

Survival and growth performances of amphidromous goby larvae (nike) during preliminary rearing experiments

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Abstract. The high demand for amphidromous nike goby fish larvae in Gorontalo waters can cause a severe imbalance in the natural population, since the larval stage is crucial for its regeneration. Domestication is one of the strategic steps to provide fish stocks without altering their natural sustainability. This research aimed to determine the survival rate, growth in length and body weight of nike fish and monitor the dynamics of rearing water quality in laboratory experiments. Twenty-five fish larvae were reared in 3 containers (size $30 \times 20 \times 20$ cm³, water volume 5 L) for 11 consecutive days. Tests of larvae's survival, length, and weight were recorded daily, while media quality measurements were carried out every 6 hours (06.00, 12.00, 18.00, and 24.00 h). The experiment revealed that the average survival rate of fish larvae during rearing was 20%. The results of single-factor ANOVA statistical testing on observations of individual larval size did not show any significant differences between observation days (p>0.05). The average total length and weight of the sampled nike fish were determined from individual larvae that survived during 11 days of the experiment: 26.36±0.49 mm and 0.15±0.01 g, respectively. The water quality parameters, namely the temperature (21.43-30.50°C), dissolved oxygen (7.32-9.01 mg L⁻¹), pH (6.65-7.50), salinity (31.93-38.00 ppt), TDS (25.63-33.33 mg L⁻¹), and conductivity (40.30-65.97 μ S cm⁻¹), were in the ranges tolerated by the nike fish. Generally, the positive allometric growth pattern shown by this amphidromous species indicates a high adaptive ability in captive environments and potential for domestication. Further experiments need to involve other factors related to the living characteristics of amphidromous species.

Key Words: aquaculture, domestication, Eleotridae, Gobiidae, Tomini Bay.

Introduction. Nike, the marine larva of goby fish species, is a notable symbol of the Gorontalo region (Pasisingi & Olii 2023). In 2020, the Ministry of Law and Human Rights of the Republic of Indonesia recognized it as the Communal Intellectual Property of Gorontalo Province (Botutihe 2020). Popular among locals, nike fish is also sought after by tourists for its taste and nutritional composition (Yusuf et al 2012; Liputo et al 2019; Husain et al 2020). It is prepared in diverse traditional dishes such as ilepa'o labia, perkedel nike, woku nike, ilepa'o, and duo tilumiti (Nasriani 2018), and has also been used as a raw material in a range of processed food products (Arisanti 2018; Abdullah & Mutia 2020). The heavy metal content in the fish tissue was recently confirmed as safe for consumption (Olii et al 2024).

Nike fish, exported outside the Gorontalo area (Soekamto et al 2018), indicate a high market demand. This research is driven by two main phenomena: the uncertain period of nike fish emergence in nature and the potential threat of their extinction. First, the uncertain timing of nike fish appearances in nature impacts the market's ability to meet the high demand. The fish in the Gorontalo Sea appear only at the beginning of the Hijriyah month (a lunar calendar based on the cycles of the moon) or for three to seven consecutive days, reappearing at the start of the following lunar phase in the Hijriyah calendar (Olii et al 2017; Pasisingi & Abdullah 2018). This unpredictability makes it difficult to find and consume consistently. Tourists and local residents must await the

natural emergence of the species to consume it in its fresh form. Second, while there is no scientific evidence proving the extinction of nike fish, their absence in Gorontalo waters during the new moon phase (Hasanah 2020) and the decreasing catches by local fishermen (Madjowa et al 2020) signals a threat to their sustainability. Therefore, early measures are needed to preserve the fish resources in Gorontalo waters. Domestication and culture are crucial, given the nike fish's role as a source of protein and a promotional symbol of Gorontalo's local specialties.

The study of live performance of nike fish in a controlled environment has never been conducted. The mariculture industry requires basic research on the application of species adaptation in cultivation and domestication, throughout their life cycle (Alimudin & Wiyono 2005). Acclimatization involves preparing fish physiologically for new environmental conditions (Arianto 2018). Through initial rearing and experimental setups, researchers can modulate the fish's capacity to adapt to controlled environments, potentially including breeding (Yulfiperius 2006). According to Helvick et al (2009), survival and growth are critical parameters for evaluating the success of larval fish rearing. Therefore, this research aimed to determine the survival rate and growth in length and weight of nike fish, along with water quality parameters during the rearing experiment.

