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## Effect of Varying Dietary Protein Level and Stocking density on Growth Performance, Survival and Feed Utilization of African Catfish (*Clarias gariepinus* - Burchell, 1822)

Sinyangwe BG<sup>1</sup>, Chirwa ER<sup>2,3</sup>, Mzengereza K<sup>3,4\*</sup>, Kapute F<sup>3</sup>, Singini W<sup>3</sup>, Chirwa BB<sup>5</sup>, Kondowe B<sup>3</sup>, Zidana H<sup>6</sup> and Jere B<sup>2,3</sup>

### Abstract

A 2 × 2 factorial experiment was conducted to determine the effect of varying stocking density and crude protein level on growth performance of the African catfish (*Clarias gariepinus*) fry reared in (1 × 1 × 1 m<sup>3</sup>, L × W × H) tanks for 40 days. African catfish fry weighing 0.01 g/fish were randomly distributed into 12 tanks at either 80 fish/m<sup>3</sup> or 120 fish/m<sup>3</sup>. The fry were fed on diets containing either 38.8% or 52.5% crude protein (CP) respectively at 10% body weight of the fish four times daily. ANOVA was performed on growth, survival and feed utilization parameters in SPSS (16). Mean body weight, percent weight gain, survival rate, and daily growth rate were positively affected by protein level and inversely affected by stocking density (P<0.05), but not affected by their interaction (P>0.05). Results showed that growth performance of fry and overall production of fingerlings of the African catfish increased when fry are stocked at 80 fish/m<sup>3</sup> and fed on 52.5% CP diet.

**Keywords:** African catfish; Stocking density; Crude protein

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### Introduction

The African catfish (*Clarias gariepinus*), family Clariidae, is an endemic fish species in most African countries. It has been introduced and commercially cultured in several countries in Europe, Asia and South America [1]. It is one of the highly demanded freshwater food fish and cultivable species because of its resistance to diseases, ability to tolerate a wide range of adverse environmental conditions, and high stocking densities under poly-culture conditions, relatively fast growth rate and good quality meat [2]. In Malawi, *C. gariepinus* was identified as the most promising candidate for aquaculture with potential yields of more than three times that of the fastest growing tilapia species, *Oreochromis niloticus* [3]. As at now, tilapia (*Oreochromis shiranus*, *Oreochromis karongae* and *Tilapia rendalli*) is the widely cultured species in Malawi notwithstanding the fact that it has premature female sexual maturation that hinders the growth rate leading to stuntedness [4]. The Malawi national annual production of catfish increased from 5 tonnes in 1996 to 17 tonnes in 2003 [5]. Despite the technical support from development partners to promote the development of catfish

in Malawi, inadequate supply of fingerlings remains a major constraint in the culture of African catfish. Poor quality feed and inconsistent stocking densities have among other myriad factors led to poor growth and low fry survival rates to fingerling

- 1 Oxfam in Malawi, Private Bag B331, Lilongwe, Malawi
- 2 Lilongwe University of Agriculture & Natural Resources, P.O. Box 219, Lilongwe, Malawi
- 3 Mzuzu University, Department of Fisheries & Aquatic Science, Private Bag 201, Mzuzu University, Malawi
- 4 Laboratory of Aquatic Animal Nutrition, Faculty of Fisheries, Kagoshima University, 4-50-20, Kagoshima, 890-0056, Japan.
- 5 Ministry of Agriculture & Food Security, Fisheries Department, P.O. Box 593, Lilongwe, Malawi
- 6 National Aquaculture Centre, P.O. Box 44, Domasi, Malawi

\*Corresponding author: Mzengereza K

✉ kumbumzenge@gmail.com

Laboratory of Aquatic Animal Nutrition, Faculty of Fisheries, Kagoshima University, 4-50-20, Kagoshima, 890-0056, Japan.

