

# Optimizing growth and shell pigmentation in *Cherax quadricarinatus* juveniles using purple sweet potato extract

<sup>1</sup>Hany Handajani, <sup>1</sup>Fathimatuz Zahroh, <sup>1</sup>Dony Prasetyo, <sup>1,2</sup>Soni Andriawan

<sup>1</sup>Department of Aquaculture, Faculty of Agriculture and Animal Science, University of Muhammadiyah Malang, Malang, East Java, Indonesia; <sup>2</sup>Biotechnology Development Center, University of Muhammadiyah Malang, Malang, East Java, Indonesia.  
Corresponding author: H. Handajani, handajani@umm.ac.id

**Abstract.** The red claw crayfish (*Cherax quadricarinatus*) is a freshwater crustacean with promising aquaculture potential due to its adaptability and economic value. This study investigated the effects of purple sweet potato (*Ipomoea batatas*) extract as a natural feed additive on the growth and shell colouration of juvenile *Cherax quadricarinatus*. The experiment employed four dietary treatments: a control diet without extract (P1) and diets supplemented with 10% (P2), 20% (P3), and 30% (P4) purple sweet potato extract over 30 days. Statistical analysis ( $p < 0.05$ ) revealed that P2 exhibited superior growth performance, with the highest absolute weight ( $0.99 \pm 0.39$  g), specific growth rate ( $1.60 \pm 1.07\%$ ), and survival rate ( $80.00 \pm 8.16\%$ ). Additionally, colour intensity was optimized at 10% extract concentration, achieving the highest values by Week 4. However, higher extract levels (20% and 30%) reduced carotenoid absorption and less efficient feed conversion ratios, indicating a dose-dependent effect. Carotenoid content decreased with increasing extract concentrations, with P1 and P4 showing the highest ( $71 \mu\text{mol g}^{-1}$ ) and lowest ( $36 \mu\text{mol g}^{-1}$ ) absorption, respectively. Water quality parameters, including temperature, pH, and dissolved oxygen, remained within optimal ranges for *C. quadricarinatus*, ensuring consistent results. The findings highlight the potential of purple sweet potato extract as a sustainable alternative to synthetic additives, enhancing growth and colour intensity while supporting environmentally friendly aquaculture practices. However, excessive supplementation may hinder carotenoid absorption and feed efficiency. This research provides valuable insights into optimizing natural additives for aquaculture feed to improve growth, pigmentation, and sustainability in crustacean farming.

**Key Words:** aquaculture feed additives, carotenoid absorption, feed formulation, natural pigment.

**Introduction.** Aquaculture has become a pivotal component of global food security, addressing the growing demand for sustainable and high-quality protein sources (Azra et al 2021). Among the numerous species cultivated within this industry, the freshwater lobster (*Cherax quadricarinatus*), commonly referred to as the red claw crayfish, has attracted significant attention due to its robust growth, adaptability, and high market value (Garza de Yta et al 2012; Pyatikopova et al 2023; Zharchynska & Hrynevych 2023). As a species native to the freshwater systems of northern Australia and Papua New Guinea, this prawn has proven to be an economically viable option for aquaculture, especially in regions looking to diversify their aquatic farming practices (Haubrock et al 2021; Hou et al 2023; Snovsky & Galil 2011).

In aquaculture, two primary factors govern the commercial success of any species: growth performance and marketability (Devlin et al 2020; Janssen et al 2017). For *C. quadricarinatus*, marketability is closely linked to the aesthetic appeal of the lobsters, particularly the brightness and vibrancy of their shell coloration (Mauro et al 2024; Rakhmawati et al 2023). Coloration, which is influenced by genetic and environmental factors, is not merely a cosmetic trait but also an indicator of the lobster's overall health and nutritional status (Ghonimy et al 2023; Leema et al 2010). Therefore, enhancing shell color holds dual importance, improving consumer appeal while simultaneously reflecting the health and vitality of the species.

Historically, synthetic carotenoids and other artificial colorants have been used to enhance the pigmentation of aquatic species (Pereira da Costa & Campos Miranda-Filho 2020). However, the increasing awareness of the environmental and health risks associated with synthetic additives has led to a shift toward exploring natural alternatives (Aguilar-Pérez et al 2023; Bolek 2022). Plant-derived pigments, particularly anthocyanins, have emerged as a promising category of natural color enhancers (Lv et al 2023; Saati et al 2022). Anthocyanins, which are responsible for the red, purple, and blue hues found in various fruits and vegetables, are not only potent antioxidants but also exhibit a range of bioactive properties that can positively influence growth and health in aquatic species (Cammareri et al 2024; Khoo et al 2017; Mohammadi et al 2024).

The purple sweet potato (*Ipomoea batatas*), a widely cultivated root vegetable, is particularly rich in anthocyanins (Farida et al 2024; Sumartini 2023). Purple sweet potato extract in aquaculture represents a novel approach to enhancing the growth and coloration of species such as *C. quadricarinatus*. The dual functionality of purple sweet potato extracts as a natural colorant and growth promoter presents a sustainable alternative to synthetic additives (Gagas et al 2023; Hartoto et al 2024). Moreover, incorporating plant-based bioactives into aquaculture feed aligns with broader goals of reducing the environmental footprint of aquaculture and meeting the rising consumer demand for naturally derived products (Chandra Mohana et al 2023; Onomu & Okuthe 2024; Presenza et al 2023).

This study aimed to examine the effects of purple sweet potato extract on the growth performance and coloration of *C. quadricarinatus* juveniles. The research was based on the hypothesis that including anthocyanin-rich extract in these lobsters' diet would lead to enhanced growth rates and improved shell coloration. By analyzing these outcomes, the study aimed to thoroughly understand the potential benefits of using purple sweet potato extract as a feed additive in aquaculture.

The significance of this research extended beyond the immediate context of *C. quadricarinatus* cultivation. As the aquaculture industry continues to grapple with sustainability challenges and environmental impact, developing natural feed additives represents a critical area of innovation. The findings of this study are expected to contribute to the growing body of knowledge on the application of plant-based bioactives in aquaculture, offering insights that could inform the practices of producers worldwide. Furthermore, by advancing the use of natural additives that enhance growth and coloration, this research could potentially improve the economic viability and market competitiveness of freshwater lobster farming, thereby supporting the industry's long-term sustainability.

**Material and Method.** This research was conducted at the Fisheries Laboratory, Faculty of Agriculture and Animal Science, Universitas Muhammadiyah Malang, from September 4<sup>th</sup> to October 18<sup>th</sup> 2023. The study employed an experimental method using a Completely Randomized Design (CRD) with four treatments and four replications, as follows:

- P1: Artificial feed without sweet potato extract;
- P2: Artificial feed supplemented with 10% sweet potato extract;
- P3: Artificial feed supplemented with 20% sweet potato extract;
- P4: Artificial feed supplemented with 30% sweet potato extract.

