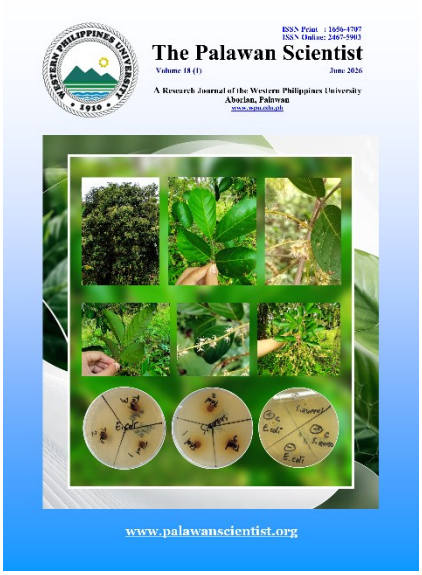

Efficacy of bamboo as an alternative substrate for cage culture of abalone *Haliotis asinina* Linnaeus, 1758

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ABSTRACT

Abalone is a highly valued marine gastropod with a declining wild population due to increased fishing pressure. To meet market demand, there is growing interest in the cage culture of abalone juveniles. However, the most commonly used substrate in cage culture, polyvinyl chloride (PVC), has been linked to environmental and health risks. This study examined the efficacy of bamboo as an alternative substrate for the bottom and suspended cage culture of tropical abalone *Haliotis asinina* Linnaeus, 1758. Two culture experiments were conducted using (1) sea bottom cages and (2) suspended sea cages in Binduyan, Puerto Princesa City, and Pamantolon, Taytay, Palawan, respectively. Abalone juveniles grown with bamboo substrate (BS) had significantly higher weight gain, specific growth rate, and shell length growth rate compared to those reared with PVC substrate (PS) ($P < 0.05$). In addition, after 90 days and 150 days of culture, abalone juveniles on BS exhibited positive allometric growth compared to those on PS, which showed isometric growth. Moreover, the survival rates of abalone juveniles with BS were not significantly different from those with PS at $P < 0.05$. These findings suggest that BS is a viable alternative to PS for abalone juvenile culture, as it is indigenous, inexpensive, and environmentally friendly. The study's results can promote sustainable aquaculture practices for abalone while raising awareness of the potential environmental and health risks associated with PVC for cultured abalone and humans consuming cultured abalone.

Keywords: allometric, bottom sea cage, growth rate, isometric, PVC, suspended sea cage

INTRODUCTION

The tropical abalone *Haliotis asinina* Linnaeus, 1758, is not only palatable but also considered a nutraceutical due to its many health benefits. It contains various bioactive molecules, such as polysaccharides, proteins, and fatty acids that provide many health benefits, including antioxidant, anti-cancer, anti-inflammatory, anti-thrombotic, anti-microbial, anti-aging, anti-arthritic, and anti-hypertensive properties (Suleria et al. 2017a, b). Moreover, its shells can be used to make jewelry and ornaments (Surtida 2000). Strong domestic and overseas demand has made abalone a highly valued marine gastropod (Setyono 2005). The high market value of abalone has led to increased fishing pressure, causing a dramatic decline with wild population (Smith 2022; Gonzales 2015). Consequently, significant interest has emerged in the cage culture of

abalone juveniles to meet market demands and their high export potential.

Since the early 1990's, *H. asinina* broodstock has been successfully bred in Philippine hatcheries to support the increasing demand for abalone juveniles (Fermin 2001). Growth and spawning occur year-round, with fast growth and high survival rates in both land-based and sea-based culture systems (Fermin 2001). Despite advancements in abalone juvenile culture in the Philippines, continuous improvements are needed for optimal results. One aspect worth examining is the use of polyvinyl chloride (PVC) substrates. An appropriate substrate provides a suitable space for abalone juveniles to shelter and forage for food, day and night (Setyono 2015). Several studies have examined the use of PVC substrates (PS) in both on land-based and sea-based abalone juvenile culture, and is currently the most commonly used substrate in commercial production (Capinpin et al. 1999; Setyono 2015; SEAFDEC/AQD 2000, 2022). However, PVC plastic has been reported to pose potential risks to human health and the environment (Osmanski 2020; Health Care Without Harm Europe 2021). Given that abalone graze on its surface for periphytic algae, the use of PVC in their culture should be reconsidered.

Previous studies on the effect of substrates in abalone juvenile culture have primarily focused on synthetic materials, such as PVC pipes, plastic baskets, corrugated fiberglass, plexiglass, rubberized canvas, and fibro-cement board (Avilles and Shepherd 1996; Gapasin and Polohan 2005; Setyono 2007). No studies have been conducted on the impact of indigenous materials, such as bamboo, as alternative substrates to synthetic materials in abalone cage culture except for abalone conditioning, as reported by De Guzman and Creencia (2014). The present study investigates the use of bamboo (*Bambusa blumeana* Schult.f.) as an alternative substrate. Bamboo substrate (BS) is an indigenous material usually abundant in rural coastal areas, costing PHP 100-150 (USD 1.72-2.58) per pole (5 m), and is environmentally friendly. Considering the environmental benefits of these indigenous materials, elucidating their effects on the growth and survival of abalone juveniles in cage culture is needed. As the Philippines' abalone juvenile culture continues to expand, this study's results have significant applications. The current generation must rise to the challenge of adopting eco-friendly products for a sustainable future. A favorable outcome from this study could encourage small-scale aquaculturists to use inexpensive, indigenous, and environmentally friendly substrates for abalone juvenile culture in both the bottom and suspended sea cages.

This study investigated the potential of BS as an alternative to PS. Specifically, this study determined the following: (1) weight growth rates, shell length (SL) growth rates, and survival rates of reared abalone in bottom sea cage culture and suspended sea cage culture experiments; (2) food conversion efficiency (FCE) of reared abalone in suspended sea cage culture experiment; and (3) length and weight (LW) relationship of reared abalone in bottom sea cage culture and suspended sea cage culture experiments, which can be used in stock evaluation models to predict weight from length.

METHODS

Study Site

This study included two experiments conducted from 2019 to 2021. The first study site was the Western Philippines University Binduyan Marine Research Station (WPU-BMRS), located at Barangay Binduyan, Puerto Princesa City, 79 km north of the capital of Palawan province. Here, the researchers conducted the bottom sea cage culture, where weight, SL, and FCE data were collected. The second study site was in Barangay Pamantolon, Taytay, Palawan, 147 km from WPU-BMRS (Figure 1), where the suspended sea cage culture experiment was conducted, with data collected on weight and SL only.

Experiment 1: Bottom Sea Cage Culture

A total of 216 abalone juveniles, with a mean (±SE) weight of 1.52 ± 0.77 g and a mean SL of 20.37 ± 3.21 mm, were obtained from the hatchery of WPU-BMRS for this experiment. This study applied two treatments (T), each with three replicate cages: T1 = BS (bamboo substrate) and T2 = PS (PVC substrate). Two sets of BS and PS, each measuring approximately 0.30 m × 0.30 m with a total surface area of 0.36 m^2, were placed in each cubical cage. Each replicate meshed (3.2 mm) cages (30 cm^3) were positioned on a platform at a depth of 8-9 m, 100 m from the shoreline, following a completely randomized design (CRD). Thirty-six (36) abalone juveniles were stocked in each cage were tagged using a laminated label pasted with Mighty Bond® glue to the posterior side of the abalone shell (Figure 2). These tagged abalone samples were used for measuring weight and SL throughout the three-month culture period. A stocking density of 100 abalone juveniles per m^2 substrate surface area was maintained. Explicit acclimation was not conducted, as the tanks where the juveniles were kept used seawater freshly pumped from the open sea, which was approximately 100 m from the cage rearing site. A summary of Experiment 1 characteristics is provided in Table 1.

