



# Maejo International Journal of Energy and Environmental Communication

Journal homepage: <https://ph02.tci-thaijo.org/index.php/MJEEC>



## ARTICLE

### Advancing spotted babylon broodstock performance and sustainable aquaculture practices through natural and formulated feeds

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#### ARTICLE INFO

##### Article history:

Received 09 February 2023

Received in revised form

28 February 2023

Accepted 04 March 2023

##### Keywords:

Spotted babylon

Broodstock feed

Natural feeds

Formulated feed

Growth performance

#### ABSTRACT

In the approach to optimize spotted babylon (*Babylonia areolata*) broodstock nutrition within hatchery environments, two distinct experiments were conducted. The first focused on broodstock, having an average weight of 43g. They were exposed to various natural feeds, with yellow stripe trevally being a notable inclusion, and they were kept at a density of 50 snails/m<sup>2</sup>. The following phase centered on juveniles, assessing the impact of replacing fishmeal with beef scraps and bonemeal in different proportions: 0%, 10%, 20%, and 30%. Yellow Stripe Trevally meat was used as a comparative control diet. These juveniles were kept at a density of 180 snails/m<sup>2</sup>. Both experimental groups were housed in standardized tanks, benefiting from a consistent 12-hour seawater supply daily. Through diligent monitoring, we observed that the trevally diet led to marked improvements in the growth and reproductive performance of the broodstock. On the other hand, for the juvenile group, the traditional diet was superior up to the 20% fishmeal substitution level. However, the growth rate diminished notably with a 30% substitution. Across the board, there was a consistent survival rate among all experimental groups. In sum, this study's findings underscore the effectiveness of yellow stripe trevally as a potent feed for broodstock. Additionally, when considering juvenile diets, fishmeal substitution with beef and bonemeal should be approached cautiously, ideally at most 20%.

## 1. Introduction

The aquaculture of spotted babylon, *Babylonia areolata*, in Thailand faces challenges, chiefly due to the inadequate seed supply and elevated production costs. Ensuring the successful conditioning of *B. areolata* broodstock is pivotal, especially for selective breeding programs (Hall et al., 2017). Given the increasing industrial significance of this species in Thailand, the

objective is to consistently yield a substantial number of high-quality eggs and larvae. Notably, considerable inconsistencies in spawning events, hatchability, and survival rates of larvae and juveniles of spotted babylon occur during the same season, even between different batches and hatcheries (Chaitanawisuti et al., 2011). This inconsistency prevails despite efforts to standardize the rearing conditions, such as managing larval density, controlling water quality, and employing specific microalgal species for

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feeding. Unfortunately, as demonstrated by Chaitanawisuti and Kritsanapuntu (1997), the results remain unpredictable with larvae quality fluctuating significantly.

A major determinant of this inconsistency is the variable quality of eggs and larvae. Factors influencing these qualities are manifold, some of which are inherent (like genotype, broodstock age and size, and egg size), while others are external, encompassing egg management, broodstock nutrition, and bacterial activity on the egg surface (Ballestrazzi et al., 2003). For many teleosts, certain nutrients - proteins, fatty acids, vitamin E, ascorbic acids, and carotenoids - play pivotal roles in reproductive processes such as gonadal maturation, gamete viability, and spawning efficacy (Fu et al., 2018). However, how these nutrients interact with reproductive processes has yet to be fully elucidated.

Multiple studies emphasize the significant impact of the type and amount of dietary lipid on the reproductive efficiency of broodstock. For instance, Ling et al. (2006) point out the potential benefits of particular dietary lipids. Similarly, Bautista-Teruel et al. (2001) found that enriching an artificial diet with crucial nutrients like proteins, lipids, and specific fatty acids enhanced the reproductive performance of abalones. Utting and Millican (1997) further underscored that the lipid content and quality of microalgal dietary supplements deeply influence egg production and fatty acid composition in marine bivalves. While it is accepted that lipids and particularly PUFA reserves like eicosapentaenoic acid play a crucial role in developing embryos and larvae (Sangsawangchote et al., 2010), the exact nutrients triggering maturation and egg production in broodstock remain unidentified, emphasizing the need for deeper investigation.

Surprisingly, given their commercial importance, there needs to be more in-depth research examining the influence of nutrition on the reproductive efficiency of spotted babylon broodstock. Recognizing this research gap, our study aspires to pioneer the understanding of dietary impacts on spotted babylon (Figure 1) broodstock development. In this study, we aim to discern the effects of both natural and formulated feeds on the growth and reproductive performance of *B. areolata*. The insights garnered will serve as foundational guidelines to curate optimized feeds tailored for the broodstock of this species.

## 2. Material and methods

### 2.1 Experimental designs

Experiment 1: The research adopted a completely randomized design (CRD) structure, comprising 4 treatments, each with 3 replications (Figure 2). The primary objective was to identify viable fresh food substitutes for yellow stripe trevally. The treatments were structured as follows:

Treatment 1: Green mussel (*Perna viridis*) meat.

Treatment 2: Blue swimming crab (*Portunus pelagicus*) meat.

Treatment 3: Pacific white shrimp (*Litopenaeus vannamei*) meat.