**Prawn preparation.** One hundred sixty *C. quadricarinatus* (3 to 5 cm) were obtained from a breeder and quarantined for three days without feeding. During the quarantine process, the prawns were provided with aeration and a heater to maintain the oxygen level and temperature within the fibre quarantine tank.

#### **Feed preparation**

**Preparation of fish silage flour.** The scad fish was obtained from the Sendang Biru Fish Auction Facility in the Sumbermanjing Wetan District of Malang, East Java. Preparing fish silage flour involved cleaning 5 kg of scad fish, grinding them, and placing them into a container. Then, 150 mL of lactic acid bacteria and 20% molasses were added, and the

mixture was fermented for 5 days. After fermentation, the mixture was dried in an oven at 50°C for 2 days, then ground with a mixer grinder and sieved to obtain a fine powder.

**Preparation of purple sweet potato extract.** The fully mature and deep purple sweet potatoes were acquired from the Sawojajar market in the Kedung Kandang District of Malang City, East Java, Indonesia. They were further utilized in the Fisheries Laboratory at the University of Muhammadiyah Malang. Afterward, they were thoroughly cleaned before 1 kg was sliced into thin or small pieces and dried under the sun until all moisture was removed. Moreover, dried sweet potatoes were blended into a powder and macerated with ethanol as a solvent, in a ratio of 1:10, in glass jars (Zubaidah et al 2021). The mixture was stirred once daily for three days, filtered, then collected in a tray and dried in an oven set to 50°C for two days. Finally, the dried material was sieved until a fine powder was achieved. The resulting extract, now a fine purple powder, was utilized as the material for this study.

**Feed preparation.** The stages involved in preparing the test feed begin with selecting and preparing raw materials, determining the nutritional content of the feed ingredients, and formulating the feed. The preparation phase for the test feed includes choosing and preparing ingredients such as fish silage meal, soybean meal flour, wheat flour, fish oil, vitamin mix, carboxymethyl cellulose (CMC), and purple sweet potato extract. Before preparing the feed, it was essential to establish the nutritional content of each ingredient through proximate analysis, as detailed in Table 1. The preparation began by weighing the raw materials according to the feed formulation in Table 2. The ingredients were added to the mixing container, starting with the smallest quantity and gradually incorporating larger amounts, ensuring a homogeneous mixture at each step. The purple sweet potato extract, used as an additive to enhance color brightness, was mixed thoroughly with the other components until evenly distributed. The final mixture was then stored in labeled plastic bags according to the specific treatments. The feed was administered to *C. quadricarinatus* using the paste method.

Table 1

Proximate composition of feed ingredients

<i>Ingredient</i>	<i>Protein (%)</i>	<i>Fat (%)</i>	<i>Ash (%)</i>	<i>NFE (%)</i>
Fish silage meal	57.76	5.93	13.52	23.86
Soybean meal flour	50.04	2.12	7.16	63.55
Wheat flour	8.90	1.30	0.00	98.75
Fish oil	0.00	100.00	0.00	0.00
Vitamin mix	0.00	0.00	0.00	0.00
CMC (Carboxymethyl cellulose)	0.00	0.00	0.00	0.00
Purple sweet potato extract	0.00	0.00	0.00	0.00

Table 2

Feed formulation

<i>Ingredient</i>	<i>Requirements (gram)</i>
Fish silage meal	30
Soybean meal flour	30
Wheat flour	30
Fish oil	4
Vitamin mix	3
CMC	3
Total	100

**The proximate analysis.** Proximate analysis evaluates feed nutrients, including protein, fat, ash, and nitrogen-free extract (NFE). The methods used included drying moisture at

105°C, determining protein via the Kjeldahl method, extracting fat by Soxhlet, incinerating ash at 550°C, and digesting fiber with acid and alkali (AOAC 2012; Paul et al 2023; Saputri & Putri 2024). NFE was calculated by subtracting other components, which ensures a comprehensive feed evaluation.

**Observation of color intensity in *C. quadricarinatus*.** The observation of color intensity in *C. quadricarinatus* was based on a color finder table with a scale ranging from 1 to 10. This method involved comparing the natural body color of freshwater prawn with a color finder chart, where each shade is assigned a specific value. The observation focused on colors closely matching the lobster's body color. The assessment begins by ranking the scores from the lowest (1) to the highest (10), representing a gradient from light brown to dark or deep brown. The score determined the level of color brightness; the lower the score, the brighter the color intensity of the lobster (Figure 1). Color intensity observations were conducted weekly, or every 7 days, over 30 days, with a specific focus on shades of brown (Budi et al 2021).

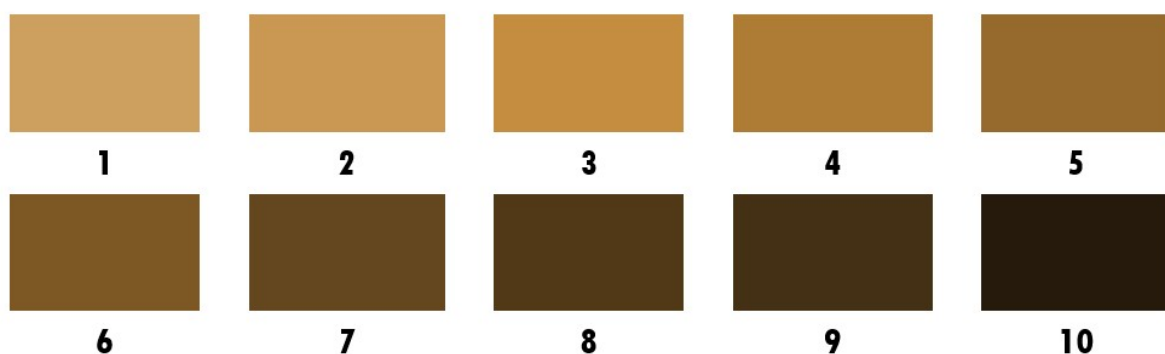


Figure 1. Freshwater lobster color finder chart.

This figure represents the color finder chart used to assess the color intensity of freshwater lobsters. The chart typically includes a range of shades from light brown to dark brown, with corresponding values from 1 to 10, which are used to evaluate the lobster's body color intensity during the observation period.

**Carotenoid analysis.** Solvents such as acetone were used for the carotenoid extraction from the feed to assess its content, contributing to coloration and providing health benefits. The extract was then purified through chromatography to separate non-carotenoid components. The purified extract was analyzed using a spectrophotometer at specific wavelengths (approximately 480, 663, and 645 nm), and the absorbance readings were utilized to determine carotenoid concentration based on a standard curve. The total carotenoid content was expressed in milligrams per kilogram ( $\text{mg kg}^{-1}$ ) of the feed (Sukarman & Hirnawati 2014). The carotenoid content in the sample is calculated using the following formula:

$$\text{Carotenoid } (\mu\text{mol g}^{-1}) = \frac{(A_{480} + 0.114 \times A_{663} - 0.638 \times A_{645}) \times V \times 10^3}{112.5 \times W}$$

Where:

A480 - absorbance value at a wavelength of 480 nm;

A645 - absorbance value at a wavelength of 645 nm;

A663 - absorbance value at a wavelength of 663 nm;

V - volume of the extract (mL);

W - weight of the sample (g).