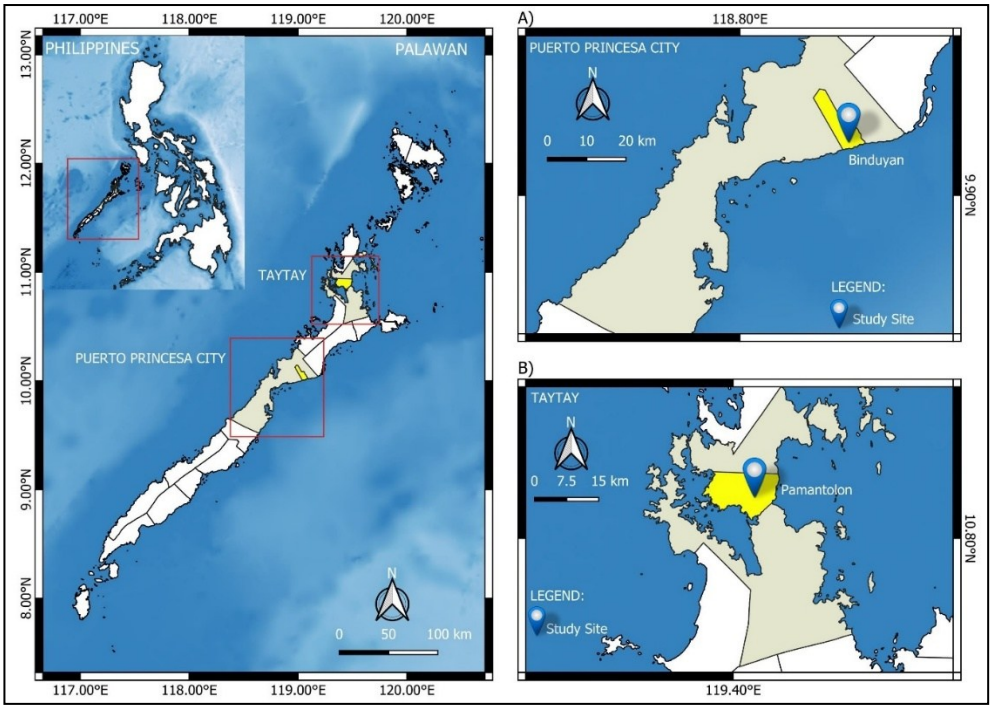

Figure 1. Map showing the study sites in (A) Binduyan, Puerto Princesa City and (B) Pamantolon, Taytay, Palawan, Philippines.

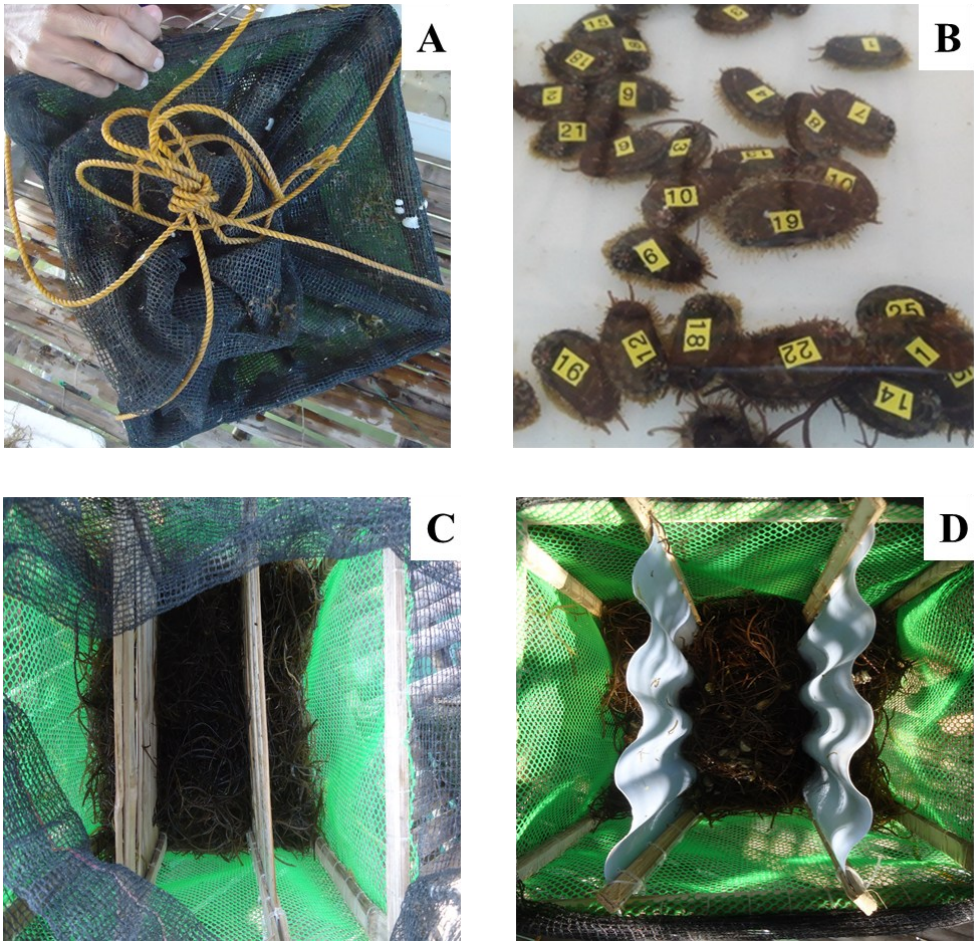

Figure 2. Photographs of the (A) covered meshed cage with rope for attachment to a bottom platform, (B) tagged abalone juveniles used in the experiment, and (C) bamboo substrates and (D) PVC substrates placed inside the cage.

Table 1. Summary of characteristics of the experiments on rearing abalone juveniles using sea cages at Palawan, Philippines.

Variables	**Experiment 1, Bottom Sea Cage Culture**		**Experiment 2, Suspended Sea Cage Culture**	
Treatment (Substrate)	1-bamboo	2-PVC	1-bamboo	2-PVC
Replicate cages	3	3	3	3
Abalone mean initial shell length (mm)	20.33	20.14	26.27	26.00
Abalone mean initial weight (g)	1.51	1.51	3.36	3.20
Dimension of cage used (cm^3)	30	30	30	30
Mesh size of cage net (mm)	3.2	3.2	3.2	3.2
Density/cage (0.36 m^{-2} surface area substrate)	36	36	60	60
Distance of cage from surface (m)	8-9	8-9	1-2	1-2
Interval of data measurement (days)	15	15	30	30
Method of retrieving the sea cages	SCUBA diving	SCUBA diving	Manual lifting	Manual lifting
Food provided in the cage (seaweed)	*Gracilaria*	*Gracilaria*	*Gracilaria*	*Gracilaria*
Interval of provision of seaweed food (days)	15	15	15	15
Feeding rate (% of body weight)	>180	>180	>100	>100
Culture period (days)	90	90	150	150
Study site	Binduyan Marine Research Station		Pamantolon, Taytay, Palawan	

Experiment 2: Suspended Sea Cage Culture

A total of 360 abalone juveniles were used in this culture, stocked at a density of 167 abalone juveniles per m^2 substrate surface area, with 60 individuals per cage. The juveniles had a mean weight of 3.26 ± 1.38 g and a mean SL of 26.13 ± 3.38 mm and were sourced from the hatchery of WPU-BMRS. The abalone juveniles were transported for approximately three hours in oxygenated bags containing cut bamboo pole substrates lined with a damp towel. Ice packs placed outside the bags in a styrofoam box kept them cool during transport. Upon arrival at the site, acclimation was conducted before placing the abalone juveniles in the suspended sea cages. Acclimation took place in a shaded hut near the longline for about one hour, during which ambient seawater was gradually sprinkled over them until they were fully submerged.

