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## Combined effects of photoperiod and temperature on growth and survival of African catfish (*Clarias gariepinus*, Burchell 1822) larvae under laboratory conditions

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#### ABSTRACT

The effect of photoperiod (24L:00D, 12L:12D, and 00L:24D) and temperature (22  $\pm$  1°C and 28  $\pm$  1°C) on performance of *Clarias gariepinus* larvae was tested. Larvae weighing 3.2  $\pm$  0.24 mg were cultured in aquaria at a stocking density of 20 fish L<sup>-1</sup> and fed twice a day on catfish starter diet (40% CP) at 10 % BW day<sup>-1</sup>. Highest mean weight gain (31.00 mg), SGR (7.56% day<sup>-1</sup>), and survival (83%) were achieved at photoperiod and temperature combination of 00L:24D and 28  $\pm$  1°C. Percent survival of larvae differed significantly (p < .05) among treatments with optimal survival of (83%) in treatment combination of 28  $\pm$  1°C and 00L:24D, while lowest survival (40%) in treatment combination of 22  $\pm$  1°C and 24L:00D.

#### **KEYWORDS**

*Clarias gariepinus*; growth; photoperiod; survival; temperature

### Introduction

The main constraint facing the culture of *C. gariepinus* is low and highly variable survival of larval stages, especially during fluctuating weather conditions (De Graaf and Janssen 1996). Growth optimization is of paramount importance for profitability of fish farming activity. Within a given species, numerous factors can influence fish growth, among which the most important is probably temperature (Gardeur et al. 2007). Temperature regulates metabolic activity, and all fish species are characterized by a range of temperature within which growth is maximal (Jobling 1996; Person-Le Ruyet et al. 2006; Bjornsson et al. 2007). Previous studies indicate a temperature range of  $25^{\circ}C-33^{\circ}C$  with  $28^{\circ}C$  being the best for *C. gariepinus*. Verreth and Den Bieman (1987) recommended  $28 \pm 1^{\circ}C$  as the optimal rearing range for *C. gariepinus* larvae. However, one factor alone may not necessarily have an effect on performance without interactions with other factors.

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Photoperiod is known to have an effect on the feeding activity, growth, and survival of fish, thus influencing overall performance (Nwosu and Holzlöhner 2000; Adewolu et al. 2008). Ogbe et al. (2001) reported *C. gariepinus* as bottom feeders feeding comfortably in the dark. The effect of photoperiod on growth and survival of *C. gariepinus* at the larval stage has been demonstrated by several authors (Britz and Pienaar 1992; Appelbaum and Kamler 2000; Appelbaum and McGeer 1998a; Almazán-Rueda et al. 2004). To date, the combined effect of photoperiod and temperature on *C. gariepinus* larval development is yet to be reported.

#### Materials and methods

Experimental fish were obtained from the hatchery at Chepkoilel University College fish farm in Eldoret, Kenya. To determine mean initial wet body weight (BW) and mean total length (L), 30 three-day-old *C. gariepinus* larvae were collected from a pool of hatchlings. They were measured for wet BW on a Sartorius<sup>\*</sup> analytical electronic balance (readability 0.001 mg, Model VI-200) and total length using a hand transparent plastic ruler to the nearest 0.1 mm. A total of 18 aquaria with a 20L capacity were each stocked with 200 catfish larvae (mean BW 3.20  $\pm$  0.24 mg) in 10L water at a stocking rate of 20 larvae L<sup>-1</sup>.

The larvae were randomly distributed in 18 glass aquaria in triplicates of three photoperiods and two rearing temperatures in a  $3 \times 2$  (photoperiod  $\times$  temperature) factorial nested design. The three photoperiods, 24 h of light (24L:00D), 12 h of light and 12 h of darkness (12L:12D), and 24 h of darkness (0L:24D) in combination with two temperatures ( $22 \pm 1^{\circ}$ C and  $28 \pm 1^{\circ}$ C) were randomly assigned to the 18 tanks. Two fluorescent tube lights of 40 W (Philips<sup>®</sup> TLD 36/54, Netherlands) providing 100 lux of light were used to give the required 24 h of continuous light for 24L:00D. To provide 24 h of darkness (00L:24D), a black cellophane polythene paper was used to cover the rearing units throughout the experiment. For the 12L:12D, the fluorescent light was available except that the rearing units were covered for half the time with the polythene paper. Temperatures of  $22 \pm 1^{\circ}$ C and  $28 \pm 1^{\circ}$ C were maintained with submersible aquarium water heaters.

