

Embryonic and Larval Development Stages of the African Catfish *Clarias gariepinus* (Burchell, 1822) (Teleostei, Clariidae) in The Ouargla, Algeria

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Abstract

This study was designed to investigate for the first time in the region of Ouargla, Algeria, the events and timing of the embryonic development of the African catfish, *C. gariepinus* (Burchell, 1822). The embryonic development was carefully monitored using a binocular magnifier with 10 and 20 lenses that reveal details on live specimens from fertilization to the first take of food. The offensive and foul odor continue to be the characteristic smell of the hatching stage. Pigmentation and continuous spread of cephalo-caudal melanophore in fry hatched on the first day. The barely hatched larvae are photophobic. The yellow reserve is reduced and leads to the ability to swim easily, and the majority starts feeding on the fourth day. The vitellin reserve is significantly reduced, allowing the larvae to feed exogenously. The events observed in our opinion demonstrate the presence of two exceptionally critical parameters: the use of a high-quality diet and the guarantee of a clean and quality source of water, meeting the standards of aquaculture farming.

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INTRODUCTION

The African catfish *Clarias gariepinus* (Burchell, 1822) is a preferred aquaculture fish in the world, due to its robustness and rapid growth rate (Behmene, 2020). "Asataf" is the local name of the African catfish in southern Algeria, and these fishes are highly valued by consumers accounting for an important source of animal protein for rural populations, especially in these Algerian regions. This is largely due to its high fecundity, growth performance, as well as hardiness to poor water quality. The residents of the Ihrir Valley, in particular

the Tuaregs of the region, have had a very important relationship with these fish for centuries (Behmene *et al.*, 2020).

Artificial breeding of *C. gariepinus* is widely practiced worldwide. However, the supply of fish fry cannot meet the growth of the aquaculture industry. This situation is generally due to low natural seed production and a lack of control over artificial breeding techniques (Behmene, 2020). As a result, observations of the embryonic and larval stages mentioned in this passage allow careful monitoring of the embryonic and larval development of

this species to improve the survival of *C. gariepinus* larvae. Early embryonic and larval development studies are imperative and correlated to successful larval rearing, for the production of Large-scale animal husbandry (Khan and Mollah, 1998; Rahman *et al.*, 2004).

This study was designed to investigate for the first time in the region of Ouargla, Algeria, the events and timing of the embryonic development of the African catfish, *C. gariepinus*.

METHODOLOGY

Place and Time

The experiment took place in June 2018, at the Aquaculture Development Saharan Station, which is located 820 km south of the capital in the commune of Hassi ben Abdellah 25 km from the chief town of the willaya of Ouargla (Figure 1).

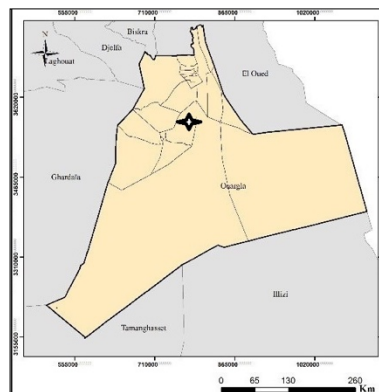


Figure 1. Geographical location of Ouargla.

Research Materials

For each dose of ovaprim hormone, a single selection test was conducted to determine the response to spawning

(Bachir-Bouiadjra *et al.*, 2021). Two females and two males were used in this study. Injection doses are shown in Table 1.

Table 1. Hormonal treatment of broodstocks (Bachir-Bouiadjra *et al.*, 2021).

Treatment	Broodstocks	Number of individuals	The hormone used	Injections (ml/kg)	Body weight (g)	Dosage of hormone (ml)	Length (cm)
T1	female	1	Ovaprim	0.4	1294.5	0.52	59
T2	female	1	Ovaprim	0.5	1152	0.58	54
T3	male	1	Ovaprim	0.25	2225	0.56	/
T4	male	1	hormone-free	0	823.3	0	53

The stages of embryonic and larval development are illustrated with microscopic tools, binocular magnifying glass (Reference 193143) and optical microscope, and compared to those in the literature (Olaniyi and Omitogun, 2013). During the absorption of yellow, the larvae were fed every two hours with frozen artemia, and, for the first time, egg yolk was added to a quality artificial food

called “Coppens” (Bachir-Bouiadjra *et al.*, 2021).

Research Design

The stages of embryonic and larval development of *C. gariepinus* are examined chronologically and described, from oocytes to fry, yellow absorption, to release of the larval stages.