Similar to Experiment 1, the same cage materials and substrate sizes were used, along with two treatments, each with three replicate cages (T1 = BS, T2 = PS). The cages were suspended on a longline in a seaweed farm, approximately 1.5 m apart and 1-2 m below the sea surface. The juveniles were reared for five months. A summary of the characteristics of Experiment 2 is provided in Table 1.

Feeding and Cage Management

Proximate analysis of *Gracilaria firma* C.F. Chang & B.-M. Xia, 1976, the seaweed utilized as a primary food source for abalone in the experiments had 13.45% crude protein, 2.15% N, 66.26 mg/g amino acid, and 49.27% amino acid in protein. The seaweed was collected from the coastal areas of Tabon, Quezon, Palawan and maintained in concrete tanks with aeration at the WPU-BMRS hatchery. Before feeding, the seaweed was weighed, and any unconsumed portions were weighed again after 15 days. In Experiment 2, the typical seaweed provision per cage on an *ad libitum* basis per cage ranged from 600 g on day 0 to 1,500 g on day 135. Every 15 days, unconsumed seaweeds were removed and recorded to estimate food consumption. The feeding rates were maintained at >180% and >100% (both on an *ad libitum* basis) with seaweed supply monitored and recorded regularly.

Weekly cages inspections were carried out to check for predator attacks, such as tears or holes in the mesh. Any attached algae, fouling organisms, or debris were removed to ensure a constant water influx into the cage. Additionally, the cages were thoroughly cleaned during this process.

Weight Growth Rates

In Experiment 1, data were collected every 15 days with the assistance of SCUBA divers, who retrieved the cages from the sea bottom. Each abalone juvenile was manually removed from the substrate using a spatula and transferred to a basin filled with seawater and aeration. The juveniles were then placed on a dry towel before their weight was measured using a digital weighing scale (Salter, 200 g).

In Experiment 2, the weight of tagged abalone juveniles was measured every 30 days using a digital weighing scale. For consistency with Experiment 1, the weight values on days 15, 45, and 75 were estimated through interpolation based on the 30-day measurement data.

After measurements were taken in both Experiment 1 and Experiment 2, all juveniles were returned to their respective cages, which were then transported back to the culture site and securely tied in place.

Weight gain (WG) was calculated using the formula:

$$\boldsymbol{WG} = \left[\frac{WT - WI}{WI}\right] x\ 100\%.$$

Specific growth rate (SGR) was computed using the formula:

$$\boldsymbol{SGR} = \frac{\ln WT - \ln WI}{T}\ x\ 100\%.$$

Daily increase in body weight (DIBW) was computed using the formula:

$$\boldsymbol{DIBW} = \frac{WT - WI}{T} \text{ (g/day) * 1000mg/g,}$$

where: W_T = weight at time T (g); W_I= initial weight (g); *ln* = natural logarithm.

Shell Length Growth Rates

In Experiment 1, the SL of abalone juveniles was measured every 15 days using the Vernier caliper. After measurement, all juveniles were returned to their respective cages, which were then transported back to the culture site.

In Experiment 2, the SL of tagged abalone juveniles was measured every 30 days using a Vernier caliper. For consistency with Experiment 1, the SL values on days 15, 45, and 75 were estimated through interpolation based on the 30-day measurement data.

After measurements were taken in both Experiment 1 and Experiment 2, all abalone juveniles were returned to their respective cages, which were then transported back to the culture site and securely tied in place.

Shell length increment (SLI) was calculated using the formula:

$$\boldsymbol{SLI} = [(SL_T - SL_I) / SL_I] \times 100\%.$$

Daily increase in shell length (DISL) was computed using the formula:

$$\boldsymbol{DISL} = \frac{SLT - SLI}{T} \text{ (mm/day) * 1000 } \mu\text{m/mm,}$$

where: SL_T = shell length at time T (mm); SL_I = initial shell length (mm); T = number of rearing days.

Survival Rates

To measure the survival rates of abalone, the researchers first counted and recorded the initial abalone stock per cage. In Experiment 1, the number of surviving abalone in each treatment was recorded every 15 days, while in Experiment 2, counts were taken every 30 days. For consistency with Experiment 1, survival rate values in Experiment 2 on days 15, 45, and 75 were estimated through interpolation based on the 30-day measurement data.

For both Experiment 1 and Experiment 2, the survival rate for each cage was calculated by comparing the live count with the initial abalone stock.

The survival rate (SR) was computed using the formula:

$$\boldsymbol{SR} = \frac{Number\ of\ live\ abalone}{Total\ initial\ stock} \times 100\%.$$

The biomass of cultured abalone was calculated using the formula:

$$\boldsymbol{B} = \text{Total stock} \times \% \text{ Survival} \times W_M$$

where: W_M = mean body weight.

Statistical analysis was conducted on the collected data to identify trends in survival rates. Descriptive statistics, including mean and standard deviation, were used to summarize survival rates for each treatment. An independent samples t-test was performed to determine significant differences in survival rates between treatments in each experiment at $P < 0.05$. Additionally, repeated measures ANOVA was used to assess survival rate trends over time within each treatment.

Feed Conversion Efficiency

Feed conversion efficiency (FCE) is the ability of abalone to convert *Gracilaria* into body weight. In this study, FCE was calculated using data on food consumption and weight gain in Experiment 2. As detailed above in the feeding management section, feed consumption was estimated based on recorded weight of food before and after 15 days, assuming that the lost amount was consumed.

The feed conversion ratio (FCR) is calculated using the formula:

$$\boldsymbol{FCR} = \textit{Feed consumed / Weight gain.}$$

The feed conversion efficiency (FCE) is computed using the formula:

$$\boldsymbol{FCE} = \textit{(Weight gain / Feed consumed) x 100.}$$

Length-Weight Relationship

To analyze the length-weight (LW) relationship, all weight and SL data from Experiment 1 and Experiment 2 were grouped into three length classes (15-25, 26-35, 35-45 mm). The LW relationship was expressed using the equation: L = SL of the abalone in mm, W = aL^b, where W = weight of the abalone in g, a = intercept, b = slope (growth coefficient) (Le Cren 1951; Najmudeen 2015). The parameters a and b were calculated using linear regression in SPSS on log-transformed SL and weight data, with the coefficient of determination (R^2) used as an indicator of linear regression quality. A t-test was performed at a confidence level of 95% to confirm if the values of *br* (calculated *b* coefficient (or slope) from linear regression) obtained by linear regression were significantly different from the isometric value, expressed by the equations below (Najmudeen 2015).

