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Induction of reproduction of fish Anasa *Nomorhampus* sp. endemic Palu, Central Sulawesi orally through hormon bioencapsulation use *Chironomus* sp.

Induksi reproduksi ikan anasa *Nomorhampus* sp. endemik Palu, Sulawesi Tengah secara oral melalui bioenkapsulasi hormon menggunakan *Chironomus* sp.

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ABSTRACT

Species *Nomorhampus* sp. with the local name Anasa fish, endemic to Palu, Central Sulawesi, has a unique superior mouth shape, beak-shaped jaws, attractive colors and patterns, becoming an export commodity with high economic value, but currently it cannot be cultivated, domestication efforts are needed to avoid extinction, by carrying out hormonal manipulation that accelerates the domestication process. This study aims to evaluate the effectiveness of Oodev® on the induction of reproduction in the fish *Nomorhampus* sp. through bioencapsulation using *Chironomus* sp. which resulted in births, five groups of fish were fed using Oodev® at a dose of 1 mL/kg parent weight and NaCl 1 mL/kg parent weight as a control group, the fish were kept for 60 days. This research focuses on assessing specific weight growth rate (LPBS), specific length growth rate (LPPS), survival rate (TKH), gonadosomatic index (IGS), gonadal histology, birth frequency, number of births, and growth performance. Fish fed Oodev® feed showed higher SGR and IGS ($p<0.05$), 40% of fish fed Oodev® supplementary feed successfully gave birth with a total of 9 fry, while no birth occurred in control fish. Histological analysis showed faster gonad development in fish fed Oodev®. Hormonal induction with Oodev® can accelerate reproduction in anasa fish in cultivation containers. These findings provide valuable insight for fish farmers regarding the effect of Oodev® on gonad development in anasa fish in both male and female parents. It is hoped that this discovery will speed up the process of domestication of Anasa fish.

Keywords: domestication, endemic, *Nomorhampus* sp., Oodev®, reproduction

ABSTRAK

Spesies *Nomorhampus* sp. dengan nama lokal ikan Anasa endemik Palu, Sulawesi Tengah, memiliki keunikan bentuk mulut superior rahang berbentuk paruh, warna dan corak menarik menjadi komoditas ekspor dengan nilai ekonomis yang tinggi, namun saat ini belum dapat dibudidayakan, perlu upaya domestikasi agar tidak terjadi kepunahan, dengan melakukan manipulasi hormormonal yang mempercepat proses domestikasi. Penelitian ini bertujuan untuk mengevaluasi efektivitas Oodev® terhadap induksi reproduksi ikan *Nomorhampus* sp. melalui bioenkapsulasi menggunakan *Chironomus* sp. yang menghasilkan kelahiran, lima kelompok ikan yang diberi pakan menggunakan Oodev® dosis 1 mL/kg bobot induk dan NaCl 1 mL/kg bobot induk sebagai kelompok kontrol, ikan dipelihara selama 60 hari. Penelitian ini fokus pada penilaian laju pertumbuhan bobot spesifik (LPBS), laju pertumbuhan panjang spesifik (LPPS), tingkat kelangsungan hidup (TKH), indeks gonadosomatik (IGS), histologi gonad, frekuensi kelahiran, jumlah kelahiran, dan kinerja pertumbuhan. Ikan yang diberi pakan Oodev® menunjukkan SGR dan IGS yang lebih tinggi ($p<0,05$), 40% ikan yang diberi pakan tambahan Oodev® berhasil melahirkan dengan jumlah total 9 ekor benih, sementara pada ikan kontrol tidak terjadi kelahiran. Analisa histologi menunjukkan perkembangan gonad yang berkembang lebih cepat pada ikan yang diberi pakan Oodev®. Induksi hormonal dengan Oodev® mampu mempercepat reproduksi pada ikan anasa di wadah budidaya. Temuan ini memberikan wawasan berharga bagi pembudidaya ikan mengenai pengaruh Oodev® terhadap perkembangan gonad pada ikan anasa baik pada induk jantan maupun betina. Penemuan ini diharapkan dapat mempercepat proses domestikasi ikan Anasa.

Kata kunci: domestikasi, endemik, *Nomorhampus* sp., Oodev®, reproduksi

INTRODUCTION

Sulawesi Island is one of the islands with high biodiversity (Andriani *et al.,* 2018). Among its diverse species are endemic ornamental fish. The uniqueness and diversity of these fish are still being explored, and Sulawesi Island serves as the center of endemic fish biodiversity in Indonesia, with their distribution remaining limited (Gani *et al.,* 2022; Hadiaty, 2018; Utama *et al.,* 2022; Wicaksono *et al.,* 2022; Zainal *et al.,* 2022). Previous explorations have successfully identified ornamental fish endemic to Southeast Asia from the Zenarchopteridae family, specifically the *Nomorhampus* sp. This fish is found in rivers and freshwater lakes in Indonesia and the Philippines. Twelve species of *Nomorhampus* sp. have been discovered on the island of Sulawesi, while seven species have been identified in the Philippines (Kobayashi *et al.,* 2020; Lawelle *et al.,* 2021).

