. A. (2017). Effect of stocking density and different feed on growth and survival of *Mastacembelus armatus* larvae. *Fundamental and Applied Agriculture*.2 (2): 237 - 242.

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