**Growth performance and survival rate analysis.** The growth performances of *C. quadricarinatus*, in addition to their survival rates, were also examined. These analyses included the growth rate (GR), the absolute length growth (AL), the feed conversion ratio

(FCR), the total feed consumption (TFC), and the survival rate (SR) (Handajani et al 2021; Robbani et al 2022).

$$GR (g) = Wt - W0$$

Where:

GR - growth rate (g);  
W0 - initial weight (g);  
Wt - final weight (g).

$$SGR (\%) = (\ln Wt - \ln W0) / t \times 100$$

Where:

SGR - specific growth rate (%);  
W0 - initial weight (g);  
Wt - final weight (g).

$$AL (cm) = Lt - L0$$

Where:

AL - absolute length (cm);  
L0 - initial length (cm);  
Lt - final length (cm).

$$FCR = \frac{\sum F}{(Wt - W0)}$$

Where:

FCR - feed conversion rate;  
F - the number of feeds (g);  
W0 - initial weight (g);  
Wt - final weight (g).

$$SR (\%) = \frac{S0}{St} \times 100$$

Where:

W0 - initial weight (g);  
Wt - final weight (g).

$$TFC = F2 - F1$$

Where:

TFC - total feed consumption (g);  
F1 - uneaten feed;  
F2 - given feed.

**Water quality parameters.** Water quality measurements were conducted to monitor the environmental conditions during feed testing, ensuring optimal conditions for the study. Temperature was measured with a calibrated digital thermometer at regular intervals. Simultaneously, pH levels were assessed using a pH meter calibrated with standard buffer solutions to maintain an acceptable range for the species studied. Dissolved oxygen (DO) levels were measured with a dissolved oxygen meter, where the probe was immersed in the water sample to ensure ample oxygen levels in the aquatic environment. These comprehensive methods helped evaluate the feed's nutritional quality, color enhancement properties, carotenoid content, and environmental conditions essential for the research (Zubaidah et al 2022).

**Statistical analysis.** The research data were statistically analysed using Analysis of Variance (ANOVA) to identify significant group differences. SPSS software version 25.0 was used to perform the ANOVA and other statistical tests, ensuring accurate interpretation of the results.

## Results

**Proximate analysis of feed.** Table 3 presents the results of the proximate analysis of the artificial feed for *Cherax quadricarinatus* (freshwater lobster) with different concentrations of purple sweet potato extract: P1 (without purple sweet potato extract), P2 (with 10% purple sweet potato extract), P3 (with 20% purple sweet potato extract), and P4 (with 30% purple sweet potato extract), showing varying results across the different treatments.

Table 3

The proximate composition for each feed treatment

Sample	Ash (%)	Crude protein (%)	Crude fat (%)	Crude fiber (%)	NFE (%)	EM (mcal)	Total	CHO	TDN
P1	10.14	35.70	7.80	1.67	44.69	3313.1	65.09	36.58	99.61
P2	0.00	25.21	5.71	0.74	68.34	3706.1	31.66	69.08	107.13
P3	6.83	21.18	5.75	1.24	65.01	3446.0	45.09	56.15	100.36
P4	0.00	18.54	4.69	1.02	75.75	3696.2	24.25	76.77	105.87

NFE-nitrogen free extract; EM-energy metabolism; CHO-carbohydrate; TDN-total digestible nutrients

The proximate analysis of the artificial feed for *C. quadricarinatus* reveals significant variations in nutritional content depending on the concentration of purple sweet potato extract added. The ash content, representing total mineral levels, is highest in P1 (1014) and P3 (683), with no detectable ash in P2 and P4, suggesting that the extract influences mineral composition. Protein content decreases as the concentration of purple sweet potato extract increases from 3570 in P1 (no extract) to 1854 in P4 (30% extract), indicating a dilution effect on protein levels. Both crude fat and fiber content also decline with higher extract concentrations, potentially impacting the feed's energy density and digestibility. In contrast, non-nitrogen free extract (NFE) and total digestible nutrients (TDN) are highest in P4, suggesting that higher extract concentrations enhance the carbohydrate and energy content of the feed. Overall, adding purple sweet potato extract reduces protein and fat while increasing carbohydrates, which could affect the growth and health of the lobsters.

**Color testing of freshwater lobsters.** The study examined color changes in freshwater lobsters subjected to varying concentrations of purple sweet potato extract (0, 10, 20, and 30%) over 4 weeks (Table 4 and Table 5). At the start of the experiment (Week 0), all groups exhibited uniform color values of  $100 \pm 0.00$ . By Week 1, a significant reduction in color values was observed, with the control group (0% extract) decreasing to  $89.0 \pm 0.75$  and the 10% extract group to  $85.9 \pm 0.07$ . The latter was comparable to the 20% and 30% extract groups, indicating that higher extract concentrations did not improve color retention during this phase. By Week 2, the color values continued to decline across all treatments, with the control group dropping to  $70.1 \pm 0.59$ . Notably, the 10% extract group exhibited the highest color value at  $75.7 \pm 0.10$ , suggesting that this concentration may optimize coloration at this stage. In Week 3, the declining trend persisted, with values ranging from  $53.9 \pm 0.32$  to  $65.0 \pm 0.13$ . The 10% extract group consistently maintained the highest color values. By the end of the study (Week 4), the 10% extract group achieved a color value of  $61.8 \pm 0.14$ , which was statistically superior to other treatments ( $p < 0.05$ ). These findings suggest that including purple sweet potato extract at a 10% concentration is optimal for enhancing and maintaining coloration in freshwater lobsters over 4 weeks. Higher concentrations (20% and 30%) did not confer additional benefits, indicating the need for precise optimization of carotenoid supplementation to maximize pigmentation.