The yellow stripe trevally fish (*Selaroides leptocephalus*) served as the control group for comparison.

Experiment 2: This study also embraced a CRD, but with 5 treatments, each replicated thrice. The intent was to evaluate formulated feeds that incorporated beef scraps and bonemeal as

potential fish meal alternatives. The broodstocks received formulated feeds based on the varying extents of fish meal substitution using beef scraps and bonemeal at intervals of 0%, 10%, 20%, and 30%, labeled as BSB0, BSB10, BSB20, and BSB30, respectively. For reference, yellow stripe trevally was utilized as the control.



Figure 1. Spotted babylon broodstock (in experiment 1)

### 2.2 Feed formulation

Experiment 1 (Flesh feed preparation): Ingredients such as yellow stripe trevally, green mussel, blue swimming crab, and Pacific white shrimp were procured from local markets. Post-purchase, these items were refrigerated at 4°C until they were ready for use.

Experiment 2 (formulated feed preparation): Beef scraps and bonemeal were integrated into the feeds to substitute fishmeal protein at varying concentrations: 0%, 10%, 20%, and 30%, resulting in formulations BSB0, BSB10, BSB20, and BSB30 respectively. The formulated feeds incorporated tuna oil as the lipid component and wheat flour as the carbohydrate base. Prior to feed preparation, the beef scraps and bonemeal were finely ground to achieve a consistent particle size. The mixing process involved thoroughly blending all dry ingredients, including poultry by-products, for 30 minutes. Subsequently, tuna oil was introduced and mixed for an additional 15 minutes.

Water, amounting to 30% of the dry ingredients' weight, was then added, and the mixture was blended for another 15 minutes. Following this, the compound was extruded and left to dry at room temperature over a span of 48 hours. For serving convenience, these feeds were shaped into small, 1.5-cm diameter round pieces, enabling easy consumption by the snails. All formulated diets were preserved in a refrigerator at 4°C until they were administered. Each diet type underwent duplicate analysis for their proximate compositions, adhering to the standardized methods delineated by AOAC (2012).

### 2.3 Broodstock and rearing system

Experiment 1: From Chumphon Province's commercial farm, 1,800 *B. areolata* broodstocks were selected, each averaging a wet weight of 43 g. After a 2-week acclimation in an indoor hatchery, 264 broodstocks were allocated across 12 tanks (22 snails/tank) with a 1:1 male-to-female ratio. Each tank, measuring 0.80x0.55x0.50 meters, had a sand base for burrowing and received

a steady flow of unfiltered seawater for 12 hours daily. The water was maintained at a depth of 30 cm with consistent aeration. Daily feeding occurred at 10:00 hours, with leftovers removed promptly. Every fortnight, tanks were cleaned and refilled with new seawater. Water conditions kept were 26.5-30.0°C in temperature and 28-30 ppt salinity, among other parameters. Tanks were exposed to natural lighting, and daily checks were made for spawning activities.



**Figure 2.** Rearing system in the experiment1 and experiment-2, experimental rearing tanks supplied a flow-through system of unfiltered natural seawater (A), The tank bottoms were covered with coarse sand as substratum for burying the snails (B), spotted babylon were burying in coarse sand (C), feeding of the spotted babylon (D)

#### 2.4 Juvenile Rearing in Experiment 2

From the Aquatic Animal Hatchery and Research Unit, 4,700 *B. areolata* juveniles, averaging 0.5 g, were chosen. Following a 24-hour acclimation, 1,200 of these were spread across 15 tanks, resulting in 80 snails per tank. These tanks, measuring 0.80 x 0.55 x 0.50 meters, were layered with coarse sand for the snails to burrow: 8 cm depth for broodstocks and 2 cm for juveniles. Seawater, flowing at 16 liters per minute for 12 hours daily, was provided with steady aeration and a consistent water depth of 30 cm. Feeding occurred twice daily, with each session lasting around 60 minutes. Post-feeding, tanks were cleaned, and unconsumed food was removed.

They were then refilled with fresh seawater. The maintained water conditions included a temperature of 26.0-29.0°C, salinity of 28-30 ppt, dissolved oxygen of 5.0-7.0 mg/l, and ammonia nitrogen up to 0.17 mg/l. The tanks operated under natural light. Periodically, growth of the snails was monitored, and survival rates were determined at the study's end.

#### 2.5 Assessing growth and reproductive metrics

Reproductive success was gauged through various measures such as spawning frequency, egg capsule count, hatching rates, larval sizes, and overall egg capsule production. Over a 3-month trial, egg capsules were daily harvested from females fed on different experimental diets. Each spawning event was noted, and capsules were either netted or handpicked. The tally of spawning broodstock and the egg capsules they produced were recorded during each feed test. Monthly averages of spawning events and egg capsule counts were calculated at 30-day marks. Upon concluding the study, every broodstock was individually measured for weight and shell dimensions. Cumulative feed consumption data, along with metrics like average weight, weight increment, feed conversion ratio (FCR), and survival rates, were analyzed.