Larvae were fed a commercial catfish starter feed at 10% of the larvae's body weight and administered in two equal portions daily at 0800 h and 1600 h. Nutrient composition of the commercial starter feed is shown in Table 1. The experiment lasted for 4 weeks after stocking. Water quality in the tanks was maintained by siphoning out the bottom debris (fecal matter, uneaten food) and replacing 1/3 of the water every morning (0700 h) before feeding. Water for replacement was obtained from two (500L) temperature-regulated storage tanks. Temperature and pH were recorded daily at 0600 h using a mercury thermometer and a pH meter, respectively. Dissolved

Nutrient	Quantity
Protein	40%
Fat	10%
Ash	9%
Fiber	1.1%
Calcium	2.4%
Phosphorus	1.5%
Lysine	3.2%
Methionine + Cystine	2%
Copper	10 mg kg <sup>-1</sup>
Vitamin A	30,000 IU kg <sup>-1</sup>
Vitamin D3	$3000 \text{ IU kg}^{-1}$
Vitamin E	400 IU kg <sup>-1</sup>
Vitamin C	$300 \text{ mg kg}^{-1}$
Selenium	0.4 m g kg <sup>-1</sup>

Table 1. Nutrient content of the commercial catfish starter feed.

oxygen, total ammonia nitrogen ( $NH_3$ -N), and nitrite-nitrogen ( $NO_2$ -N) were measured at intervals of 14 days using the indophenol and colored azo methods respectively (Boyd and Tucker 1992).

Growth was measured weekly for a period of 4 weeks. Each time, 30 larvae were randomly sampled from each tank and measured for growth (total length and wet body weight). Mortalities were recorded before every feeding (0800 and 1600 h). Mean weight per tank was used to adjust weekly food rations. Specific growth rate (SGR), % weight gain, relative condition factor, and percent survival of the larvae was calculated with the following formulae:

Specific growth rate SGR =  $100(\ln W_t - \ln W_0/t)$ 

where:  $-(\ln = \text{Natural logarithm}, W_0 = \text{Initial weight (g)}, Wt = \text{Final weight (g)}, and t = \text{Time (days)}.$ 

Weight gain(%) = (Final weight of fish – Initial weight of fish)  $\times$  100/Initial weight of fish.

Relative condition factor =  $W/\hat{W}$ 

(where W = observed weight of the fish,  $\hat{W}$  = predicted weight of the fish from length – weight relationship (log W = log a + b log L).

Percent survival % = Initial number of larvae in the tank

- Number of dead larvae

 $\times$  100/Initial number of larvae in the tank.

The combined effect of photoperiod and temperature on survival was determined after counting the remaining fish in the tank at the end of the experiment. Data were tested for normality using Shapiro-Wilk's W test and analyzed using MINITAB version 13. Statistical comparison of 20 😔 P. S. ORINA ET AL.

data among treatments for mean growth and survival were made with two-way analysis of variance (ANOVA) at p = .05. Duncan's multiple range test was used to identify specific differences among treatments. Correlation analysis was conducted to establish relationship between treatments (photoperiod, temperature) and growth and survival of catfish larvae.

## Results

Growth responses for C. gariepinus under the different treatments are shown in Table 2. Final mean BW, L, and SGR ranges were 11.52-1.00 mg, 12.91-17.46 mm, 4.57–7.56% day<sup>-1</sup>, respectively. Final mean wet body weight, total length, and specific growth rates differed among treatments. Significant interactions (p < .05) between temperature and photoperiod were evident in growth performance parameters mean BW, length gain, and SGR. Mean (and percentage) weight and length gains as well as SGR of C. gariepinus were significantly higher at a combined treatment of 28 ± 1°C and 00L:24D (total darkness) temperature and photoperiod, respectively, than the other treatments. Mean growth in weight and SGR in C. gariepinus larvae at  $22 \pm 1^{\circ}$ C and 00L:24D did not significantly differ from that at  $28 \pm 1^{\circ}$ C and 12L:12D. Poorest SGR ( $4.57 \pm 0.92$ ), mean weight gain ( $8.31 \pm 1.74$  mg), and mean gain in length (5.63  $\pm$  0.42 mm) were obtained at 22  $\pm$  1°C and 24L:00D temperature and photoperiod regimes respectively. Generally, at each of the temperatures tested ( $22 \pm 1^{\circ}C$  and  $28 \pm 1^{\circ}C$ ), total darkness gave the best growth performance (Table 2).

*C. gariepinus* larvae growth curves are shown in Figures 1 and 2 for length and weight gain respectively. Both temperature and photoperiod significantly (p < .05) affected fish growth. Compared to other treatments, growth in total darkness (00L:24D) was significantly faster (p < .05). Continuous light (24L: 00D) produced significantly (p < .05) slower growth throughout the experiment.

