

Captive breeding practices and insights on four indigenous *Betta* species of Sarawak

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ABSTRACT

Sarawak, located on the island of Borneo, is home to several indigenous *Betta* species, many of which face threats from habitat degradation. Despite their ecological and ornamental significance, limited information exists on their captive breeding potential. This study investigated the reproductive behavior and larval development of four native *Betta* species (*Betta brownorum*, *Betta ibanorum*, *Betta macrostoma*, and *Betta taeniata*) under controlled conditions. A total of 46 wild specimens were collected from four locations across Sarawak and subjected to species-specific breeding trials in aquaria. *B. brownorum*, a bubble-nester, exhibited nest-building and egg deposition behavior, with hatching observed within 3–4 days. In contrast, the three mouthbrooding species exhibited paternal incubation, which lasted 3–4 weeks. Larval development was monitored over time, showing steady increases in total length and distinct developmental stages were documented. These findings confirm the feasibility of captive breeding in all four species and highlight differences in reproductive strategy and larval growth patterns. The outcomes provide valuable baseline data for conservation planning and the development of ex-situ breeding programs for Sarawak's threatened *Betta* species.

1. Introduction

Sarawak is recognized as a biodiversity hotspot, home to several endemic *Betta* species, including *B. brownorum*, *B. ibanorum*, *B. macrostoma*, and *B. taeniata*. However, Kamal et al. [1] stated that the anthropogenic pressures, including deforestation, mining and agricultural expansion, threaten their natural habitats, underscoring the urgent need for conservation measures.

The genus *Betta* comprises over 70 species of freshwater fish distributed primarily in Southeast Asia [2]. According to Panijpan et al. [3], wild *Betta* species can adapt and thrive in various environments, including freshwater, brackish water and peat swamps. *Betta* species have a specialized respiratory system comprising a labyrinth organ, pseudobranch, and gills, enabling them to survive in environments with limited water availability Apriliani et al., 2019[4]. These species are renowned for their vibrant coloration, territorial behavior Lichak et al., 2022[5]. Fabanjo & Abdullah, 2021[6](2021) stated that *Betta*

fish hold significant economic value due to their vibrant body coloration and distinctive fin morphology, making them highly sought after in the ornamental fish trade. Despite being well-known in the ornamental fish trade, their wild population is threatened by habitat loss, pollution, and overfishing.

Betta fish have two reproductive strategies which include mouthbrooding and bubble nesting. According to Tan & Ng [7], *B. brownorum* is the only bubble nester while the other three (*B. ibanorum*, *B. macrostoma* and *B. taeniata*) are mouthbrooders. Paternal mouthbrooding involves the male parent placing eggs/larvae in the buccal cavity to protect the offspring from biotic and abiotic stresses Abecia et al., 2022[8]. Alternatively, the male 'nester' creates a 3D convex lens-shaped bubble nest for fertilized eggs and their offspring Panijpan et al., 2017[9]. These behavioral differences highlight the need for species-specific conservation strategies to ensure effective protection and management.

Captive breeding involves breeding wild animals to protect

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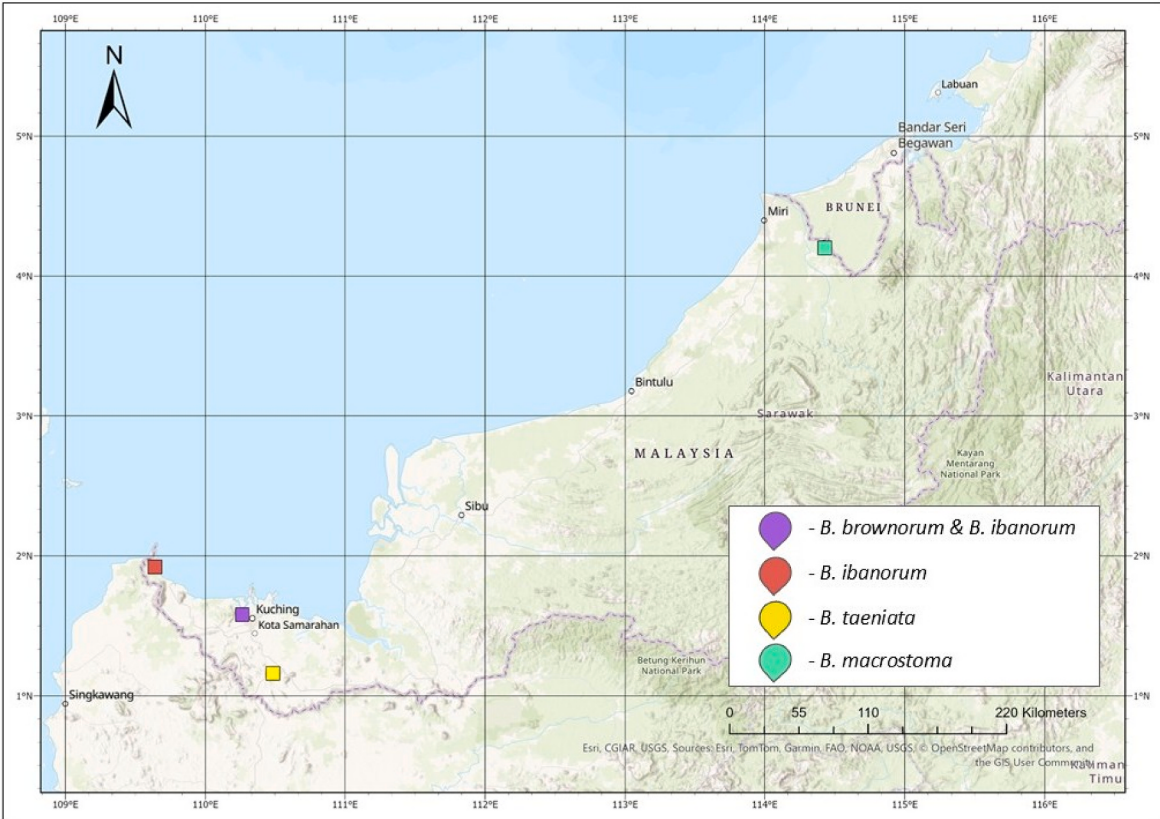


Fig. 1. Sampling locations for *Betta* species in Sarawak [7].

endangered species from extinction in their natural habitats Wakchaure & Ganguly, 2016[10]. Captive breeding programs have emerged as a critical tool for preserving threatened fish species. By providing a controlled environment that mimics natural conditions, researchers can study species-specific requirements, reproductive behaviors, and environmental tolerances. Captive breeding data can aid in decision-making for reintroductions, increasing the likelihood of success [11]. These insights are invaluable for designing conservation initiatives, particularly for species with limited wild populations.