#### 2.6 Feed formulations

Based on the results from Table 1, various fresh foods were analyzed to determine their protein, lipid, moisture, and ash content. The data is represented as the mean  $\pm$  standard deviation. Further, data for *P. pelagicus* and *L. vannamei* demonstrated their respective values within the same parameters. Notably, values within the same column with different superscripts signify statistically significant differences ( $P < 0.05$ ).

Table 2 highlights the analysis of different ingredients used in feed formulation. Ingredients like yellow stripe trevally fish, Fishmeal, Beef scraps and bonemeal, soybean meal, Wheat flour, Shrimp head meal, and Wheat gluten were investigated. Each of these ingredients was evaluated for their protein, lipid, moisture, and ash content. Similar to Table 1, the results are represented as the mean  $\pm$  standard deviation, and significant differences are marked by different superscripts.

Table 3 detailed the compositions of various treatments, showcasing the quantities of each ingredient incorporated into the experimental feeds. Each treatment had a specific blend of ingredients like yellow stripe trevally fish, fish meal, beef scraps and bonemeal, among others. For instance, while Treatment 1 exclusively used yellow stripe Trevally Fish, other treatments employed a mix of ingredients. Additionally, each feed composition included a fixed amount of Vitamin and Mineral mixes. The vitamin mix contained specific quantities of retinal (A), cholecalciferol (D3), tocopherol (E), and several other vitamins per kilogram of diet. Similarly, the Mineral mix incorporated elements like Ca, Mg, Cu, and others in designated proportions.

In Table 4, formulated feeds from different treatments underwent proximate analysis. Each treatment, namely Treatment 1 (control) through Treatment 5 (BSB30), was studied for their protein, lipid, moisture, and ash content. As before, results are illustrated as the mean  $\pm$  standard deviation, and values in the same column bearing different superscripts represent significant differences ( $P < 0.05$ ). For instance, Treatment 1(control) and Treatment 2(BSB0) displayed distinct values for each parameter, highlighting the variability in the nutritional content of the formulated feeds.

**Table 1.** Proximate analysis of the flesh feeds (dry weight basis)

Fresh food	Protein (%)	Lipid (%)	Moisture (%)	Ash (%)
<i>S. leptolepis</i>	82.43±0.50 <sup>d</sup>	8.90±0.20 <sup>d</sup>	4.43±0.19 <sup>a</sup>	5.86±0.45 <sup>b</sup>
<i>P. viridis</i>	62.40±0.50 <sup>a</sup>	6.69±0.20 <sup>c</sup>	14.88±0.52 <sup>c</sup>	11.05±0.15 <sup>d</sup>
<i>P. pelagicus</i>	70.74±0.93 <sup>b</sup>	2.33±0.06 <sup>a</sup>	9.96±0.33 <sup>b</sup>	10.11±0.05 <sup>c</sup>
<i>L. vannamei</i>	78.99±0.27 <sup>c</sup>	4.80±0.06 <sup>b</sup>	9.98±0.11 <sup>b</sup>	4.13±0.05 <sup>a</sup>

Data represented as Mean±SD; Value in the same column with different superscripts are significantly different ( $P<0.05$ )

**Table 2.** Proximate analysis of raw materials for formulated feeds (dry weight basis)

Ingredients	Protein (%)	Lipid (%)	Moisture (%)	Ash (%)
Yellow stripe trevally fish	82.43±0.50	8.90±0.20	5.86±0.45	4.43±0.19
Fishmeal	58.63±0.86	10.95±0.20	5.16±0.08	30.73±0.33
Beef scraps and bonemeal	57.90±0.09	10.94±0.20	5.47±0.15	22.25±0.11
Soybean meal	44.52±0.44	2.24±0.06	10.33±0.23	6.95±0.01
Wheat flour	13.28±0.44	1.38±0.07	12.12±0.24	0.65±0.02
Shrimp head meal	47.18±0.99	6.05±0.31	12.16±0.53	19.98±0.13
Wheat gluten	75.84±0.58	1.98±0.10	0.81±0.02	10.42±0.07

**Table 3.** Composition of experimental formulated feeds (g/100g diet)

Ingredients	Treatment				
	1	2	3	4	5
Yellow stripe trevally fish	100	-	-	-	-
Fish meal	-	40	30	20	10
Beef scraps and bonemeal	-	0	10	20	30
Soybean meal	-	5	5	5	5
Wheat flour	-	24	24	24	24
Shrimp head meal	-	5	5	5	5
Wheat gluten	-	15	15	15	15
Tuna oil	-	5	5	5	5
Vitamins mix <sup>1</sup>	-	2	2	2	2
Minerals mix <sup>2</sup>	-	2	2	2	2
Calcium hydrogen phosphate	-	2	2	2	2
Total	100	100	100	100	100

<sup>1</sup>Vitamin mix (g/diet 1 kg): Retinal(A) 150,000,000 IU; Cholecalciferol(D3) 3,000,000 IU; Tocopherol(E) 27.5; Menadione sodium bisulfate(K3) 4.67; Thiamine(B1) 25; Calcium pantothenate 5; Folic acid 0.4; Ascorbic acid 143