Hypothesis: H_0: b0 = 3 (Isometric growth); H_A: $br \neq 3$ (Allometric growth)

The t-test formula to test the hypothesis: $\boldsymbol{t} = \frac{br - b0}{SE}$

where:

br – calculated *b* coefficient (or slope) from linear regression

b0 = 3

SE – standard error of *b* coefficient from linear regression

Data Analysis

Data analyses were conducted using SPSS version 26.0 for Windows. Descriptive statistics were applied to summarize and organize the dataset. An independent samples t-test was conducted to determine whether there were significant differences in WG, SGR, DIBW, SLI, DISL, SR, FCR, and FCE between treatments at $P < 0.05$.

To ensure consistency and reliability, the initial sample sizes for each treatment were standardized. The values of weight and SL were initially averaged across individual abalone juveniles within each cage, subsequently averaged across the three replicate cages per treatment to determine overall treatment effects. This method of averaging was maintained throughout the experiment.

In Experiment 1, the BS treatment utilized initial sample weights ranging from 0.41 to 3.31 g, with corresponding initial sample lengths between 13.60 and 26.60 mm. Meanwhile, the PS treatment had initial sample weights ranging from 0.45 to 3.29 g, with initial sample lengths from 14.10 to 26.20 mm. The final weights and lengths for each treatment group are detailed in Table 1.

In Experiment 2, the BS treatment featured initial sample weights between 1.00 and 5.95 g, with initial sample lengths ranging from 19.80 to 33.10 mm. The PS treatment, on the other hand, included initial sample weights between 1.30 and 5.65 g and initial sample lengths ranging from 20.00 to 32.20 mm.

RESULTS

Weight Growth Rates

Experiment 1, bottom sea cage culture. After 90 days of culture, abalone juveniles grown in bottom sea cages with BS and PS exhibited an increasing growth pattern (Figure 3). The weight mean of abalone juveniles with BS increased from 1.51 ± 0.70 g to 6.13 ± 2.17 g, while with PS increased from 1.51 ± 0.73 g to 4.77 ± 1.88 g (Table 2). A t-test revealed that abalone juveniles with BS gained weight significantly higher than those with PS. The mean weight gain of abalone juveniles with BS after 90 days was 358.13 ± 186.85%, compared to 279.71 ± 182.92% for those with PS (Table 2).

Abalone juveniles in both treatments exhibited the faster specific growth rate (SGR) after 45 days of culture, which then gradually declined. In general, the t-test showed that abalone juveniles with BS had significantly higher SGR ($P < 0.001$) compared to those with PS (Table 2).

Experiment 2, suspended sea cage culture. After 150 days of culture, abalone juveniles grown in suspended sea cages with BS and PS followed a similar increasing growth pattern, as shown in Figure 3. The mean weight of abalone juveniles with BS increased from 3.09 ± 1.19 g to 7.64 ± 2.20 g, while those with PS increased from 3.02 ± 1.04 g to 7.37 ± 2.28 g (Table 3). Abalone juveniles cultured with BS exhibited significantly higher mean weights from day 60 to day 120 ($P < 0.05$). Nevertheless, on day 150, a t-test showed no significant difference ($P = 0.27$) in mean weight between juveniles with BS and PS.

On day 60, abalone juveniles in the suspended sea cages reached their fastest SGR in both substrates, but this rate gradually declined. Based on the t-test results, there was no significant statistical difference ($P = 0.61$) in the growth rates of abalone juveniles with BS and PS.

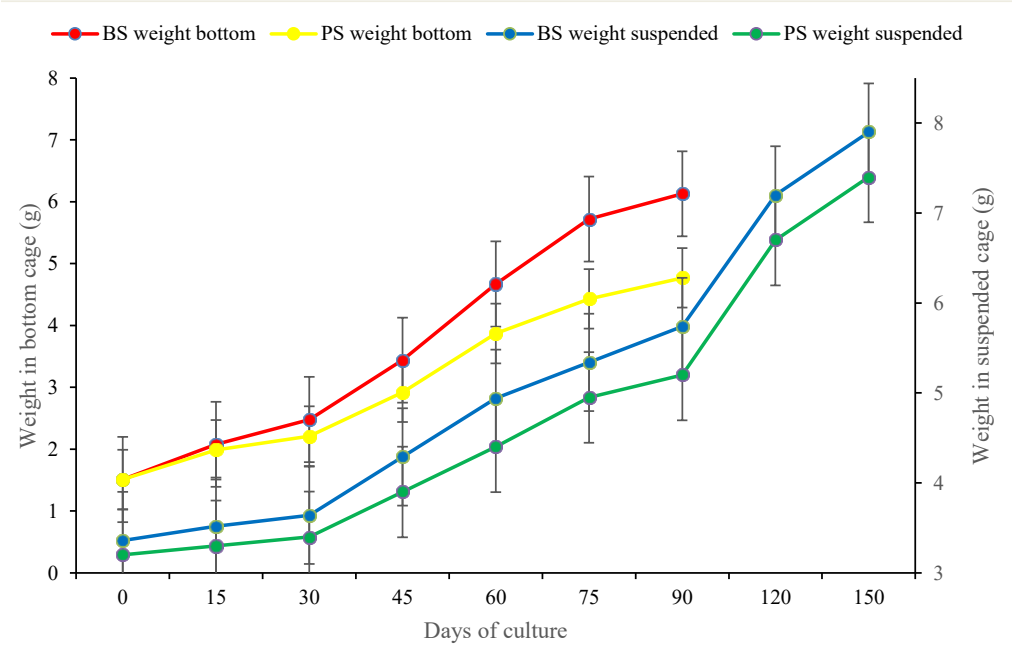

Figure 3. The mean (±SE) weight growth pattern of abalone juveniles with time across the two treatments: bamboo substrate (BS) and PVC substrate (PS) in the bottom and suspended sea cages. (Note: The BS and PS weights in suspended cages on days 15, 45, and 75 were estimated through interpolation.)

Table 2. Weight growth rates of abalone *Haliotis asinina* in bottom sea cages with bamboo substrate and PVC substrate fed with seaweed *Gracilaria firma* for 90 days (W_I – initial weight, W_F – final weight, W_G – weight gain, DIBW – daily increase in body weight, SGR – specific growth rate). *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$ (t-test).

Treatment	Mean W_I (g)	Mean W_F (g)	W_G (%)	DIBW (mg/day)	SGR (%)	Biomass (g)
Bamboo substrate	1.51 ± 0.70	6.13 ± 2.17***	358.13 ± 186.85**	51.13 ± 19.67***	1.61 ± 0.43***	631.17 ± 2.16
PVC substrate	1.51 ± 0.73	4.77 ± 1.64	279.71 ± 182.92	36.43 ± 15.00	1.37 ± 0.50	486.41 ± 1.64

Table 3. Weight growth rates of abalone *Haliotis asinina* juveniles in suspended sea cages with bamboo substrate and PVC substrate fed with seaweed *Gracilaria firma* for 150 days (W_I – initial weight, W_F – final weight, W_G – weight gain, DIBW – daily increase in body weight, SGR – specific growth rate).

Treatment	Mean W_I (g)	Mean W_F (g)	W_G [a] (%)	DIBW (g/day)	SGR (%)	Biomass (g)
Bamboo substrate	3.09 ± 1.15	7.64 ± 2.20	161.83 ± 103.83	0.03 ± 0.01	0.60 ± 0.24	1,336.45 ± 2.43
PVC substrate	3.02 ± 1.04	7.37 ± 2.28	155.96 ± 95.35	0.03 ± 0.01	0.58 ± 0.23	1,290.57 ± 2.31

Shell Length Growth Rates

Experiment 1, bottom sea cage culture. The SL in both substrates increased over 90 days of sea cage culture (Figure 4). The mean SL increased from 20.33 ± 3.22 mm to 32.06 ± 3.49 mm with BS, and from 20.14 ± 3.35 mm to 29.57 ± 3.21 mm with PS. A t-test revealed that abalone juveniles with BS had a significantly higher increment than those with PS (Table 4).

