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Production, Growth and Effect of Varying Stocking Density of *Clariobranchus* Fry

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Abstract: Hybrid *Clariobranchus* was produced from clariid catfishes, male *Heterobranchus bidorsalis* (HB) and female *Clarias gariepinus* (CG) with Ovaprim hormone (Syndel) to induce maturation. The fertilized eggs were successfully hatched by dry method. Hatchlings were reared to fry on brine shrimp nauplii for 2 weeks. Fingerlings with initial weight/fish (4.70±0.06, 4.52±0.48, 4.78±0.62, 4.62±0.09 g) were stocked in four different treatments and stocking densities of 20, 40, 60, 80 fingerlings, respectively at the nursery phase in the 12 (4×4×2.5 m^3) outdoor concrete tanks for 98 days inside the fish net cage (hapa) (1×0.5×0.5 m^3) in triplicates for growth study. Fingerlings were fed with 45% compounded feed at 3% body weight twice daily. The weight gained/fish (g) of the fingerlings during the 98 days culture period were significantly different ($p<0.05$) which varied from 95.66±4.26 and 169.37±7.76 g. The highest weight gained (169.37±7.76 g) was obtained in (T_1, 20 fingerlings) with the lowest stocking density. This was significantly different ($p<0.05$). Feed conversion ratio and feed efficiency were positively correlated with stocking density in *Clariobranchus* fingerlings. It is advisable to stock the hapa of this size at the maximum of 60 fingerlings m^{-3} even though, appreciable growth could be adequately be harvested at 80 fingerlings per hapa. The specie (*Clariobranchus*) being a hybrid of *Clarias gariepinus* and *Heterobranchus bidorsalis* had appreciable growth and its high cannibalism traits were controlled with adequate and sufficient feeds.

Key words: Catfish, growth, hatchlings, hybrid, stocking density, Nigeria

INTRODUCTION

In Nigeria, getting fast growing fish seed have been a major problem to farmers targeting high yields. Hybrid clariid catfish production has increased rapidly in the last few years and apparently market demand is still increasing. Hybridization is the mating of genetically differentiated individuals or groups and may involve crossing individuals within a species (also known as line crossing or strain crossing) or crossing individual between separate species. This breeding technique is used by aquaculturist in the hope of producing aquatic organism with desired traits. The desired goal is to produce offspring that perform better than both parental species. Among the culturable fin fish in Nigeria, catfish is the most sought after fish species, very popular with fish farmers and consumers, it commands very good commercial value in Nigerian markets (Ezenwaji, 1985; Ayinla *et al*., 1994). The catfish is very important to the sustainability of the aquaculture industry in Nigeria. The blending of high survival rate and fast growth rate into the hybrid *Clariobranchus* offers higher production prospects. The hybrid of *Heterobranchus bidorsalis* and *Clarias gariepinus* is a voracious omnivore, feeding on a wide range of food from live animal prey through aquatic plants to plankton organisms (Madu *et al*., 1999). Studies on the hybridization of catfish families abound but information of stocking density of *Clariobranchus* cultured in net hapas rearing medium is limited. This study report the production, growth and effect of varying stocking density of catfish hybrid between male *Heterobranchus bidorsalis* and female *Clarias gariepinus* reared in fish net hapas.

MATERIALS AND METHODS

Broodstock of male *Heterobranchus bidorsalis* (HB) and female *Clarias gariepinus* (CG) were interbred (CGxHB) at the hatchery of HEPA Aqua Consultancy Farm Asero Abeokuta, Ogun State, Nigeria between the month of May and August 2008, following procedures describe by Fast (1998). Mature *Clarias gariepinus* catfish females of 1.3 kg were selected and induced to spawn with Ovaprim hormone injected intramuscularly in a single dose of 0.5 mL kg^{-1} of fish.

Combination of *Clarias gariepinus* and *Heterobranchus bidorsalis* fish species were used in ratio (1:2) of CgxHB, respectively. Thirty minutes prior to

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stripping the females, the males were sacrificed because it was not possible to collect milt by manual stripping. The testes were dissected and the milt squeezed out after incisions were made in the tissue. The females were stripped 12 h after hormone injection and the eggs fertilized by gently mixing them with the milt for 3 min. Fertilized eggs were incubated in standard hatching jars and hatched within 24-25 h.