<sup>2</sup>Mineral mix (g/diet 1 kg): Ca 11.22; Mg 2.48; Cu 1.25; Zn 2.14; Na 106.22; squid liver powder 100

**Table 4.** Proximate analysis of formulated feeds (dry weight basis)

Treatment	Protein (%)	Lipid (%)	Moisture (%)	Ash (%)
Treatment 1 (Control)	82.43±0.50 <sup>c</sup>	8.90±0.20 <sup>a</sup>	5.86±0.45 <sup>a</sup>	4.43±0.19 <sup>a</sup>
Treatment 2 (BSB0)	41.60±0.53 <sup>a</sup>	9.33±0.49 <sup>b</sup>	7.878±0.12 <sup>b</sup>	16.28±0.09 <sup>c</sup>
Treatment 3 (BSB10)	41.85±0.87 <sup>ab</sup>	10.29±0.14 <sup>bc</sup>	7.74±0.07 <sup>b</sup>	15.63±0.16 <sup>d</sup>
Treatment 4 (BSB20)	42.34±0.22 <sup>ab</sup>	11.42±1.19 <sup>c</sup>	7.18±0.13 <sup>b</sup>	14.63±0.13 <sup>c</sup>
Treatment 5 (BSB30)	42.88±0.77 <sup>b</sup>	11.27±0.85 <sup>c</sup>	7.67±0.09 <sup>b</sup>	13.73±0.06 <sup>b</sup>

Data represented as Mean±SD; Value in the same column with different superscripts are significantly different ( $P<0.05$ )

##### 5. Statistical analysis

Data is presented as mean ± SD. Significant differences among

treatments were identified using one-way ANOVA. When differences were found, Duncan's test was applied to distinguish between means at a significance level of  $P<0.05$ .

**Table 5.** Initial weight, length, width and the final weight, length, width, and survival rate of spotted babylon broodstock fed with different diets during 6 months in experiment 1

Treatment	Weight (g)		Shell length (cm)		Shell width (cm)		Survival rate (%)
	Initial	Final	Initial	Final	Initial	Final	Final
T1	43.82±0.33 <sup>a</sup>	44.81±0.05 <sup>a</sup>	6.70±0.01 <sup>a</sup>	6.76±0.02 <sup>a</sup>	4.20±0.01 <sup>a</sup>	4.27±0.00 <sup>a</sup>	90.9±0.00 <sup>a</sup>
T2	43.92±0.64 <sup>a</sup>	43.36±0.43 <sup>a</sup>	6.70±0.00 <sup>a</sup>	6.70±0.01 <sup>a</sup>	4.18±0.04 <sup>a</sup>	4.25±0.01 <sup>a</sup>	100 <sup>b</sup>
T3	43.44±0.66 <sup>a</sup>	46.47±1.84 <sup>a</sup>	6.70±0.01 <sup>a</sup>	6.72±0.02 <sup>a</sup>	4.18±0.00 <sup>a</sup>	4.22±0.04 <sup>a</sup>	100 <sup>b</sup>
T4	43.70±0.70 <sup>a</sup>	44.73±1.37 <sup>a</sup>	6.66±0.06 <sup>a</sup>	6.69±0.05 <sup>a</sup>	4.21±0.03 <sup>a</sup>	4.29±0.03 <sup>a</sup>	93.9±5.2 <sup>a</sup>

Data represented as Mean±SD

Value in the same column with different superscripts are significantly different ( $P<0.05$ )

T1: yellow stripe trevally, T2: green mussel, T3: blue swimming crab, T4: pacific white shrimp

**Table 6.** Reproductive performance and survival rate of juvenile spotted babylon snails fed with different diet during 6 months in experiment 1

Treatment	egg capsule length(cm)	egg capsule width(cm)	fecundity (egg/capsule)	egg size (µm)	Hatching rate (%)
T1	2.11±0.08 <sup>a</sup>	0.99±0.02 <sup>a</sup>	661±22.10 <sup>c</sup>	303.3±0.51 <sup>a</sup>	57.0±9.7 <sup>a</sup>
T2	2.09±0.02 <sup>a</sup>	0.92±0.01 <sup>a</sup>	548±21.66 <sup>b</sup>	306.1±0.30 <sup>a</sup>	58.7±11.0 <sup>a</sup>
T3	2.01±0.04 <sup>a</sup>	0.95±0.05 <sup>a</sup>	498±31.38 <sup>a</sup>	305.4±0.39 <sup>a</sup>	62.3±9.7 <sup>a</sup>
T4	2.01±0.06 <sup>a</sup>	0.95±0.12 <sup>a</sup>	475±47.82 <sup>a</sup>	314.8±0.77 <sup>a</sup>	70.1±7.9 <sup>a</sup>

Data represented as Mean±SD

Value in the same column with different superscripts are significantly different ( $P<0.05$ )

T1: yellow stripe trevally, T2: green mussel, T3: blue swimming crab, T4: pacific white shrimp

