# Effect of Stocking Density on Monosex Nile Tilapia Growth during Pond Culture in India

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**Abstract**—Stocking density is considered one of the important factors affecting fish growth. But, information related to impact of stocking density on growth performance of monosex tilapia population under the ecological conditions of Gangetic plains in West Bengal, India is limited. The aim of our study was to compare the growth potential of monosex tilapia at various stocking densities and to determine an ideal stocking density for culture of all-male monosex fish. The males were isolated by examination of genital papilla region and were stocked separately in 0.01 ha earthen ponds at different stocking densities (5000, 10000, 15000, 20000, 25000 and 30000 fingerlings/ha). It was found that the highest weight, length, daily weight gain, growth rate and protein content were observed for the 20000 fish/ha density class. Thus, culture of monosex tilapia at a density of 20000 fish/ha can be considered ideal for augmented production of the fish under Indian context.

*Keywords*—Growth potential, Nile tilapia, Pond culture, Stocking density.

# I. INTRODUCTION

WITH increase in human population and diminished natural fisheries resources, aquaculture is rapidly gaining importance. Pond aquaculture is growing fast in many resource-constrained Asian countries [1]. In order to maintain the present per capita supply of aquatic products in the future, further growth of aquaculture production is needed as the supply through capture fisheries cannot grow any more [2]. But, fish culture on a small-scale basis has often failed due to inadequate knowledge regarding ideal stocking density of fish [3]. In aquaculture, 'stocking density' should denote the concentration at which fish are initially stocked into a system. However, it is generally used to refer to the density of fish at any point of time. It is considered to be one of the important factors that affect fish growth, feed utilization and gross fish yield [4]. The full utilization of space for maximum fish production through intensive culture can improve the profitability of the fish farm. Fish intensification by increasing stocking density is also found suitable to overcome the problem of land shortage [5].

On the other hand, several studies have indicated an inverse relationship between the stocking density and growth rate of tilapia [6]. Again, relationship between the survival of the fish and stocking density is not found consistent [7]. In practice, the densities at which farmers keep their stock are based on experience and institution, with codes of practice and handbooks being used as a guide. Information regarding effect of stocking density on fish performance during intensive tilapia culture is limited, inconsistent and sometimes controversial [7]. Thus, the impact of stocking density of the fish during the traditional pond culture system needs to be addressed in a coherent manner.

The Nile tilapia, Oreochromis niloticus (Linnaeus, 1758) is a widely cultured species because it grows and reproduces in a wide range of environmental conditions and tolerates stress induced by handling [8]. It is currently ranked second only to carps in global production [6]. The efficiency of reproduction in tilapia has paradoxical consequences. This aptitude allows easy and rapid propagation of the fish in various environmental conditions, but can as well be a source of problem. Within a limited environment, uncontrolled multiplication of the fish not only reduces the faunal diversity of the system but also produces dwarf fish population of poor market value [9], [10]. Monosex culture of male tilapia was postulated to solve this problem and manual sexing by visual examination of genital papillary region of fingerlings is considered a potent method for production of all-male tilapia population [11]. The predominant advantage of monosex culture can be achieved in such aquaculture situations where one sex displays marked growth superiority, as in tilapia [12]. Thus, culture of monosex tilapia might prove effective to induce a positive approach towards tilapia culture in India.

Nile tilapia production occurs primarily in semi-intensive ponds using fertilizers to increase fish yields at low levels of production [13]. It has been widely introduced in the shallow and seasonal ponds of eastern region of India. The fish can form a readily available source of animal protein in the diets of rural and urban dwellers belonging to the lower

socioeconomic strata. Hence, new techniques for maintenance of high growth rate of tilapia are the need of the day. Considering these aspects, the present study aims to evaluate the growth performance of the all-male monosex tilapia population at various stocking densities and determine an ideal stocking density for tilapia culture under the climatic and ecological conditions prevailing in the Gangetic plains of West Bengal, India.