This study aims to address these challenges by documenting the captive breeding requirements of *B. brownorum*, *B. ibanorum*, *B. macrostoma* and *B. taeniata*. Through detailed observations of breeding behaviors and reproductive outputs, this research seeks to provide a foundation for future conservation efforts. The findings highlight the potential of captive breeding as a complementary strategy for conservation, ensuring the long-term survival of Sarawak’s unique *Betta* species.

2. Materials and methods

2.1. Sampling periods and locations

From March 2022 to July 2023, *Betta* fish specimens were collected from four districts in Sarawak namely Kuching, Lundu, Serian, and Miri (Fig. 1).

A total of 46 individuals representing four species (*B. brownorum*, *B. ibanorum*, *B. macrostoma*, and *B. taeniata*) were sampled from four specific sites. Detailed descriptions of the sampling sites are presented in the supplementary section.

All collection activities were conducted under the Sarawak Forestry Corporation permit number SFC.810–4/6/1 (2022) – 005. Fish were captured using scoop nets or light fishing rods, depending on habitat accessibility and species behavior. The specimens were temporarily

Table 1
Morphometric measurement of *Betta* broodstocks.

Species	Total length (cm) Mean ± Standard Error (Range)		Body Weight (g) Mean ± Standard Error (Range)	
	Male	Female	Male	Female
<i>B. brownorum</i>	2.78 ± 0.15 (2.70–2.90)	2.12 ± 0.05 (2.00–2.3)	0.17 ± 0.07 (0.16–0.1)	0.13 ± 0.07 (0.08–0.18)
<i>B. ibanorum</i>	7.80 ± 1.66 (5.60–11.00)	8.93 ± 1.85 (6.80–10.50)	5.61 ± 3.10 (1.61–11.74)	7.50 ± 1.71 (3.98–11.32)
<i>B. macrostoma</i>	7.66 ± 0.34 (6.10–8.60)	7.42 ± 0.37 (5.90–8.70)	4.03 ± 0.51 (1.94–5.68)	3.72 ± 0.45 (2.57–5.62)
<i>B. taeniata</i>	6.44 ± 0.14 (6.00–6.90)	6.08 ± 0.24 (5.40–6.80)	2.71 ± 0.20 (2.31–3.36)	2.35 ± 0.21 (1.69–2.93)

stored in labeled plastic bags and transported to the Faculty of Resource Science and Technology (FRST), UNIMAS for further analysis.

2.2. Fish acclimatization and rearing

Upon arrival, the fish were quarantined for 14 days to examine their health and acclimate to confinement. Acclimatization involved gradually adjusting the water temperature by floating the transport bags in the rearing aquaria for approximately 10 min. The samples of male and female fish were raised in different aquariums.

Each tank was enriched with dried catappa leaves to imitate the species’ natural soft, tannin-rich blackwater habitat. Water and leaves were replenished weekly. Fish were fed grindal worms (*Enchytraeus buchholzi*) twice a day (0900 h and 1600 h) until satiation. There was no mortality during the acclimatization phase. The fish acclimatization was conducted based on Permana et al. [12] method.

The broodstock consisted of 10 *B. brownorum* (5 males, 5 females),

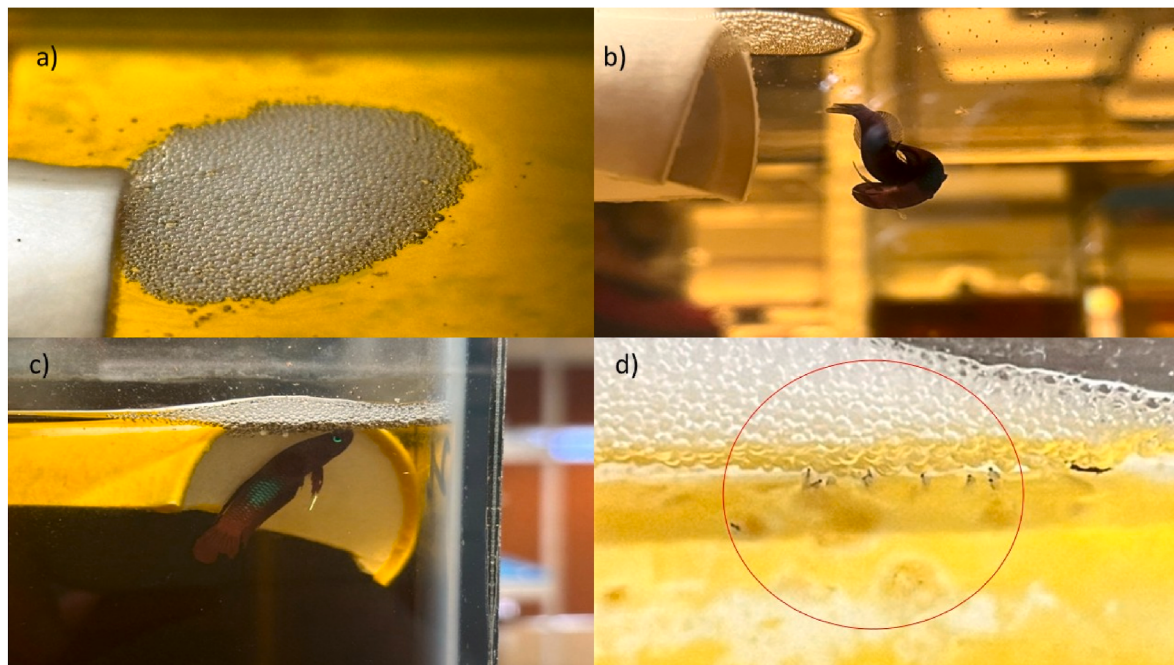


Fig. 2. Breeding behaviour of *B. brownorum*: a) Bubble-nest construction, b) *B. brownorum* during the spawning process, c) Male *B. brownorum* guarding the eggs, d) Hatched larvae hanging at the bubble nest.