The *Nomorhampus* sp. species has a distinctively superior mouth shape and a beak-like jaw, along with attractive colors and patterns, making it suitable as a freshwater ornamental fish (Kusumah *et al.,* 2016). The reproductive type of Zenarchopteridae fish is generally viviparous (Downing & Burns, 1997; Kanou *et al.,* 2019), with the embryonic development phase occurring within the parent's body (Parawangsa *et al.,* 2019). A limitation of *Nomorhampus* sp. is its low reproductive output, as a single parent can produce a maximum of only 22 offspring. *Nomorhampus* sp. fish has become an export commodity for foreign markets, including species such as *Nomorhampus ebrardti, Nomorhampus celebensis,* and *Nomorhampus liemi.* Its body shape, resembling a miniature arowana, along with its historical value as an endemic fish, makes it highly attractive to ornamental fish collectors in Europe and China.

One species of *Nomorhampus sp.* found in the Palu River in Central Sulawesi, locally known as the anasa fish, also has the potential to become an export commodity for the ornamental fish market. Demand for this fish is high, but since it is still obtained from wild catches, there is a risk of overfishing, which could threaten its survival and the sustainability of resources. Therefore, domestication efforts are necessary to prevent extinction and support the development of aquaculture commodities that can be cultivated in captivity. Domestication is carried out by moving animals from their natural habitat to captivity (Milla *et al.,* 2021) so that they can adapt to environmental conditions outside their original habitat.

The ultimate goal is to understand the characteristics of fish and modify their offspring to produce more productive and efficient individuals (Houston *et al.,* 2020; Teletchea, 2021). The domestication of aquatic animals has progressed rapidly due to the development of commercial aquaculture, efforts to conserve endangered populations, and the introduction of new commodities in the aquaculture industry (Augusta, 2016; Zadmajid, 2016). Domestication activities consist of three stages, and Anasa fish are already in the second stage. The first stage involves selecting and collecting fish for domestication and adapting them to the new environment (Milla *et al.,* 2021). After transportation, the second stage consists of acclimatization to ensure the fish can grow and develop.

The third stage focuses on reproduction, enabling the fish to produce the first generation from wild fish spawning. Additionally, reproductive technology is applied to facilitate fish cultivation (Hara *et al.,* 2016; Husen *et al.,* 2021) according to specific targets and needs. The adaptability of fish affects their reproductive ability during domestication (Susatyo *et al.,* 2022). Fish reproduction is directly influenced by environmental factors such as rainfall, temperature changes, petrichor, substrate, and water quality, including pH, hardness, and dissolved oxygen (DO) (Mellisa *et al.,* 2019). Differences between the natural environment of fish and captivity can create obstacles that lead to reproductive failure, as environmental factors influence the physiological reproductive system of fish (Tahapari & Dewi, 2013). Environmental differences and the extended adaptation period required can increase the risk of failure in natural spawning. This issue can be addressed through hormonal manipulation, which aims to shorten the domestication period during the reproductive stage.

The addition of the required reproductive hormones can accelerate the maturation process in Anasa fish (*Nomorhampus* sp.). One commercial product that helps stimulate gonad maturation is Oodev® (Oocyte Developer). Oodev® contains a combination of Pregnant Mare Serum Gonadotropin (PMSG) and Anti-Dopamine (AD). PMSG is a complex glycoprotein derived from pregnant mare serum, which mimics the effects of Luteinizing Hormone (LH) and Follicle-Stimulating Hormone (FSH). FSH plays a role in

activating the gonads to synthesize estradiol-17β, which stimulates the formation of vitellogenin (Brzuska, 2021; Nagahama & Yamashita, 2008). PMSG also has a longer-lasting effect compared to other gonadotropin hormones due to its high carbohydrate content, particularly in the sialic acid group (Wahyuningsih *et al.,* 2013).

The anti-dopamine component consists of neurotransmitter chemical compounds that inhibit dopamine receptor activity and increase GnRH secretion. Research on the use of Oodev® as a hormone that stimulates gonad maturation and significantly affects the reproduction of various fish species has been conducted in several studies. These include research on tiger prawns (Laining *et al.,* 2016), red fin shark species (Islami *et al.,* 2017), snakehead fish (Anwar *et al.,* 2018), and kelabau fish (Asiah *et al.,* 2021), with an optimal dose of 0.5–1 mL/kg of broodstock weight. Based on the description above, this study aims to evaluate the effectiveness of using Oodev® on reproduction, including gonad maturation in males and females, as well as birth in Anasa fish (*Nomorhampus* sp.), to achieve reproductive targets outside their natural habitat more quickly and successfully.