Figure 3 indicates that survival response of *C. gariepinus* larvae was significantly affected by interaction between temperature and photoperiod (p < .05). Regardless of the photoperiod,  $28 \pm 1^{\circ}$ C resulted in significantly higher survival than  $22 \pm 1^{\circ}$ C (Figure 3). Survival of *C. gariepinus* larvae was highest (83%) in aquaria combination of  $28 \pm 1^{\circ}$ C and total darkness. The lowest survival (40%) occurred at  $22 \pm 1^{\circ}$ C and continuous light (24L:00D). Unlike fish in half (12L:12D) and continuous darkness (00L:24D), larvae in continuous light tended to congregate in aquaria corners, seemingly nervous and/or in search of dark areas.

Water total ammonia (TA), pH, total ammonia nitrogen (TAN), dissolved oxygen (DO), and nitrite (NO<sub>2</sub>-N) were within the acceptable levels for growth of *C. gariepinus*. All measured parameters except DO presented no

			Treat	ment*		
Parameter	T11	T12	T13	T21	T22	T23
Initial mean weight (mg)	$3.20 \pm 0.23$	$3.20 \pm 0.24$	3.20 ± 0.24	3.22 ± 0.25	$3.19 \pm 0.23$	3.23 ± 0.24
Final mean weight (mg)	$11.52 \pm 1.66^{a}$	$15.00 \pm 0.57^{b}$	$20.10 \pm 2.77^{c}$	$13.15 \pm 1.16^{a}$	$18.27 \pm 2.75^{c}$	$31.00 \pm 2.28^{d}$
Mean weight gain (mg)	$8.31 \pm 1.74^{a}$	11.80 ± 2.32 <sup>b</sup>	$16.90 \pm 1.79^{c}$	$10.03 \pm 1.99^{a}$	$14.35 \pm 2.49^{c}$	27.27 ± 2.62 <sup>d</sup>
% weight gain	$259.5 \pm 96.4^{a}$	$368.31 \pm 116.8^{b}$	$527.5 \pm 272.8^{c}$	$321.61 \pm 40.5^{a}$	$465.8 \pm 181.9^{\circ}$	856.7 ± 456.6 <sup>d</sup>
SGR (% day <sup>-1</sup> )	$4.57 \pm 0.92^{a}$	5.51 ± 1.22 <sup>b</sup>	$6.56 \pm 0.99^{\circ}$	$5.14 \pm 1.44^{a}$	$6.23 \pm 0.85^{\circ}$	7.56 ± 1.77 <sup>d</sup>
Initial mean length (mm)	$7.27 \pm 0.28$	7.27 ± 0.29	7.127 ± 0.24	$7.26 \pm 0.26$	$7.28 \pm 0.28$	$7.25 \pm 0.29$
Final mean length (mm)	$12.91 \pm 0.62^{a}$	13.63 ± 0.47 <sup>b</sup>	$15.93 \pm 0.52^{d}$	$13.98 \pm 0.57^{\rm b}$	$14.96 \pm 0.24^{c}$	$17.46 \pm 0.31^{e}$
Gain in length (mm)	$5.63 \pm 0.42^{a}$	6.33 ± 0.49 <sup>b</sup>	8.77 ± 0.47 <sup>d</sup>	$6.72 \pm 0.49^{b}$	$7.68 \pm 0.58^{c}$	$10.22 \pm 0.69^{e}$
% gain in length	$77.4 \pm 10.2^{a}$	87.1 ± 13.4 <sup>b</sup>	$123.1 \pm 15.3^{d}$	$92.6 \pm 11.0^{b}$	$105.5 \pm 19.1^{\circ}$	$140.9 \pm 21.2^{e}$
*Treatments: T11 = 22°C, 24L:0	0D; T12 = 22°C, 12L:12D	; T13 = 22°C, 00L:24D; T2	.1 = 28°C, 24L:00D; T22 =	= 28°C, 12L:12D; T23 = 28	3°C, 00L:24D.	

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**Figure 1.** Mean total length (±SE) of *C. gariepinus* larvae under different photoperiod and temperature combinations during a 4-week experiment period.



**Figure 2.** Mean wet weight (±SE) of *C. gariepinus* larvae under different photoperiod and temperature combinations over a 4-week experiment period.

significant differences (p > .05) within and among treatments (Table 3). Aquaria at 28 ± 1°C had a significantly (p < .05) lower DO than aquaria at 22 ± 1°C. Nevertheless, an interaction between photoperiod and temperature did not have any significant effect (p > .05) on TA, pH, TAN, DO, and NO<sub>2</sub>-N among treatments.