12 *B. ibanorum* (6 males, 6 females), 12 *B. macrostoma* (6 males, 6 females) and 12 *B. taeniata* (6 males, 6 females). Individuals were sexed based on secondary sexual characteristics (fin morphology, coloration), measured for total length (TL), and their body weight was recorded. The TL and body weight of the broodstocks were recorded in Table 1.

Breeding trials were conducted according to the breeding strategies, which included mouthbrooding and bubble-nesting. Each pair with fully developed males and females was placed in the breeding tanks, along with dried catappa leaves, to achieve the soft, brown water condition that was favorable for spawning. Each breeding trial involved one pair of males and females per aquarium, replicated five times per species. For the bubble-nester (*B. brownorum*), a halved polystyrene cup was provided as a substrate for bubble nesting. For mouthbrooders (*B. ibanorum*, *B. macrostoma*, *B. taeniata*), breeding tanks were left unobstructed to allow natural spawning behavior.

2.3. Water quality monitoring

Water quality parameters were monitored throughout the acclimation, breeding, and larval rearing periods. The water temperature was maintained between 26 and 28 °C, with pH levels ranging from 5.5 to 6.5. Dissolved oxygen levels were maintained above 6 mg/L using sponge filtration. Partial water changes (50 %) were conducted every three days, with complete water replacement conducted weekly.

2.4. Larval development

After successful breeding, fertilized eggs and developing larvae were monitored daily. The larvae were fed with *Artemia* nauplii after they reached active feeding. For each species, 10 larvae were sampled to assess morphological changes and growth. Measurements of total length (TL) were taken using a stereomicroscope (OLYMPUS SZX7) connected to a computer with a measurement accuracy of ± 0.01 mm. The data are presented as means $\pm$ standard deviation (SD) unless otherwise specified.

Larval developmental stages were classified based on morphological features, such as yolk sac absorption, fin formation, pigmentation, and operculum development. Terminology follows Priyadi et al. [13],

wherein the term “larvae” refers to individuals from hatching until the completion of yolk sac absorption and initial fin development, while “juveniles” refers to individuals with fully developed fins, pigmentation, and functional lateral line systems.

3. Result and discussion

3.1. Habitat

46 *Betta* fish samples in total were successfully caught during the sampling process. *B. ibanorum* were found in peat swamps at the Matang area (Kuching) and the stream located by Sematan road (Lundu). *B. brownorum* was also found in peat swamps in the Matang area. *B. taeniata* were found in the stream located at Kampung Lachang in Serian. Lastly, *B. macrostoma* was found in peat swamps near Rumah Gadong in Marudi (Miri). The general descriptions of the habitat of the *Betta* species were documented in the supplementary section. Table 1 shows the morphometric measurements of *Betta* broodstocks.

3.2. Breeding behaviors

The tank setup for the breeding trials was based on their breeding methods (bubble nest and mouthbrooder). According to Tan & Ng [7], *B. brownorum* is a bubble nester while the others are mouthbrooders.

3.3. Bubble nesting species

B. brownorum exhibited typical bubble-nesting behavior with the male *B. brownorum* constructing buoyant nests using bubbles near the polystyrene cup. Courtship was a rapid process involving circling and embracing, after which the male would carefully deposit eggs within the nest. An average of 15 fertilized eggs was observed per breeding cycle in *B. brownorum*, from which 11 larvae were obtained. The males were responsible for nest maintenance, ensuring egg adhesion and regulating air circulation. Based on the observation, the eggs placed in the bubble nest began to hatch after three to four days. The hatched larvae were noticed to be hanging at the bubble nest. Fig. 2 illustrates the *B. brownorum*'s behaviors.

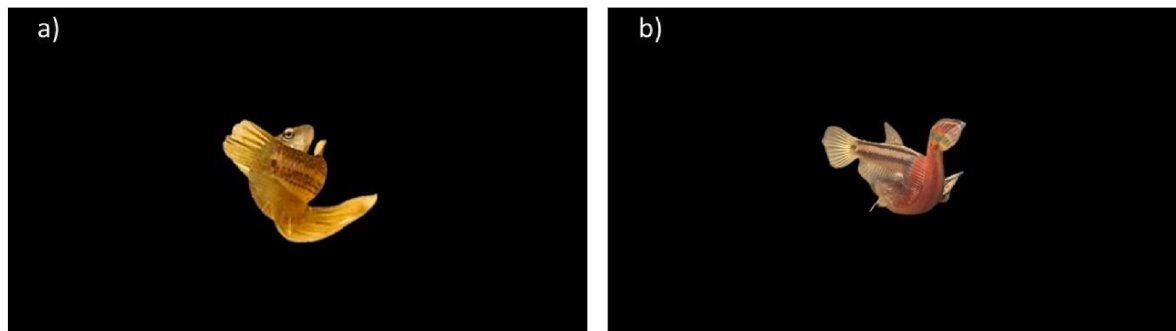


Fig. 3. a) *B. ibanorum* during the spawning process, b) *B. macrostoma* during the spawning process.