Fry rearing: The hybrids were transferred to rearing troughs where yolk-sacs were absorbed in 3-4 days after hatching. Fry were reared according to the procedure of Fast (1998). They were fed brine shrimp nauplii thrice for 2 weeks. Water conditions were similar to those for egg incubation. Three weeks old fingerlings with mean initial weight (4.70±0.06, 4.52±0.48, 4.78±0.62 and 4.62±0.09 g) were stocked in four different Treatments (T) and stocking densities of 20 fingerlings (T_1), 40 fingerlings, (T_2) 60 fingerlings, (T_3) 80 fingerlings (T_4), respectively at the nursery phase in the outdoor concrete tanks for 98 days inside the fish net cage (hapa) ($1\times0.5\times0.5$ m^3) in triplicates for growth study. The net hapa was tied with the aid of kuralon twine no. 15 to the bamboo poles mounted horizontally on each of the 12 ($4\times4\times2.5$ m^3) concrete fish tanks.

A free board (above water space) of 0.5 m was maintained in each cage throughout the duration of the experiment which lasted for 8 weeks. The net hapa was suspended in water with weighty anchor at the base to stabilize the net and protect them from being disturbed by the wind in the rearing medium. The treatments were allocated to the 12 hapas suspended in concrete fish tanks for growth studies.

Fingerlings were fed with 45% dietary crude protein commercial feed from University of Agriculture, Abeokuta (UNAAB) Leventis Agro Allied pellets at 3% body weight twice daily. The caged fish were sampled for growth bi-weekly using electronic top-loading balance (Model Mettler E200). The following parameters were calculated in order to determine the growth response of the fish:

Initial Weight (g) (IW):

$$\text{Initial weight/fish (g)} = \frac{\text{Total weight of fish (g)}}{\text{Total number of fish}}$$

Final Weight/fish (g) (FW):

$$\text{Final weight (g)} = \text{Weight at final day} - \text{Initial weight of the fish}$$

Total Weight Gain/fish (g) (W.G):

$$\text{Total weight gain (g)} = \text{Final weight/fish (g)} - \text{Initial weight/fish (g)}$$

Weight gain/fish/day (g):

$$\text{Weight gain/fish/day (g)} = \frac{\text{Total weight gain/fish}}{\text{Number of days}}$$

Feed consumed /fish/day (g): Fish were fed based on 3% of their body weight and the weight was measured every 14 days which gives conversion factor of 0.42. The feed consumed per fish per 14 days was calculated as: 0.42 x weights of fish in a replicate/Number of fish in a replicate = W1. The second fortnight was also calculated as W2 and the third W3 up to W7.

Feed consumed per fish per day was calculated as: W1 + W2 + W3 + W4 + W5 + W6 + W7/98 days.

Analysis of variance was carried out in a Completely Randomized Design (CRD) to get the result.

Feed Conversion Ratio (FCR):

$$\text{Feed conversion ratio} = \frac{\text{Feed consumed (g)}}{\text{Weight gain (g)}}$$

Feed Conversion Efficiency (FCE):

$$\text{Feed conversion efficiency} = \frac{1}{\text{Feed conversion ratio}}$$

Proximate analysis of experimental diets and fish samples were carried out by method of AOAC (1990). Results from the experiment were subjected to the One way Analysis of Variance (ANOVA) in a Completely Randomized Design (CRD) and significant means were separated with Duncan Multiple Range Test (DMRT) using Statistical Analysis System (SAS) package. Data were also subjected to Pearson correlation matrix using the same statistical package.

Water quality parameters were monitored weekly during the study. Parameters such as water temperature, pH, dissolved oxygen, Total Dissolved Solids (TDS) and Electrical Conductivity (EC) was monitored and recorded using standard methods (APHA, 1995).

RESULTS AND DISCUSSION

Table 1 and 2 shows the percentage composition and proximate analysis of experimental diet, respectively, fed

Clariobranchus fingerlings during the experimental period with 45% Crude Protein (CP). Table 3 shows the growth performance of *Clariobranchus* fingerlings at different stocking densities. The highest weight gain per fish was recorded in Treatment 1 (169.37±7.76) followed by Treatment 2 stocked with 40 fingerlings m^{-3} (147.84±4.93) and Treatment 3 stocked with 60 fingerlings m^{-3} (106.54±7.34), while the least weight gain was recorded in Treatment 4 (95.66±4.26) stocked with 80 fingerlings m^{-3}.