MATERIALS AND METHODS

Test fish and maintenance

The test fish were collected in January 2023 from the inlet river basin of Lake Lindu, Central Sulawesi. The collected fish measured 5–6.5 cm in length, and the female fish had a stomach circumference of less than 2.5 cm, which aligns with the size of new broodstock based on previous research. A total of 12 males and 13 females were successfully obtained. The fish were maintained separately by sex for three weeks to allow them to adapt to the cultivation container and to ensure that no females were pregnant or giving birth. This maintenance was conducted at the Wet Aquatic Organism Reproduction and Genetics Laboratory, Faculty of Fisheries and Marine Sciences, Bogor Agricultural University. The study used aquarium containers measuring 30×25×25 cm^3, with a total of 10 aquariums. A recirculation (closed) system was implemented, equipped with foam and filtration to maintain water quality. The stocking density was 9 L per fish, with each aquarium containing one male and one female.

Feed enrichment with bioencapsulation of *Chironomus* sp.

The administration of hormones to Anasa fish (*Nomorhampus* sp.) through natural *Chironomus* sp. feed is carried out using the bioencapsulation method by soaking the feed in Oodev® through several stages. First, *Chironomus* sp. is cleaned by washing and then drained using a sieve. Next, *Chironomus* sp. is soaked in a salt solution at a concentration of 2 ppm to induce dehydration, enhancing hormone absorption. The test fish are weighed to determine their weight and calculate the required dose of Oodev®. The hormone is then added to the enrichment container according to the predetermined dosage. The Oodev® dose used is 1 mL/kg of parent fish weight, based on previous studies on Red Fin Shark (Islami *et al.,* 2017), Snakehead Fish (Anwar *et al.,* 2018), and Kelabau Fish (Asiah *et al.,* 2021), which found an optimal dose of 0.5–1 mL/kg of parent weight.

Chironomus sp. was treated with hormones at the specified dose, stirred until evenly distributed, then soaked and left for 60 minutes before being washed again with clean water. It was then drained and stored frozen in a freezer at -20°C. Feed stock preparation was carried out every three days to maintain the quality of the feed and the hormones administered. The control feed was processed in the same way using a NaCl solution at a dose of 1 mL/kg of fish weight.

Enrichment feeding

The treated enrichment feed is given at a feeding rate of 5% and is provided three times a day: in the morning at 08.00 WIB, in the afternoon at 13.00 WIB, and in the evening at 17:00 WIB.

Birth observation

Fish births are observed daily by monitoring their behavior. If a fish appears calm and stays in the corner of the aquarium, with an enlarged abdominal circumference, it may be close to giving birth. Aquarium checks are conducted every morning at 07.00 WIB, in the afternoon at 13.00 WIB, and in the evening at 19.00 WIB. If a birth occurs, the larvae are immediately transferred to a new container.

Test parameters

Gonadosomatic index (GSI)

The gonadosomatic index was measured at the beginning of the study (Day 0) and at the end of the maintenance period (Day 60). This parameter

is useful for assessing gonad development in fish. The gonadosomatic index (GSI) can be calculated using the following formula:

$$GSI\ (\%) = \frac{\text{Weight of gonad organs (g)}}{\text{Body weight of test fish (g)}} \times 100$$

Note:
GSI = Gonadosomatic index (%)

Gonad histology

Histological observation of the gonads was conducted at the end of the study (H60) due to the limited number of fish and the challenging and restricted sampling location. This parameter was also examined to determine the stage of gonad development in the test fish and assess the effect of hormone administration. The histological analysis of the gonads involved several stages, including fixation, dehydration, clearing, infiltration, embedding, cutting, deparaffinization, staining, dehydration, clearing, coloration, and histological observation and documentation using a binocular microscope. This observation was conducted on both male and female test fish. For female gonads, the analysis included the development of oocyte eggs and embryogenesis of *Nomorhampus versicolor*, while for male gonads, the spermatogonia, secondary spermatocytes, spermatids, and spermatozoa phases were examined.

Birth interval and number of fish at the same birth

The birth interval refers to the time between one birth and the next, measured in days. This parameter is calculated to determine the interval between successive births in the same individual. A total of 10 female parents were observed under two treatments with five replications. The number of offspring per birth refers to the number of offspring produced in a single birth by each female *Nomorhampus versicolor* during the treatment. This parameter is used to assess the reproductive output of hormonally induced and non-treated parents. Observations and calculations were conducted over a 60-day maintenance period.

Specific weight growth rate (SWGR)

The calculation of the specific weight growth rate in test fish was conducted at the beginning and end of the study on both broodstock and larvae. The measurement of these test parameters was performed using the formula calculation provided by Yu *et al*. (2020).

$$SWGR\ (\%/day) = \frac{\ln W_t - \ln W_0}{t} \times 100$$

Note:
SWGR = Specific weight growth rate (%/day)
W_t = Average weight of fish at the end (g)
W_0 = Average weight of fish at the start (g)
t = Maintenance period (day)

Specific length growth rate (SLGR)

The specific length growth rate was calculated on days 0 and 60 at the end of the maintenance period using the NRC formula (1983).

$$SLGR\ (\%/day) = \frac{LnLt - LnL0}{t} \times 100$$

Note:
SLGR = Specific length growth rate (%/day)
L_t = Average length of fish at the end (cm)
L_0 = Average length of fish at the start (cm)
t = Maintenance period (day)

Survival rate (SR)

The survival rate calculation test parameters were assessed throughout the study and calculated from the beginning to the end of broodstock and larval maintenance using the formula provided by Yu *et al*. (2020).

$$SR\ (\%) = \frac{N_t}{N_0} \times 100$$

Note:
SR = Survival rate (%)
N_t = Final number of test fish (ekor)
N_0 = Start number of test fish (ekor)

Larvae growth performance

The parameters of the larval growth performance test include the initial and final weight, initial and final length, and the survival rate of the larvae. The survival rate was assessed throughout the study, from birth until 30 days of larval maintenance.