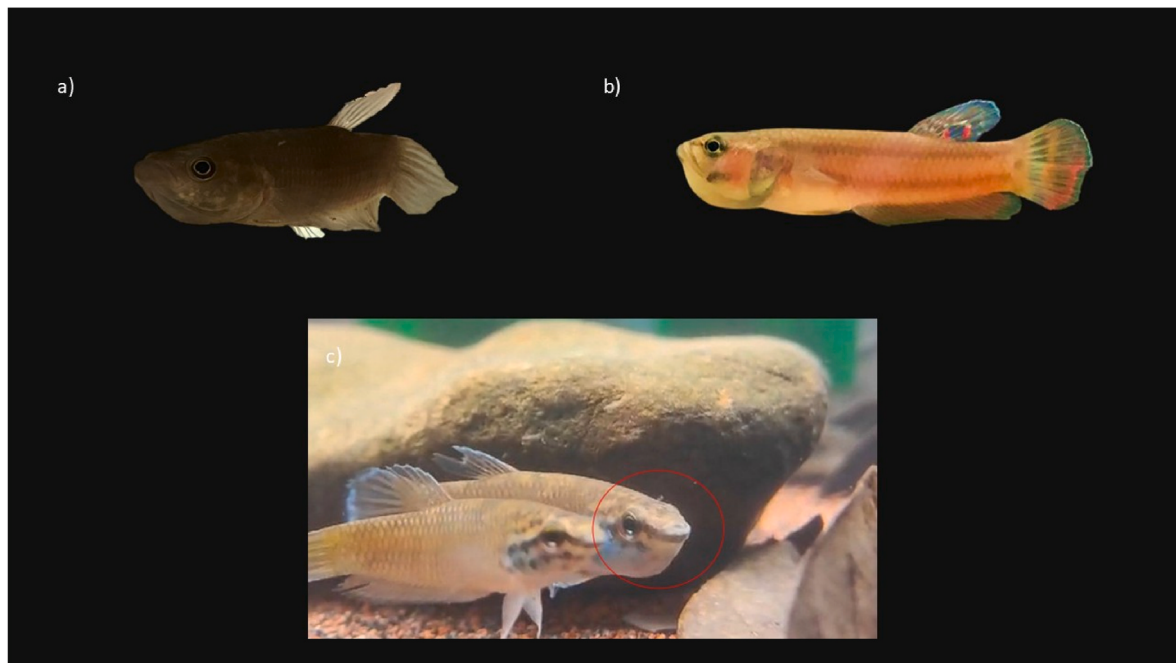


Fig. 4. a) *B. ibanorum* during mouthbrooding, b) *B. macrostoma* during mouthbrooding, c) *B. taeniata* during mouthbrooding.

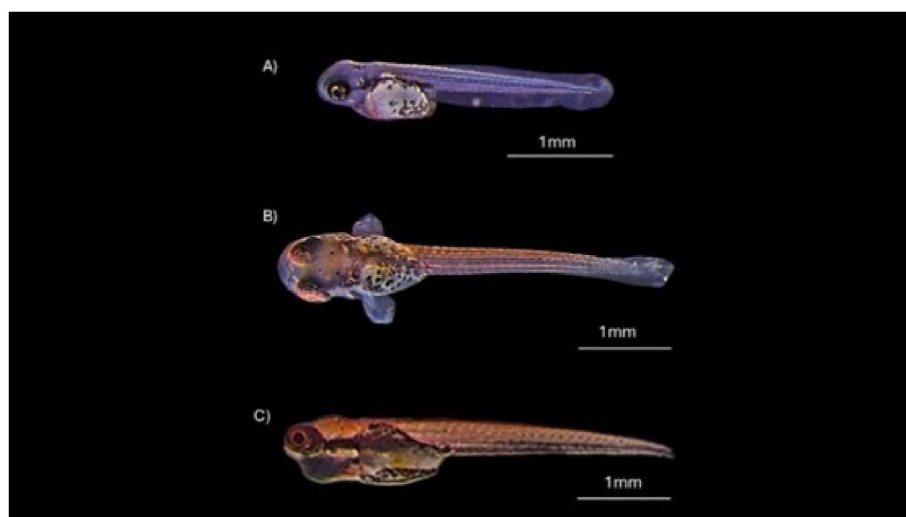


Fig. 5. The newly hatched *B. brownorum* was observed under the microscope. A) Larvae *B. brownorum* Day 1 under microscope (2.5 $\times$), B) Top view of larvae *B. brownorum* Day 3 under microscope (1.5 $\times$), C) Side view of larvae *B. brownorum* Day 3 under microscope (1.5 $\times$).

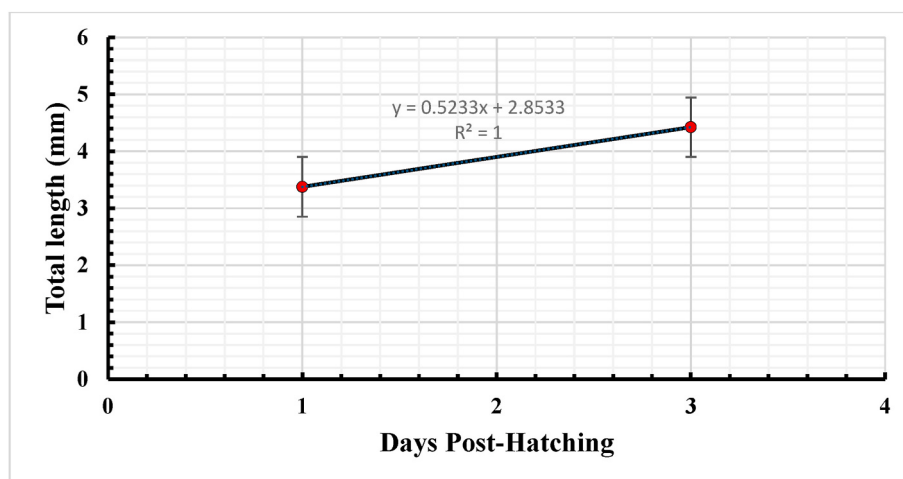


Fig. 6. Growth of *B. brownorum*'s larvae under captive conditions. The graph shows mean total length (TL) over time post-hatching. A linear regression line ($y = 0.5233x + 2.8533$, $R^2 = 1$) is shown, $n = 10$ larvae per timepoint.