Work Procedure

A normal embryonic development follow-up of African catfish *Clarias gariepinus* was carefully monitored using microscopic tools (MOTIC B3-220-ASC 1000x) from fertilization to the first take of food. We used the protocol of Olaniyi and Omitogun (2013), as a reference for monitoring and observations at the laboratory level.

Data Analysis

Length records were obtained from live specimens. The age was recorded in hours post-fertilization (hpf), counting the time of fertilization as 0 h and the day of fertilization as the first day.

RESULTS AND DISCUSSION

Stage of Fertilization

After the fusion of the sperm cells, the adhesion of the eggs is increased to allow binding to the substrate (Figure.2). The results of this study, mentioned above, are similar to those of Sule *et al.* (2001), which has an average diameter of 1.3 ± 0.16 mm for *C. gariepinus* spawners ranging from 900 to 1099g. Variation in egg size is a fish characteristic, and may be due to the quality of the strain (Puvaneswari *et al.*, 2009), and the size of the broodstock (Sule *et al.*, 2001); moreover, these same results corroborate the embryological work on *C. gariepinus* (Legendre and Teugels, 1991; Haylor and Mollah, 1995).

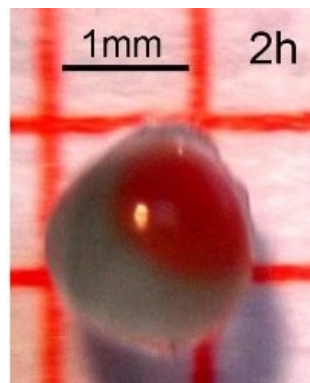


Figure 2. Stages of embryonic development at 2h.

Blastodiscus Stage

The fertilized egg is characterized by a red spot (blastodiscus) immediately after fertilization which is visible to the naked eye (Olaniyi and Omitogun, 2013). Then there is an accumulation of vitellin mass (plant pole) at one extreme pole which is entirely covered with granular cytoplasm pole (animal) at the other end pole (Figure. 2).

Size is a very interesting criterion for determining egg quality (Bromage and Roberts, 1995). The egg diameter reported in this study is slightly smaller and identical to that of the prickly catfish, *Heteropneustes fossilis*, 1.3–1.5 mm (Puvaneswari *et al.*, 2009), 1.03 ± 0.03 mm, but differs from the conclusions of Korzelecka-Orkisz *et al.* (2010) which

state that the eggs of Fossil Heteropneusts are 1.03 ± 0.03 mm, and are smaller than those of *C. gariepinus*.

For Iswanto *et al.* (2015), the egg diameter increased by 1.39-1.52 mm (1.47 ± 0.03 mm on average) for African catfish from Egypt. The biological process of developing the fertilized egg at hatching depends on temperature, i.e., the higher the water temperature, the faster the eggs hatch and the better the survival of the larvae (Haylor and Mollah, 1995; de Graaf and Janssen, 1996).

Morula Stadium

At the animal pole, the blastodisc looked like a mulberry; this embryonic development stage is called the morula. Several other cell divisions have led to

many blastomeres (Olaniyi and Omitogun, 2013). The many cells at the stage are called mulberry (Figure 2).

Blastula to Gastrula Stage

The blastoderm expanded, marking the transition from blastula to gastrula. The beginning of the epibole began as gastrulation, with the random displacement of a transition wave (Behmene *et al.*, 2020).

Embryos that do not engage in this movement are dead and their color becomes dark. This movement occurs on the yellow surface of the sphere, allowing the embryo to move in the peri-vitellin spaces, and ultimately leads to a mixture of the peri-vitellin fluid. This led to the formation of epiblasts and hypoblasts; the closure of the blastopore marked the end of this movement and morphogenesis began, causing the establishment of an embryonic axis (Olaniyi and Omitogun, 2013) (Figure 3).

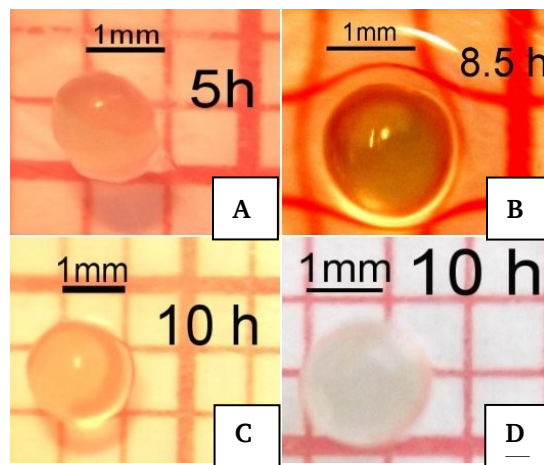


Figure 3. Embryonic stages of development 5-10 hours. A: 5h; B: 8.5 h, C and D: 10h.