After 60 days, abalone juveniles in the bottom sea cages reached their highest SL growth rate. Thereafter, the SL growth rate gradually declined across all substrates. In general, abalone juveniles with BS had significantly higher DISL than those with PS (Table 4).

Experiment 2, suspended sea cage culture. The SL of abalone juveniles increased over 150 days of sea cage culture (Figure 4). The mean SL increased from 25.71 ± 3.05 mm to 33.88 ± 2.96 mm with BS, and from 25.77 ± 2.76 mm to 33.22 ± 2.96 mm with PS (Table 5). A t-test revealed no statistically significant difference in SL increment between juveniles with BS and PS ($P = 0.76$).

The DISL of abalone juveniles was nearly identical for both BS and PS. The results of the t-test confirmed that there was no statistically significant difference in DISL between two treatments ($P = 0.24$).

Survival Rates

Experiment 1, bottom sea cage culture. The survival rates of abalone juveniles in bottom sea cages with BS and PS were relatively high, at 95.37% and 94.4%, respectively (Figure 5). Mortality in this study was primarily due to mishandling during the removal of abalone juveniles from the substrates. However, the t-test showed no significant difference in the survival rates between the two substrates ($P = 0.76$).

Experiment 2, suspended sea cage culture. Similarly, in suspended sea cage culture, survival rates of abalone juveniles with BS and PS were relatively high. Figure 6 shows that the survival rates for BS and PS were 92.81% and 95.93%, respectively. Mishandling was again the primary cause of mortality, as juvenile abalone are difficult to remove from substrates. However, a t-test showed no significant difference in the survival rates between the two substrates ($P = 0.21$).

Table 4. Shell length growth rates of abalone *Haliotis asinina* juveniles in bottom sea cages with bamboo substrate and PVC substrate fed with seaweed *Gracilaria firma* for 90 days (SL_I-initial shell length, SL_F-final shell length, SLI-shell length increment, DISL-daily increase in shell length). *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$ (t-test).

Treatment	SL_I (mm)	SL_F (mm)	SLI (%)	DISL (µm/day)
Bamboo substrate	20.33 ± 3.22	32.06 ± 3.49	59.65** ± 22.37	129.35*** ± 34.52
PVC substrate	20.14 ± 3.35	29.57 ± 3.21	50.46 ± 22.67	105.84 ± 34.77

Table 5. Shell length growth rates of abalone *Haliotis asinina* juveniles in suspended sea cages with bamboo substrate and PVC substrate fed with seaweed *Gracilaria firma* for 150 days (SL_I-initial shell length, SL_F-final shell length, SLI-shell length increment, DISL-daily increase in shell length).

Treatment	SL_I (mm)	SL_F (mm)	SLI (%)	DISL (mm/day)
Bamboo substrate	25.71 ± 3.05	33.88 ± 2.96	31.76 ± 15.82	0.053 ± 0.021
PVC substrate	25.77 ± 2.76	33.22 ± 2.96	29.49 ± 16.91	0.049 ± 0.022

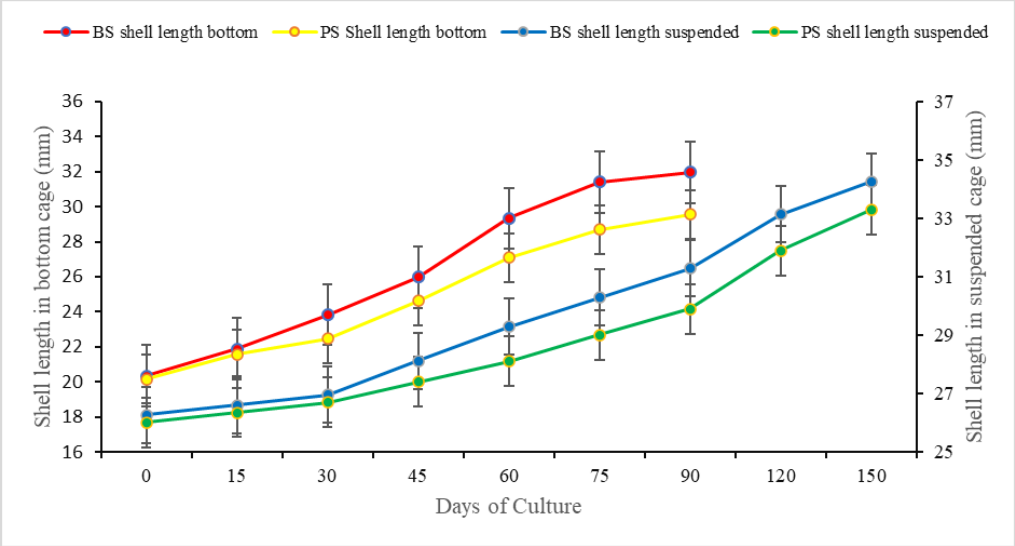

Figure 4. The mean (±SE) shell length growth pattern of abalone juveniles with time across the two treatments (bamboo substrate (BS) and PVC substrate (PS) in the bottom and suspended sea cages. (Note: The BS and PS shell length in suspended cages on days 15, 45, and 75 were estimated through interpolation.)

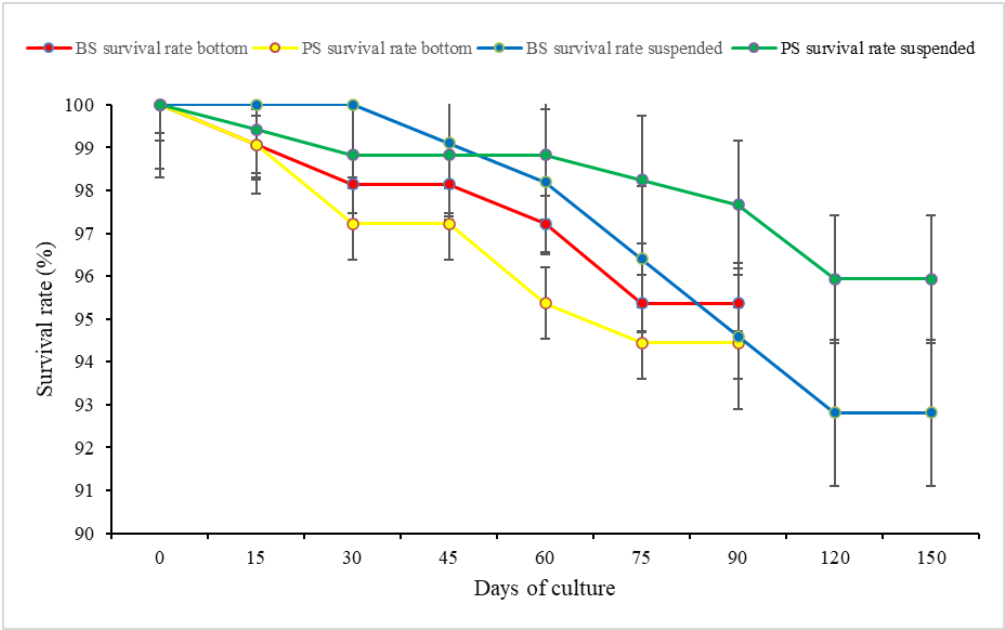

Figure 5. The survival rate (±SE) of abalone juveniles with time across the two treatments: bamboo substrate (BS) and PVC substrate (PS) in the bottom and suspended sea cages. (Note: The BS and PS survival rates in suspended cages on days 15, 45, and 75 were estimated through interpolation.)