RESULTS AND DISCUSSION

Result

Gonadosomatic index (GSI)

The Oodev® bioencapsulation treatment, administered at a dose of 1 mL/kg of broodstock weight through *Chironomus* sp., effectively increased the gonadosomatic index of the test fish. The highest average value reached 35.50% in

female Oodev® fish and 15.15% in male Oodev® fish. Furthermore, the gonadosomatic index of anasa fish (*Nomorhampus* sp.) was analyzed using an independent t-test to determine whether there were significant differences between treatments. The test results confirmed significant differences between treatments.

Birth interval and number of fish at the same birth

Births occurred during the treatment trial, with the first birth taking place on the 42nd day of maintenance in the Oodev® test fish pair, repeat 1 (Individual O1), producing a total of two offspring with a birth interval of no more than five minutes. This pair gave birth again after a two-day interval, on the 44th day, producing three larvae

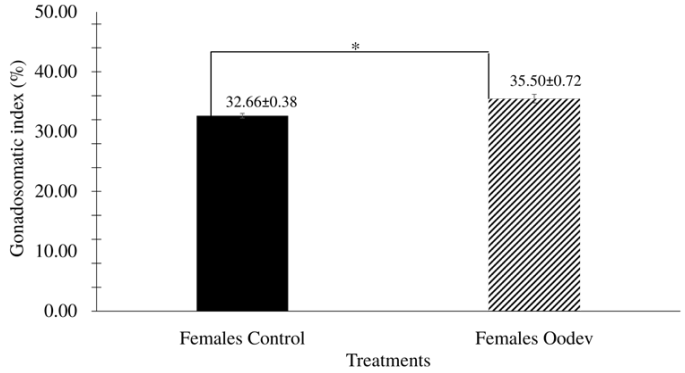

Figure 1. Gonosomatic index of *Nomorhampus* sp. females in Oodev® bioencapsulation treatment at a dose of 1 mL/ kg of broodstock weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.

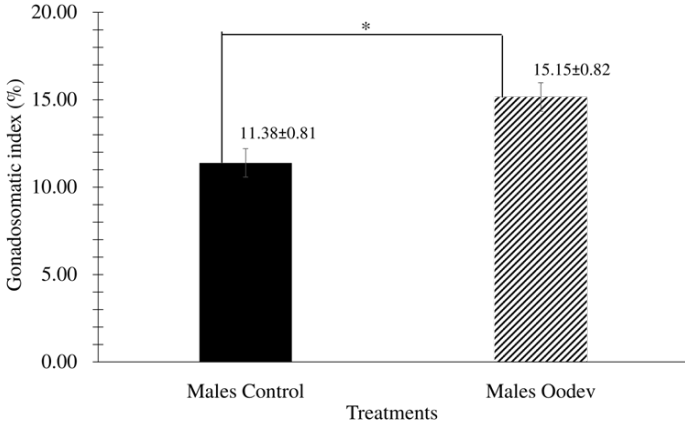

Figure 2. Gonosomatic index of *Nomorhampus* sp. males in the bioencapsulation treatment of Oodev® at a dose of 1 mL/ kg female weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.

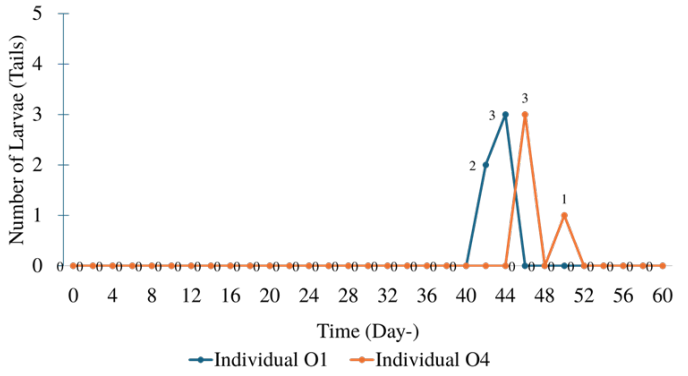

Figure 3. Birth interval and larval count *Nomorhampus* sp. In the bioencapsulation treatment of Oodev® at a dose of 1 mL/ kg parent weight using *Chironomus* sp. for 60 days.

within a birth duration of 10 minutes. Another birth occurred on the 46th day with the Oodev® test fish pair, repeat 4 (Individual O4), resulting in three offspring with a 10 minutes interval. In the control treatment, no births were observed; therefore, the data presented in the graph table only includes individuals that gave birth.