**Tel:** +888998466

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stage, culminating in overall low fingerling production. Low fry to fingerling survival rate of less than 10% has been common in Malawi catfish culture [6]. Efforts to improve survival rates have been sporadic and inconsistent. In 2000, the National Aquaculture Centre succeeded in improving the survival rate to about 30% and produced over 20,000 fingerlings [7]. However, in 2001, the number of fingerlings decreased to 16,905 due to irregularities in feed quality and stocking of fry [6]. Catfish are omnivorous as well as predatory and therefore have insatiable appetite for high protein to satisfy their requirements. Diets with low protein that do not meet the catfish requirements triggers hierarchal cannibalistic behaviour which degenerate into low survival at fry to fingerling stage. Ultimately, the overall fish production is jeopardised with low economic returns at commercial scale. In intensive aquaculture systems, dietary protein sources must be maximised because of low to absent primary production that fosters secondary zooplankton growth abundant in pond based extensive systems. According to Sirakov, optimum stocking density ensures that feed is utilised by the fish more efficiently and ensure maximum space utilization [8]. Thus, for optimal fry growth and survival and overall fingerling production, catfish fry must be stocked at optimal density and fed the right protein level - variables that are not optimised in Malawi's catfish farming industry at present. The objective of this study was to determine the effect of varying stocking density and crude protein level on the growth of African catfish fry stocked in concrete tanks.

## Materials and Methods

### Production of test fish

Catfish fry for the experiment were obtained by artificial reproduction of mature brood stock following a method recommended by de Graaf and Jensen [9]. Two sexually mature females ( $400 \pm 0.1$  g) and two males (each  $300 \pm 0.1$  g) were randomly selected from pond-harvested fish at the National Aquaculture Centre (NAC) and transported to the hatchery where spawning was induced by hypophysation. The two female and two male catfishes were injected with ova prim and put into a breeding tank with air stone aerators at a regulated water temperature of 26°C.

Egg collectors locally made from fertilizer-sack material were placed in breeding tanks to collect the eggs spawned by the fish. The collected eggs hatched 48 h after fertilization. After complete absorption of the yolk sac, three-day old swim up larvae were transferred to holding tanks.

Larvae were reared inside the hatchery for 7 days while fed on chicken eggs during which egg yolk absorption was monitored under a microscope. After 3 days eggs yolk was completely depleted and coincided with anal opening marking a transition from larvae to swim up fry. The fry were then fed on cattle blood to stimulate digestive activity before being randomly allocated to 1 m<sup>3</sup> experimental tanks.

### Experimental design

The experiment was a 2 × 2 factorial with Complete Randomization. There were two factors, stocking density and crude protein

level, each with two levels, thus a total of four treatments run in triplicate allotted to 12 tanks. Treatment 1 (T1) comprised of fry stocked at 120 fry/m<sup>2</sup> and fed on 38.8% CP diet. Treatment 2 (T2) had fry stocked at 120 fry/m<sup>2</sup> fed with 52.5% CP, Treatment 3 (T3) had fry fed at 38.8%CP stocked at 80 fry/m<sup>2</sup>. Treatment 4 (T4) was stocked at 80 fry/m<sup>2</sup> and fed on 52.5% CP diet.

### Fish stocking, feeding protocol and management

Catfish fry ( $0.01 \pm 0.0$  g) were randomly stocked in twelve tanks each with 1 × 1 × 1 m<sup>3</sup>, L × W × H capacity. The fish were hand fed by broadcasting four times a day (06.00, 10.00 h, 14.00 and 18.00 h) in split-rations at 10% body weight. The JICA catfish feed (**Table 1**) commonly used in Malawi recommended 38.8% CP while trial and error method was used to formulate the 52.5% CP diet (**Table 2**) by carefully selecting feed ingredients that could provide a feed that meet nutrient requirement of catfish (50-55%CP). Catfish fry were fed basal diet one week after stocking as acclimatization to the experimental tanks. After one week of acclimatization, the catfish fry were weaned from basal diet and introduced to experimental feed which lasted for 40 days. Uneaten feed was siphoned out every day before putting in the new feed.

### Sample collection

The initial body weight (g) and total number of catfish fry were recorded at the onset of the experiment during stocking and 30% of the stocked fry in each of the twelve tanks were sampled every ten days to monitor growth and survival. Volumetric comparison method was used to count the fry. A representative sample of fry was counted using a fine-mesh scoop net and then placed in a graduated cylinder containing a pre-measured quantity of water, without adding more water taken together with the fry. Change in water level when the fry was added was then recorded. The total number of fry was estimated by placing total number of fry in a graduated measuring container, recording the water level change, and then comparing the two numbers. The sampled fry were weighed using an electronic digital scale (METTER TOLEDO PG500Z-S Delta Range). Critical water quality parameters (temperature, DO and pH) were recorded using a HANNA Multi-parameter HI 8424 m (HANNA Company, USA)