**Table 7.** Water qualities in broodstock rearing tanks throughout 6 months of experiment 1

Treatment	Temp(°C)	Salinity(ppt)	pH	DO (mg/l)	Alkalinity (mg/l)	NH <sub>3</sub> -N (mg/l)
T1	28.63±0.14 <sup>a</sup>	29.27±0.63 <sup>a</sup>	7.71±0.02 <sup>a</sup>	5.91±0.03 <sup>a</sup>	163.10±1.05 <sup>a</sup>	0.15±0.02 <sup>a</sup>
T2	28.69±0.04 <sup>a</sup>	29.76±0.02 <sup>a</sup>	7.70±0.01 <sup>a</sup>	5.76±0.05 <sup>a</sup>	167.10±2.80 <sup>a</sup>	0.09±0.04 <sup>a</sup>
T3	28.67±0.03 <sup>a</sup>	29.76±0.01 <sup>a</sup>	7.70±0.01 <sup>a</sup>	5.77±0.02 <sup>a</sup>	167.59±1.38 <sup>a</sup>	0.12±0.01 <sup>a</sup>
T4	28.56±0.15 <sup>a</sup>	29.66±0.23 <sup>a</sup>	7.77±0.09 <sup>a</sup>	5.80±0.18 <sup>a</sup>	168.13±0.69 <sup>a</sup>	0.05±0.02 <sup>a</sup>

Data represented as Mean±SD

Value in the same column with different superscripts are significantly different ( $P<0.05$ )

T1: yellow stripe trevally, T2: green mussel, T3: blue swimming crab, T4: pacific white shrimp

### 3. Results and discussion

#### 3.1 Experiment 1: analysis of dietary impact on spotted babylon broodstock

The growth trajectory and survivability of the Spotted Babylon broodstock, delineated through initial and final measurements of weight, length, and width over a span of 6 months, is detailed in Table 5. A pivotal observation was that while weight gain percentages, and growth increments in length and width, alongside the monthly growth rate, displayed no discernible variation ( $P>0.05$ ), a pronounced disparity was evident in the survival rates across the four distinct feeding treatments ( $P<0.05$ ). Treatments 2 and 3 exhibited an impeccable survival rate of 100%, starkly contrasting the reduced rates of 90.9% and 93.9% observed in

treatments 1 and 4 respectively ( $P<0.05$ ).

Furthermore, the fecundity or reproductive potential showcased significant variances across the different flesh feeds ( $P<0.05$ ). Notably, broodstock under treatments 3 and 4 depicted a parity in their fecundity levels ( $P>0.05$ ). An intriguing pattern emerged with broodstock from treatment 1 leading the fecundity charts, averaging at 661±22.10 eggs/capsule. They were sequentially followed by those from treatments 2, 3, and 4, registering fecundity rates of 548±21.66, 498±31.38, and 475±47.82 eggs/capsule, respectively.

Diving deeper into the ecological traits of spotted babylon, it's worth noting their intrinsic scavenger behavior, feeding primarily on deceased marine organisms settled on the ocean floor (Kritsanapuntu et al., 2006). Furthermore, specimens of this

species exceeding 3 cm in shell length typically exhibit a decelerated growth trajectory. This implies that the sheer volume of food consumed and its protein composition might not be the cardinal determinants influencing the growth dynamics of the *B. areolata* broodstocks.

Reproductive metrics and survival rates of the spotted babylon broodstock, derived from various dietary regimens spanning 6 months, are articulated in Tables 5-6. Surprisingly, metrics like egg capsule dimensions, egg calibrations, and hatching propensities showcased no significant fluctuations across the four experimental feeds (Sreejaya, 2008). Yet, the unpredictable nature of larvae culture yields can be attributed to the inconsistent quality spectrum of eggs and larvae. This quality is invariably governed by a confluence of endogenous parameters like genetic makeup, broodstock age and size, and egg dimensions.

Exogenous variables also play a role, encompassing facets like egg management protocols, dietary regimes for broodstocks, and microbial colonization on egg surfaces, as deduced by Ballestrazzi et al. (2003). It's well-documented in teleost studies that specific nutrients, ranging from proteins, and essential fatty acids, to vitamins like E, ascorbic acids, and carotenoids, have profound implications on reproductive trajectories, encompassing processes like gonadal maturation, gamete vitality, and spawning dynamics (Haitana et al., 2016).

Yet, the intricate nexus between nutrient profiles and reproductive mechanics remains an enigma. A myriad of scholarly pursuits, like those of Ling et al. (2006), underscores the paramount importance of both the quantum and caliber of dietary lipids in shaping broodstock reproductive outcomes. In a similar vein, Bautista-Teruel et al., (2001) illustrated how an enriched content of crucial nutrients, particularly proteins, lipids, and pivotal unsaturated fatty acids, catalyzed augmented reproductive outputs in abalones, specifically *Haliotis asinina*.

### 3.2 Experiment 2: an analytical dive into the growth performance of juvenile snails

Comparative metrics & feeding regimes: The juvenile snails, irrespective of their varied dietary inclusions, exhibited analogous growth metrics in terms of overall weight, shell dimensions including length and width (as visualized in Figures 3-5). A pronounced discrepancy was evident in these parameters among juveniles subjected to different formulated feeds and carangid fish ( $P < 0.05$ ). In an intriguing revelation, the cohort fed with carangid fish (T1) outpaced their counterparts in growth metrics, registering superior measurements ( $P < 0.05$ ).