Gonad histology

The histological results from the non-birthing mothers showed that the Oodev® bioencapsulation treatment, administered at a dose of 1 mL/kg through *Chironomus* sp., led to uniform gonad development at the oogenesis stage and faster, more even spermatozoa development. In contrast, the control treatment also showed development but was uneven and slower.

Specific weight growth rate

The Oodev® bioencapsulation treatment, administered at a dose of 1 mL/kg of broodstock weight through *Chironomus* sp., can increase the specific weight growth rate, with the highest average value reaching 0.46% per day in female Oodev® fish and 0.40% per day in male Oodev® fish. Furthermore, the specific weight growth rate was analyzed using an independent t-test to determine whether there were significant differences between treatments. The test results confirmed a significant difference.

Specific length growth rate

The growth response of fish tested with Oodev® bioencapsulation treatment at a dose of 1 mL/kg of broodstock weight, administered through *Chironomus* sp., showed that the specific length growth rate did not differ significantly between treatments for both females and males. The highest average growth value was 0.11% per day in female Oodev® test fish and 0.12% per day in male Oodev® test fish. Furthermore, the specific length growth rate was analyzed using an independent t-test to determine whether there was a difference between treatments. The test results showed no significant difference between treatments (P value >0.05).

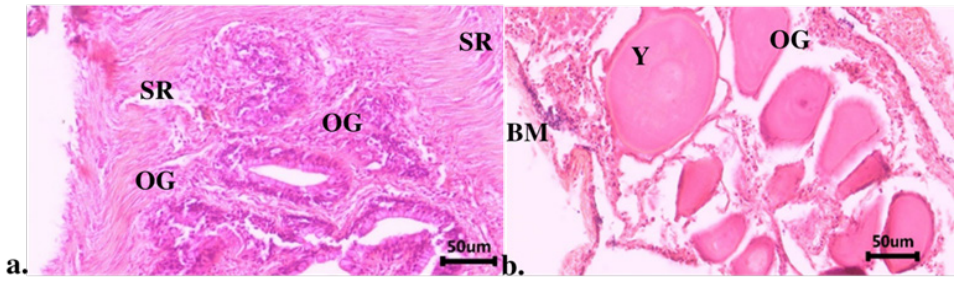

Figure 4. Female gonadal histology *Nomorhampus* sp. who did not give birth on the 60th day of the control treatment. (b) Female gonadal histology *Nomorhampus* sp. who did not give birth on the 60th day after the bioencapsulation treatment of Oodev® at a dose of 1 mL/kg weight through *Chironomus* sp.
Note: Basement membrane (BM), germinal epithelium (GE), nucleus (N), developing oocyte (OC), oogonium (OG), stroma (SR), yolk (Y), zona pellucida (ZP).

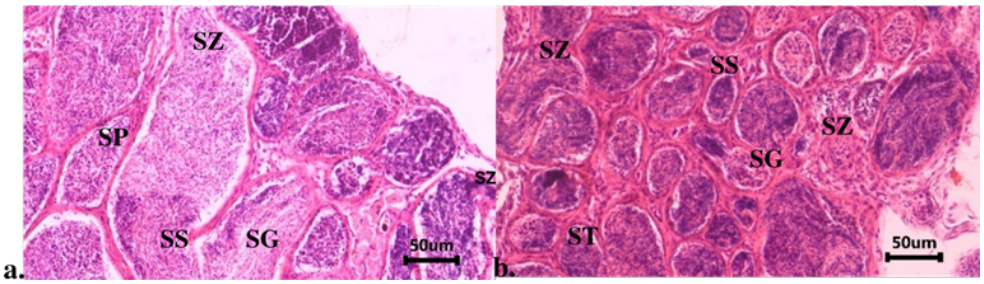

Figure 5. Male gonadal histology *Nomorhampus* sp. on the 60th day after the control treatment. (b) Male gonadal histology *Nomorhampus* sp. on the 60th day after bioencapsulation treatment of Oodev® at a dose of 1 mL/ kg parent weight using *Chironomus* sp.
Note: Spermatogonia (SG), primary spermatocytes (SP), secondary spermatocytes (SS), spermatids (ST), spermatozoa (SZ).

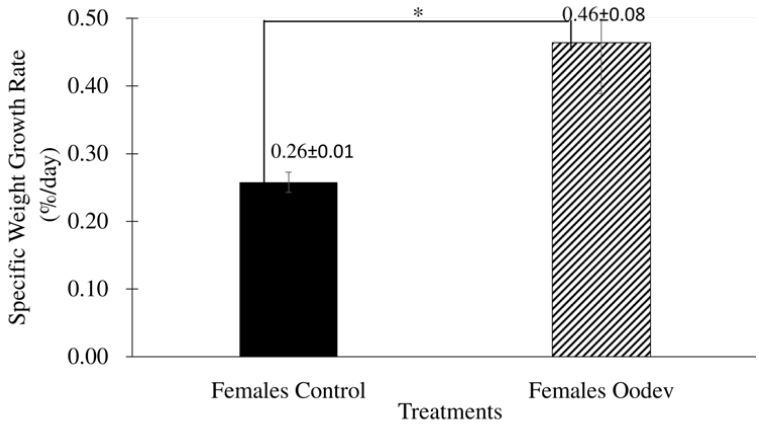

Figure 6. Specific weight growth rate of *Nomorhampus* sp. females in Oodev® bioencapsulation treatment at a dose of 1 mL/ kg brood weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.*) Significance effect at the α=0.05 level.