**Table 1** Ingredient composition of the adopted JICA catfish feed.

Feed Ingredient	Composition (%)	CP (%)
Fish meal	50	26.8
Soya meal	12.5	5.5
wheat flour	25	3.9
Milk powder	10	2.6
vitamin mix	2	0
Mineral mix	0.5	0
<b>Total</b>	<b>100</b>	<b>38.8</b>

**Table 2** Proximate composition of African catfish feed ingredients.

Feed ingredient	Composition (%)	CP (%)	CP in feed (%)
Broiler starter	50	40	20
Fish meal	50	65	32.5
<b>Total</b>	<b>100</b>		<b>52.5</b>

twice per day in the morning (09.00) and afternoon (14:00). Mean Weight Gain (MWG), Feed Conversion Ratio (FCR), Daily Growth Rate (DGR) and Survival Rate (SR) were calculated using formulae as described by Abd Al-Hakim et al. [10] and Pechsiri and Yakupitiyage [11].

### Evaluation of growth performance and feed utilization

MWG (g) was calculated using the formula:

Mean Weight Gain (g) = Mean final weight (g) – Mean initial weight (g)

$$FCR = \frac{\text{Weight of feed given (g)}}{\text{Final weight} - \text{Initial weight (g)}}$$

$DGR = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Experimental days}} \times 100$

$SR (\%) = \frac{\text{Number of fish survived}}{\text{Total number of fish stocked}} \times 100$

### Statistical analysis

Data was analyzed in SPSS version 16.0 using two-way ANOVA at 5% level of significance. Significant differences between means were evaluated using Duncan's multiple range test (DMRT).

## Results

### Growth of African catfish fry

Mean Body Weight (MBW) of fish stocked at 80 fish/m<sup>3</sup> was significantly higher than those stocked at 120 fish/m<sup>3</sup> when fed with either 38.8 or 52.5 %CP level ( $p < 0.001$ ). This difference was not apparent in the first ten days of the rearing period, but continued to show from the end of the first half of the rearing period to the end of the experiment (**Figures 1 and 2**).

Daily growth rate (DGR) and specific growth rate (SGR) were significantly higher and feed conversion ratio significantly lower when the fry were stocked at 80 fish/m<sup>3</sup> and fed on 52.5% CP ( $p < 0.001$ ) (**Table 3**).

Water quality parameters were within normal range recommended for optimal growth of the catfish fry (**Table 4**).

**Table 3** Growth parameters (Mean  $\pm$  SE) for catfish fry fed on 38.8% and 52.5% CP, stocked at 80 and 120 fry/m<sup>3</sup>.

Growth parameter	Crude protein/Stocking density			
	38.8% CP		52.5% CP	
	80 fry/m <sup>3</sup>	120 fry/m <sup>3</sup>	80 fry/m <sup>3</sup>	120 fry/m <sup>3</sup>
Initial MBW (g)	0.01 $\pm$ 0.00 <sup>a</sup>	0.01 $\pm$ 0.00 <sup>a</sup>	0.01 $\pm$ 0.00 <sup>a</sup>	0.01 $\pm$ 0.00 <sup>a</sup>
Final MBW (g)	1.60 $\pm$ 0.11 <sup>a</sup>	1.09 $\pm$ 0.06 <sup>b</sup>	2.07 $\pm$ 0.13 <sup>c</sup>	1.34 $\pm$ 0.08 <sup>d</sup>
MWG (g)	1.59 $\pm$ 0.11 <sup>a</sup>	1.09 $\pm$ 0.06 <sup>b</sup>	2.06 $\pm$ 0.13 <sup>c</sup>	1.33 $\pm$ 0.08 <sup>d</sup>
DGR (g/fish/day)	0.04 $\pm$ 0.00 <sup>a</sup>	0.02 $\pm$ 0.00 <sup>b</sup>	0.05 $\pm$ 0.005 <sup>c</sup>	0.04 $\pm$ 0.006 <sup>a</sup>
FCR	2.01 $\pm$ 0.12 <sup>a</sup>	4.1 $\pm$ 1.04 <sup>b</sup>	1.62 $\pm$ 0.12 <sup>c</sup>	3.01 $\pm$ 0.95 <sup>d</sup>
SR (%)	53.33 <sup>a</sup>	49.45 <sup>a</sup>	79.58 <sup>b</sup>	56.66 <sup>a</sup>