From the third to sixth month mark, juveniles subjected to formulated diets inclusive of varying proportions (0%, 10%, 20%) of beef scrap and bonemeal (BSB0, BSB10, and BSB20) mirrored each other in their growth statistics ( $P > 0.05$ ), albeit trailing behind the T1 (control) cohort. Conversely, the subset nurtured on a diet steeped in 30% beef scrap and bonemeal (30%BSBM) trailed at the bottom of the growth leaderboard, depicting the lowest metrics in weight and shell dimensions ( $P < 0.05$ ) as illustrated in Figures 3, 4,

and 5.

The conclusion of this experimental phase spotlighted the undeniable superiority of the carangid fish diet (T1), which championed in promoting weight, length, and width gain, vis-a-vis other dietary treatments ( $P < 0.05$ ) - a testament documented in Table 8. A probable theory behind this growth acceleration is the protein-rich, reduced ash content of carangid fish, paired with a less pungent fishy odor that proved enticing for the snails, echoing findings from Daroonchoo et al. (2005).

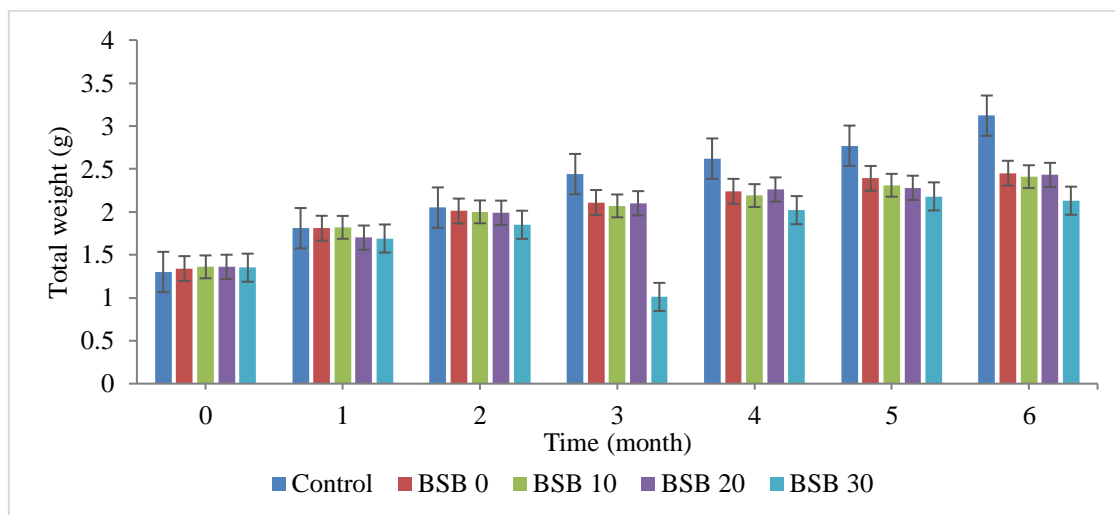
Cross-comparison with previous research: The weight augmentation of the T1 cohort was congruent with the findings of Chaitanawisuti et al. (2011), who reported analogous weight gain trajectories in juvenile snails, both in canvas and earthen ponds. Furthermore, Dobson et al. (2020) showcased a comparable weight progression in snails nurtured on trash fish, observing a weight transition from  $0.90 \pm 0.38$  g to  $4.93 \pm 1.44$  g over a 92-day period. With regard to protein content, Paibulkichakul (2008) advocated an optimal protein content of 38%, juxtaposed against the experimental diet here, which ranged from 41-43% protein, albeit with elevated ash levels.

This brings to mind Moutinho et al.'s (2017) study, where disparate growth performances were registered in gilthead seabream juveniles based on the inclusion levels of meat and bone meal in their diet. Feeding efficiency and methodology: The Feed Conversion Ratio (FCR) analysis revealed that snails from the T1 and BSB30 cohorts had an elevated FCR compared to their BSB0, BSB10, and BSB20 counterparts ( $P < 0.05$ ). A unique feeding mechanism was observed in the T1 cohort; these snails would puncture the fish meat with their proboscis, secrete gastric enzymes to initiate digestion, and then siphon the pre-digested nourishment.