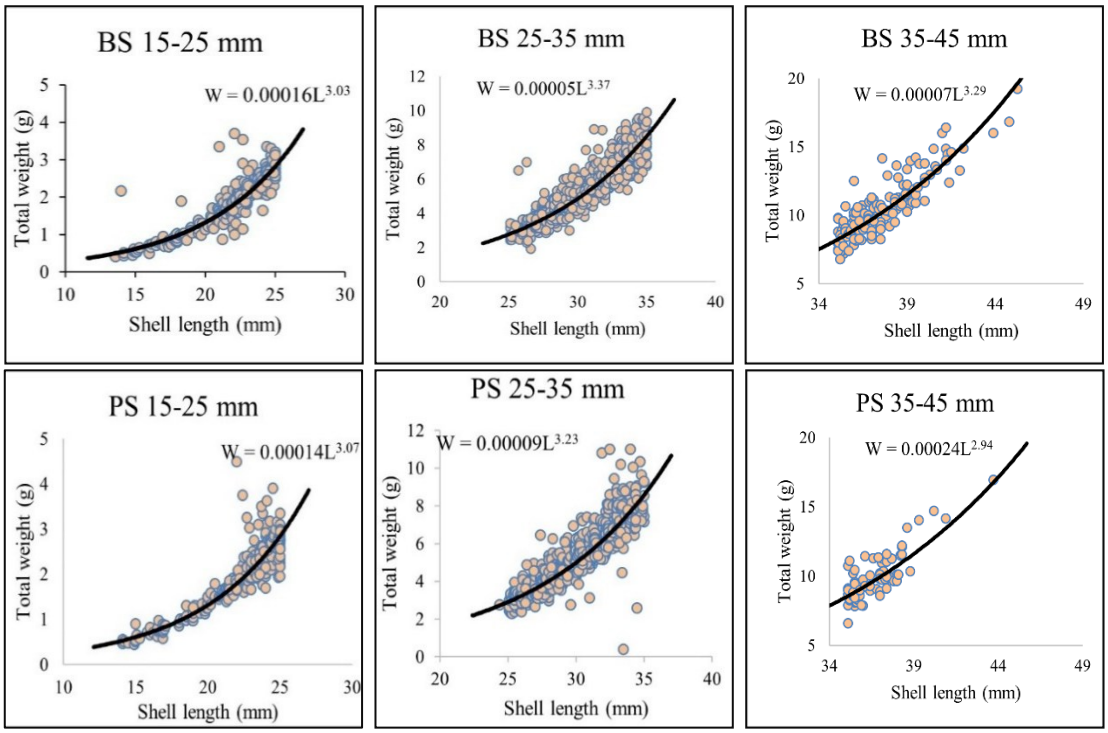

Figure 6. Length-weight relationship of abalone *Haliotis asinina* cultured with bamboo substrate (BS) and PVC substrate (PS) for 15-25 mm shell length (BS: b = 3.03, isometric, PS: b = 3.07, isometric), 25-35 mm shell length (BS: b = 3.37, allometric +, PS: b = 3.23, allometric +), and 35-45 mm shell length (BS: b = 3.29, isometric, PS: b = 2.94, isometric).

Food Conversion Efficiency

The results demonstrated that FCE was higher for abalone juveniles with BS (11.86%) than for those with PS (9.86%). In terms of FCR, approximately 10.10 kg of *Gracilaria* algal food was needed to produce 1 kg of abalone with PS, compared to a relatively lower FCR of 8.43 for abalone with BS (Table 6).

Length and Weight Relationships

Experiment 1, bottom sea cage culture. Abalone juveniles reared with BS (b = 2.89) and PS (b = 3.00) initially showed isometric growth. However, after 90 days, only abalone juveniles with BS (b = 3.15) transitioned to positive allometric growth, while those with PS (b = 2.95) continued to exhibit isometric growth. The LW relationship analysis showed a reasonably good fit for both substrates: BS (SL 24–42 mm, R^2 = 0.93) and PS (SL 22–38 mm, R^2 = 0.779). The relative condition factor (Kn) remained nearly unchanged from the start of the experiment (BS = 1.03, PS = 1.01) to the end of the experiment (BS = 1.0, PS = 1.02) (Table 7).

Experiment 2, suspended sea cage culture. Abalone juveniles reared with BS (b = 2.61) initially exhibited negative allometric growth, while those with PS (b = 3.03) showed isometric growth. However, by the end of the experiment, abalone juveniles with BS had transitioned to positive allometric growth, whereas those with PS continued to exhibit isometric growth. The LW relationship analysis indicated a reasonably good fit for both substrates: BS (SL 26–42.20 mm, R^2 = 0.878) and PS (SL 24.40–43.70 mm, R^2 = 0.861). The Kn at the beginning of the experiment showed good health conditions (BS = 1.02, PS = 1.04), and both treatments maintained relatively good health conditions by the end (BS = 1.00 and PS = 1.04) (Table 8).

Figure 6 illustrates the LW relationships of *H. asinina* across different size groups. The 15-25 mm size group showed isometric growth in both BS and PS. The Kn values were similar across substrates, indicating healthy growth. The R^2 values were high, indicating that the model was a good fit for the data (Table 8).

For the 25-35 mm size group, the growth shifted to a positive allometric+ for both substrates, with a higher b-value or allometric exponent in BS than in PS. A higher b indicates a faster growth rate for the BS. The Kn values continued to indicate healthy growth. The R^2 values remained high for both substrates, indicating a good fit of the model to the data (Table 8).

For the 35-45 mm size range, the growth reverted to isometric for both BS and PS. The b

coefficient was higher for BS than PS, which is supposed to indicate faster growth, but the t-test revealed that both groups still exhibited isometric growths. The Kn value remained 1.01 for both substrates, which still indicates healthy growth. The R^2 value was a good fit for BS, but lower for PS, indicating a weaker correlation among the data (Table 8).

Table 6. Food conversion ratio and food conversion efficiency of abalone *Haliotis asinina* juveniles in suspended sea cages with bamboo substrate and PVC substrate fed with *Gracilaria firma* for 45 days (FCR-feed conversion ratio, FCE-feed conversion efficiency).

Treatment	Length Range (mm)	Weight Gained (g)	Food Consumed (g)	FCR	FCE (%)
Bamboo substrate	1.16 – 4.68	54.6	460.32	8.43	11.86
PVC substrate	1.28 – 3.44	50.02	505.42	10.10	9.86

Table 7. Length-weight relationship parameters of the abalone *Haliotis asinina* juveniles between the two substrates: bamboo substrate and PVC substrate in bottom sea cages for 90 days culture (*a*–the line intercept, *b*–the slope/constant, Kn–the relative condition factor).

Substrate	N	Days of Culture	Length Range (mm)	*a*	*b*	Kn	SE (b)	R^2	P-value	Growth Type
Bamboo substrate (BS)	108	0	13.6 – 26.60	0.00023	2.89	1.03	0.113	0.861	< 0.001	Isometric
PVC substrate (PS)	108	0	14.10 – 26.20	0.00017	3.00	1.01	0.069	0.947	< 0.001	Isometric
Bamboo substrate (BS)	103	90	24.0-42.0	0.00011	3.14	1.0	0.089	0.930	< 0.001	Allometric +
PVC substrate (PS)	102	90	22.0-38.0	0.00021	2.95	1.02	0.166	0.779	< 0.001	Isometric

Table 8. Length-weight relationship parameters of the abalone *Haliotis asinina* juveniles between the two substrates: bamboo substrate and PVC substrate in suspended sea cages for 150 days culture (*a*–the line intercept, *b*–the slope/constant, Kn–the relative condition factor).