Values without common superscripts in a row are significantly different ( $P < 0.05$ )

**Table 4** Water quality parameters (Mean  $\pm$  SE) in tanks during the experiment.

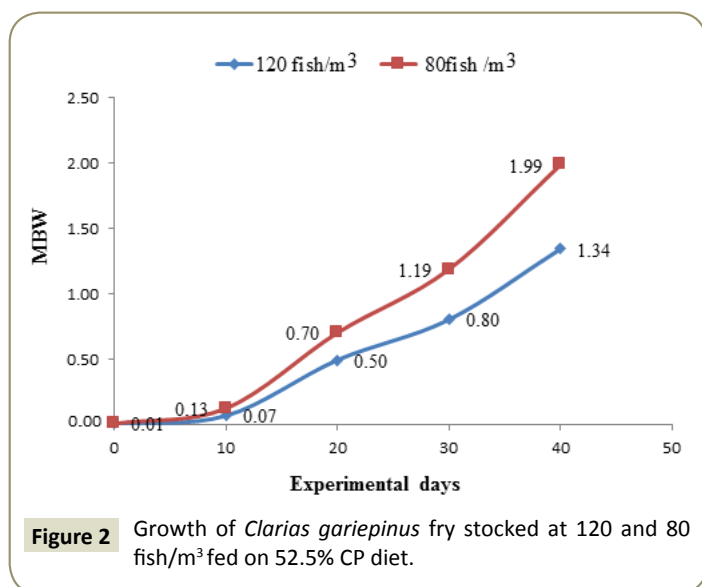
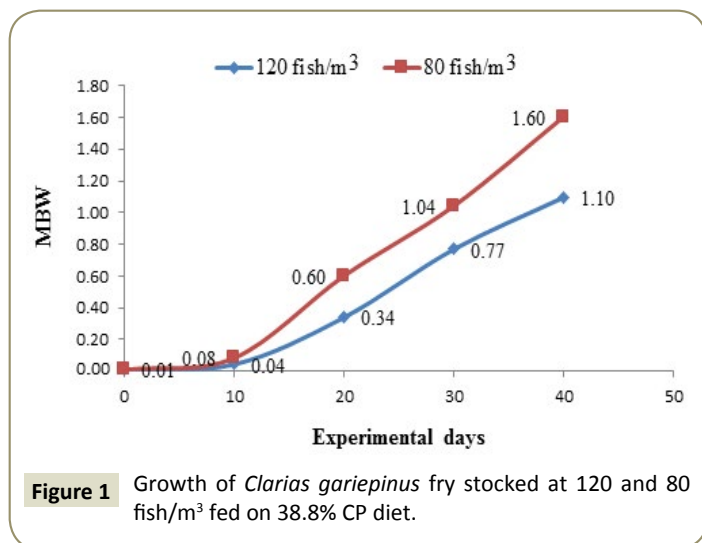
	Mean water temp. ( $^{\circ}$ C)	Mean DO (mg/l)	Mean pH
Minimum	25.65 $\pm$ 0.36	6.74 $\pm$ 0.06	7.29 $\pm$ 0.08
Maximum	28.51 $\pm$ 0.41	7.08 $\pm$ 0.04	7.55 $\pm$ 0.06

## Discussion

Growth is an index of energy gains and losses within an environment. Weight gain is one of the important parameters for measuring growth. Stocking density is one of the main factors determining fish growth. Stocking density had significant effect on growth (DGR and MWG), feed utilisation (FCR) and survival rate (SR) of the catfish fry. Under crowded conditions or higher stocking densities, fish suffer stress as a result of aggressive feeding interaction and eat less, resulting in growth retardation [12]. Suzuki et al. reported increase in stocking density as a result of higher energy requirements for stress-induced activity causing a reduction in growth rate and feed utilization [13]. In addition, high stocking densities entails biological overcrowding, a scenario that increases demand for oxygen to support many physio-chemical systems such as respiration and metabolism. As a result, fish may succumb to suffocation at sublethal or anoxic oxygen levels. It is therefore important, that stocking density commensurate with carrying capacity of aquaculture systems. In the present study, the higher stocking densities might have reduced access to adequate oxygen and subsequently slowed down metabolism hence lower growth than in lower stocking density. The increased FCR (**Table 3**) at high stocking density (120 fish/m<sup>3</sup>) may imply that high stocking density reduced feed efficiency. Similar results have been reported in the larvae of Common carp (*Cyprinus carpio*) and Golden mahseer (*Tor putitora*) [12,14]. As a result of competition for feed resources, there is low feed intake resulting in low energy levels to drive metabolic processes that convert nutrients into flesh. Conversely, more energy is expended on stress management hence poor feed utilization efficiency. High mortality of the fry at high stocking density could be due to insufficient food arising from competition for food resources among the fish [15-18]. Results of the present study on survival (**Table 3**) show that higher stocking densities have lower survival rates than lower stocking densities ceteris paribus. At higher stocking densities, increased biological stress of overcrowding is a precursor to stress related mortalities and vice versa.