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# II. MATERIALS AND METHODS

The study was conducted at the fish farm in Rajarhat, North 24 parganas, West Bengal, India in 18 newly excavated 0.01 ha freshwater earthen ponds. Initially, the ponds were completely drained, tilled, dried and leveled. Next the ponds were limed at the rate of 0.25 tonnes/ha maintaining a water level at 10 cm. One week after liming, organic fertilizer in the form of cattle manure was applied in heaps at different locations in the ponds at the rate of 1.5 tonnes/ha. After completion of liming and fertilization, the water levels in the ponds were increased to about 1 m. Juveniles of Nile tilapia were collected and sexed by the examination of genital papilla to isolate the males. The isolated males (n=3150; mean weight 35.15  $\pm$  0.36 g; mean length  $13.03 \pm 0.23$  cm) were acclimatized and released in the 18 ponds at six different stocking densities of 5000, 10000, 15000, 20000, 25000 and 30000 fingerlings/ha corresponding to density classes I, II, III, IV, V and VI, respectively, in three replicates. During the next 90 days, the fish were fed a commercial diet (crude protein content 30% and total digestible energy  $3000 \pm 400$  kcal/Kg food) at a rate of 5% body weight per day. The total daily feed ration was divided into four equal portions. Throughout the entire culture period different water quality parameters like temperature, DO<sub>2</sub>, free CO<sub>2</sub>, pH, total alkalinity and turbidity were monitored daily using the standard procedures of American Public Health Association [14].

Fish from each pond were measured individually for weight and length every 15 days and at the end of the 3 months trial. Besides, growth parameters like specific growth rate (SGR), relative growth rate (RGR), daily weight gain (DWG), food conversion ratio (FCR) and protein efficiency ratio (PER) were measured according to standard formulation [15] at the end of the culture period. Crude muscle protein content was also determined using Kjeltec system 1026 distilling unit taking equal amount of wet muscle tissue from 10 fish from every density class. Final harvesting was made through draining of water by water pump.

The data were expressed in terms of mean  $\pm$  standard error (SE). One way ANOVA was performed to compare the survival percentage and growth parameters of different density classes. When appropriate, Duncan's multiple tests (at 5%) [16] was applied to evaluate the differences among means and the statistically homogenous means were denoted by similar alphabets.

# III. RESULTS

The DO<sub>2</sub> values were varied between 7.1 mg/l to 7.4 mg/l in all the different density class ponds (Table I). The temperature was varied between 31.0<sup>o</sup>C in density class III ponds to 32.7<sup>o</sup>C in density class VI ponds (Table I). The pH values in the ponds ranges from 7.5 to 8.0 with the maximum pH values recorded in the density class III ponds (8.0 ± 0.1) (Table I). The Secchi disk readings varies between 28.2 cm in density class V ponds to 40.4 cm in density class I ponds (Table I). The highest (8.0 ± 0.3 mg/l) and the lowest (3.2 ± 0.5 mg/l) free CO<sub>2</sub> concentration were observed in ponds of density classes VI and III, respectively (Table I). The alkalinity values of the ponds were observed between 123.9 mg/l to 144.4 mg/l (Table I).

Density	Physicochemical parameters									
classes	Temperature ( <sup>0</sup> C)	DO <sub>2</sub> (mg/l)	Free CO <sub>2</sub> (mg/l)	рН	Turbidity (cm)	Alkalinity (mg/l)				
Ι	$31.2\pm0.8$	$7.1\pm0.08$	$4.1\pm0.6$	$7.5\pm0.08$	$40.4\pm0.3$	125.6 ± 3.6				
Π	$31.6\pm0.8$	$7.3\pm0.08$	$6.6\pm0.4$	$7.8\pm0.1$	$35.0\pm2.2$	139.7 ± 2.0				
III	$31.0\pm0.7$	$7.1 \pm 0.1$	$3.2\pm0.5$	$8.0\pm0.1$	$35.4\pm0.3$	$144.4 \pm 1.7$				
IV	$31.7\pm0.7$	$7.1\pm0.09$	$5.4 \pm 1.0$	$7.5\pm0.08$	$30.1 \pm 1.0$	125.9 ± 4.6				
V	$32.2\pm0.8$	$7.4\pm0.1$	$7.5\pm0.4$	$7.5\pm0.09$	$28.2\pm0.8$	123.9 ± 2.5				
VI	32.7 ± 1.0	$7.1 \pm 0.1$	$8.0 \pm 0.3$	$7.8\pm0.09$	$28.9 \pm 1.0$	132.1 ± 4.2				

 TABLE I

 Weekly Values of Physicochemical Parameters of Different Density Class Ponds during Monosex Tilapia Culture.

