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# Influence of temperature on survival, yolk utilization, growth, and morphometric anomaly rates in post-embryonic *Clarias jaensis* under controlled conditions

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**Key words:** *Clarias jaensis*, Temperature, Survival, Growth, Morphometric anomalies

**Abstract**

Captive breeding of *Clarias jaensis* remains limited due to a lack of knowledge regarding optimal temperature conditions to ensure larval survival and early development. This study evaluated the impact of different temperatures on survival, yolk absorption, linear growth, and the rate of morphometric anomalies in post-embryos. A total of 580 newly hatched post-embryos were evenly distributed in 10 trays, placed in pairs in five polyester tanks. Each tank was subjected to one of the five experimental temperatures: 22°C, 25°C, 27°C, 29°C, and 31°C. Survival and anomaly rates were analyzed using the Kaplan-Meier test, while the evolution of yolk sac volume and larval length was studied using a one-factor ANOVA. The results show that the best survival rates were obtained at 25°C (96.5 ± 3.5%), 27°C (91.5 ± 6.4%), and 22°C (90 ± 2.8%). No survival was observed at 29°C and 31°C after three and two days post-hatching, respectively. Yolk absorption was significantly faster at 27°C (98.92 ± 0.58%), while differences in linear growth were not significant between 22°C, 25°C, and 27°C. The most frequent morphometric anomalies included pericardial edema, yolk edema, and skeletal deformities, with a lower malformation rate at 25°C (4.5%) compared to 22°C and 27°C. Based on these results, it is recommended to stabilize the breeding temperature at 25°C to maximize survival, and at 27°C to promote rapid growth and yolk absorption.

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## Introduction

*Clarias jaensis*, an African catfish prized for its hardiness, rapid growth, and nutritional and cultural value, shows great potential for aquaculture (Angoni *et al.*, 2016; Zango *et al.*, 2016; Tiogue *et al.*, 2020). However, despite this potential, mastering its rearing in controlled environments remains a major challenge, forcing fish farmers to rely on fry sourced from the wild (Pouomogne, 2008; Kenfack *et al.*, 2019). This practice, in addition to limiting aquaculture production, places significant pressure on natural stocks, which are already weakened by overfishing and climate change. The provision of sufficient fry to meet the growing demand of fish farmers, therefore, depends on mastering the complete life cycle of *C. jaensis*, particularly its larval development in controlled conditions. Temperature is a key environmental factor in the early development of fish, significantly influencing their survival, growth, and morphogenesis (Cahu *et al.*, 2003; Fontaine and Le Bail, 2004; Gatesoupe *et al.*, 1999). The embryonic and larval stages, which are particularly vulnerable to environmental fluctuations, are especially sensitive to thermal variations. Understanding the influence of temperature on larval development is essential for optimizing farming practices and ensuring successful production in captivity. Previous studies on other Clariidae species, such as *C. gariepinus* (Legendre and Teugels, 1991; Chebel *et al.*, 2005) and *Heterobranchus bidorsalis* (Olaniyi and Omitogun, 2013, 2014), have highlighted the significant impact of temperature on larval survival, growth, and development. However, the specific thermal tolerance of *C. jaensis* remains largely unknown. This study aims to fill this gap by rigorously evaluating the influence of temperature on the survival, yolk absorption, growth, and occurrence of morphometric anomalies in *C. jaensis* post-embryos. By exposing the larvae to different temperatures in a controlled environment, we aim to identify the optimal thermal range that promotes their development and to understand the underlying mechanisms of the observed effects. The results of this research will have a dual impact. On one hand, they will provide basic information for improving the farming practices of *C. jaensis* in captivity, allowing for the adaptation of incubation and larval rearing conditions to maximize survival, growth, and fry quality. On the other hand, they will contribute to a better understanding of the biology of this species and its sensitivity to temperature variations, which is essential for the sustainable management of its natural populations in the face of climate change challenges.

## Materials and methods

*Study period and location*

This study was conducted in September 2023, during the rainy season, at the experimental unit of the Cameroonian Cooperative for Fisheries Development (COOPCADHA) in Bafoussam, located in the Mifi department, West Region of Cameroon (5°27'10.2"N 10°25'33.5"E).