Substrate	N	Days of Culture	Length Range (mm)	*a*	*b*	Kn	SE (b)	R^2	P-value	Growth Type
Bamboo substrate	167	0	19.8 – 33.10	0.00061	2.61	1.04	0.150	0.646	< 0.001	Allometric -
PVC substrate	172	0	20.00 – 32.20	0.00015	3.03	1.02	0.082	0.890	< 0.001	Isometric
Bamboo substrate	155	150	26.0 - 42.20	0.00009	3.23	1.00	0.098	0.878	< 0.001	Allometric +
PVC substrate	165	150	24.40 - 43.70	0.00011	3.15	1.04	0.099	0.861	< 0.001	Isometric

DISCUSSION

Growth Rates

This study provides valuable insights into the use of bamboo substrate (BS) for rearing abalone juveniles, highlighting its potential as an effective alternative to PVC substrate (PS). The results showed that abalone juveniles raised with BS exhibited significantly higher weight gain and SL growth rates than those reared with PS. These findings are consistent with the previous study by Creencia et al. (2021), which suggested that BS could serve as a viable substrate for abalone juvenile culture in bottom sea cages.

In the two experiments, the researchers investigated the impact of BS on the growth of abalone juveniles in both bottom and suspended sea cage culture at different stages of development. Although periphytic algae abundance was not directly measured in this study, the periphyton presence on the bamboo slats was consistently observed throughout the experiment. Previous studies (e.g. Keshavanath et al. 2004; Zhang et al. 2013; Creencia et al. 2019) have supported the influence of periphytic algae in

enhancing abalone growth, which demonstrated higher periphyton biomass on BS than other materials. Future studies should quantify periphyton abundance to better understand its contribution to abalone growth performance. This periphyton biomass may have served as an additional food source, promoting the enhanced growth of abalone juveniles. This observation is consistent with the study of Keshavanath et al. (2004), which found that periphyton on bamboo poles could supplement fish feed in tilapia culture, resulting to increased yields compared to systems without bamboo poles. Similarly, Wahab et al. (1999) reported that the use of BS significantly ($P < 0.05$) increased growth and production of Indian carp in ponds culture.

Compared to the previous study on a 90-day culture period, the experiment using BS in bottom sea cage culture yielded higher DIBW (51.13 ± 19.67 mg/day) and SGR (1.61 ± 0.43%) than the findings of Bautista-Teruel and Millamena (1999), who used PS, tank culture, different algal (*Gracilaria bailinae* J.F.Zhang & B.M.Xia, 1994) feeds, and a smaller initial stocks (DIBW = 10 mg/day; SGR = 0.06 ± 0.05). In contrast, the results were lower than those reported by Capinpin and Corre (1996) (DIBW = 67.1 mg/day; DISL = 192.9 µm/day). The faster growth rate in Capinpin and Corre's (1996) study could be attributed to their smaller initial stock, different algal (*Gracilaria heteroclada* J.Feldmann & G.Feldmann, 1943) feed, and lower stocking density. Smaller juveniles generally grow faster due to higher feeding rates per unit biomass (Capinpin et al. 1999; Minh et al. 2010). Abalone juvenile growth decreases as stocking density increases due to density-dependent competition for space or food, which affects feeding rates and movement efficiency during feeding (Capinpin et al. 1999). The algal (*G. heteroclada*) feed used by Capinpin and Corre (1996) could also be a factor, which has a higher crude protein content of 17.32% compared to only 13.4% in the algal (*G. firma*) feed used in this study (Table 1). Although the protein content of the food may affect abalone growth rates, this may not always be the determining factor. Bautista-Teruel and Millamena (1999) reported lower growth rates despite using feed with higher protein content (*G. bailinae* – 17.56%) and a smaller size of initial stock. These results indicate that culture conditions, including initial size, stocking density, type of food, temperature, water quality, and substrate, significantly influence abalone juvenile growth (Capinpin et al. 1999; Steinarsson and Imsland 2003; Alcantara and Noro 2005; Minh et al. 2010). These factors need to be considered when comparing growth results across different studies. Differences in culture methods across studies makes it difficult to draw general conclusions about abalone juvenile growth performance. Therefore, it is essential to clearly describe rearing methods when comparing results to ensure consistency in evaluations. By doing so, the scientific community can better understand the factors influencing abalone juvenile growth and make informed decisions about abalone farming practices.

In the suspended sea cage experiment, abalone juveniles were grown in suspended cages near the water surface using BS and PS. The results showed that weight gain was significantly higher with BS than with the PS after 90 days of culture ($P < 0.01$). However, after 150 days, the difference became statistically insignificant ($P < 0.05$). In terms of SL, the abalone juveniles raised with BS maintained a significant advantage over those with PS throughout the 150-day culture period, consistent with the bottom sea cage experiment. These findings suggest that BS can serve as a cost-effective and locally available alternative to PS for the grow-out culture of abalone juveniles in long-line aquaculture systems.

Both experiments showed a notable decline in the growth rate of abalone juveniles towards the later stages of culture. This decline in daily growth rates is likely attributed to the onset of gonad maturation, which requires substantial energy allocation. At the end of the study, the SL range of the abalone juveniles ranged from 32.06 mm to 34.27 mm, approaching the size threshold for sexual maturity (Capinpin et al. 1998). It is well established that the growth rate of abalone juveniles decrease after reaching sexual maturity, as energy is redirected towards gonad development rather than somatic growth (Capinpin and Corre 1996; Mai et al. 1996).

Survival Rates

The handling of abalone juveniles during sampling may have affected their growth and immunity due to stress caused by manual detachment and placement in plastic containers without aeration (Hooper et al. 2011; Daw 2022). Mishandling during sampling was one of the causes of the mortality of abalone juveniles in this study. Additionally, fluctuations in water quality due to adverse weather conditions may further influence their survival rate. Despite these challenges, the survival rates for BS in both experiments remained relatively high, ranging from 92.78% to 99.17%. Another contributing factor to abalone mortalities in this study was the presence of crabs inside the cage. Aspe et al. (2019) reported that swimming crabs (*Thalamita crenata* Rüppell, 1830 and *Charybdis natator* Herbst, 1794*)* prey on abalone juveniles in the Pamantolon area of Taytay, Palawan, where the suspended cage experiment was conducted. Predators in enclosed systems, such as cages, pose a significant threat to abalone juvenile farming, as the cultured animals have no means of escaping. Additionally, cage damage may allow predators to enter or juveniles to escape, both of which negatively affect production. Schiel and Welden (1987), in their study on the response of red abalone (*Haliotis rufescens* Swainson, 1822) to predators, found that crabs and lobsters consumed more abalone than sea

stars. Furthermore, Tegner et al. (1989) identified predation as one of the leading causes of failed attempts to enhance natural abalone populations. The use of black netting or double netting in cages was observed in the experiments to be an effective deterrent against potential abalone juvenile predators. The high survival rates in cages with BS substrate suggest that BS may serve as a viable, cost-effective, and sustainable alternative for abalone juvenile culture.

