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Morphological Investigation of Larval Development in *Maylandia estherae* (Konings, 1995), an Endemic Cichlid Species of Lake Malawi

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ABSTRACT

This study investigates the morphological aspects of larval development in Maylandia estherae, an endemic cichlid species from Lake Malawi. Larvae obtained from mature broodstock were sampled daily from hatching until the juvenile stage, and their morphological development was observed under a microscope. Important morphological features such as mouth opening, exogenous feeding, free swimming, yolk sac, and pigmentation were observed and photographed. The findings indicate that *M. estherae* larvae reached the juvenile stage within approximately 20-25 days. Throughout larval development, it was determined that total length and mouth gape gradually increased, the yolk sac was depleted within 12-13 days, and exogenous feeding commenced alongside the development of free-swimming ability. Additionally, significant changes in fin differentiation and pigmentation were observed in the larvae. The larval development duration and morphological characteristics of M. estherae were found to be similar to those of other Malawi cichlid species. The findings of this study contribute to the understanding of the ontogenetic processes of *M. estherae* and have implications for aquaculture practices. Furthermore, the results provide a significant contribution to the literature on the larval development of Malawi cichlids and offer insights for future research.

INTRODUCTION

Cichlid species from Lake Malawi have garnered significant attention in the aquarium industry and hold a substantial market share in recent years (Monticini, 2010). Lake Malawi, located in eastern Africa, is a unique ecosystem considered to be one of the world's largest and deepest lakes. The lake covers an area of approximately 29,600 km² and has a maximum depth exceeding 700 meters (Bootsma & Hecky, 2003). Lake Malawi is renowned for its extraordinary biodiversity, with cichlid fish constituting the vast majority of the lake's species (Snoeks, 2000). Research has revealed that Lake Malawi is home to approximately 1000 endemic cichlid species (Monticini, 2010; Konings, 2016). Many

cichlid species have become popular among aquarium hobbyists due to their color diversity, interesting behaviors, and reproductive strategies. One of the most sought-after and commercially valuable species in the aquarium industry is the *Maylandia estherae* (Konings, 2001).

M. estherae is a cichlid species that inhabits the rocky coastal regions of Lake Malawi (Ribbink et al., 1983). Like many other cichlid species, adult individuals engage in mouth brooding. Mouth brooding is defined as the process in which female individuals carry and protect fertilized eggs in their mouths, ensuring their development (Keenleyside, 1991). This reproductive strategy significantly increases the survival chances of larvae (Barlow, 2000). However, there are limited studies in the literature regarding the larval development of *M. estherae*.

Examining the larval development of fish is of great importance for understanding ontogenetic processes, elucidating reproductive biology, and conducting aquaculture studies (Kendall et al., 1984). The larval stage is a critical phase in the life cycle of fish, characterized by the fastest morphological, physiological, and behavioral changes (Fuiman & Werner, 2009). Therefore, a detailed examination of larval development provides valuable information about the biology, ecology, and evolutionary processes of species. Furthermore, knowledge of larval development is crucial for fish culture and stock management.

M. estherae, being an endemic species of Lake Malawi and its popularity in the aquarium industry, requires further research on its biology and ecology. This study aimed to morphologically investigate the larval development of *M. estherae*. The findings will contribute to understanding the ontogenetic processes of the species and support aquaculture studies. Additionally, the study results will significantly contribute to the knowledge of larval development in Malawi cichlids and provide insights for future research.

MATERIAL AND METHODS

Broodstock Selection and Aquarium Conditions

In this study, M. estherae individuals older than one year, having reached reproductive maturity and actively breeding, were selected as broodstock. The broodstock was housed in rectangular glass aquariums measuring 80×40×45 cm with a water capacity of approximately 125-130 liters and a water depth of 40 cm. Each aquarium was stocked with a total of 18 individuals, consisting of 3 males and 15 females. Aquarium aeration was provided only by sponge filters, and no decorative materials were added. Throughout the broodstock maintenance, spawning, and larval development processes, the water temperature was maintained at 29±0.5°C, and the pH was kept between 7.5-8. To ensure a stable water temperature, the room temperature was kept controlled, and no heaters were used in the aquariums.

Egg Collection and Incubation Process

The spawning process lasted for several hours, and fertilized eggs were mouth-brooded by female individuals. The swelling on the ventral side of eggcarrying females was used to detect the presence of eggs. To monitor larval development, eggs were collected from females identified as carrying eggs in their mouths one day after spawning and transferred to an artificial incubation unit. The eggs hatched approximately 4 days after fertilization, and the hatching day was considered as the 1st day of larval age.