*Production of post-embryos*

The broodstock, collected in January 2023 from the Mifi River (5°31'05.3"N 10°21'18.2"E), were kept in above-ground tanks and fed a high-protein diet (45%). The post-embryos were obtained through artificial reproduction, following a method similar to that described by Zango *et al.* (2016), with a few modifications. Eight males (mean weight: 374.9 ± 97.6 g; mean total length: 41.6 ± 5.6 cm) and six females (mean weight: 284 ± 39 g; mean total length: 35.3 ± 2 cm) showing signs of sexual maturity were selected. The females received an injection of Ovaprim® (1 ml/kg of body weight) to induce ovulation, and they were then placed in holding tanks at 25°C. Ovulation began, on average, 26 hours after the injection.

One hundred and twenty grams of mature oocytes were collected individually from the females and placed in a bowl. In vitro fertilization was performed by adding 5 ml of viable milt, diluted according to the technique of Nguenga *et al.* (1996). After fertilization and rinsing, the eggs were spread in a single layer on mesh frames immersed in closed-circuit hatching tanks (BEF kit). Incubation took place at a constant

temperature of 25 ± 0.6°C, and the eggs hatched approximately 26 hours later. Unfertilized eggs and non-viable embryos were regularly removed from the trays.

**Fig. 1.** Experimental setup
Polyester tanks from the BEF kit (47x35x27 cm)
Tray (2 liters)

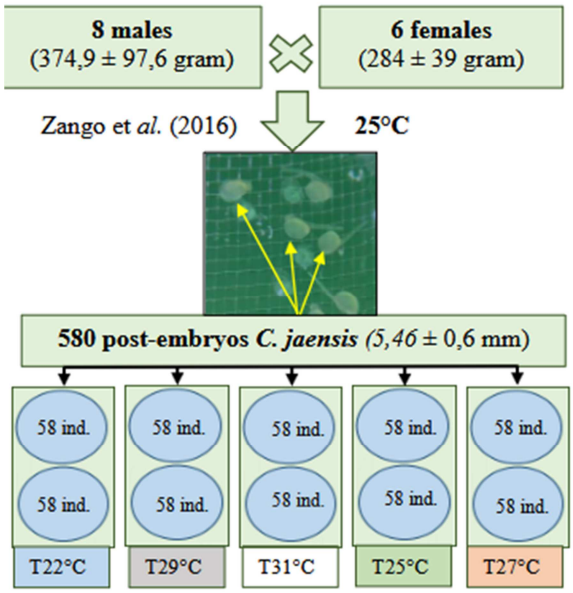



**Fig. 2.** Randomized experimental design
05 treatments, duplicated per treatment

*Experimental design and conduct of the trial*

Hatching was considered successful when 90% of the larvae were viable (Shan *et al.*, 2008). Five hundred eighty newly hatched post-embryos were evenly distributed in 10 trays, placed in pairs in five polyester tanks of the BEF kit (Fig. 1). To avoid thermal shock, the water in the tanks was initially maintained at 25°C (hatching temperature). Then, using a thermostat, the temperature of each tank was gradually adjusted to reach one of the five target values: 22°C, 25°C, 27°C, 29°C, and 31°C. These temperatures were kept constant throughout the yolk sac resorption period (7 days). Each tank had an effective volume of 35 liters and was equipped with a water circulation system (Fig. 2). A partial water renewal (50%) was carried out on days 3 and 6 of the experiment. The larvae were not fed during this period. Water temperature, pH, conductivity, nitrates ($NO_3$), and carbonate hardness (KH) were measured once daily in the morning.

*Studied characteristics and collected data*

*Monitoring of survival and anomalies*

Mortality was recorded daily in each replicate. These data were used to calculate the cumulative survival rate at the end of the experiment according to Adewolu *et al.* (2009), Ts (%) = ((No - Nt) / No)*100, where Ts is the survival rate in percentage, No is the initial number of larvae, and Nt is the cumulative mortality at a given time. Simultaneously, daily observations were made to identify and quantify morphometric anomalies using a binocular magnifier. The criteria for identifying anomalies were based on the works of Bordin *et al.* (2022), Hossain *et al.* (2021), and Jezierska *et al.* (2000). The cumulative morphometric anomaly rate was calculated daily according to the following formula: Tan (%) = (Na / No)*100, where Tan is the cumulative anomaly rate, Na is the total number of larvae that presented at least one anomaly by day j, and No is the total initial number of larvae.