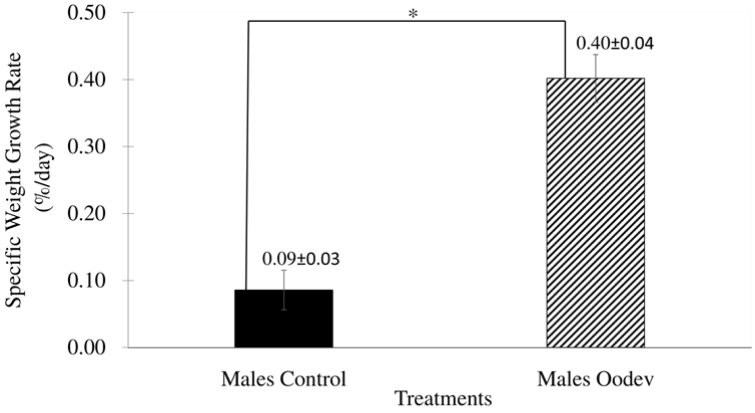

Figure 7. Specific weight growth rate of *Nomorhampus* sp. males in the bioencapsulation treatment of Oodev® at a dose of 1 mL/ kg female weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.*) Significance effect at the α=0.05 level.

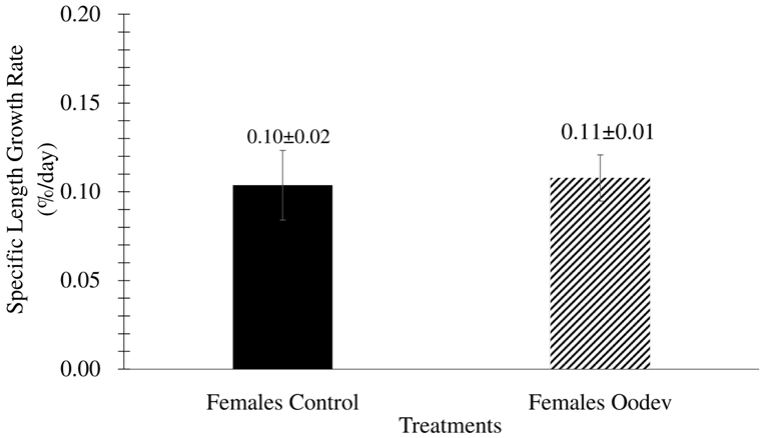

Figure 8. The growth rate of the specific length of the fish *Nomorhampus* sp. females in Oodev® bioencapsulation treatment at a dose of 1 mL/kg brood weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.

Survival rate (SR)

The survival rate of female and male anasa fish (*Nomorhampus* sp.) broodstock treated with Oodev® bioencapsulation at a dose of 1 mL/kg of broodstock weight through *Chironomus* sp. was 100%, with no significant difference between treatments.

Larval growth performance

The growth performance of anasa fish larvae (*Nomorhampus* sp.) from Oodev-treated broodstock was evaluated using a total of nine larvae, maintained for 30 days under controlled conditions and fed three times a day with natural foods, including *Artemia* and silk worms. The

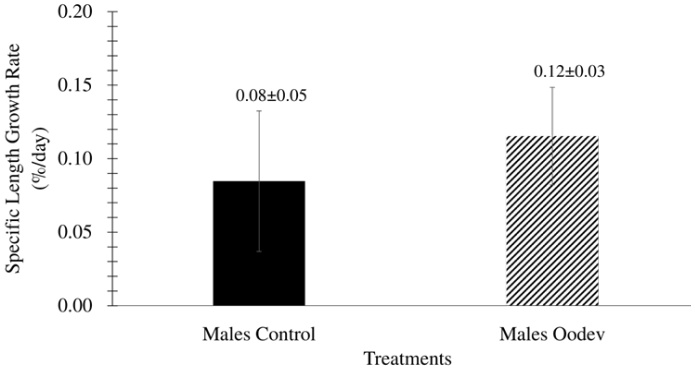

Figure 9. The growth rate of the specific length of the fish *Nomorhampus* sp. males in the bioencapsulation treatment of Oodev® at a dose of 1 mL/kg female weight using *Chironomus* sp. for 60 days.
Note: The values listed are the average ± standard deviation.