Material and Method

Time and location of research. This experiment conducted in August 2021 comprised two stages: preparation and observation. The maintenance of test biota was conducted at the Hydrobioecology Laboratory, Faculty of Marine and Fisheries Technology, Universitas Negeri Gorontalo. Observations of the survival and growth of the larvae, as well as measurements of the water quality of the rearing media, were carried out over eleven consecutive days.

Preparation. The experiment utilized three sets of aquariums with aerator, each measuring $30 \times 20 \times 20 \text{ cm}^3$, each containing 5 L of seawater from the natural habitat of the larva. Approximately 250 nike fish were caught in Gorontalo Sea waters using nets during their natural emergence season. Live samples were placed in plastic containers, treated with oxygen, and transported to the laboratory. Upon arrival, all fish larvae were transferred to an initial holding aquarium with a dimension of 50 x 30 x 30 cm³, containing seawater, and were aerated for approximately 24 hours to ensure physiological homogeneity. From this container, 25 fish larvae were randomly selected and transferred to each of the three experimental aquariums, where water quality parameters were measured and recorded. The fish was unfed, assuming that nike's natural diet would be present and photosynthesized in the rearing medium.

Observation. The survival rates and the length and weight of test larvae were recorded every morning at 07:00 h, following the measurement of water quality parameters. Larvae length and weight were measured using a digital caliper (accuracy of 0.01 mm) and a digital scale (accuracy of 0.001 g), respectively. Temperature, dissolved oxygen (DO), pH, salinity, total dissolved solids (TDS), and conductivity (DHL) were measured every six hours at 06:00, 12:00, 18:00, and 24:00 h using a water quality tester device.

Data analysis. The survival rate of the fish was calculated using the following equation by Effendie (1979):

SR (%) =
$$(N_t/N_0) \times 100$$

Where:

SR - survival rate (%); N₀ - number of living test biota at the start of the experiment (initial); N_t - number of living test biota at the end of the experiment (final). Data from water quality measurements were visualized in graphical form and interpreted descriptively. A single factor ANOVA (95% confidence level) was conducted to test the significance of size changes during the observation period, for the data on alteration in length and body weight of individual larvae. This analysis designated the "observation day" as the variable for levels and "the number of rearing containers" as the variable for repetitions. Determining population growth patterns begins with analyzing the length-weight relationship using (Robertis & Williams 2008) equation below:

$$W = aL^b$$

Where: W - body weight (g); L - total length (mm); a - equation constant; b - growth coefficient.

The above equation is logarithmically transformed into the following linear equation:

$\ln W = \ln a + b \ln L$

A further t-test was applied to the b value in the equation to determine the growth pattern. According to (De Guzman & Rosario 2020), a value of b>3 fish growth patterns is positive allometric, b=3 isometric growth patterns, while b<3 means the biota has a negative allometric growth pattern.

Results. The average survival rate of nike fish larvae over the 11-day rearing period was 20%, with an average of 2 individuals dying each day. The highest mortality rate occurred on the second day of maintenance. Subsequently, the number of deaths remained relatively low until the last three days of maintenance, as depicted in Figure 1.



Figure 1. Survival of test biota larvae ('nike") during rearing experiment.

The average range of total length and weight of larvae spanned from 25.84-27.35 mm and 0.14-0.17 g, respectively. Individually, larvae measured on average 26.36 ± 0.49 mm in length and weighed 0.15 ± 0.01 g. Statistical analysis revealed no significant changes in the test biota's length or weight over the 11-day rearing period (p>0.05), as shown in Figure 2. At a coefficient of determination of 70%, the equation of the length-weight relationship showed a positive allometric growth pattern during the experiment. This

indicated that the rate of body weight increase was faster than the increase in body length (Figure 3).



Figure 2. Average changes in total length (a) and weight (b) of test specimens from 3 rearing containers, during the experiment.



Figure 3. Relationship between length and weight of nike larvae during the experiment.