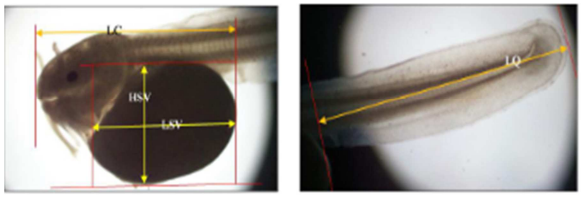
*Yolk absorption and linear growth*

At hatching (day 0), then at 1 day post-hatching (dph) and every 48 hours until 7 dph, a sample of 7 to 9 larvae per replicate and treatment was taken to measure the length and height of the yolk sac, as well as the total body length. Each larva was hydrated, placed on a slide, and aligned along its longitudinal axis (Fig. 3), then scanned with an Olympus D21-CB camera connected to an Olympus BX51 microscope. Measurements were made using OlyVIA software (precision: 0.001 mm). These data allowed for the calculation of:

Yolk Sac Volume (YSV) using the formula described by Bagarinao (1986) for an elongated spheroid YSV = $\pi/6 \times LSV \times HSV^2$, where LSV is the yolk sac length and HSV is the yolk sac height.

Yolk Absorption Rate (YAR): YAR (%) = (ln $YSV_0$ - ln $YSV_t$) / t × 100 %, where $YSV_0$ is the initial yolk sac volume, $YSV_t$ is the final yolk sac volume, and t is the time elapsed.

Total Length Gain (TLG) over different periods, calculated using the formula:
TLG = $Lt_t$ - $Lt_0$, where $Lt_t$ and $Lt_0$ are the average lengths on the final and initial days, respectively.




LSV: Yolk sac length, HSV: Yolk sac height,
LQ+LC=LTC: Total body length

**Fig. 3.** Measured morphometric characteristics

*Statistical analysis*

The collected data were analyzed using SPSS 20.0 software. Survival rate was analyzed using the Kaplan-Meier test, while differences in the proportions of morphometric anomalies were evaluated with the Chi-square test. The evolution of yolk sac volume and total larval length was studied using repeated measures ANOVA. All analyses were performed with a significance threshold of 0.05.

**Results**

*Variation of water parameters*

The average values of the physical and chemical water parameters during the first seven days of post-hatching life for *Clarias jaensis* post-embryos are presented in Table 1. The results show that, apart from the temperature parameter, no other water parameter analyzed showed a significant difference ($p > 0.05$) between the different treatments.

*Thermosensitivity of C. jaensis post-embryos*

The survival rate of *C. jaensis* post-embryos varied independently of their age and the temperature of the rearing environment (Fig. 4). The cumulative survival rates during the first three days post-hatching at 22°C (98±1.4%), 25°C (100±0%), and 27°C (99.5±0.7%) differed significantly ($p<0.01$) from those recorded at 31°C (0%) and 29°C (4.5±2.1%). The latter decreased until reaching zero by the second and third days post-hatching, respectively. However, the highest survival rate over time was recorded at a temperature of 25°C (96.5±3.5%), although it was not statistically different ($p > 0.05$) from that observed at 27°C (91.5±6.4%) and 22°C (90±2.8%).

**Table 1.** Variation of the physico-chemical parameters of the water in the rearing environment of *C. jaensis* post-embryos

| Treatment | Water parameters | | | | |
|---|---|---|---|---|---|
| | Tc (°C) | pH | Cond. (µs) | $NO_3$ (mg/l) | KH (mg/l) |
| 22°C | 22.15±0.46 | 6.53±0.12 | 95.17±4.6 | 0 | 34.28±14.5 |
| 25°C | 25.6±0.3 | 6.51±0.13 | 98.84±3.3 | 0 | 34.28±14.5 |
| 27°C | 27.3±0.5 | 6.46±0.2 | 101±8.6 | 0 | 34.28±14.5 |
| 29°C | 29.2±0.9 | 6.45±0.08 | 102.34±1.8 | 0 | 40±0 |
| 31°C | 31.1±0.28 | 6.51±0.11 | 95.5±2.8 | 0 | 40±0 |

*Yolk absorption rate*

From the first day post-hatching (dph), larvae reared at 27°C stood out significantly ($p < 0.05$) with a yolk absorption rate of 24.17 ± 5.6%, compared to those reared at 22°C (11.18 ± 3.65%) and 25°C (10.91 ± 4.07%), as illustrated in Fig. 5. By the 3rd day post-hatching, these same larvae reared at 27°C reached a 60% efficiency in yolk utilization, while the yolk utilization was lower at 25°C (24.41 ± 3.04%) and 22°C (20.94 ± 9.6%). This trend continued until the 5th dph, where larvae reared at 25°C (72.43 ± 3.32%) showed slightly higher yolk utilization than those

reared at 27°C (67.10 ± 1.67%). Larvae reared at 22°C continued to show slower utilization (53.29 ± 1.06%). By the end of the experimental period (7 dph), larvae reared at 27°C had almost completely consumed their yolk (98.92 ± 0.58%). Although significant, yolk consumption was less complete in larvae reared at 25°C (91.3 ± 5.3%) and 22°C (86.3 ± 7.38%).