Morphological Investigation of Larval Development

To examine larval development, eggs were collected from the mouths of broodstock 1-2 days after spawning was observed, and artificial incubation was performed. The morphological development of larvae was monitored daily from hatching to the juvenile stage. Sampled larvae were kept in an incubation unit at a constant room temperature and a water temperature of $29\pm0.5^{\circ}$ C.

Morphological investigations were performed using an Olympus BX51 research microscope (Tokyo, Japan), and larvae were photographed using a Q Imaging Micropublisher 3.3 RTV camera (Canada) attached to the microscope. After the photography process was completed, live specimens were returned to the incubation unit. The day the larvae hatched from the eggs was defined as Day 1.

RESULTS

The morphological appearance of the larvae between day-1 and day-13 is presented in Figure 1.

1 DAH (1st day after hatching): On the first day, the larva has a yolk sac close to the size of the egg. The head part is distinct. The eye structure has formed, but the eye lenses are indistinct. Development is ongoing. The body is in the form of a primordial fin. The formation of the pelvic fin can be clearly observed. During this period, the larva is completely on the bottom. Occasional short-term tail movements are observed. It is mostly motionless throughout the day.

3 DAH: The yolk sac has slightly decreased in size compared to the first day. However, it still maintains a large volume. The head shape has become slightly more distinct. The eye appears to have a normal eye structure. The primordial fin has started to develop.

During these days, the larva can exhibit more rapid tail movements. However, it still cannot exhibit freeswimming behavior.

4-5 DAH: During these days, the yolk sac has slightly decreased in size compared to the first 3 days. It is not small enough in volume for the larva to carry for free swimming. The larva can perform short-term swimming movements. However, it is still mostly on the bottom throughout the day. The mouth is open.

6-8 DAH: The yolk sac has further decreased in size. The larva's short-term free-swimming time has extended. The larva is capable of feeding from external sources. However, even if no food is given to the larva during this period, the yolk sac can provide the energy support for the larva to sustain its life. The dorsal and caudal fins have started to take shape. Pigmentation has intensified on the head and dorsal parts of the body. However, the overall body is still transparent in appearance.

9-11 DAH: The yolk sac has significantly decreased in size. However, it has not been completely consumed. The dorsal fin has taken shape. The anal fin has become distinct. Free-swimming movements have accelerated. During this period, external feeding should be initiated.

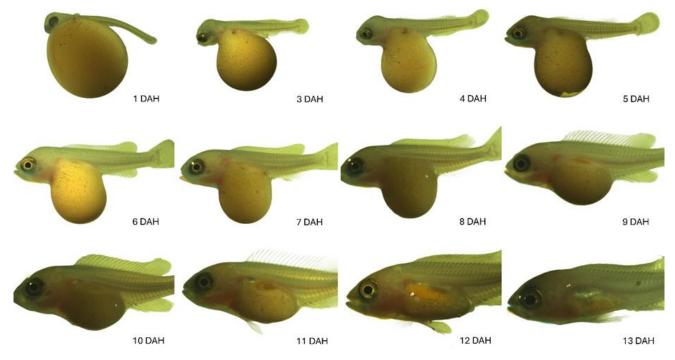


Figure 1. Morphological development of *M. estherae* larvae from the first to 13th days after hatching (DAH: Days After Hatching)

12-13 DAH: The yolk sac has been completely consumed. The shapes of the dorsal, anal, and caudal fins are much more distinct. Pigmentation has spread throughout the body. The transparent appearance of the body is transforming into a colored form. During these days, free-swimming movements have become much more rapid.

13-25 DAH: During this period, the body color of the larva becomes more distinct with each passing day. Gradually, the orange-reddish body color appearance of the parents is forming. Similarly, the rays and shapes of the dorsal, anal, and caudal fins are also transforming into the adult form. The behaviors of external feeding and swimming are like juvenile fish. During this period, the transition from the larval stage to the juvenile stage occurs. The timing of this transition process is directly related to water temperature.

DISCUSSION

In this study, the larval development of *M. estherae* was investigated morphologically and the findings were compared with other Malawi cichlid species. It was determined that *M. estherae* larvae exhibited rapid development from the first day after hatching and reached the juvenile stage in approximately 21 days. Similarly, it has been reported that larval development lasts 21-28 days in other Malawi cichlids such as *Labeotropheus fuelleborni, Pseudotropheus zebra, Cynotilapia afra, and Aulonocara stuartgranti* (Balon, 1977; Holden & Bruton, 1994; Msiska & Costa-Pierce, 1999).