Table 4

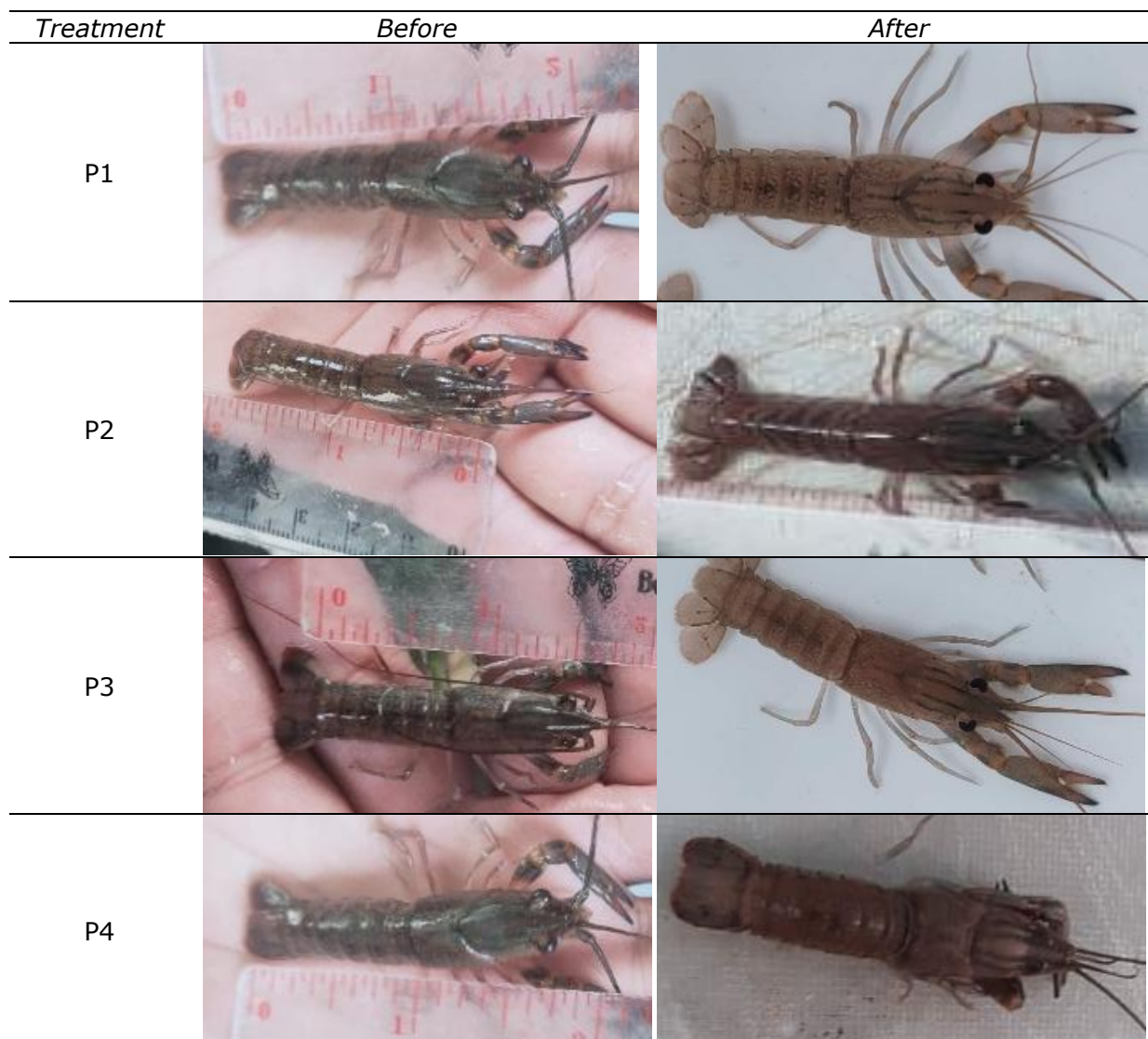
The impact of various doses of purple sweet potato extract (%) on changes in the colour values of freshwater lobsters over weeks

Time week	Dosage of purple sweet potato extract (%)			
	0 (P1)	10 (P2)	20 (P3)	30 (P4)
0	10.0±0.00 <sup>a</sup>	10.0±0.00 <sup>a</sup>	10.0±0.00 <sup>a</sup>	10.0±0.00 <sup>a</sup>
1	8.90±0.75 <sup>b</sup>	8.59±0.07 <sup>ab</sup>	8.10±0.09 <sup>a</sup>	8.23±0.10 <sup>a</sup>
2	7.01±0.59 <sup>ab</sup>	7.57±0.10 <sup>b</sup>	6.79±0.40 <sup>a</sup>	6.82±0.20 <sup>a</sup>
3	5.44±0.07 <sup>a</sup>	6.50±0.13 <sup>b</sup>	5.39±0.32 <sup>a</sup>	5.66±0.27 <sup>a</sup>
4	4.67±0.23 <sup>a</sup>	6.18±0.14 <sup>c</sup>	4.89±0.13 <sup>a</sup>	5.29±0.21 <sup>b</sup>

Data are presented as mean ± SD (n=3); different letters indicate significant differences among treatments at each time point (p<0.05).

Table 5

Differences in lobster color before and after the study



**Carotenoid analysis.** Figure 2 illustrates the carotenoid content ( $\mu\text{mol g}^{-1}$ ) in *Cherax quadricarinatus* across four treatment groups during a 30-day feeding trial. The control group (P1), lacking purple sweet potato extract, demonstrated the highest carotenoid absorption at  $71 \mu\text{mol g}^{-1}$ . In contrast, the group receiving the highest extract concentration (P4) showed the lowest absorption at  $36 \mu\text{mol g}^{-1}$ . Intermediate levels

were observed in P3 (62  $\mu\text{mol g}^{-1}$ ) and P2 (44  $\mu\text{mol g}^{-1}$ ), indicating a decline in carotenoid uptake with increasing extract concentration. The results suggest that adding purple sweet potato extract does not consistently enhance carotenoid absorption in *C. quadricarinatus*, with higher concentrations potentially inhibiting uptake.

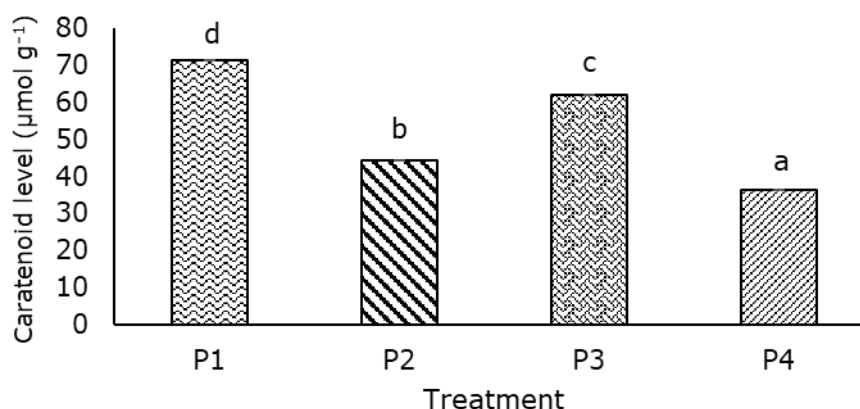


Figure 2. The average carotenoid content of *Cherax quadricarinatus*.