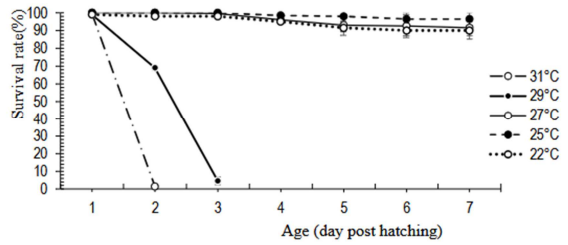



**Fig. 4.** Evolution of the survival rate of *C. jaensis* post-embryos in terms of temperature

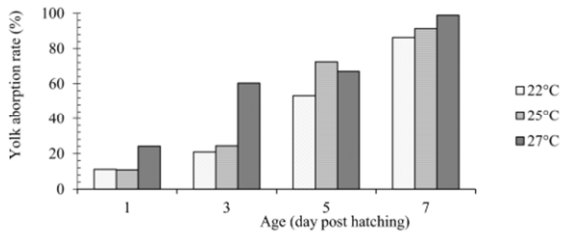



**Fig. 5.** Yolk absorption rate in *C. jaensis* in terms of age and temperature

*Linear growth in C. jaensis post-embryos*

Post-embryos reared at 25°C showed a higher average length (6.88 ± 0.34 mm) compared to those at 22°C (5.72 ± 1.04 mm) and 27°C (6.42 ± 0.34 mm). By the 3rd day post-hatching (dph), a similar trend was observed, with more pronounced growth at 27°C (8.43 ± 0.00 mm) compared to 25°C (8.20 ± 0.23 mm) and 22°C (7.96 ± 0.27 mm). This progression continued throughout the observation period, culminating on the 7th dph, where post-embryos reared at 27°C reached an average length of 10.73 ± 0.25 mm, slightly exceeding those reared at 25°C (10.26 ± 0.60 mm) and 22°C (10.07 ± 0.89 mm). Although post-embryos reared at 25°C showed significant growth over time, the results indicate that those at 27°C exhibited the best linear growth performance over the experimental period. In contrast, larvae reared at 22°C showed slower growth, with a developmental delay compared to the other experimental groups. By the end of the experimental period, post-embryos reared at 27°C had achieved the greatest length, although the difference from those reared at 25°C was not statistically significant ($p > 0.05$).

*Types and rates of anomalies observed in C. jaensis post-embryos*

Two main categories of anomalies were observed in *C. jaensis* post-embryos: edema and skeletal deformities. Edema was classified into three types: pericardial (around the heart), yolk sac (in the yolk sac), and subcutaneous (observed in 7-day-old post-embryos). Pericardial and yolk sac edema progressed in some larvae to become generalized edema around the yolk sac. Embryos exhibiting these anomalies predominantly developed skeletal malformations, particularly in the caudal fin.

The morphometric anomaly rate remained significantly ($p<0.05$) the lowest at a temperature of 25°C (0 to 4%), regardless of the age considered. In contrast, it was higher and significant ($p<0.05$) at 31°C (17.5±0.7%) and 29°C (16.5±0%) respectively on the second and third days following hatching (Fig. 4). However, no significant difference ($p>0.05$) was observed between the malformation rates recorded on the 3rd, 6th, and 7th days post-hatching for temperatures of 22°C and 27°C.

**Discussion**

The results obtained in this study reveal that temperature is a determining factor affecting the survival of *Clarias jaensis* post-embryos. The pH, dissolved oxygen, hardness, and conductivity remained constant throughout the experiment, indicating that the variations in survival, growth, and anomalies observed are primarily due to temperature, as also observed by Legendre and Teugels (1991) in *Clarias gariepinus*.

The results of this study reveal a higher survival rate at 25°C (96.5%), followed closely by 27°C (91.5%), with no statistically significant difference between

these two temperatures. These results are consistent with those obtained by Chebel *et al.* (2005) for *C. gariepinus*, where temperatures between 25°C and 28°C resulted in survival rates above 90%. In contrast, temperatures above 29°C had a detrimental effect on larval survival, with a total mortality rate at 31°C and only 4.5% survival at 29°C by the third day. These observations align with the studies of Kamler (1992), who demonstrated that excessively high temperatures (>30°C) cause thermal stress in fish larvae, impairing their metabolism and reducing their ability to utilize dissolved oxygen. Similar results were reported by Olaniyi and Omitogun (2013) for *Heterobranchus bidorsalis* larvae, where temperatures of 30°C and above led to 100% mortality within the first few days post-hatching.