Water quality remained relatively stable throughout the 11 days of observation, with notable exceptions in pH and salinity parameters during the last three days (Figure 4). Table 1 presents the range of water quality parameters measured during the experiment. The lowest temperature (21.43°C) and the highest (30.50°C) were recorded on observation day seven at 24:00 h and day five at 12:00 h, respectively. The lowest dissolved oxygen (DO) level (7.32 mg L⁻¹) and the highest (9.01 mg L⁻¹) were observed on day eight at 18:00 h and day two at 18:00 h, correspondingly. The lowest pH (6.65) and the highest (7.50) were detected on day 11 at 12:00 h and 24:00 h, respectively. Similarly, the lowest (31.93 ppt) and highest (38.00 ppt) salinity averages were recorded consecutively on day 11, at 16:00 h and 24:00 h. The lowest average total dissolved solids (TDS) value (25.63 mg L⁻¹) was observed on day six at 12:00 h, while the highest value (33.33 mg L⁻¹) was measured on day four at 18:00 h. Lastly, the lowest average conductivity value (40.30 μ S cm⁻¹) was recorded on day seven at 06:00 h, while the highest (65.97 μ S cm⁻¹) was measured on day two at 24:00 h.



Figure 4. Average measurement values at 6:00, 12:00, 18:00, and 24:00 h for water quality parameters: temperature (a), dissolved oxygen (b), pH (c), salinity (d), Total Dissolved Solid (e), and conductivity (f) in 3 nike rearing mediums for 11 observation days.

Table 1

The environmental parameters during larva rearing time (measured at 6:00, 12:00, 18:00, and 24:00 h)

Parameters	Unit	Range (min-max)	Average
Temperature	°C	21.43-30.50	27.97±1.42
DO	mg L ⁻¹	7.32-9.01	8.09±0.53
рН	-	6.65-7.50	6.97±0.14
Salinity	ppt	31.93-38.00	35.49±1.46
TDS	mg L ⁻¹	25.63-33.33	29.11±1.68
Conductivity	µS cm⁻¹	40.30-65.97	57.54±4.18

Discussion. The life cycle of nike fish presents a unique case, as it falls within the category of amphidromous fish (Pasisingi & Abdullah 2018; Pasisingi et al 2021; Olii

& Pasisingi 2022a). The term "nike fish" refers to a group of small fish that were originally thought to be native exclusively to the waters of Gorontalo, North Sulawesi (Ibrahim et al 2024). Genetic studies recognize that nike fish encompass a group of goby larval stages comprising multispecies, as reported by (Sahami et al 2019; Sahami & Habibie 2021). These fishes begin their life in seawater and migrate to freshwater to develop and spawn (Olii et al 2017; Baid et al 2024). The adult stadia of this goby species are not referred to as nike fish, as emphasized by Pasisingi et al (2020) and Sahami & Habibie (2020). Unlike their larval counterparts, the adults are less favored for consumption by the public. This is attributed to their relatively larger size, complete limbs, hardened bones (Pasisingi et al 2024), and fin completeness (Sinulingga et al 2024) as they transition to freshwater (Pasisingi et al 2020).

Marine fisheries cultivation, particularly the availability of seeds, emerges as a viable alternative to alleviate fishermen's reliance on traditional fishing methods, as highlighted by Cahyanurani et al (2022). Domestication represents a significant effort to acclimate fish to captive conditions, fostering accelerated growth rates and reproductive potential (Haryono et al 2020). This demands a series of intricate, long-term, and high-cost experiments to achieve success. Moreover, the migratory and growth patterns of nike fish entail gradual adaptations to changing water salinity and various other external factors. However, the initiation of nike fish rearing in seawater media in this research holds promise, offering insights into the potential for successful domestication of nike. This parallels the achievements in salmon, a group of amphidromous fish with similar migration characteristics, culture as demonstrated by Barrett et al (2020) and Frühe et al (2020).

This study marks the inaugural documentation of nike larvae-rearing experiments. Achieving domestication necessitates a comprehensive understanding of the fish's life cycle in captivity, irrespective of its natural habitat, as asserted by Kristanto (2022). Hence, both adult and larval stages must be integral to these initial efforts. However, the core focus of this research, preliminary rearing, is solely limited to nike larval stages, renowned for their high economic value, as noted by Handoko et al (2017). This strategic limitation is grounded in prioritizing the sustainability of the experiment. Even in scenarios where the likelihood of successful nike domestication is deemed low, the success in maintaining the larval stage in captivity for a specified duration presents an alternative for ensuring a supply of fresh nike during non-emergence seasons.

