Sexual Dimorphism and Gonadal Observation of Short-finned Eel *Anguilla bicolor* Reared Under Control Condition in Indonesia

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Introduction

Figure 1: Short-finned eel, *Anguilla bicolor*

Anguilla bicolor, known as the Indonesian shortfin eel, is a unique catadromous fish found in Southeast Asia's freshwater ecosystems. It spawns in deep tropical waters, grows in freshwater and estuaries, then returns to the ocean to spawn again. With high demand for eel products in Asian markets, *A. bicolor* shows great potential for aquaculture due to its culinary value and market demand. Efforts to produce eel seeds focus on artificial egg collection, but obtaining mature males capable of releasing sperm remains challenging. This research aims **to understand gonad development in both male and female of *A. bicolor***, which is crucial for successful artificial seed production. These studies will provide information on *A. bicolor* gonad development, aiming to refine seed production techniques and ensure a reliable supply for eel farming. The ultimate goal is to meet market demands and support sustainable eel cultivation in Southeast Asia.

Materials and Methods

Ethical approval

This experiment was conducted per Universiti Malaysia Sabah's guidelines for animal care in scientific research.

Fish procurement

This study was conducted at Aquaculture Research Centre (ARC) of PT Suri Tani Pemuka, Banyuwangi, Indonesia. The A. ;'bicolor used were obtained from the wild from glass eel stage and cultured in control condition for 1.5 years.

Morphometric measurement, body indices and sex determination

- A total of 51 tails of *A. bicolor* specimens were carefully selected, consisting of 23 tails with body weight ranging from 100 to 200 g, 16 tails with body weight ranging from 200 to 500 g, and 12 tails weighing from 500 to 1000 g were selected.
- All specimens were sacrificed and dissected to determine the body indices and sex determination. Specimens were sacrificed by using iced water immersion method.
- Before dissection, body weight, BW (g), total length, TL (cm) and body round, BR (cm) were measured. Each fish was photographed with a ruler as reference scale. From the photograph recorded, standard length, SL (cm), body height, BH (cm), head length, HL (cm), and eye diameter, ED (cm) were measured using Image-J software.
- Other morphometric measurement features such as the presence of genital pore and genital papillae were also photographed. Biopsy analysis of gonad was done for all specimen to confirm the presence of eggs and sperm. This involved capturing microscopic images of gonadal tissue.

Figure 2: Research activities throughout the study.

Results

Morphology, body indices and sex determination

From 51 individuals of *A. bicolor,* the sex ratio of male to females was at **29:22**. From Figure 3, male was identified ranging from **96 to 300 g**. Meanwhile for female, body weight is ranging from **166 to 1950 g**. The graph shows goodness-of-fit, R2=0.9564 and negative allometry length-weight growth with b=3.2661 (Figure 4).

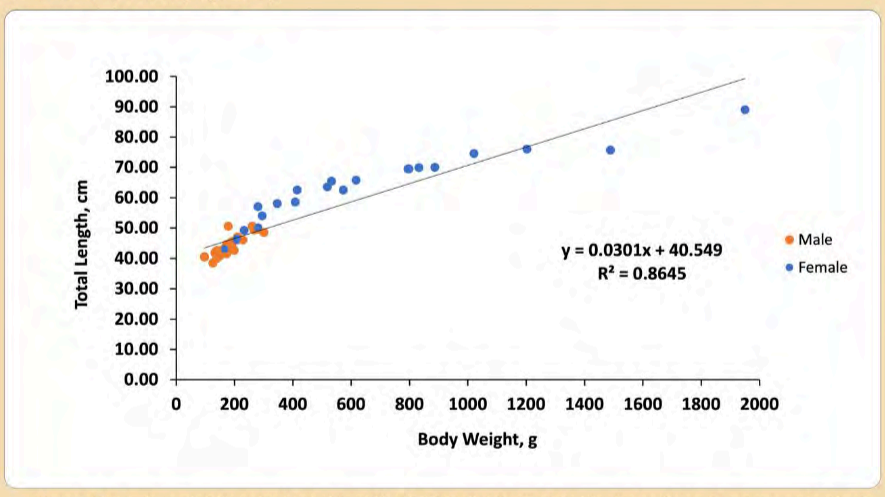

Figure 3: Body weight against total length of 51 individual of *A. bicolor* cultured under controlled condition in Indonesia.

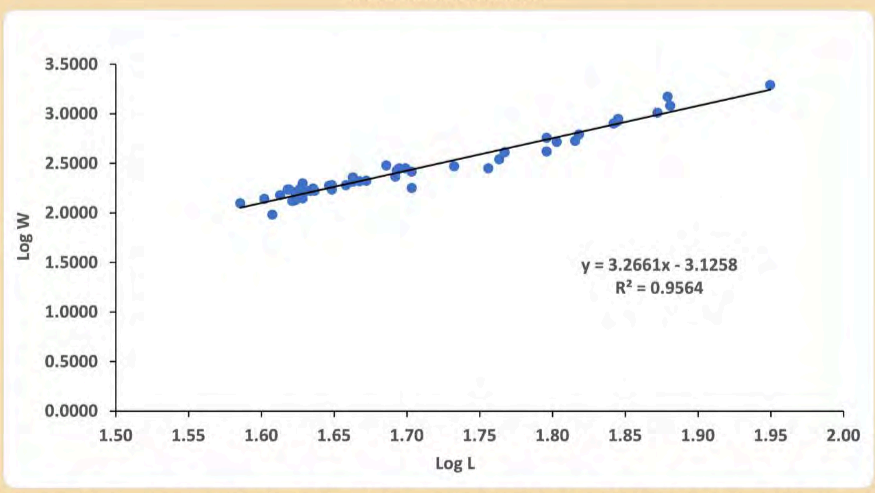

Figure 4: Regression equation for *A. bicolor* showing ln BW=3.2661, ln TL=3.1258 shows goodness-of-fit with R2=0.9564 when cultured under controlled condition in Indonesia.