Figure 3. Mean survival trend of *C. gariepinus* larvae under different photoperiod and temperature combinations during a 4-week experiment period.

**Table 3.** Mean ( $\pm$ SE) water quality parameters in various treatments under laboratory condition over 28-day period. Means within a row lacking a common superscript are significantly different (p < .05). SE = standard error, calculated from the mean-square for error of the ANOVA, TA = total alkalinity, TAN = total ammonia nitrogen, and DO = dissolved oxygen.

	Treatment*							
Variable	T11	T12	T13	T21	T22	T23		
TA (mg CaCO <sub>3</sub> $I^{-1}$ )	$108.1 \pm 3.4^{a}$	$100.5 \pm 6.9^{a}$	$102.2 \pm 8.4^{a}$	$73.9 \pm 7.9^{a}$	$73.9 \pm 7.9^{a}$	$73.9 \pm 7.9^{a}$		
pН	$7.5 \pm 0.8^{a}$	7.1 ± 0.7 <sup>a</sup>	$7.2 \pm 0.7^{a}$	$7.0 \pm 0.7^{a}$	$7.6 \pm 0.8^{a}$	$6.9 \pm 0.7^{a}$		
TAN (mgl <sup>-1</sup> )	$0.6 \pm 0.1^{a}$	$0.6 \pm 0.1^{a}$	$0.5 \pm 0.1^{a}$	$0.6 \pm 0.1^{a}$	$0.6 \pm 0.1^{a}$	$0.6 \pm 0.1^{a}$		
Dawn DO (mgl <sup>-1</sup> )	4.2 ± 1.4 <sup>b</sup>	4.2 ± 1.4 <sup>b</sup>	4.2 ± 1.4 <sup>b</sup>	$3.0 \pm 0.3^{a}$	$3.1 \pm 0.5^{a}$	3.0 ± 1.4 <sup>b</sup>		
$NO_2-N (mgl^{-1})$	< 0.03 <sup>a</sup>	< 0.03 <sup>a</sup>	< 0.02 <sup>a</sup>	< 0.02 <sup>a</sup>	< 0.02 <sup>a</sup>	< 0.02 <sup>a</sup>		

\*Treatments: T11 = 22°C, 24L:00D; T12 = 22°C, 12L:12D; T13 = 22°C, 00 L: 24D; T21 = 28°C, 24L: 00D; T22 = 28°C, 12L:12D; T23 = 28°C, 00L:24D.

#### Discussion

These results corroborate previous findings that *C. gariepinus* larvae are photophobic and eat more effectively in darkness (Hogendoorm 1980; Britz and Pienaar 1992; Bruton 1979a, 1979b; Appelbaum and McGeer 1998b). Behavioral studies have illustrated that avoidance of light and preference for dark areas is an innate behavior in young *C. gariepinus* (Britz and Pienaar 1992).

Rather than increased light, making it easier for the *C. gariepinus* larvae to spot the feed, the larvae were probably stressed by the presence of light and could therefore not feed comfortably (Adewolu et al. 2008). Under continuous light, there are higher chances of developing aggressive behavior, thus limiting feed intake. This is in line with Britz and Pienaar (1992), Appelbaum and Kamler (2000), and Verreth and Van Tongeren (1989), who observed that swimming and aggressive behavior of *C. gariepinus* larvae increased with

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prolonged exposure to light. This eventually results in poor growth, which is attributed to less resting time for the fish. Lower weight gains may be attributed to the fish being more stressed and aggressive under continuous light conditions, compared to those under 12D:12L and 24D:00L. This corroborates earlier studies on photoperiod and temperature (Almazán-Rueda et al. 2004; Almazán-Rueda et al. 2005).

Study findings indicate low-light regimens and low temperatures lead to reduced rates of cannibalism, low activity, and high feeding rates of African catfish larvae. The higher survival found in fish cultured in the dark regardless of the temperature are in agreement with previous findings (Appelbaum and McGeer 1998a; Appelbaum and Kamler 2000; Adewolu et al. 2008) that demonstrated that larval behavior ensures growth in an environment most conducive for survival. Further studies indicate that photoperiod and temperature are among factors that regulate such behavior (Coward and Bromage 2000; Bromage et al. 2001). In nature, the African catfish is known to spawn in flooded streams, and fertilized eggs attach to reeds and grasses, providing darker conditions. Thus the combined effect of reduced photoperiod and increased temperature appears to be nature's way of ensuring greater survival and growth of *C. gariepinus* larvae.

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