3.4. Mouthbrooding species

B. ibanorum, *B. macrostoma*, and *B. taeniata* displayed mouthbrooding behavior. Based on observation, both female and male individuals of *B. ibanorum* and *B. macrostoma* entangle with each other during the spawning process, exhibiting courtship behavior (Fig. 3). Despite the lack of photographic evidence for *B. taeniata*, its courtship was presumed to be in the same pattern of *B. ibanorum* and *B. macrostoma*. During the spawning, the female *Betta* released eggs and the male *Betta* incubated the eggs in their mouth. The incubation lasted for three to four weeks. An average of 36 fertilized eggs was observed per breeding cycle in *B. ibanorum* (29 larvae were obtained), 14 fertilized eggs for *B. taeniata* (11 larvae were obtained) and 17 fertilized eggs for *B. macrostoma* (13 larvae were obtained). Fig. 4 illustrates *B. ibanorum*, *B. macrostoma*, and *B. taeniata* during mouthbrooding.

3.5. Larval development

3.5.1. *Betta brownorum*

The early development of *B. brownorum* larvae was monitored to assess their growth post-hatching. On Day 1, observations revealed that the larvae still retained and consumed their yolk sacs, which served as their primary source of nutrition, as supported by Permana et al. [12] and Riddle & Hu [14]. This is a critical stage as the yolk provides essential nutrients required for the initial development of the larvae, which supports their growth until they can transition to external food sources (*Artemia* nauplii), as evidenced by the increment in total length by the third day.

By the third day, the larvae had transitioned to external feeding by consuming *Artemia* nauplii. This shift to an external food source supports continued growth and development. The retention of yolk sacs on the first day is a typical characteristic of *B. brownorum* larvae indicating a developmental phase where endogenous resources are crucial for sustaining early growth. The introduction of *Artemia* nauplii on the third day provides a rich source of nutrients, further promoting larval growth and development as suggested by Refs. Policar et al., 2007[15]. Fig. 5 shows the larval development of *B. brownorum*.

The total length of *B. brownorum*'s larvae was measured over two days to observe growth patterns. The results indicate an increase in the larval length from Day 1 to Day 3. The average length of the larvae increased from 3.38 mm to 4.42 mm. This value was close to 3.30 mm on Day 1 post-hatching, reported by Ref. [16]. This demonstrated a notable growth in the larvae over the two days. A positive linear relationship was observed between days post-hatching and the TL of *B. brownorum*'s

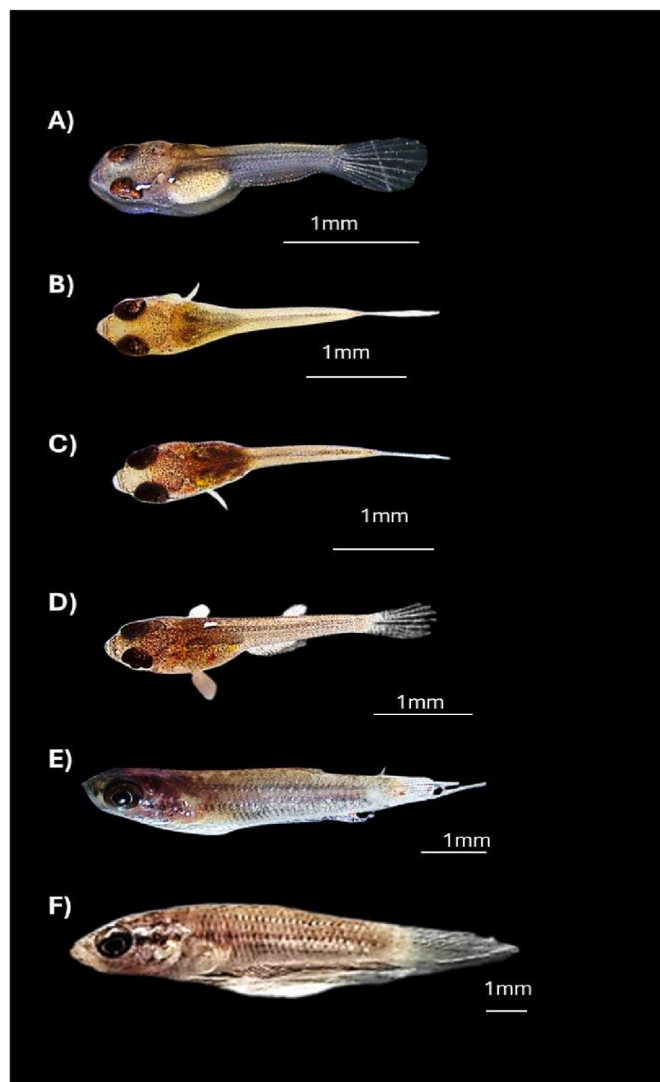


Fig. 7. *B. ibanorum* larvae under microscope: A) Day 7 (2.5×), B) Day 14 (2.5×), C) Day 21 (2.5×), D) Day 28 (2.5×), E) Day 120 (preserved, 0.8×), F) Day 150 (preserved, 2.5×).