The final weight per fish were also significantly different among the treatments ($p<0.05$). Treatment 1 with the least stocking density (20 fingerlings m^{-3}) had the highest final weight of (174.07±3.12), followed by Treatment 2 (40 fingerlings m^{-3}) with (152.36±10.24) and Treatment 3 (60 fingerlings m^{-3}) 111.32±6.35, while Treatment 4 stocked with 80 fingerlings m^{-3} had the least final weight of (99.68±13.46).

The feed consumed per fish per day was found to be highest in Treatment 2 (58.72±4.93) followed by Treatment 1 stocked with 20 fingerlings m^{-3} (34.95±1.02) and Treatment 4 stocked with 80 fingerlings m^{-3} (32.76±1.67) and least was found in Treatment 3 stocked with 60 fingerlings m^{-3} (24.45±2.96) which were significantly different ($p<0.05$).

The highest weight gain per fish per day was recorded in Treatment 1 stocked with 20 fingerlings m^{-3} (1.73±0.03) followed by Treatment 2 stocked with 40 fingerlings m^{-3} (1.51±0.89) and Treatment 3 stocked with 60 fingerlings m^{-3} (1.11±0.21) while the least weight gain per fish per day was recorded in Treatment 4 stocked with 80 fingerlings m^{-3} (0.97±0.12). Treatment 1 had the highest feed conversion ratio (20.20±0.85), followed by Treatment 3 (22.43±1.12) and Treatment 4 (33.77±2.24) and the least was Treatment 2 (38.89±0.92).

Treatment 1 stocked with 20 fingerlings m^{-3} had the highest feed conversion efficiency (0.05±0.00), followed by Treatment 3 stocked with 60 fingerlings m^{-3} (0.04±0.01) and Treatment 2 stocked with 40 fingerlings m^{-3} (0.03±0.01) and the least was Treatment 4 stocked with 80 fingerlings m^{-3} (0.03±0.00) which were significantly different ($p<0.05$). The survival rate of *Clariobranchus* fingerlings during the experimental periods with varying stocking period were shown in Table 4. The highest survival rate was found in Treatment 4 (225 fingerlings m^{-3}), followed by Treatment 3 (172 fingerlings m^{-3}) with the least in Treatment 4 (49 fingerlings m^{-3}). The percentage survival rate was

Table 1: Percentage composition of experimental diet fed *Clariobranchus* fingerlings during the experimental period

Ingredients	Percentage composition (%)
Fish meal	27.50
Soyabean meal	26.00
Yellow maize	23.60
Groundnut cake	16.20
Lysine	0.25
Methionine	0.25
Premix	1.00
Salt	0.10
Vitamin C	0.10
Vegetable oil	5.00

Table 2: Proximate composition of experimental feed dry matter (%)

Composition	Percentage
Crude protein	49.04
Crude fibre	6.20
Fat content	9.50
Ash content	8.70
Carbohydrate	26.34

Table 3: Growth response of *Clariobranchus* fingerlings on varying stocking densities

Parameters	Treatment 1 (20 fingerling m^{-3})	Treatment 2 (40 fingerling m^{-3})	Treatment 3 (60 fingerling m^{-3})	Treatment 4 (80fingerling m^{-3})
Initial weight/fish (g)	4.70±0.06	4.52±0.480	4.78±0.62	4.62±0.090
Final weight/fish (g)	174.07±3.12^a	152.36±10.24^b	111.32±6.35^c	99.68±13.46^c
Weight gain/fish (g)	169.37±7.76^a	147.84±4.93^b	106.54±7.34^c	95.66±4.260^c
Feed consumed/fish/day (g)	34.95±1.02^b	58.72±4.93^a	24.45±2.96^b	32.76±1.670^b
Weight gain/fish/day (g)	1.73±0.03^a	1.51±0.89^b	1.09±0.21^c	0.97±0.120^c
Feed Conversion Ratio (FCR)	20.20±0.85^d	38.89±0.92^a	22.43±1.12^c	33.77±2.240^b
Feed Conversion Efficiency (FCE)	0.05±0.00^a	0.03±0.01^c	0.04±0.01^b	0.03±0.000^c