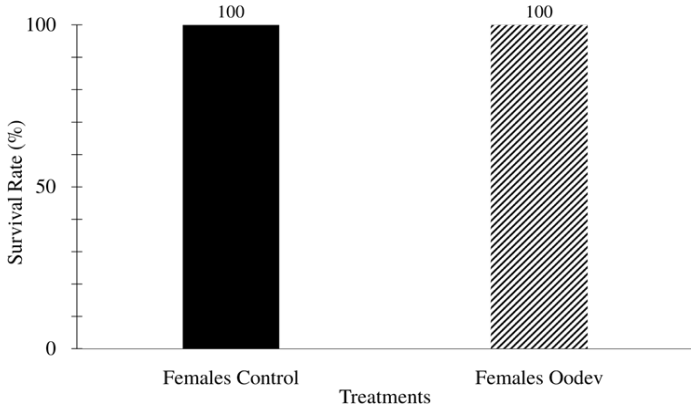

Figure 10. The survival rate of *Nomorhampus* sp. females in Oodev® bioencapsulation treatment at a dose of 1 mL/kg brood weight using *Chironomus* sp. for 60 days.

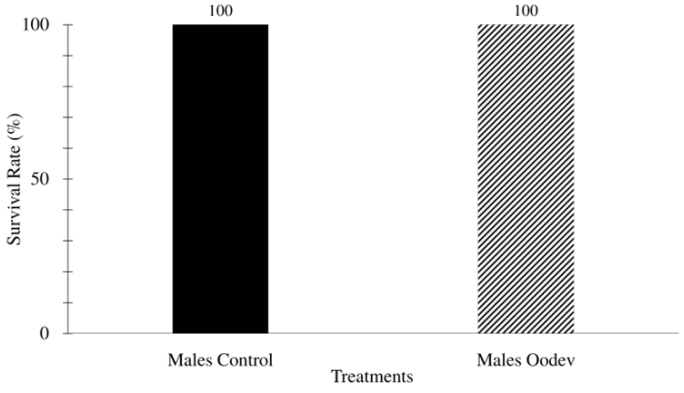

Figure 11. The survival rate of *Nomorhampus* sp. males in the bioencapsulation treatment of Oodev® at a dose of 1 mL/kg female weight using *Chironomus* sp. for 60 days.

results showed that the larvae exhibited good growth performance, with a SR of 100%, an SLGR of 0.7% per day, and an SWGR of 3.3% per day. Detailed larval growth data can be found in Table 1.

Discussion

The anasa fish (*Nomorhampus* sp.) is an endemic ornamental species with high potential. According to personal interviews, exporter demand in the Jabodetabek area reaches 1,000–1,500 fish per month, with prices ranging from IDR 15,000.00 to IDR 60,000.00 per fish. At the time this research was conducted, the supply of anasa fish was still sourced from natural catches, which poses a risk of overfishing and threatens the species' survival and resource sustainability. Therefore, domestication efforts are necessary to prevent extinction and support the development of aquaculture commodities. However, as of this study, domestication efforts for anasa fish had only progressed to the collection and adaptation stage in a controlled environment (Kraemer *et al.,* 2019).

This study focused on reproductive efforts to achieve the final stage of domestication. The reproduction of fish outside their natural habitat requires stimulation through environmental factors such as temperature, salinity, and the addition of specific materials (Kucharczyk *et al.,* 2022; Mylonas *et al.,* 2017). The administration of Oodev® via bioencapsulation in natural *Artemia* feed at a dose of 0.5 mL/kg has been successfully applied in seahorses, resulting in improved reproductive performance (Alaufa *et al.,* 2019). This success serves as the basis for using the bioencapsulation method in anasa fish (*Nomorhampus* sp.). Due to the limited availability of these fish, their small size, and high sensitivity to anesthesia, the bioencapsulation method is a more effective approach for reducing mortality risks and enhancing the success of hormone manipulation in *Nomorhampus* sp.

The gonadosomatic index value serves as a quantitative reference for describing changes used as indicators of gonad development and maturity in the reproductive process. This value increases in correlation with the growth in gonad weight and size in both male and female fish (Mukti *et al.,* 2020). The development of the gonadosomatic index in male and female test fish reflects the formation of egg cells in females and sperm in males. In this study, the gonadosomatic index value serves as an indicator that Oodev®, administered at a dose of 1 mL/kg of broodstock weight through *Chironomus* sp. feed, can accelerate the maturation process in anasa fish (*Nomorhampus* sp.). This effect is attributed to Oodev®'s composition, which includes a combination of Pregnant Mare Serum Gonadotropin (PMSG) and anti-dopamine (AD) (Susilo & Tuti, 2019).

Pregnant mare serum gonadothropin (PMSG) is a complex glycoprotein derived from pregnant mare serum that exhibits effects similar to luteinizing hormone (LH) and follicle-stimulating hormone (FSH), with FSH having a greater effect than LH. FSH and LH regulate oocyte maturation in female fish and control the mitosis phase in spermiogenesis and spermiation (Lestari *et al.,* 2016; Bertolini *et al.,* 2020; Mellisa *et al.,* 2019; Molés *et al.,* 2020). The FSH hormone in PMSG stimulates the hypothalamus to produce GnRH, while antidopamine blocks dopamine activity in the hypothalamus, leading to increased GnRH production. This, in turn, stimulates the pituitary

Table 1. Growth performance of *Nomorhampus* sp. fish larvae for 30 days.