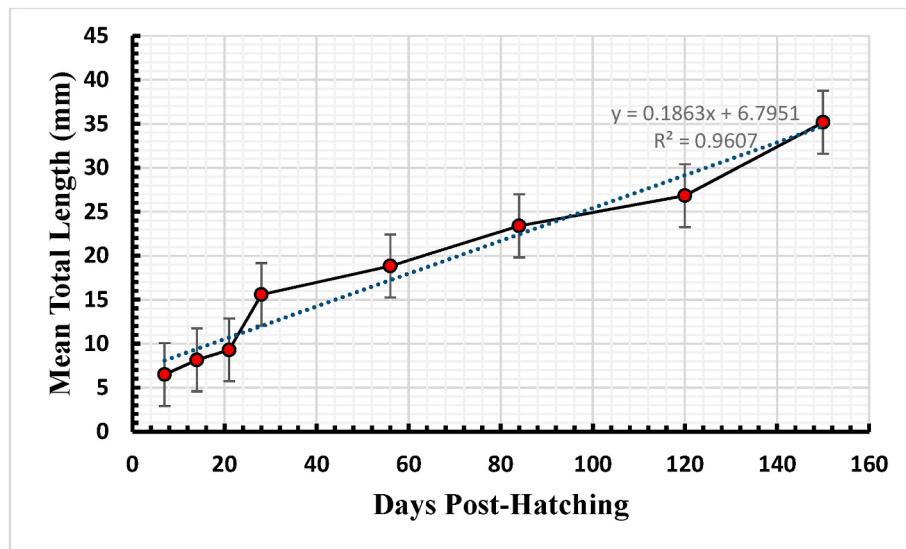


Fig. 8. Growth of *B. ibanorum*'s larvae under captive conditions. The graph shows mean total length (TL) over time post-hatching. A linear regression line ($y = 0.1863x + 6.7951$, $R^2 = 0.9607$) is shown, $n = 10$ larvae per timepoint.

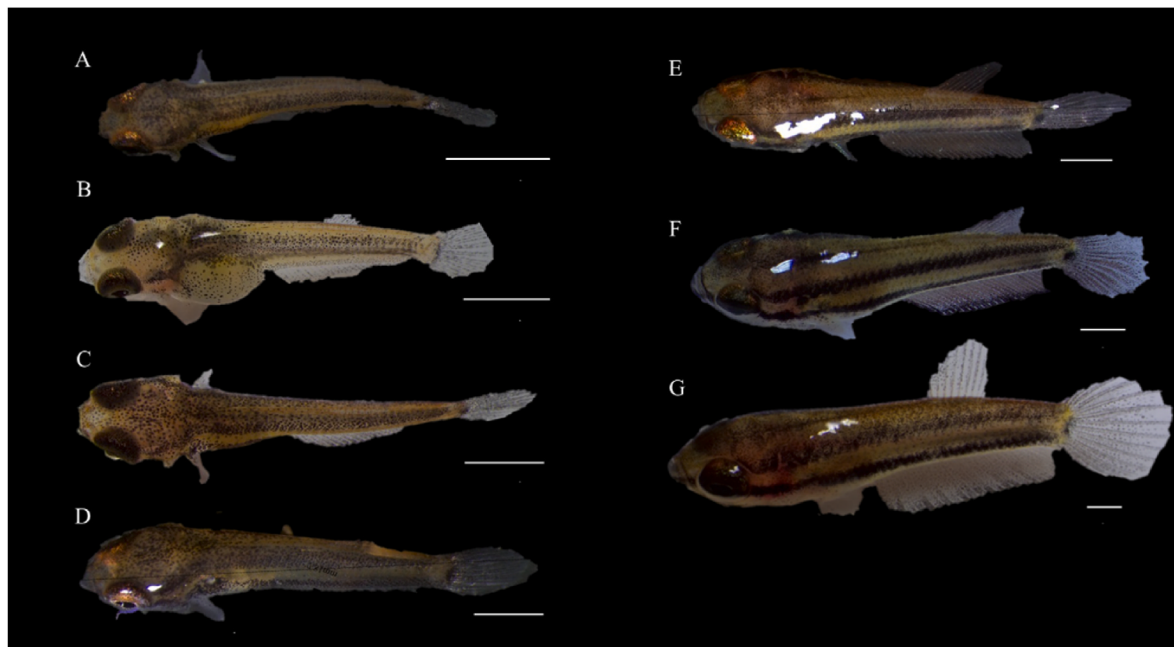


Fig. 9. Documentation of larvae *B. macrostoma*. A) Newly hatched *B. macrostoma*. B) Flexion stage of *B. macrostoma*. C) Post-flexion stage of *B. macrostoma*. D) End of the post-flexion stage. E) Pre-juvenile stage. G) Juvenile stage. Scale bar = 1 mm.

larvae, as indicated by regression analysis $y = 0.5233x + 2.8533$ as shown in Fig. 6.

3.5.2. *Betta ibanorum*

The larval development of *B. ibanorum* is illustrated in Fig. 7. In Stage A (newly hatched larva) from Fig. 7, the occiput, eye, mouth, yolk sac, primordial fin, notochord, anal fin and tail fin were present. The yolk sac was notably large, indicating a reliance on yolk for nutrition during early development. By Stage B (preflexion stage), the yolk sac had diminished, and pectoral fins began to develop, suggesting a transition to more active feeding behaviors. In subsequent stages, significant morphological changes included the development of the swim bladder in Stage C (preflexion stage), allowing for improved buoyancy control. The dorsal fin became more defined in Stage D (flexion stage),

contributing to better maneuverability. In Stage E (post-flexion stage), the operculum was formed, enhancing respiratory efficiency and by Stage F (juvenile stage), the lateral line system was visible.

The observed developmental stages of *B. ibanorum* align with previous studies on *B. rubra* Priyadi et al., 2024 [12]. Although certain adaptations, such as the rapid development of the operculum, suggest unique evolutionary responses to their specific habitats in Northwestern Borneo. The formation of the lateral line in Stage F highlights the importance of sensory adaptations in predator detection and social interactions Mogdans, 2019[17].