The overall average survival of tilapia in the six density classes was found to be 73.92%. But, stocking density had

significant effect (P-value < 0.01) on the survival percentage of Nile tilapia during pond culture (Fig. 1). The maximum (84.5  $\pm$ 

1.55%) and the minimum ( $62 \pm 1.08\%$ ) survival were found in

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Density	Growth parameters										
classes	Weight (g)	Length (cm)	DWG (g/day)	RGR (%)	SGR (%)	FCR	PER	Protein content (% wet weight)			
Ι	$192.7^{\rm f}\pm2.3$	$21.2^{\rm f}\pm 0.2$	$1.74^{\rm f}\pm 0.02$	$448.43^{f} \pm 0.6$	$1.9^{\text{d}} \pm 0.04$	$2.5^{\text{a}}\pm0.04$	$1.3^{\text{d}}\pm0.03$	$13.8^{\rm e}\pm0.04$			
Π	$231.0^{d}\pm2.3$	$22.6^{\rm d}\pm0.2$	$2.19^{d}\pm0.08$	$557.48^{d}\pm0.6$	$2.1^{\rm c}\pm0.06$	$2.3^{\rm b}\pm0.02$	$1.4^{\rm c}\pm0.02$	$14.18^{d}\pm0.09$			
III	$291.6^b\pm2.4$	$24.7^{\text{b}}\pm0.1$	$2.87^{\text{b}} \pm 0.02$	$729.95^b\pm0.3$	$2.3^{a}\pm0.05$	$1.9^{\rm d}\pm0.03$	$1.5^{\mathrm{b}} \pm 0.01$	$16.03^{\text{b}}\pm0.1$			
IV	$309.6^{a}\pm2.6$	$25.4^{\rm a}\pm 0.2$	$3.13^{a}\pm0.03$	$780.22^{a}\pm0.5$	$2.4^{a}\pm0.03$	$2.1^{\rm c}\pm0.03$	$1.6^{a} \pm 0.01$	$16.58^a\pm0.09$			
v	$261.5^{\rm c}\pm1.6$	$23.5^{c}\pm0.1$	$2.49^{\text{c}}\pm0.02$	$644.15^{\rm c}\pm0.3$	$2.2^{\text{b}}\pm0.02$	$1.8^{\text{e}} \pm 0.02$	$1.5^{\text{b}}\pm0.02$	$15.73^{\rm c}\pm0.09$			
VI	$208.9^{\text{e}} \pm 2.2$	$21.8^{e}\pm0.1$	$1.93^{\text{e}} \pm 0.02$	$494.23^e\pm0.4$	$1.9^{\rm d}\pm 0.02$	$2.4^{a}\pm0.03$	$1.4^{c}\pm0.02$	$14.15^{e}\pm0.06$			

 TABLE II

 GROWTH PERFORMANCES OF MONOSEX NILE TILAPIA AT VARIOUS STOCKING DENSITIES DURING POND CULTURE. SIMILAR ALPHABETS DENOTE HOMOGENOUS

the density classes I and VI, respectively (Fig. 1). The highest growth was observed in the density class IV corresponding to 20,000 fish/ha (Table II). This density class showed the highest weight, length, RGR, SGR, PER and protein content among all the density categories (Table II). Interestingly, FCR was maximum for the density class I where stocking density was 5000 fish/ha and minimum for density class V corresponding to 25,000 fish/ha (Table II). The overall average production of all the density groups were  $3195.6 \pm 287.8$  kg/ha. Stocking density had significant effect (P-value < 0.01) on the total production of fish during culture (Fig. 2). Among the six different density categories, ponds of class IV showed maximum yield while the minimum yield was observed in density class I (Fig. 2).

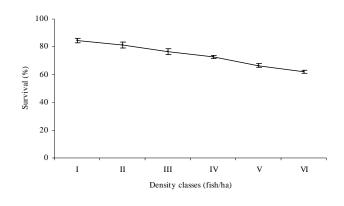


Fig. 1 Survival percentage of monosex tilapia under different stocking density classes. Class I: 5000 fish/ha; Class II: 10,000 fish/ha; Class III: 15,000 fish/ha; Class IV: 20,000 fish/ha; Class V: 25,000 fish/ha; Class VI: 30,000 fish/ha.