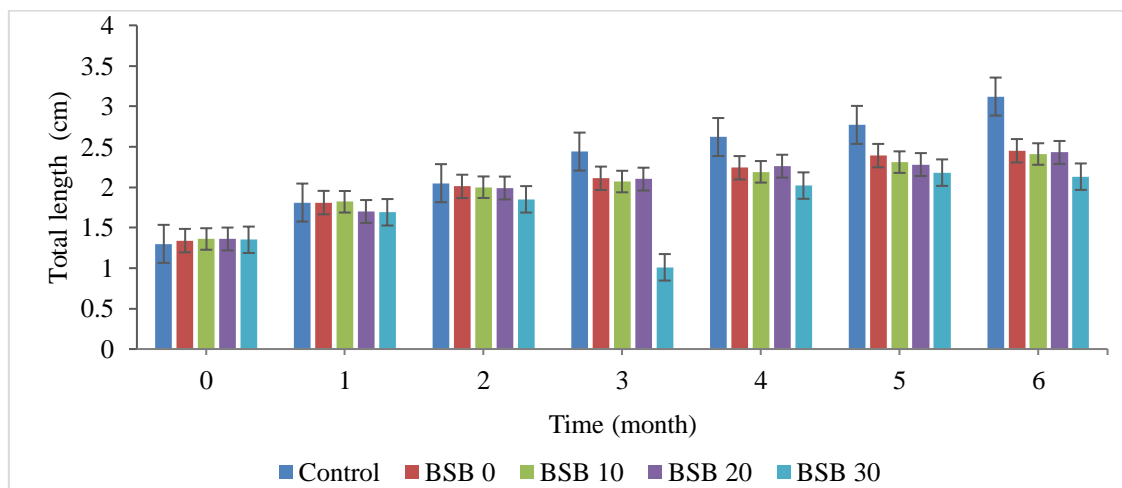
This implies that fish meat may be more susceptible to degradation than pellet diets, an observation underpinned by Chaitanawisuti and Kritsanapantu (2011). Yet, the FCR for BSB30 was in line with the T1 group, potentially attributed to the heightened ash content in the 30%BSBM diet, echoing insights from Moutinho et al., 2017. A critical dimension of our research focused on the sustainability and survival adaptability of *B. areolata* across varied dietary treatments. Survival metrics: Our research on the *B. areolata*'s survival adaptability across different diets revealed consistent survival rates regardless of dietary variations ( $P > 0.05$ ), as detailed in Table 8. These results align with Chaitanawisuti et al. (2011), who also found steady survival rates for snails in varied environments, such as canvas and earthen ponds.

Their study meticulously documented consistent survival rates for snails, reinforcing our findings, and establishing a strong correlation between our contemporary research and historical data (Table 9). The consistent data highlights the snail's resilience and adaptability across diverse dietary and environmental conditions. Further studies could delve into the biological reasons behind this consistent survival rate. Prospect endeavors might explore the underlying biological mechanisms that render this consistency, enabling a more nuanced understanding of the species' resilience.

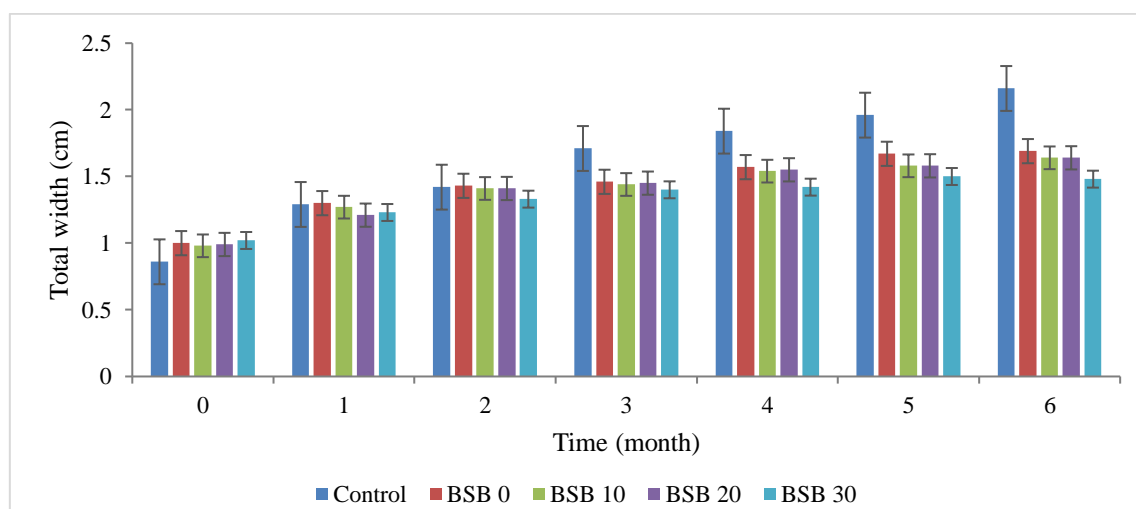




**Figure 3.** Total weight of juvenile spotted babylon snails fed with different formulated feeds during 6 for experiment



**Figure 4.** Total length of juvenile spotted babylon snails fed with different formulated feeds during 6 months experiment



**Figure 5.** Total width of juvenile spotted babylon snails fed with different formulated feeds during 6 months experiment

**Table 8.** Total weight gain, length gain, width gain, survival rate and FCR of juvenile spotted babylon snails fed with different formulated feeds throughout 6 months experiment