According to the graph in Fig. 8, the average growth measurements of *B. ibanorum*'s larvae showed a continuous increase from Day 7 to Day 150. Starting from an average of 6.49 mm on Day 7, the measurements rose steadily to 35.17 mm by Day 150. This progressive increase

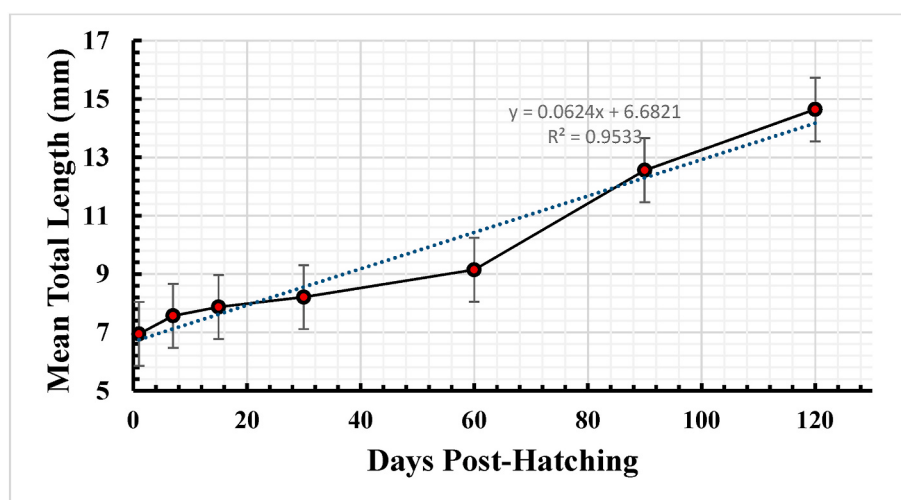


Fig. 10. Growth of *B. macrostoma*'s larvae under captive conditions. The graph shows mean total length (TL) over time post-hatching. A linear regression line ($y = 0.0624x + 6.6821$, $R^2 = 0.9533$) is shown, $n = 10$ larvae per timepoint.

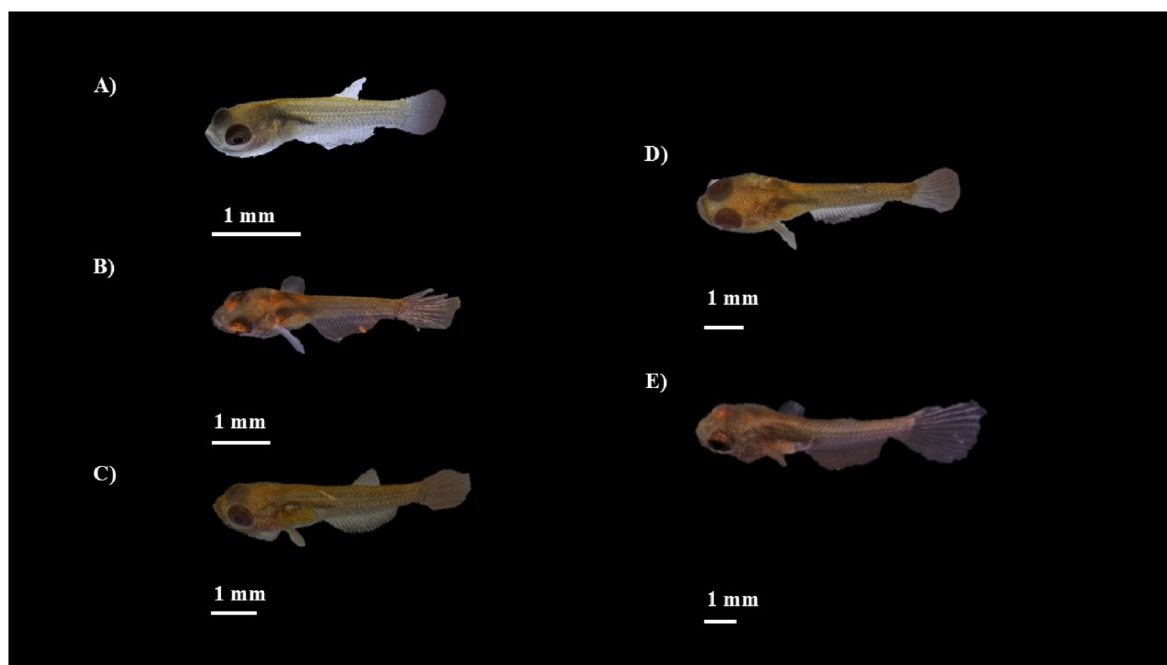


Fig. 11. Documentation of larvae *B. taeniata*: A) Newly hatching of *B. taeniata*, B) Flexion stage, C) End of flexion stage, D) Post-flexion stage, E) End of Post-flexion stage.

indicates significant growth over the 150 days. A positive linear relationship was observed between days post-hatching and the TL of *B. ibanorum*'s larvae, as indicated by regression analysis $y = 0.1863x + 6.7951$ in Fig. 8.

3.5.3. *Betta macrostoma*

Fig. 9 depicts the larval development of *B. macrostoma*, with gradual morphological changes from early to late stages. In the early stages (A-D), the larvae have a spherical body with underdeveloped fins, little coloration and remains of the yolk sac, which provides important nutrition before active feeding begins. As development advances (E-G), the body lengthens, fins become more prominent and distinct pigmentation patterns. There was also the emergence of its stripes. These alterations represent the transition from yolk-dependent larvae to free-swimming juveniles, indicating the species' growth and adaptation for

independent survival.

Fig. 10 shows that the average growth measurement of *B. macrostoma*'s larvae increased steadily from 6.94 mm (Day 1) to 14.64 mm (Day 120). This progressive increase shows significant growth over the span of 120 days. This steady growth, beginning at an average length of 6.94 mm on Day 1 and extending to 14.64 mm by Day 120, demonstrates the species' significant growth potential under experimental conditions. These results are in line with the study done by Priyadi et al. [13] on *B. rubra*, which also exhibits continuous growth over prolonged periods. A positive linear relationship was observed between days post-hatching and the TL of *B. macrostoma*'s larvae, as indicated by regression analysis $y = 0.0624x + 6.6821$ as shown in Fig. 10.