**Growth performance and survival rate analysis.** The growth performance of *C. quadricarinatus* was significantly influenced by the supplementation of sweet potato extract (Table 6), as indicated by the statistical analysis ( $p < 0.05$ ). The control group (P1) exhibited the lowest values for all measured parameters, including absolute weight ( $0.59 \pm 0.15$  g), specific growth rate ( $1.18 \pm 0.50$ ), absolute length ( $0.25 \pm 0.05$  cm), and survival rate ( $40.00 \pm 8.16\%$ ), accompanied by the highest feed conversion ratio (FCR) of  $1.96 \pm 1.06$ . In contrast, treatment P2 showed superior growth performance, with an absolute weight of  $0.99 \pm 0.39$  g, a specific growth rate of  $1.60 \pm 1.07$ , and a significant enhancement in survival rate ( $80.00 \pm 8.16\%$ ). Treatment P3 also demonstrated notable growth, recording an absolute weight of  $0.87 \pm 0.38$  g and a survival rate of  $80.00 \pm 11.55\%$ . However, the FCR for P3 ( $1.61 \pm 0.23$ ) was higher than that of P2, indicating less efficient feed utilization in comparison. Treatment P4, while showing moderate performance with an absolute weight of  $0.74 \pm 0.58$  g and a survival rate of  $67.50 \pm 9.57\%$ , also exhibited an FCR ( $1.80 \pm 0.51$ ) similar to that of the control group.

Regarding this discovery, sweet potato extract supplementation significantly improved *C. quadricarinatus* growth, with treatment P2 showing the best results in weight, growth rate, and survival. Higher concentrations in treatments P3 and P4 led to less efficient feed utilization, indicating that moderate supplementation is optimal for growth and feed efficiency.

Table 6  
The growth performance of *Cherax quadricarinatus* after sweet potato extract treatments

Treatment	Absolute weight (g)	Specific growth rate (%)	Absolute length (cm)	SR (%)	FCR
P1	$0.59 \pm 0.15^a$	$1.18 \pm 0.50^a$	$0.25 \pm 0.05^a$	$40.00 \pm 8.16^a$	$1.96 \pm 1.06^c$
P2	$0.99 \pm 0.39^b$	$1.60 \pm 1.07^b$	$1.10 \pm 0.01^b$	$80.00 \pm 8.16^c$	$1.23 \pm 0.05^a$
P3	$0.87 \pm 0.38^b$	$1.30 \pm 0.21^a$	$0.42 \pm 0.26^a$	$80.00 \pm 11.55^c$	$1.61 \pm 0.23^b$
P4	$0.74 \pm 0.58^b$	$1.21 \pm 0.70^a$	$0.31 \pm 0.19^a$	$67.50 \pm 9.57^b$	$1.80 \pm 0.51^c$

Data are presented as mean  $\pm$  SD (n=3); different letters indicate significant differences among treatments at each time point ( $p < 0.05$ ). SR-survival rate; FCR-feed conversion ratio.

**Water quality analysis.** Table 7 presents the environmental parameters of temperature, pH, and dissolved oxygen across four treatments (P1, P2, P3, and P4), compared to the optimal levels established for freshwater species. The recorded temperature values across treatments ranged from  $22.3^\circ\text{C}$  to  $28.3^\circ\text{C}$ , with P3 exhibiting the widest variation ( $22.3^\circ\text{C}$  to  $28.3^\circ\text{C}$ ). Although these temperatures were slightly below

the optimal range of 24°C to 29°C, they might still support metabolic and physiological processes; however, they could limit peak efficiency in growth and immune response, as higher temperatures within the optimal range are known to enhance enzymatic activity and overall metabolism in aquatic species.

Table 7

Results of water quality measurements during the study

Parameter	Treatment				Optimum level
	P1	P2	P3	P4	
Temperature (°C)	23.1-28.2	22.3-27.9	22.3-28.3	23.1-27.9	24-29*
pH	6.8-8.5	7.1-8.5	7.1-8.5	6.9-8.4	6.5-9*
DO (mg L <sup>-1</sup> )	4-5.7	3.5-5.7	4.1-5.7	4-5.7	3-6*

The pH values across all treatments varied from 6.8 to 8.5, aligning well with the recommended optimum range of 6.5 to 9. Treatments P2 and P3 recorded slightly higher ranges (7.1 to 8.5), favorable for maintaining water alkalinity and stable physiological conditions. Consistently optimal pH levels regulate osmotic balance and metabolic function, preventing stress in freshwater organisms. DO concentrations across treatments ranged from 3.5 to 5.7 mg L<sup>-1</sup>, generally within the optimal 3 to 6 mg L<sup>-1</sup>. However, P2 exhibited the lowest minimum value (3.5 mg L<sup>-1</sup>), which might pose a risk of hypoxic stress, particularly during periods of high metabolic demand, such as feeding or increased activity. Adequate dissolved oxygen levels are essential for aerobic respiration and efficient nutrient assimilation, and suboptimal DO levels could hinder growth and survival rates. Overall, although the recorded parameters stay within acceptable ranges for freshwater species, slight deviations from the optimum, especially in temperature and DO levels, could affect the lobsters' growth and health performance. These findings emphasize the need for precise environmental monitoring and management to sustain favorable conditions for optimal growth and physiological efficiency.

**Discussion.** The study found that a 10% purple sweet potato extract was optimal for enhancing freshwater lobster coloration, achieving the highest color values by Week 4 ( $61.8 \pm 0.14$ ;  $p < 0.05$ ), while higher concentrations provided no additional benefits (Tables 4 and 5). These findings align with other studies emphasizing the importance of carotenoid sources in enhancing crustacean pigmentation. For instance, Wang et al (2021) demonstrated that dietary astaxanthin supplementation improved pigmentation in shrimp, with optimal concentrations yielding significant benefits compared to higher or lower doses, which often resulted in diminished effects. Lower doses, such as 20 mg kg<sup>-1</sup>, have also shown positive effects on pigmentation but may not achieve the same intensity as higher doses (Zhang et al 2022). Similarly, a review by Wade et al (2017) on crustaceans noted that moderate concentrations of natural carotenoids improved color retention and immune response. In contrast, excessive supplementation inhibited carotenoid absorption due to saturation effects in the digestive system. Moreover, purple sweet potato extract, rich in anthocyanins and natural pigments, offers additional antioxidative benefits, as highlighted by Li et al (2019) and Baron et al (2008), who found that anthocyanin-rich diets improved stress tolerance and overall pigmentation in ornamental fish. These synergistic properties may explain the superior performance of the 10% concentration observed in the current study. Conversely, higher concentrations, such as the 30% extract group, may lead to negative interactions or reduced bioavailability, as excessive anthocyanins could interfere with carotenoid metabolism. These findings conclude that a 10% purple sweet potato extract effectively enhances freshwater lobster coloration, potentially benefiting aquaculture and ornamental fisheries.

Moreover, the study also found carotenoid content in *C. quadricarinatus* over 30 days. The control group (P1) exhibited the highest absorption (71 μmol g<sup>-1</sup>), while the highest extract concentration (P4) had the lowest (36 μmol g<sup>-1</sup>), indicating that higher concentrations may reduce carotenoid uptake (Figure 2). These results suggest that adding purple sweet potato extract does not necessarily enhance carotenoid absorption.