Somite Stadium

Early formation of somite can be noticed, with the couple blocks of cells that developed along the back of the embryo giving the spine. The formation of Somite is then developed by the cephalo-caudal route (Figure 3), at this stage until the next (hatching), characterized by a specific odor.

Ola-Oladimeji (2015) had earlier stated that relative equality in fertilization rates is a strong indication of similarities in egg and milt qualities of broodstocks used. The lack of means, the absence of closed circuits and a technological filtering system, which does not comply with aquaculture standards, forced us to ensure the change of water manually (see 'siphoning'), with constant change and supply of fresh water to the aquariums, which facilitated the hatching, survival and development of the embryo through

constant and rigorous cleaning of eggshell impurities at hatching. During the formation of the somite, and shortly before the outbreak, an important observation was noted concerning the offensive odor, which reflects embryogenesis activity. In our observations, the first outbreak in our captive specimens of *C. gariepinus* from natural areas in the southern region occurred at 18 h to 28 °C.

Hatching Stage

Hatching shows the rupture of the embryo out of the chorion, or the egg capsule through the tail. According to Olaniyi and Omitogun (2013), shortly before hatching, an occasional embryo, the peristaltic movement was observed, (mean of nine contraction/min movements at first then became more frequent) (Behmene, 2020) (Figure 4).

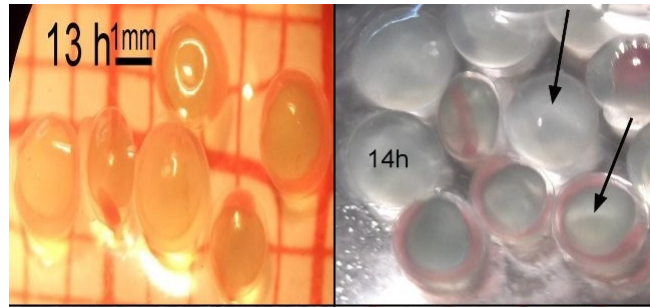


Figure 4. Stages of embryonic development at 13 h.

Then, the first breath was noticed with a heart rate of 115 to 160/ min, then the caudal bud, which surrounds the yolk sac slightly disengaged from the middle part.

The brain head was well-attached to the yolk sac, although the anterior part of the head was elongated and bulbous. Frequent successive jerks eventually broke it, that is to say, at hatching, the tail bud beat and broke the chorion or egg capsule/ coated membranous (Figure 5). According

to Olaniyi and Omitogun (2013), in Nigeria, it was reported at 17 h to 28.5 ± 0.5 which confirms our results, despite the precarious conditions of breeding, the poor quality of water too rich in iron. In Brazil, hatching took place within 30 hours of incubation at a water temperature of 25°C (Kipper *et al.*, 2013). Immediately after hatching, body pigmentation and onset of blood circulation were observed.

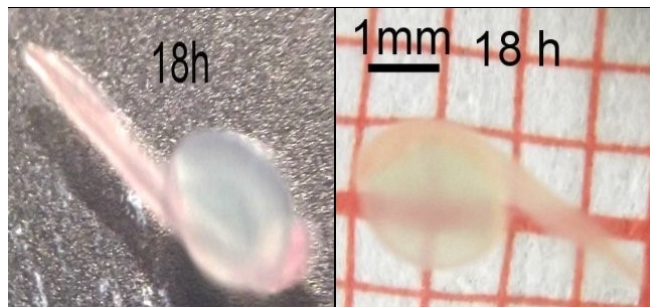


Figure 5. Stages of embryonic development at 18 h.

The offensive and foul odor continue to be the characteristic smell of the hatching stage (Olaniyi and Omitogun, 2013).

Larval Stage (Hatched Fry)

This stage (Figure 6) is characterized by a series of developmental activities such as Pigmentation and continuous spread of cephalocaudal melanophore. According to Olaniyi and Omitogun (2013), at hatching, the just-hatched larvae are translucent and

measure 5.0 ± 0.5 mm medium length. The eyes are not yet formed. The barely hatched larvae are photo-phobic. Almost an hour after the outbreak, blood circulation begins. The excretory system is rudimentary. The mouth is closed and the yolk bag full. The optical primordia and the otic vesicle, with its otoliths are easily visible and develop, to provide the visual and auditory functions necessary for the development of the larvae. Swimming movements in larvae are established 48 hours after hatching.