Feed Conversion Efficiency

Regarding FCE, the presence of BS provided a significant advantage by promoting more efficient food utilization and growth in abalone juveniles compared to PS. The improved FCE of abalone juveniles observed with BS can be attributed to several factors. The BS provides attachment to facilitate feeding, maximizes accessibility to food resources (Setyono 2015), and serves as a habitat for microscopic food sources, such as benthic diatoms and other organisms that settle and attach to the substrate. These organisms contribute to the overall food availability and enhance the nutritional quality of the abalone diet (Wahab et al. 1999; Keshavanath et al. 2004; Zhang et al. 2013). Furthermore, the substrate creates a sheltered environment for abalone, reducing stress and providing a sense of security. This favorable environment likely enhances their feeding efficiency and overall growth performance (Setyono 2015). It has been suggested that the growth of *H. asinina* is influenced by the availability of attachment space or shelter (Fermin and Buen 2002). By offering a designated substrate for attachment, abalones have increased access to food resources, which may result in improved FCE.

Additionally, BS offers advantages in terms of its composition and degradation properties. As a biodegradable material, BS provides a natural and environmentally friendly option compared to the PS, a non-biodegradable synthetic material. The BS creates a more natural and suitable habitat for abalone, mimicking their natural environment and promoting their overall well-being. This may positively influence their feeding efficiency and subsequently improve FCE (Setyono 2015).

It is worth noting that PS has been widely employed in abalone culture due to its durability and practicality. However, this study's findings suggest that BS offers advantages in terms of FCE, promoting more efficient feed utilization and growth.

Length-Weight Relationships

The LW relationship analysis assessed the growth performance of abalone juveniles reared with BS compared to PS. This study found that abalone juveniles with both BS and PS initially showed isometric growth, meaning there was a proportional increase in weight and SL. However, after 90 days in the bottom cage experiment, abalone juveniles with BS showed a positive allometric growth pattern (b = 3.14), while abalone juveniles with PS continued to show isometric growth (b = 2.95) (Table 7). The isometric growth of abalone juveniles with PS (SL range = 22-38 mm, mean = 29.57 mm) is consistent with the study of Najmudeen (2015), which also reported isometric growth of abalone juveniles in the SL size range of 25–35 mm. Interestingly, the abalone juveniles with BS (SL range = 24–42 mm, mean = 32.06 mm) showed a positive allometric growth pattern in this study.

A LW model was created to further verify this growth pattern based on all the weight and length data in two experiments. The result of the LW model for abalone juveniles with a SL of 26-35 mm in BS ($W=0.00005L^{3.37}$) and PS ($W=0.00009L^{3.23}$) showed positive allometric growth (Table 8). The SL 26-35 mm LW model also showed a good fit for both substrates, with high R^2 values (BS = 0.87, PS = 0.75) indicating a strong correlation between weight and SL. Additionally, the Kn remained consistently good for both treatments from the initial day of culture until the end of the experiment (BS = 1.0, PS = 1.02) (Table 9).

Table 9. Length-weight relationship of reared abalone *Haliotis asinina* juveniles between the two substrates: bamboo substrate and PVC substrate in various size groups (*a*–the line intercept, *b*–the slope/constant, Kn–the relative condition factor).

Substrate	Length Range (mm)	*a*	*b*	Kn	SE (b)	R^2	P-value	Growth Type
Bamboo substrate	15 - 25	0.00016	3.03	1.03	0.102	0.76	< 0.001	Isometric
PVC substrate	15 - 25	0.00014	3.07	1.01	0.064	0.90	< 0.001	Isometric
Bamboo substrate	25 - 35	0.00005	3.37	1.01	0.043	0.87	< 0.001	Allometric +
PVC substrate	25 - 35	0.00009	3.23	1.05	0.065	0.75	< 0.001	Allometric +
Bamboo substrate	35 - 45	0.000068	3.29	1.01	0.194	0.65	< 0.001	Isometric
PVC substrate	35 - 45	0.00024	2.94	1.01	0.443	0.38	< 0.001	Isometric

The suspended cage experiment revealed that the abalone juveniles reared with BS (b = 2.61) exhibited negative allometric growth initially, while those reared with PS showed isometric growth (BS = 3.03). However, towards the end of the experiment, the abalone juveniles with BS displayed positive allometric growth, while those with PS remained isometric. The LW relationship analysis indicated a reasonably good fit for both substrates. The Kn at the beginning of the experiment showed good health conditions for both treatments. By the end of the experiment, both treatments maintained relatively good health conditions. One possible explanation for the observed differences in growth rates between the BS and PS treatments is the development of periphytic algae on the bamboo slats used in BS. Periphytic algae can provide an additional food source for abalone juveniles, which could contribute to faster growth rates (Zhang et al. 2013; Creencia et al. 2019). Additionally, the supply of drift algae, which can vary in composition depending on location and water movement, could also affect the growth rate of abalone juveniles in different treatments (Tutschulte and Connel 1988). Previous studies have shown that the composition of drift algae can influence the diet and growth of abalone juveniles (Poore 1972a, 1972b), and areas with higher water movement tend to have higher growth rates, although there is an optimal degree of water movement for feeding (Shepherd 1973; McShane et al. 1988).

Generally, the results suggest that both BS and PS are suitable for abalone juvenile culture, as the abalone maintained good health throughout the culture period. However, using bamboo slats may have contributed to faster growth rates due to the development of periphytic algae, highlighting the potential for using alternative substrates or modifying substrate design to improve abalone juvenile growth performance. These results suggest substrate choice can significantly impact abalone juveniles' growth and health. The BS may be more favorable for abalone juvenile growth and development than the PS. However, the suitability of the substrate may change as the abalone juveniles grow and develop. Therefore, a careful evaluation of substrate choice is necessary throughout the different stages of the abalone juvenile culture. Further studies are needed to investigate the underlying mechanisms of how substrates affect abalone juvenile growth and development.

This study highlights the potential of using BS as an alternative to PS for abalone juvenile rearing. The results show that abalone juveniles reared with BS had significantly higher weight and SL growth rates than those raised with PS. They also had comparable survival rates and feed conversion efficiency with PS. This suggests that BS can be a cost-effective and sustainable alternative for abalone juvenile culture in cages.

Overall, using BS in abalone juvenile rearing could be a promising approach for sustainable and cost-effective abalone juvenile culture. This could encourage farmers to use low-cost and locally available materials for abalone juvenile grow-out culture. The results of these experiments suggest that the choice of substrate can significantly impact the growth and health of abalone juveniles. Additionally, the LW relationship analysis can provide useful insights into the growth patterns of abalone juveniles under different rearing conditions.

These findings have significant implications for sustainable abalone juvenile culture development, as using locally available materials can reduce costs and improve efficiency while promoting environmental sustainability. Ultimately, these findings can contribute to the development of a sustainable and eco-friendly abalone culture, benefiting both the environment and the economy.

Further research is needed to confirm these findings and evaluate the effectiveness of BS in different ecological contexts. To address these limitations, future research should confirm the study's findings by measuring differences in the abundance and nutritional quality of periphytic algae in BS and PS. It should also include qualitative methods and involve stakeholders in similar systems to provide insights into unexpected interactions and feedback loops.

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GENERATIVE AI STATEMENT

Artificcial intelligence was not used in the conduct and writing of this study exceot for minor grammatical checking.

ETHICAL CONSIDERATIONS

This study followed all institutional and national ethical guidelines for the care and use of experimental abalone juveniles.

DECLARATION OF COMPETING INTEREST

The authors declare that there are no competing interests.

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