In fact, excessive inclusion may inhibit absorption, a finding that aligns with several studies. Some papers demonstrated that while moderate carotenoid supplementation boosts pigmentation in shrimp, high concentrations decrease bioavailability, likely due to metabolic saturation (Boonyaratpalin et al 2001; Díaz-Jiménez et al 2019; Wade et al 2017). Similarly, Jiang et al (2024) and Zhao et al (2023) found that crayfish exhibited reduced carotenoid absorption when supplementation exceeded optimal levels. As a result, these studies indicate that carotenoid absorption follows a dose-dependent curve, where excessive supplementation can overwhelm absorption mechanisms. Furthermore, a study by Hien et al (2022) and Huggins et al (2010) on fish highlighted similar trends, showing that excessive carotenoid levels could lead to reduced pigmentation due to impaired absorption. Additionally, research on *Pennaeus monodon* further supports these findings, revealing that excess carotenoids could disrupt digestive processes (Wade et al 2017) and reduce overall bioavailability (van het Hof et al 2000). These findings underscore the importance of optimizing carotenoid supplementation in aquaculture diets. While carotenoid-rich ingredients like purple sweet potato extract can enhance pigmentation and health, excessive inclusion may diminish their effectiveness. Therefore, precise dietary formulation tailored to the species' absorption capacity is crucial for maximizing carotenoid uptake and achieving desired physiological outcomes. Sweet potato extract also could enhance *C. quadricarinatus* growth, with P2 performing best, followed by P3 and P4, while P1 had the lowest growth. These findings align with previous research on using plant-based additives in aquaculture. For instance, natural plant extracts, such as those from ginger, enhance shrimp growth and feed conversion, though the optimal effects were observed at intermediate concentrations (Shahraki et al 2021; Tu et al 2023; Venkatramalingam et al 2007). Similarly, Arulvendhan et al (2024) and Muralisankar et al (2018) reported improvements in growth and survival in freshwater prawns supplemented with plant extracts, highlighting the importance of concentration in optimizing these effects. This study suggests that moderate supplementation with sweet potato extract (P2) offers the most favorable growth performance for *C. quadricarinatus*, with improved weight gain and feed conversion efficiency compared to higher or lower dosages. This supports the growing body of literature indicating the beneficial role of plant-based additives in crustacean aquaculture.

Finally, the results demonstrate that *C. quadricarinatus* thrives within the optimal environmental parameters of temperature (24°C to 29°C), pH (6.5 to 9), and dissolved oxygen (DO) (3 to 6 mg L<sup>-1</sup>), which are critical for its growth and health. Ferrer-Chujutalli et al (2024) emphasized that maintaining temperatures within this species-specific optimal range maximizes growth and immune responses in freshwater crustaceans. They suggested keeping temperatures closer to 24°C to 29°C would benefit this species. This aligns with several studies showing improved growth and survival of crayfish when pH levels are maintained within the optimal range, ensuring enzymatic efficiency and proper ion regulation (Haddaway et al 2013; Maoxiao et al 2018; Siewert & Buck 1991). Furthermore, Qiu et al (2022) and Ma et al (2021) highlighted that DO levels toward the upper end of the range (5 to 6 mg L<sup>-1</sup>) are crucial for supporting higher energy metabolism and better feed conversion efficiency. In contrast, DO levels falling below 4 mg L<sup>-1</sup> may lead to hypoxic stress, hindering growth and performance. Overall, the environmental conditions observed in this study closely match the recommended ranges for freshwater crustaceans. However, slight deviations in temperature and DO levels in some treatments may limit the species' performance, emphasizing the importance of maintaining ideal environmental conditions for optimal health and growth.

**Conclusions.** The study demonstrates that including purple sweet potato extract in the diet of *C. quadricarinatus* positively influences growth and coloration when applied at optimal levels. Specifically, a 10% extract concentration significantly enhanced absolute weight, specific growth rate, survival rate, and shell pigmentation, indicating its efficacy as a natural feed additive. Higher extract concentrations (20% and 30%) reduced carotenoid uptake and feed efficiency, highlighting a dose-dependent response. Environmental parameters, including temperature, pH, and dissolved oxygen, remained within the optimal range for *C. quadricarinatus*, ensuring reliable results. These findings

underscore the potential of purple sweet potato extract as a sustainable alternative to synthetic additives in aquaculture, offering economic and environmental benefits. Future research should focus on refining the dosage and exploring its long-term effects on physiological health and marketability. This study contributes valuable insights into using natural bioactives to enhance aquaculture practices sustainably.

**Acknowledgements.** The authors are grateful to the Laboratory of Aquaculture, Department of Aquaculture, University of Muhammadiyah Malang, Indonesia. The Faculty of Agriculture and Animal Sciences fund supported this research.

**Conflict of interest.** The authors declare that there is no conflict of interest.

## References

- Aguilar-Pérez K. M., Ruiz-Pulido G., Medina D. I., Parra-Saldivar R., Iqbal H. M., 2023 Insight of nanotechnological processing for nano-fortified functional foods and nutraceutical—opportunities, challenges, and future scope in food for better health. *Critical Reviews in Food Science and Nutrition* 63(20):4618-4635.
- Arulvendhan V., Bhavan P. S., Anista M. A., Thirunavukkarasu V., Gayathri V., Muralisankar T., Karthik M., Moorthi P. V., 2024 Effect of prawn fed with *Catharanthus roseus* ethanolic extract incorporated diet in the protein profile approaches in gene expressions related and immune characteristics in giant freshwater prawn (*Macrobrachium rosenbergii*). *Aquaculture International* 32:8287–8327.
- Azra M. N., Okomoda V. T., Tabatabaei M., Hassan M., Ikhwanuddin M., 2021 The contributions of shellfish aquaculture to global food security: assessing its characteristics from a future food perspective. *Frontiers in Marine Science* 8: 654897.
- Baron M., Davies S., Alexander L., Snellgrove D., Sloman K., 2008 The effect of dietary pigments on the coloration and behaviour of flame-red dwarf gourami, *Colisa lalia*. *Animal Behaviour - ANIM BEHAV* 75:1041-1051.
- Bolek S., 2022 Consumer attitudes toward natural food additives. In *Natural additives in foods*. In: Aquaculture microbiology. Thomas J., Amaesan N. (eds), pp. 325-341, Springer Protocols Handbooks, Humana, New York, USA.
- Boonyaratpalin M., Thongrod S., Supamattaya K., Britton G., Schlipalius L., 2001 Effects of  $\beta$ -carotene source, *Dunaliella salina*, and astaxanthin on pigmentation, growth, survival and health of *Penaeus monodon*. *Aquaculture Research* 32:182-190.
- Budi S., Mardiana M., Geris G., Tantu A. G., 2021 [Color changes in goldfish *Cyprinus carpio* with the addition of extra nutmeg *Myristica argantha* at different doses]. *Jurnal Ilmiah Ecosystem* 21(1):202-207. [in Indonesian]
- Camareri M., Frary A., Frary A., Grandillo S., 2024 Genetic and biotechnological approaches to improve fruit bioactive content: a focus on eggplant and tomato anthocyanins. *International journal of molecular sciences* 25(12):6811-6852.
- Chandra Mohana N., Nethravathi A., Achar R. R., Anil Kumar K., Siddesha J. M., 2023 Preparation of feed and characterization of feed supplemented with phytocompounds. In: *Aquaculture microbiology*. Thomas J., Amaesan N. (eds), pp. 167-179, Springer Protocols Handbooks, Humana, New York, USA.
- Devlin R. H., Leggatt R. A., Benfey T. J., 2020 Chapter 7 - Genetic modification of growth in fish species used in aquaculture: Phenotypic and physiological responses. In: *Fish physiology*. Benfey T. J., Farrell A. P., Brauner C. J. (eds.), pp. 237-272, Academic Press, Canada.
- Díaz-Jiménez L., Hernández-Vergara M. P., Pérez-Rostro C. I., Ortega-Clemente L. A., 2019 The effect of astaxanthin and  $\beta$ -carotene inclusion in diets for growth, reproduction and pigmentation of the peppermint shrimp *Lysmata wurdemanni*. *Latin American Journal of Aquatic Research* 47(3):559-567.