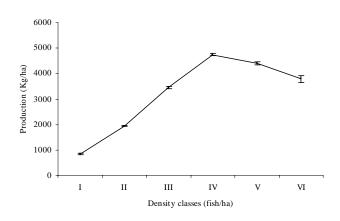


Fig. 2 Total production of monosex tilapia under different stocking density classes. Class I: 5000 fish/ha; Class II: 10,000 fish/ha; Class III: 15,000 fish/ha; Class IV: 20,000 fish/ha; Class V: 25,000 fish/ha; Class VI: 30,000 fish/ha.

#### **IV. DISCUSSION**

Different physicochemical parameters of water like temperature, DO<sub>2</sub>, free CO<sub>2</sub>, transparency and pH are generally considered to have primary importance in fish culture [17]. The optimum temperature for tilapia culture is reported to be 20- $30^{\circ}$ C or above [17]. The temperatures in all the different density class ponds were more or less stable during the culture (Table I). The ideal DO<sub>2</sub> level for tilapia culture is 4-5 mg/l and the present study showed higher DO<sub>2</sub> values for all the density class ponds attributing good environment for tilapia culture. Free CO<sub>2</sub> is another factor that negatively affects feed intake and therefore fish growth [18]. But, Nile tilapia can tolerate CO<sub>2</sub> concentration above 20 mg/l and is unlikely to have an adverse effect on fish in intensive culture systems unless free CO<sub>2</sub> concentration reaches 100 mg/l [18]. The pH values of the ponds in all the density classes (Table I) were within the permissible optimum range of 6.5 to11.0 for tilapia culture and the high pH values (around 7.5) in the ponds correspond to the low accumulation of toxic  $CO_2$  in those systems. In the present investigation, the transparency of the pond water for all the density groups are around the ideal value limits.

Stocking density is highlighted as an area of particular concern in the welfare of intensively farmed fish. The average survival of monosex tilapia in this study corroborates well with other tilapia culture experiments [17]. Different authors had found limited effect of stocking density on fish survival and demonstrated that cannibalism could be a main cause of tilapia fry mortality at high stocking densities [7]. But, the result of this study indicates an inverse relationship between fish survival and stocking density (Fig. 1). Stocking fish beyond an optimum level can cause a significant increase in fish mortality leading to reduced production (Fig. 2).

Moreover, the growth performance of tilapia is significantly related with the stocking density of the fish (Table II). The results emanating from this study shows that there is an inverse relationship between the density and growth potential of the fish when it is stocked at a very high density. But interestingly, at a very low density also the fish has a poor growth rate. The maximum growth of the fish is obtained at an intermediate stocking level of 20,000 fish/ha (Table II) that also provides a reasonably well survival percentage (Fig. 1).

The negative correlation between growth rates and stocking density of fish fry has been postulated by a number of authors. It was observed that increased fish biomass of Nile tilapia in cages had a significant negative effect on the final mean body weight [19]. Monosex Nile tilapia stocked in ponds at a low density showed better growth than at a higher density [20]. The lower growth performance of tilapia at higher stocking density could have been caused by voluntary appetite suppression, more expenditure of energy because of intense antagonistic behavioural interaction, competition for food and living space [20] and increased stress [21]. It was also reported that increasing stocking density of Nile tilapia fry might have lead to diminishing social dominance, resulting in lower individual growth rates [22]. In addition, the resting plasma cortisol concentrations of Nile tilapia fingerlings were found to rise with increased stocking density [23]. Such high cortisol concentration can be considered as a 'chronic stress response' attributed to 'social stresses' caused by increased density. This, in turn, leads to impaired fish growth, presumably due to the mobilization of dietary energy by the physiological alterations evoked by the stress response [24]. Furthermore, stress due to reduction in space availability was reported to be the primary factor for growth inhibition in other fish like Summer flounder (Paralichthys dentatus) stocked at high densities [25]. Increase in stocking density may also cause deterioration in water quality, resulting in stressful conditions [26], [27].

On the other hand, at the low stocking densities, lack of competition for food and/or social hierarchy can lead to decreased feed utilization efficiency resulting in stunted growth. Here, the difficulty of tracing food particles may have lead to the reduction of feed consumption, and to the flushing of uneaten food with the drainage water, causing the deterioration of feed utilization efficiency. Such wastage of feeding materials can increase the production cost to a great extent and the gross yield at such low densities is also small to compensate the cost. Hence, an optimum density level in terms of economic viability of tilapia culture must be established. Thus, this study enables us to postulate an optimum stocking density level of tilapia for maximum utilization of food and space with minimum stress and energy expenditure resulting in higher growth potential of the fish.

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