Test Parameters	Average±Stdev
W_0 (g)	0.5 ± 0.07
W_{30} (g)	1.2 ± 0.07
P_0 (cm)	1.3 ± 0.04
P_{30}(cm)	2.0 ± 0.06
PP (cm)	0.7 ± 0.03
SLGR (%/day)	1.5 ± 0.06
SWGR (%/day)	3.3 ± 0.43
SR (%)	100 ± 0

Note: The number of larvae (n) was 9 individuals. The individual initial weight (W0); individual final weight (Wt); individual initial length (P0); individual final length (Pt); survival rate (SR); specific daily growth rate (SWGR); specific length growth rate (SLGR); and length growth (PP) were measured.

gland to synthesize more endogenous FSH, which acts on the gonads (Ahlina *et al.,* 2014; Caldas *et al.,* 2021; Swanson *et al.,* 2003; Zohar *et al.,* 2010). Consequently, hormone administration to anasa fish (*Nomorhampus* sp.) significantly influences spermatogenesis maturation in males and enhances oogenesis, ultimately leading to successful reproduction in female anasa fish.

Histological analysis of the gonads in test fish was conducted to determine the stages of gonad development microscopically. The results showed advancements in oogenesis maturation in female test fish, with gonads exhibiting faster and more uniform egg development. Test fish treated with Oodev® through bioencapsulation of *Chironomus* sp. displayed gonads in the vitellogenic formation phase, characterized by cytoplasm dominated by oil globules. Additionally, Oodev® treatment was associated with a predominance of the early vitellogenic phase, marked by nucleus development, which would become the center of the oocyte. In contrast, female test fish in the control group exhibited slower and uneven egg development, with no vitellogenesis process observed in several replicates.

Histological analysis of male test fish also revealed differences in gonad development. The spermatocyte stage in the Oodev®-treated fish showed a more dominant and synchronized spermatozoa phase, indicating a mature gonadal state (Nurhidayat *et al.,* 2017), whereas the control group remained in the secondary spermatocyte stage with fewer spermatozoa. Reproductive problems occur not only in female fish but also due to low sperm quality during spawning (Mehdi & Ehsan, 2011). Histological results show a direct correlation with the gonadosomatic index (GSI) value, where a higher GSI indicates increased gonad development as body weight increases (Hutagalung *et al.,* 2015).

A study on the maintenance and treatment of Oodev® bioencapsulation through *Chironomus* sp. for 60 days, using broodstock caught from the natural habitat of Lake Lindu, Central Sulawesi, showed positive results. The test fish exhibited a specific weight growth rate, as illustrated in the specific weight growth rate graph. This demonstrates that Oodev® has a significant effect on fish growth, aligning with previous research on various species that received Oodev® through feed applications, including Giru fish (Tomasoa *et al.,* 2018) and Peres fish (Rahayu *et al.,* 2021). Additionally, the weight growth in the test fish confirms that the provided feed was accepted and consumed. The specific length growth rate in test fish did not differ significantly between the treatments listed. This is related to the energy allocation in test fish that have become broodstock, as their growth focuses on gonadic development, causing slower somatic growth and resulting in no significant difference in specific length growth rates between treatments.

However, test fish given Oodev® exhibited a higher specific length growth rate than those in the control treatment. Weight gain in test fish is generally associated with gonadal maturation, suggesting that increased weight is also due to increased gonad weight (Sheridan, 2021). During the trial period, the test fish demonstrated a high survival rate, with no mortality from the beginning to the end of the study. This confirms that Oodev® bioencapsulation through *Chironomus* sp. does not affect the survival rate of anasa fish (*Nomorhampus* sp.). Growth performance data in Table 1 shows a larval survival rate of 100% over a 30-day maintenance period. The high survival rate indicates that Oodev® bioencapsulation through *Chironomus* sp. does not negatively affect larval quality, with PMSG playing a role in improving egg quality during development (Tahapari & Dewi, 2013).

The addition of hormones also serves to enhance both the quantity and quality of larvae (Nainggolan *et al.,* 2014). The study found that Oodev® bioencapsulation at a dose of 1 mL/kg of broodstock weight through *Chironomus* sp. positively affects the gonad maturity of anasa fish (*Nomorhampus* sp.), promoting spermatogenesis in males and oogenesis in females, leading to the production of healthy, growing larvae. This accelerates the domestication process and enhances the species' potential as a high-value ornamental fish, reducing the risk of extinction. Several studies have also demonstrated the positive effects of Oodev® supplementation through feed on gonad maturity in different fish species, such as peres fish (Rahayu *et al.,* 2021) and ornamental clownfish (Tomasoa *et al.,* 2018). Further research is needed to explore Oodev® bioencapsulation with various dose comparisons to determine the optimal dosage for anasa fish (*Nomorhampus* sp.).

CONCLUSION

The administration of Oodev® at a dose of 1 mL/kg of broodstock weight, with an average broodstock length of 5–6.5 cm, through

bioencapsulation in *Chironomus* sp., effectively accelerates the early reproduction of anasa fish (*Nomorhampus* sp.). This is evidenced by a 40% increase in births, with an average of 4–5 offspring per broodstock and a 100% survival rate in 30-day-old fry.

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