- Farida S., Saati E. A., Damat D., Wahyudi A., Van Minh N., 2024 Identification of functional groups and types of anthocyanin pigments of purple sweet potato cv. Antin 2 and cv. Antin 3. *BIO Web of Conferences* 104:1-13.
- Ferrer-Chujutalli K., Sernaqué-Jacinto J., Reyes-Avalos W., 2024 Optimal temperature and thermal tolerance of postlarvae of the freshwater prawn *Cryphiops (Cryphiops) caementarius* acclimated to different temperatures. *Heliyon* 10(5):e25850.
- Gagas W., Junaidi M., Setyono B. D. H., 2023 Effect of giving purple sweet potato (*Ipomea batatas* L.) extract in feed to increase the color and growth of koi fish (*Cyprinus carpio*). *Asian Journal of Fisheries and Aquatic Research* 25(2):35-42.
- Garza de Yta A., Davis D. A., Rouse D. B., Ghanawi J., Saoud I. P., 2012 Evaluation of practical diets containing various terrestrial protein sources on survival and growth parameters of redclaw crayfish *Cherax quadricarinatus*. *Aquaculture Research* 43(1):84-90.
- Ghonimy A., Greco L. S. L., Li J., Wade N. M., 2023 An hypothesis on crustacean pigmentation metabolism: l-carnitine and nuclear hormone receptors as limiting factors. *Crustaceana* 96(10):939-956.
- Haddaway N. R., Mortimer R. J., Christmas M., Dunn A. M., 2013 Effect of pH on growth and survival in the freshwater crayfish *Austropotamobius pallipes*. *Freshwater Crayfish* 19(1):53-62.
- Handajani H. A., Gilang R., Warkoyo, 2021 Enrichment of commercial feed with plant proteins for *Oreochromis niloticus* diet: digestibility and growth performance. *AACL Bioflux* 14(5):2894-2904.
- Hartoto S., Hariyanto A., Maratus S. A., Putera S. H. P., Kusuma D. A., Firmansyah A., Fikri M. D., 2024 Purple sweet potato extract and aerobic exercise reduce lipid profiles in hyperlipidemic rats model. *Retos: Nuevas Tendencias en Educación Física, Deporte y Recreación* (55):756-768.
- Haubrock P. J., Oficialdegui F. J., Zeng Y., Patoka J., Yeo D. C. J., Kouba A., 2021 The redclaw crayfish: A prominent aquaculture species with invasive potential in tropical and subtropical biodiversity hotspots. *Reviews in Aquaculture* 13(3):1488-1530.
- Hien T. T. T., Loc T. V., Tu T. L. C., Phu T. M., Duc P. M., Nhan H. T., Liem P. T., 2022 Dietary effects of carotenoid on growth performance and pigmentation in bighead catfish (*Clarias macrocephalus* Günther, 1864). *Fishes* 7(1):37-53.
- Hou Y., Jia R., Sun W., Ding H., Li B., Zhu J., 2023 Red claw crayfish *Cherax quadricarinatus* cultivation influences the dynamics and assembly of benthic bacterial communities in paddy fields. *Environments* 10(10):178.
- Huggins K. A., Navara K. J., Mendonça M. T., Hill G. E., 2010 Detrimental effects of carotenoid pigments: the dark side of bright coloration. *Naturwissenschaften* 97:637-644.
- Janssen K., Berentsen P., Besson M., Komen H., 2017 Derivation of economic values for production traits in aquaculture species. *Genetics Selection Evolution* 49(1):5.
- Jiang X., Zhu S., Zhang G., Gong Y., Wu X., 2024 Effect of dietary  $\beta$ -carotene supplementation on growth and antioxidant capability of pre-adult male Chinese mitten crab, *Eriocheir sinensis*. *Aquaculture Reports* 35:101968.
- Khoo H. E., Azlan A., Tang S. T., Lim S. M., 2017 Anthocyanidins and anthocyanins: Colored pigments as food, pharmaceutical ingredients, and the potential health benefits. *Food & Nutrition Research* 61(1):1361779.
- Leema J. M., Kirubakaran M. V., Remany M., Kumar T., Babu T., 2010 Effects of intrinsic and extrinsic factors on the haemocyte profile of the spiny lobster, *Panulirus homarus* (Linnaeus, 1758) under controlled conditions. *Journal of the Marine Biological Association of India* 52:219-228.
- Li A., Xiao R., He S., An X., He Y., Wang C., Yin S., Wang B., Shi X., He J., 2019 Research advances of purple sweet potato anthocyanins: Extraction, identification, stability, bioactivity, application, and biotransformation. *Molecules* 24(21):3816.
- Lv Y., Ai Y., Fang F., Liao H., 2023 Development of active packaging films utilized natural colorants derived from plants and their diverse applications in protein-rich food products. *Food Innovation and Advances* 2(3):203-216.