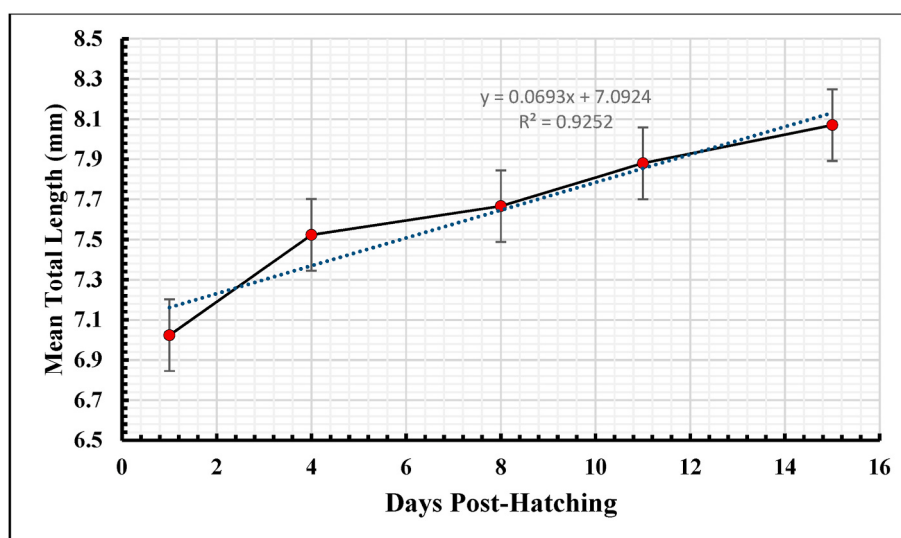


Fig. 12. Growth of *B. taeniata*'s larvae under captive conditions. The graph shows the mean total length (TL) over time post-hatching. A linear regression line ($y = 0.0693x + 7.0924$, $R^2 = 0.9252$) is shown, $n = 10$ larvae per timepoint.

3.5.4. *Betta taeniata*

The larval development of *B. taeniata* goes through different morphological changes, as illustrated in Fig. 11. In the early stage (A), the larva appears translucent with underdeveloped fins, huge eyes and little pigmentation, which indicates that it is still in the early post-hatch phase. As growth progresses (B-C), the pigmentation deepens, the fins begin to expand and the body structure becomes more defined, indicating a shift towards greater mobility and feeding capabilities. In the latter stages (D-E), the larvae have well-formed fins, increased colouring patterns and more prominent eyes that resemble juveniles. These gradual modifications, such as fin development, body elongation and enhanced pigmentation, demonstrate the species' adaptation to independent survival.

The growth performance of *B. taeniata*'s larvae was shown in Fig. 12. On day 1, the measurement of the larvae was 7.02 mm. The growth performance rose steadily to 8.07 mm on day 15. This study aligns with the findings of Priyadi et al. [13] on *B. rubra* where it also showed a progressive growth. The growth performance of *B. taeniata* shows significant potential for captive breeding under controlled environments. A positive linear relationship was observed between days post-hatching and the TL of *B. taeniata*'s larvae, as indicated by regression analysis $y = 0.0693x + 7.0924$, as shown in Fig. 12.

The observed survival rate of larvae in this study ranged from Day 3 to Day 150, suggesting variability in post-hatching viability across species and developmental stages. One possible factor influencing survival outcomes is the use of *Artemia* nauplii as the primary feed during the early active feeding stage. While *Artemia* is widely used due to its availability and general nutritional adequacy, it originates from marine environments and may not be optimal for freshwater larvae such as *Betta* spp. Dhont & Van Stappen, 2003[18]. In contrast, freshwater live feeds like *Moina* and *Daphnia* are ecologically compatible and offer several advantages, including higher digestibility due to softer exoskeletons, natural motility that stimulates feeding behavior, and superior profiles of essential fatty acids such as EPA and DHA, which are crucial for larval growth and immune development Bhosle et al., 2020[19]. Moreover, *Artemia* nauplii, if not consumed rapidly, may contribute to water quality deterioration due to uneaten biomass and molted exoskeletons, which decompose and increase ammonia concentrations Hamre et al., 2013[20]. In small rearing systems with limited filtration, such fluctuations in water quality can lead to stress and higher mortality in sensitive larvae. Therefore, integrating freshwater live feeds like *Moina* or *Daphnia* into the feeding regime may enhance larval survival by

providing nutritionally appropriate prey and maintaining better water quality, and is recommended for future *Betta* breeding protocols.

4. Conclusion

This study focuses on the successful captive breeding of four indigenous *Betta* species in Sarawak (*B. brownorum*, *B. ibanorum*, *B. macrostoma* and *B. taeniata*), illustrating the value of controlled settings in promoting reproduction and early development. The breeding behaviour and larval development were also documented. The larval development of all four species showed progressive growth, indicating significant potential for captive breeding. Captive breeding not only provides significant insights into species-specific requirements, but it is also a viable conservation approach for mitigating the effects of habitat loss and overexploitation. Future studies should focus on improving breeding methods, determining long-term survival rates and investigating reintroduction options to help natural populations. This information served to be the baseline to help Sarawak's unique *Betta* species be managed and preserved more sustainably by combining captive breeding with larger conservation activities.

Ethical Statement

All research activities involving *Betta* species were conducted in compliance with the guidelines of the Faculty of Resource Science and Technology, Universiti Malaysia Sarawak (UNIMAS) for the care and use of animals in research. Fish sampling was carried out under a permit issued by the Sarawak Forestry Corporation (Permit No. SFC.810-4/6/1 (2022)-005). No endangered or protected species were harmed, and all specimens were handled in a manner that minimised stress and mortality.

CRediT authorship contribution statement

Ahmad Syafiq Ahmad Nasir: Writing – review & editing, Validation, Supervision, Methodology, Investigation, Formal analysis, Conceptualization. **Mohd Armeen Zulkanaini:** Writing – original draft, Formal analysis. **Jeffery Mahmud:** Writing – original draft, Formal analysis. **Ruhana Hassan:** Writing – review & editing, Validation, Supervision, Methodology, Investigation, Formal analysis, Conceptualization. **Mohamad Faizul Mat Isa:** Investigation, Resources. **Samsur Mohamad:** Validation, Investigation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.repbre.2025.08.002>.

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