Treatment	weight gain (g)	length gain (cm)	width gain (cm)	Survival rate (%)	FCR
T1(control)	6.45±0.35 <sup>c</sup>	1.82±0.03 <sup>c</sup>	1.30±0.00 <sup>c</sup>	83.75±5.73 <sup>a</sup>	1.40±0.06 <sup>b</sup>
T2(BSB0)	2.90±0.26 <sup>b</sup>	1.11±0.03 <sup>b</sup>	0.69±0.05 <sup>b</sup>	89.58±2.89 <sup>a</sup>	0.80±0.08 <sup>a</sup>
T3(BSB10)	2.49±0.30 <sup>b</sup>	1.05±0.13 <sup>b</sup>	0.66±0.05 <sup>b</sup>	82.08±9.46 <sup>a</sup>	0.96±0.07 <sup>a</sup>
T4(BSB20)	2.63±0.24 <sup>b</sup>	1.07±0.08 <sup>b</sup>	0.67±0.03 <sup>b</sup>	84.17±8.04 <sup>a</sup>	0.85±0.16 <sup>a</sup>
T5(BSB30)	1.65±0.30 <sup>a</sup>	0.73±0.11 <sup>a</sup>	0.47±0.05 <sup>a</sup>	75.83±12.27 <sup>a</sup>	1.36±0.34 <sup>b</sup>

Data represented as Mean±SD.

Value in the same column with different superscripts are significantly different ( $P<0.05$ )

**Table 9.** Average water qualities in broodstock rearing tanks of the second experiment

Treatment	Temp(°C)	Salinity(ppt)	pH	DO. (mg/l)	Alkalinity (mg/l)	NH <sub>3</sub> -N (mg/l)
<b>T1</b>	27.93±0.02 <sup>c</sup>	28.59±0.00 <sup>b</sup>	7.82±0.00 <sup>a</sup>	5.46±0.04 <sup>a</sup>	154.22±0.92 <sup>b</sup>	0.10±0.04 <sup>a</sup>
<b>T2</b>	27.53±0.00 <sup>a</sup>	28.35±0.00 <sup>a</sup>	7.79±0.01 <sup>a</sup>	5.26±0.02 <sup>a</sup>	145.56±5.30 <sup>a</sup>	0.13±0.03 <sup>a</sup>
<b>T3</b>	27.55±0.02 <sup>a</sup>	28.33±0.03 <sup>a</sup>	7.79±0.01 <sup>a</sup>	5.38±0.08 <sup>b</sup>	147.56±0.98 <sup>a</sup>	0.14±0.06 <sup>a</sup>
<b>T4</b>	27.55±0.02 <sup>a</sup>	28.35±0.00 <sup>a</sup>	7.78±0.01 <sup>a</sup>	5.38±0.04 <sup>b</sup>	147.00±0.73 <sup>a</sup>	0.19±0.06 <sup>a</sup>
<b>T5</b>	27.58±0.02 <sup>b</sup>	28.35±0.00 <sup>a</sup>	7.77±0.07 <sup>a</sup>	5.52±0.07 <sup>c</sup>	147.44±1.36 <sup>a</sup>	0.15±0.04 <sup>a</sup>

Data represented as Mean±SD.

Value in the same column with different superscripts are significantly different ( $P<0.05$ )

### 3.3 Advancing biocircular practices in spotted babylon aquaculture

As we usher in a new era in aquaculture, sustainable practices and efficient resource use are vital. In the realm of spotted babylon, there's potential in harnessing industry waste products for feed development, aligning with the goals of reducing waste and cost-effective solutions (Kritsanapuntu and Chaitanawisuti, 2015). Viewing waste as a beginning, not an end, initiatives can convert aquaculture waste into valuable products like organic fertilizers (Stickney and Gatlin, 2022). The breeding of spotted babylon demands sustainable hatchery methods with minimal environmental impact, achieved through energy-efficient systems and renewable energy. Shifting to local feed production reduces the carbon footprint and boosts local economies, reflecting a circular economy.

The biocircular economy emphasizes resource regeneration, suggesting potential in reprocessing aquaculture waste (Ahmad et al., 2022). Products from Spotted Babylon should have extended lifecycles through innovative techniques, ensuring sustainability and reduced waste. Precision aquaculture, optimizing inputs like feed and energy, is essential (Wang et al., 2021). Modern business models might focus on shared resources, cooperative ventures, or leasing systems for maximum utility. Embracing the biocircular economy principles offers numerous opportunities for the spotted babylon aquaculture industry, marking a shift towards sustainable seafood and industry evolution.

## 4. Conclusion

Upon rigorous evaluation, it is empirically established that carangid fish (*S. leptolepis*) emerges as the most effective flesh food for *B. areolata* broodstocks, yielding superior growth and reproductive performance relative to other dietary options. However, seasonal considerations, particularly during monsoon

seasons when carangid fish become scarce, necessitate viable alternatives. In such circumstances, mussels (*P. viridis*) stand out as an effective substitute. Furthermore, based solely on weight growth performance, both swimming crabs (*P. pelagicus*) and Pacific white shrimps (*L. vannamei*) present themselves as feasible dietary options for *B. areolata* broodstocks. Transitioning to juvenile culture, while beef scrap and bonemeal can be incorporated as replacements for fish meal in their formulated diet, it's imperative to limit their inclusion to a maximum of 20% to ensure optimal growth outcomes. This reiterates the paramount importance of a holistic approach in choosing dietary components, always prioritizing both growth and overall health of the *B. areolata* species.

## Conflict of Interest Declaration

The authors assert that there are no conflicts or personal affiliations that might be construed as impacting the outcomes shared in this research.

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