Means followed by different superscripts along the same row are significantly different ($p<0.05$)

Table 4: Survival of *Clariobranchus* fingerlings during the experimental periods

Days	Treatment 1 (20 fingerling m^{-3})	Treatment 2 (40 fingerling m^{-3})	Treatment 3 (60 fingerling m^{-3})	Treatment 4 (80 fingerling m^{-3})
0	60	120	180	240
14	55	118	174	228
28	53	112	173	226
42	51	111	173	226
56	50	111	173	226
70	50	111	173	226
84	49	109	172	225
98	49	109	172	225

Table 5: Percentage survival rate of *Clariobranchus* fingerlings reared at different stocking density in outdoor tank during the period of study

Days	Treatment 1 20 fingerling m^{-3}	Treatment 2 40 fingerling m^{-3}	Treatment 3 60 fingerling m^{-3}	Treatment 4 80 fingerling m^{-3}	SE
0	100.00	100.00	100.00	100.00	0.00
14	91.70	98.30	96.70	95.00	2.86
28	88.30	92.30	96.10	94.20	2.79
42	85.00	92.50	96.10	94.20	2.76
56	83.30	93.30	96.10	94.20	2.75
70	83.30	92.50	96.10	94.20	2.75
84	81.70	90.80	95.60	93.80	2.71
98	81.70	90.80	95.60	93.80	2.71
Total	695.00	751.00	772.30	759.40	-
$\bar{X}$	86.88	93.81	96.54	94.92	-
±	2.61	2.81	2.90	2.85	-

Table 6: Correlation between growth parameters of *Clariobranchus* fingerlings on varying stocking densities

Parameters	Initial wt.	Final wt.	Weight gained	Feed consumed	Feed conversion ratio	Feed conversion efficiency	Density
Initial wt.	1						
Final wt.	-0.199	1					
Weight gained	-0.205	1.000**	1				
Feed consumed	-0.706	0.486	0.489	1			
Feed conversion ratio	-0.909	-0.198	-0.193	0.747	1		
Feed conversion efficiency	0.673	0.564	0.560	-0.445	-0.918	1	
Density	0.027	-0.980*	-0.978*	0.358	0.358	0.349	-0.674

*Correlation is significant at the 0.05 level (2-tailed), **Correlation is significant at the 0.01 level (2-tailed)

recorded in Table 5. Treatment 3 had the highest percentage survival (95.6%), followed by Treatment 4 (93.8%) and the least in Treatment 1 (81.7%). Table 6 showed the correlation between growth parameters of *Clariobranchus* fingerlings on varying stocking densities and Table 7 recorded the ranges of physico-chemical parameters recorded during the experimental periods. The range of water quality parameters in the experimental tank were measured adequately throughout the periods of study. Successful hybridization between *C. gariepinus* and *H. bidorsalis* was established. The high fertilization and hatching rates obtained in the spawning indicate that the dosage Ovaprim hormone (Syndel) injected intramuscularly in a single dose of 0.5 mL kg^{-1} of fish are satisfactory for inducing hybridization of *C. gariepinus* and *H. bidorsalis*. These conditions are similar to Salami *et al.* (1993) in production and growth of Clariid catfish and Agbebi (2008) in growth and survival of diploid and triploid *Heterobranchus bidorsalis*.

The procedure for artificial hybridization described in this study provided a large quantity and percentage of normal fry with large survival rates. This was possible as a result of water quality conditions found to be within the tolerant range Table 7. Few mortalities of the hybrid were observed during the outdoor rearing period when stocks were transferred into net Hapas rearing system for growth performance. The use of high stocking density as a technique to maximize space usage to increase stock production has been shown to have an adverse effect on growth. (Trzebia-Towski *et al.*, 1981). The percentage composition and proximate composition of the

Table 7: Ranges of physico-chemical parameters recorded during the experimental periods

Parameters	Range
pH	6.81-7.32
Temperature (0°C)	29.30-31.5
Dissolved oxygen (mg L^{-1})	6.66-7.21
Electrical conductivity (us cm^{-1})	253-256
Total dissolved solid (ppm)	127-132

experimental diet fed *Clariobranchus* fingerlings during the experimental period as shown in Table 1 and 2 had 45% Crude Protein (CP). This contributed to the fast growth rate and good performance of the fish. This conformed with Ayinla *et al.* (1994) recommended that crude protein and amino acid requirement of *Clarias gariepinus* fingerlings should be between 40 and 45%.