Yolk absorption is essential for larval survival before transitioning to exogenous feeding. In this study, larvae reared at 27°C absorbed nearly all of their yolk (98.92%) after 7 days, while those at 25°C and 22°C absorbed 91.3% and 86.3%, respectively. These differences show that higher temperatures accelerate yolk absorption. Similar results were reported by Legendre and Teugels (1991) for *Clarias gariepinus*, where larvae reared at 27°C consumed their yolk in 6-7 days, compared to 10 days at 22°C. Kamler (1992) also showed that temperatures of 25-28°C activate digestive enzymes such as cathepsin D, accelerating yolk digestion. At 25°C, yolk absorption was slightly slower than at 27°C, but still relatively rapid. Fiogbé *et al.* (2003) observed complete absorption in 8-9 days at this temperature for *Heterobranchus longifilis*. At 22°C, absorption was slower, as noted by Rahman *et al.* (2011) for *C. batrachus*, where larvae took up to 12 days to absorb their yolk. However, Olaniyi and Omitogun (2014) found different results with *Heterobranchus bidorsalis*, where at 30°C; larvae absorbed their yolk in 5 days, though this temperature increased larval mortality. These variations may be due to differences between species and rearing conditions of broodstock.

Linear growth of larvae was also influenced by temperature. At 27°C, larvae reached 10.73 mm after 7 days, while those at 25°C and 22°C reached 10.26 mm and 10.07 mm, respectively. Even though the difference between 25°C and 27°C was not statistically significant, this indicates that higher temperatures promote slightly faster growth. Hecht and Appelbaum (1988) observed that *C. gariepinus* larvae reared at 28°C reached an average length of 11.5 mm by the 7th day post-hatching, suggesting that temperatures slightly higher than 27°C could further enhance linear growth in some species. At 22°C, growth was slower, as reported by Haylor and Mollah (1995), where *C. gariepinus* larvae only reached 9.5 mm by the 7th dph at this temperature. Honji *et al.* (2012) found that at temperatures of 26-28°C, *Steindachneridion parahybae* absorbed its yolk in 7 days, but at temperatures above 28°C, morphological anomalies and slower growth were observed. These differences between studies may be explained by species-specific thermal tolerances. *Clarias jaensis* and *C. gariepinus* seem to have a slightly lower optimal thermal range compared to *Heterobranchus bidorsalis*, which better tolerates higher temperatures according to Olaniyi and Omitogun (2014). Additionally, other factors like water quality may also play a role. Woynarovich and Horvath (1980) showed that fluctuations in pH or dissolved oxygen can affect growth, even if temperature is stable.

The morphological anomalies observed in *Clarias jaensis* post-embryos, mainly pericardial edema and skeletal deformities, significantly increased at higher temperatures. The anomaly rate reached 17.5% at 31°C, while it was 16.5% at 29°C. In comparison, at 25°C, this rate remained low, at 4%. These results are similar to Kamler's (1992) observations, who reported an anomaly rate of 15% in fish larvae reared at temperatures above 30°C. Woynarovich and Horvath (1980) also observed that high temperatures promote developmental anomalies in larvae due to thermal stress, a finding consistent with our results for *C. jaensis*. In *Clarias gariepinus*, Chebel *et al.* (2005) reported similar anomaly rates, around 15%, when larvae were reared at temperatures above 28°C. This suggests a detrimental effect of excessive heat on the normal development of Clariidae larvae.

Furthermore, Honji *et al.* (2012) observed that temperatures below 25°C or above 28°C slowed growth and increased skeletal malformations in *Steindachneridion parahybae* larvae.

**Conclusion**

At the conclusion of this study, which aimed to evaluate the influence of temperature on survival, yolk absorption, linear growth, and the rate of morphometric anomalies, it was demonstrated that temperature has a significant impact on the early development of *Clarias jaensis*. A temperature of 25°C was found to be optimal for survival, while 27°C promoted faster growth and more rapid yolk absorption, essential for larval survival before transitioning to exogenous feeding. Temperatures of 29°C and 31°C were found to be lethal. The study also revealed a significant increase in morphometric anomalies, such as edema and skeletal deformities, at these higher temperatures. These results underscore the importance for fish farmers to maintain a stable temperature of 25°C to optimize the survival of *C. jaensis* post-embryos, and to aim for 27°C to promote faster growth and accelerated yolk absorption.

**Acknowledgments**

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