A. bicolor exhibits **sexual dimorphism** (Figure 5), where males are generally smaller than females. This size difference is a key characteristic that distinguishes the sexes, even though their genitalia do not provide clear differentiation.

Figure 5: (A) Male and (B) Female of *A. bicolor* cultured in control condition for 1.5 year.

Gonad observation

Figure 5 shows visual comparison between the ovary and testis in *A. bicolor*, highlighting their respective positions within the body (Figure 6). In the male *A. bicolor* (Figure 6(B)), **presence of visceral fat was observed surrounding the testis**. The visceral fat appears as a thick, whitish layer enveloping the gonadal tissue.

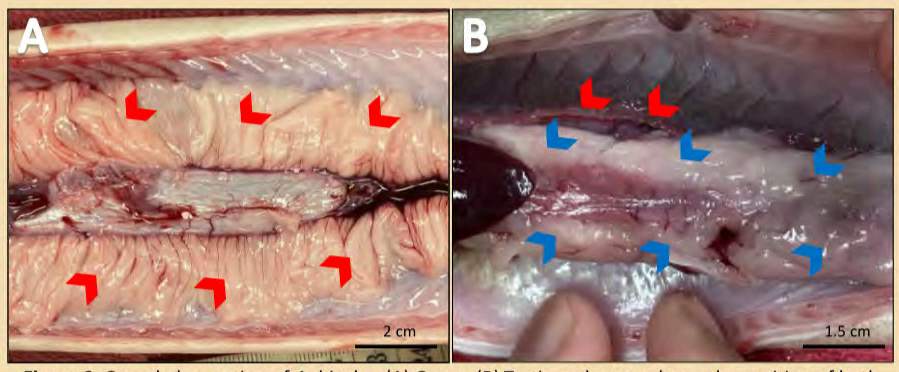

Figure 6: Gonad observation of *A. bicolor*. (A) Ovary; (B) Testis, red arrow shows the position of both ovary and testis in *A. bicolor*; blue arrow shows visceral fat near gonad.

Figure 7 shows microscopic images of gonads from both male and female *A. bicolor* at different BW. This study indicates that **male gonads do not develop significantly**, while **female gonads show slight development as body weight increases**.

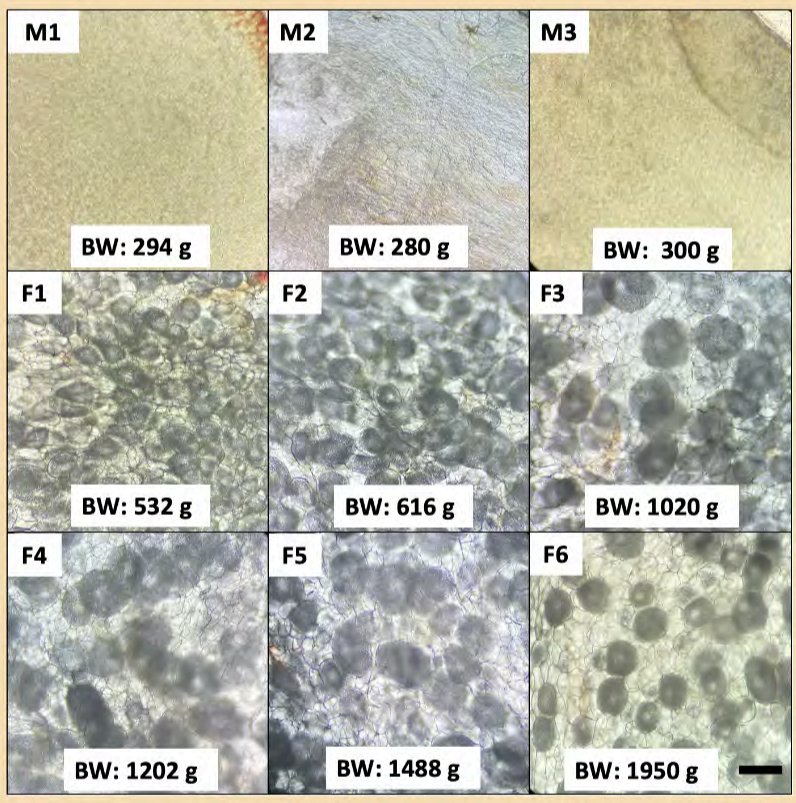

Figure 7: Photomicrographs of *A. bicolor* gonads at various range of BW. Males (M1-M3) and females (F1-F6) are shown. Scale: 50 µm.

Discussion

- All *A. bicolor* individuals weighing less than 200 g were males.
- Above 200 g, most of them were females but up to around 300g, there are possibility of existence of males.
- No males were observed over 400 g, suggesting individuals above this weight are females.
- Eels over 500 g were definitively female.
- These findings align with recent research indicating that female eels grow larger than males. (Davey *et al.*, 2005))

- Gonads of males were underdeveloped, making them unsuitable for broodstock purposes.
- The inability to use underdeveloped male gonads for egg collection is a significant challenge.
- Excess visceral fat in cultured fish, such as catfish, inhibits gonadal development; a similar issue affects *A. bicolor* males. (Zhou *et al.*, 2022)

- Strategies such as exercise or reduced feed intake might be necessary to manage visceral fat levels and promote gonadal development in male eels.

Conclusion

This study identified weight-based sexual dimorphism in *A. bicolor*, with males under 200 g and females over 400 g. However, underdeveloped male gonads present a challenge for breeding, highlighting the need for strategies to manage visceral fat and improve gonadal development in aquaculture practices.

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