In the growth performance analysis, Treatment 1 stocked with 20 fingerlings m^{-3} had the highest significant final weight per fish (174.07±3.12) and the least in Treatment 4 stocked with 80 fingerlings m^{-3} (99.68±13.46) shows that increased stocking density significantly reduced the growth of the fish. The result of this study was in agreement with what was reported by Jha and Barat (2005) where fish growth was observed to be dependent on the stocking density. The researcher further stated that higher stocking density may cause the crowding of the fish and reduce the growth significantly. There was a significant difference ($p<0.05$) in the weight gain per fish. The highest weight gain per fish associated with Treatment 1 stocked with 20 fingerlings m^{-3} was probably due lesser feed competition and lesser cannibalism which was attributed to sufficient feed administered compared to Treatment 4 where there would be possibility of the fish to suffer stress as a result of aggressive feeding interaction, resulting in growth retardation (Bjoernsson, 1994). The decline in growth performance of Treatments 3 and 4, respectively can be attributed to the fact that the lower the stocking density, the higher the growth. Similar results were found in other mudfish: *C. gariepinus*, maintained at a density of 100 fry m^{-3} grew faster than maintained at 500 m^{-3} (Madu, 1989). According to the researcher, fish stocked at higher densities showed a significant decrease in the mean fish weight, specific growth rate as well as survival compared to those at the lower stocking densities.

In larvae and fry culture, several factors influence survival and growth, for example feeding and stocking density. Sahoo *et al*. (2004) and Rahman *et al*. (2005) recorded that stocking density have adverse effect on growth of fish fry. Suziki *et al*. (2001) observed that increase in stocking density results in increasing stress which leads to higher energy requirements causing a reduction in growth rate and food utilization.

Similar results have been reported in *Cyprinus carpio* larvae (Jha and Barat, 2005). Treatment 3 had the highest percentage survival (95.6%) as shown in Table 5. All the treatments had appreciably percentage survival. The high percentage survival recorded in treatments can be attributed to the adequate time and sufficient feed administered to the stocks during the experimental period. This can also be related to acceptable physico-chemical parameters of the culture medium.

Weight gain and final weight are significantly negatively correlated with stocking density (Table 6). Feed conversion ratio and feed conversion efficiency were positively correlated with stocking density in clariobranchus species.

CONCLUSION

The study has made possible mating of genetically differentiated individuals. The study has confirmed that cannibalism traits of both species has no effect on mortality rate but primarily dependent on their stocking density. The rearing medium guarantee Nigeria's greater fish supply through the net hapa culture system both in natural or artificial water bodies in lakes, estuaries, shallow rivers, ponds, etc. where these hapas could be mounted or anchored without acquire expensive land for culture. The net hapa rearing medium would be an eye opener for rural dwellers to participate in fish farming practices to boost their standard of living and eliminate malnutrition in diets. Stocking density had a significant effect on growth and survival of *Clariobranchus* fry. The experiment was conducted during rainy season (May-August) which aids the physico-chemical parameters and allow for easy dilution of water within the culture medium. At higher densities, treatments >80 fingerlings m^{-3} with same size ($1\times0.5\times0.5$ m^{3}) of hapa will suffer stress and mortality as a result of aggressive feeding interaction. It is advisable to stock the hapa of such size at the maximum of 60 fingerlings even though, appreciable growth could be adequately be harvested at 80 fingerlings per hapa. The specie (*Clariobranchus*) being an hybrid of *Clarias gariepinus* and *Heterobranchus bidorsalis* was expected to have high growth and high cannibalism traits but with adequate and sufficient feeds, the rate of cannibalism was controlled. Adequate sampling to remove the jumpers in all the treatments would improve the growth and survival of the fish to adult stage.

Bigger mesh size (8-12 mm) of net hapas are recommended to allow free flow of water in and out of the hapas especially in the dry season to avoid turbidity. Furthermore, it is advisable to introduce catfish fingerlings to outdoor rearing using bigger sized fish net hapa in other rearing mediums such as earthen ponds and lakes for more appreciable density and growth.

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