Larvae represent a highly vulnerable and pivotal life stage for fish species, as underscored by Iida et al (2021). Within seawater environments, temperature emerges as a critical environmental determinant influencing the survival of amphidromous larvae, as noted by Teichert et al (2021). Nike fish, characterized by their resilience against prevailing conditions, are presumed to exhibit a better tolerance to extreme environmental parameters (Olii et al 2017). Parameters such as dissolved oxygen (DO), pH, salinity, ammonia, and nitrate are fundamental in shaping fish survival (Ismi et al 2018; Perwito et al 2015; Panggabean et al 2016). However, the notable exclusion of current as a crucial parameter in this research represents a significant limitation. Water current is an important factor in the development and survival of amphidromous species. The escalating stress experienced by larvae is evident in the consistent increase in length and body weight during rearing. Kristanto (2022) highlighted that transferring fish from their natural habitat to breeding containers often induces stress, diminishing survival rates in the new environment. Additionally, the larvae allocate substantial energy towards adapting to the experimental setting, resulting in a mere 20% survival rate on the 11th day of observation. Iida et al (2010) conducted experiments demonstrating that the survival rate of newly hatched goby larvae, specifically Sicyopterus japonicus, was significantly higher in saline water than in freshwater. Moreover, survival rates were higher in saline water than in freshwater at 23°C and 28°C. Notably, S. japonicus exhibits more remarkable adaptation to brackish and seawater environments than to freshwater habitats.

The water quality parameters of Muara Bone in Gorontalo, a nike's natural habitat, as shown by Olii & Pasisingi (2022b), exhibit ranges of 27.67-30.82°C for the temperature, 7.3–7.8 mg L⁻¹ for the dissolved oxygen (DO), 5.0–8.4 for the pH, 0–14 ppt

for the salinity, 1353.0–2294.2 mg L⁻¹ for the total dissolved solids (TDS), and 2705– 3856 μ S cm⁻¹ for the conductivity. Comparing these values with the findings of this research reveals similar ranges, except for the salinity, TDS, and conductivity. This disparity can be attributed to the interrelation among these three physical parameters. Conductivity and total dissolved solids, as outlined by Rusydi (2018), are correlated water quality parameters related to the salinity levels. Notably, TDS exhibits the highest correlation with conductivity, as highlighted by (Kothari et al 2021). The observed tendency for increasing salinity in the rearing media can be attributed to water evaporation, a phenomenon likely occurring during the experiment. In the context of seawater, where salinity typically measures 35 ppt, the larval development of unfed *Sicyopterus lagocephalus* larvae was significantly impacted by temperature, with survival time remarkably reduced in warmer waters (Teichert et al 2021). High salinity levels influence water conductivity due to ionized salt content within the water medium. The elevated TDS value can also be attributed to the absence of dilution stemming from freshwater input, a characteristic typically observed in nike's natural habitat waters.

The positive allometric growth pattern in this experiment indicates that the size of the larvae tends to be fat. A similar growth pattern was shown by nike larvae in Gorontalo waters on the appearance in March, April, and May 2018 (Pasisingi et al 2020), on 1 and 3 July 2021 and 1 September 2021 (Olii et al 2023), on days 2, 4, 7, 10, and 11 during August 2022 (Olii & Pasisingi 2023). The survival and growth performance data of goby larvae from this preliminary study indicate that this amphidromous species possesses high adaptability and potential for domestication.

Conclusions. This study has successfully documented the baseline data necessary to domesticate nike fish. The average survival percentage of nike fish during 11 days of rearing is 20%. The statistical test results showed that the test biota's length and weight did not change significantly (p>0.05) during the experiment. The range of total length and average individual weight was 25.84-27.35 mm and 0.14-0.17 g, respectively. The range of measured values of temperature, DO, pH, salinity, TDS, and conductivity of maintenance media water were $27.97\pm1.42^{\circ}$ C, $8.09\pm0.53 \text{ mg} \text{ L}^{-1}$, 6.97 ± 0.14 , $35.49\pm1.46 \text{ ppt}$, $29.11\pm1.68 \text{ mg} \text{ L}^{-1}$, and $57.54\pm4.18 \mu\text{S cm}^{-1}$, respectively. The survival and growth performance of nike in this preliminary research shows that this amphidromous fish has high adaptability and potential to be domesticated.

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