### Effect of crude protein level on growth performance, survival and feed utilization of African catfish fry

It is widely reported that protein is the most important and expensive constituent of the feed that should be supplied in adequate amounts to support good growth with minimal cost [19,20]. In the present study, growth of catfish fry was significantly affected by different dietary protein levels. Growth (final MBW, MWG, DGR) increased at higher dietary CP level (**Table 3 and Figure 1**). Similar growth trends resulting from varying protein level were observed in earlier studies [18]. According to growth



depression due to lower dietary levels of protein intake has been observed in various fish species under captivity. Inadequate protein amount in a diet of fish depresses its growth due to reduction in the available energy for growth [21,22]. According to a report by Brett and Groves, fish being aquatic obtain sufficient energy from chemical breakdown of proteins than land animals, thus underscoring the importance of protein in the diets of fish [23]. The lowest growth obtained with the diet with 38.8% (**Figure 1**) of protein can be due to the fact that most of the protein was used for maintenance making it unavailable for growth. The 52.8% CP (**Figure 2**) diet gave higher growth, higher mean weight gain and better protein utilization in catfish fry. This CP level is close to protein requirements recommended for other carnivorous fish fry: 48-53% for *Salmo trutta*; 45-53% for *Carassius auratus*; 42-56% for *Clarias gariepinus* and *Heterobranchus longifilis*; 50-55% for hybrid catfish *H. bidorsalis* × *C. anguillar* [24-27]. It is therefore important to formulate

fish feeds that satisfy the nutrition requirements of the target fish species, for instance most of the carnivorous fish studied show a relatively high dietary protein requirement in the range of 35-70% [28]. Holding all factors including the varied stocking density of the present study, results (**Table 3**) show that dietary protein has a marked influence on feed utilization. Diet with 52.8% CP had better FCR than diets with 38.8%CP. A similar trend has also been found by different authors for other freshwater carnivorous fish species including pikeperch, Sander *lucio*perca and snakehead [29,30]. Higher pressure on body protein in order to satisfy the dietary needs for tissue building, repair, and metabolism has been suggested as the reason for poor growth and dietary utilization observed in fish fed sub-optimal dietary protein levels [31]. This meant that fish fed the high-protein diets (52.8% dietary protein) used dietary protein more efficiently than fish fed the low-protein diet (38.8%). Reference to **Table 3** in the present study, survival rate increases with increased dietary protein level, this means low protein diets might have not met the nutrition requirements of catfish fry and thereby leading to nutrient-deficient related mortalities [32-34].

### Water quality parameters

Water quality parameters (**Table 4**) observed during the experimentation were within the range not toxic to fish, thus any differences on growth parameters are attributable to variation in stocking density and crude protein level. Temperature values were within recommended limits for African catfish larvae and fry growth and survival. Barton et al. [33] recommends that temperatures should be within 23°C-30°C corroborating with FAO. Optimum temperature for maximum growth African catfish is 28°C as reported by Hecht [35]. There is fish growth depression at temperatures above or below the optimum temperature [36] and thus the growth possible higher growth rates of fry than presently reported in this study, if temperature was regulated values recorded during the experiment were within allowable range of 6.0-9.0 and similarly DO levels were within optimal levels as they were above 6 mg/l in all treatments as recommended by FAO [33,35-37].

### Conclusion

Both crude protein level and stocking density have significant effect on growth and survival of *Clarias gariepinus* fry in tanks. Based on the result of this experiment, starter fish feed of 52.5% crude protein and fry stocking density of 80 fish/m<sup>3</sup> are needed for higher catfish fingerling production. Similar studies need to be conducted in other culture facilities such as ponds and cages.

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