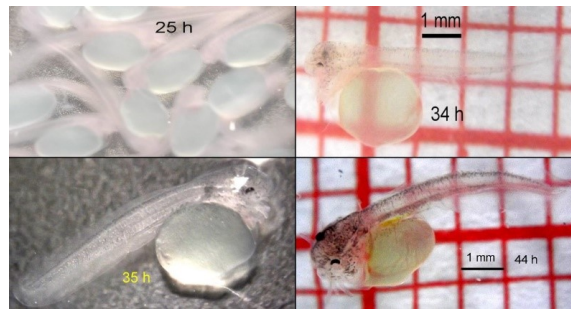


Figure 6. Stages of embryonic development at 25 h to 44 h.

Second Day Post-Embryonic Stage

The larvae were 6 ± 0.5 mm long on day 2 (Figure 7). Melanophore is pigmented toward the dorsal areas. The larval system is increased for feeding and the yellow content decreased. The mouth

is formed, revealing upper and lower parts (jaw). The food channel is distinct and becomes more important. The surgical and caudal rays are still very rudimentary but visibly recognized (Olaniyi and Omitogun, 2013).

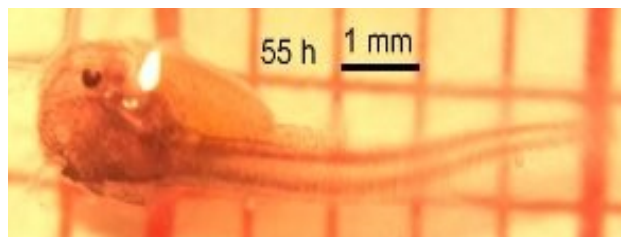


Figure 7. Embryonic development stages at 55 h.

Third Day Post-Embryonic Stage

The ocular musculature is now well-developed. The operculum is fully developed and visibly flapping the branchial arch (Behmene, 2020) (Figure 8). The yellow reserve is reduced and leads to the ability to swim easily (larvae).

Very few larvae began to be exogenously fed on the third day while the majority began feeding on the fourth day. According to Olaniyi and Omitogun (2013), the recorded larval measurements are 8.7 ± 0.5 mm which is well confirmed in our results from Figure 8.



Figure 8. Embryonic development stages at 73 h.

Four-Day Larvae

The oral and branchial systems are well-developed and vascularized. The dumbbells have increased in length, and are segmented, namely two pairs of mandibles and a pair of premaxillary and nasal. Melanophore is widespread throughout the cerebral-caudal body

(Behmene, 2020). The vitellin reserve is considerably reduced, allowing the larvae to feed exogenously (Figure 9). The digestive system is well-developed, the genito-urinary orifice has regained its functionality and some larvae are visible and relaxed. These larvae are 9.3 ± 0.5 mm long (Figure 9).

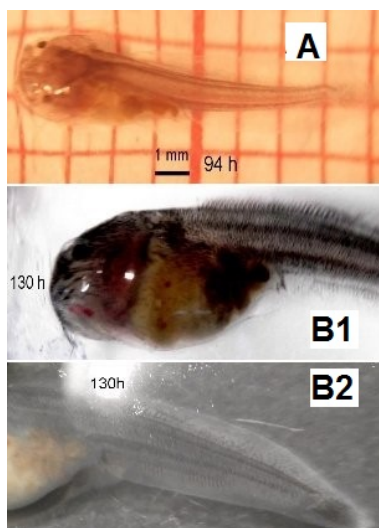


Figure 9. Embryonic developmental stages from 94 h (A) to 130 h (B1, B2).

The rays of the caudal fin are visible (Figure 9). By the second week, growth has progressed considerably and organs such as the pectoral fin are visibly well-developed.

CONCLUSION

Observations on embryos, egg hatching, and larval stages are important, and largely ensure the quality of fry for aquaculture programs. The follow-up of these different stages of embryogenesis to larval development observed in our case is very similar; however, the egg diameter is differently described in results and discussion as to the cases reported in the literature.

Hopefully, this study can encourage artificial reproduction, especially on the spawners of our water bodies in the extreme south of Algeria, which host two main species reported: the *Clarias gariepinus* and the *Clarias anguillaris*.

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