The total lengths and mouth openings of *M. estherae* larvae gradually increased during larval development. Studies on other Malawi cichlids such as *Maylandia zebra, Metriaclima lombardoi, Labidochromis caeruleus* and *Pseudotropheus socolofi* have also reported that larval growth follows a similar course and total length reaches 10-12 mm in 20-25 days (Msiska & Costa-Pierce, 1999; Carleton et al., 2005; Fujimura & Okada, 2007).

Similar results are observed when the larval development of other cichlid species outside Lake Malawi is examined. For instance, it has been reported that larval development lasts approximately 20 days

in *Amphilophus citrinellus*, a species living in Central America, and larvae reach an average length of 12 mm at the end of this period (Balon, 1977). Similarly, it has been reported that larval development lasts 25-30 days in *Cichlasoma dimerus*, a species living in South America, and larvae reach a length of 15-20 mm (Meijide & Guerrero, 2000). It has been determined that the larval development duration and growth rates of commercially important cichlid species living in other African lakes, such as *Oreochromis niloticus*, *Sarotherodon galilaeus*, and *Tilapia zillii*, are similar to those of Malawi cichlids (Fujimura & Okada, 2007).

The morphological changes observed in M. estherae larvae are similar to those of other Malawi cichlids. In the first days of larval development, it was observed that larvae carried a yolk sac and their free-swimming ability had not yet developed. It was determined that the yolk sac was completely depleted between the 5th and 7th days and larvae started to feed externally. Similar findings have been reported in Labeotropheus trewavasae, Metriaclima callainos, Dimidiochromis compressiceps and Copadichromis borleyi (Balon, 1977; Msiska & Costa-Pierce, 1999; Konings, 2001). It is known that the yolk sac is an important food source in the early stages of larval development in other cichlid species and larvae start to feed externally with the development of free-swimming ability. For example, it has been reported that the yolk sac is depleted in 3-4 days and larvae start to feed externally from the 5th day in Apistogramma cacatuoides, a species living in South America (Pandolfi et al., 2009). Similarly, it has been reported that the yolk sac is depleted in 6-7 days and larvae start to feed externally from the 8th day in Amphilophus rostratus, a species living in Central America (Molina, 2011).

During larval development, important morphological changes such as fin differentiation and pigmentation were observed in M. estherae larvae. In the first days after hatching, the caudal fin of larvae was homocercal, while it transformed into a heterocercal structure in the later days of development. Similarly, it was determined that the dorsal and anal fins gradually differentiated during larval development. Pigmentation on the body started to become evident from approximately the 10th day of development and continued to increase until the 21st day. These findings are consistent with the larval development characteristics observed in other Malawi especially in *Pseudotropheus* cichlids, elegans, Melanochromis auratus and Petrotilapia nigra (Msiska & Costa-Pierce, 1999; Konings, 2001; Fujimura & Okada, 2007). It is known that fin differentiation and pigmentation are important morphological changes during the larval development process in other cichlid species. For example, it has been reported that during larval development in Geophagus brasiliensis, a species living in South America, the caudal fin transforms from a homocercal to a heterocercal structure, dorsal and anal fins differentiate, and body pigmentation increases (Perini et al., 2010). Similarly, it has been reported that similar morphological changes are observed during larval development in Etroplus suratensis, a species living in Asia.

CONCLUSION

The larval development of M. estherae was examined morphologically and the findings were compared with other Malawi cichlids. It was determined that M. estherae larvae show similarities with other Malawi cichlids, especially with species belonging to the genera Labeotropheus, Pseudotropheus, Melanochromis, and Petrotilapia, in terms of development duration, growth rate, and morphological characteristics. In addition, similar results were obtained when examining the larval development of other cichlid species outside Lake Malawi. It was observed that the larval development duration varies between 20-30 days among species. Growth rates are similar across the examined species. The yolk sac is depleted in early stages, and external feeding starts with the development of freeswimming ability. Additionally, fin differentiation and pigmentation are important morphological changes during the larval development process. However, it is thought that larval development may show small differences between species and these differences may be related to ecological and evolutionary processes. It is recommended that future studies investigate the causes and consequences of similarities and differences in larval development of different cichlid species in more detail.

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Compliance with Ethical Standards

Authors' Contributions

- İÇ: Conceptualization, Data curation, Formal Analysis, Writing original draft.
- PÇ: Conceptualization, Investigation, Methodology, Writing – review & editing.

All authors read and approved the final manuscript.

Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical Approval

For this type of study, formal consent is not required.

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Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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