- Ma J.-L., Qiang J., Tao Y.-F., Bao J.-W., Zhu H.-J., Li L.-G., Xu P., 2021 Multi-omics analysis reveals the glycolipid metabolism response mechanism in the liver of genetically improved farmed Tilapia (GIFT, *Oreochromis niloticus*) under hypoxia stress. *BMC Genomics* 22:1-16.
- Maoxiao P., Bo Y., Xiaojun L., Donghong N., Tianyi L., Zhiguo D., Jiale L., 2018 Effects of alkalinity and pH on survival, growth, and enzyme activities in juveniles of the razor clam, *Sinonovacula constricta*. *Frontiers in Physiology* 9(552):1-14.
- Mauro M., Di Grigoli A., Maniaci G., Hornsby L. B., Badalamenti G., Chirco P., Arizza V., Gargano C., Bellini P., Arculeo M., 2024 *Cherax destructor* (Clark, 1836) and *Cherax quadricarinatus* (von Martens, 1868): Biochemical parameters and preliminary analysis of food quality. *Aquaculture Reports* 36:102162.
- Mohammadi N., Farrell M., O'Sullivan L., Langan A., Franchin M., Azevedo L., Granato D., 2024 Effectiveness of anthocyanin-containing foods and nutraceuticals in mitigating oxidative stress, inflammation, and cardiovascular health-related biomarkers: a systematic review of animal and human interventions. *Food & Function* 15(7):3274-3299.
- Muralisankar T., Bhavan P. S., Radhakrishnan S., Santhanam P., 2018 Dietary supplement of medicinal herbal leaf powder on growth performance, digestive enzymes activities, energy utilization and vitamin levels of the freshwater prawn *Macrobrachium rosenbergii*. *Proceedings of the Zoological Society* 71:265-271.
- Onomu A. J., Okuthe G. E., 2024 The role of functional feed additives in enhancing aquaculture sustainability. *Fishes* 9(5):167.
- Paul J., Islam M. M., Al Farabi A., 2023 Proximate analysis of unconventional feed available in Boalkhali Upazila, Chattogram. *American Journal of Pure and Applied Biosciences* 5(4):74-81.
- Pereira da Costa D., Campos Miranda-Filho K., 2020 The use of carotenoid pigments as food additives for aquatic organisms and their functional roles. *Reviews in Aquaculture* 12(3):1567-1578.
- Presenza L., Teixeira B. F., Galvão J. A., de Souza Vieira T. M. F., 2023 Technological strategies for the use of plant-derived compounds in the preservation of fish products. *Food chemistry* 419:136069.
- Pyatikopova O., Bedritskaya I., Tangatarova R., Ankesheva B., 2023 Biotechnical indicators of red-clawed crayfish commercial cultivation in pond aquaculture in the conditions of the South of Russia. *Vestnik of Astrakhan State Technical University. Series: Fishing industry* 2023(4):72-82.
- Qiu X.-L., Jiang Y.-L., Cai Y.-S., Chen H., Lyu X.-J., Lin L., Li J.-T., 2022 Behavioral and physiological responses to hypoxia stress in male and female *Macrobrachium rosenbergii*. *The Journal of Applied Ecology* 33(10):2836-2844.
- Rakhmawati R., Aprilia T., Indariyanti N., Saputra A., Eulis M., 2023 Color quality improvement of *Cherax quadricarinatus* with dragon fruit peel meal utilization. *Jurnal Ilmu-Ilmu Perairan, Pesisir dan Perikanan* 12(3):314-319.
- Robbani A. A., Ariyanti O., Calvin D., Sutarjo G. A., Handajani H., Andriawan S., 2022 The combination of tofu dregs and *Rhodospirillum rubrum* on *Oreochromis niloticus* growth performances. *AAFL Bioflux* 15(4):2113-2124.
- Saati E. A., Wahyudi V. A., Dyah A., Andriawan S., 2022 Anthocyanin extract of *Rosa* sp. as a natural preservative in *Euthynnus affinis*. *AAFL Bioflux* 15(1):136-146.
- Saputri N. F., Putri A. A., 2024 Proximate analysis on Nile tilapia artificial feed from shrimp waste. *Eduvest-Journal of Universal Studies* 4(10):9601-9606.
- Shahraki N., Imanpour M., Akbary P., Safari R., Jafari V., 2021 Dietary administration of aqueous *Zingiber officinale* extract on growth performance, antioxidant activity and resistance of shrimp *Litopenaeus vannamei* against *Photobacterium damsela*. *Iranian Journal of Fisheries Sciences* 20(1):32-44.
- Siewert H. F., Buck J. P., 1991 Effects of low pH on survival of crayfish (*Orconectes virilis*). *Journal of Freshwater Ecology* 6(1):87-91.
- Snovsky G., Galil B., 2011 The Australian redclaw crayfish *Cherax quadricarinatus* (von Martens, 1868) (Crustacea: Decapoda: Parastactidae) in the Sea of Galilee, Israel. *Aquatic Invasions* 6(1):S29-S31.

- Sukarman H. R., Hirnawati R., 2014 [Alternative carotenoid synthesis (astaxanthin) to improve the color quality of goldfish (*Carassius auratus*)]. *Widyariset* 17(3): 333-342. [in Indonesian]
- Sumartini S., 2023 Identification of anthocyanidins in anthocyanin extract of purple sweet potato (*Ipomoea batatas* L), black rice (*Oryza sativa* L), and black glutinous rice (*Oryza sativa* v. *glutinosa*). *Journal of Functional Food and Nutraceutical* 5(1):25-32.
- Tu P. T. C., Lien N. T. K., Diep D. X., Ly T. H., 2023 Effect of ginger, *Zingiber officinale* extract on growth performance, digestive enzyme and stress tolerance of whiteleg shrimp, *Litopenaeus vannamei* juveniles. *Israeli Journal of Aquaculture–Bamidgeh* 75(2):1-8.
- van het Hof K. H., Weststrate J. A., West C. E., Hautvast J. G., 2000 Dietary factors that affect the bioavailability of carotenoids. *The Journal of Nutrition* 130(3):503-506.
- Venkatramalingam K., Christopher J. G., Citarasu T., 2007 *Zingiber officinalis* an herbal appetizer in the tiger shrimp *Penaeus monodon* (Fabricius) larviculture. *Aquaculture Nutrition* 13(6):439-443.
- Wade N. M., Gabaudan J., Glencross B. D., 2017 A review of carotenoid utilisation and function in crustacean aquaculture. *Reviews in Aquaculture* 9(2):141-156.
- Wang W., Liu M., Fawzy S., Xue Y., Wu M., Huang X., Yi G., Lin Q., 2021 Effects of dietary *Phaffia rhodozyma* astaxanthin on growth performance, carotenoid analysis, biochemical and immune-physiological parameters, intestinal microbiota, and disease resistance in *Penaeus monodon*. *Frontiers in Microbiology* 12:762689.
- Zhang S., Chang Z., Wang W., Zheng Y., Li J., 2022 Dietary supplement of microalgal astaxanthin extraction improved shell pigmentation and nutritional value of *Litopenaeus vannamei* in an indoor industrial aquaculture system. *Aquaculture Nutrition* 1:3981246.
- Zhao Z., Zhou Y., Zou Q., Qin J., Yang F., Ou Y., Lin Y., Yi Y., 2023 Transcriptome and metabolome profiling unveil the responses of crayfish to dietary astaxanthin. *Aquaculture Reports* 33:101871.
- Zharchynska V., Hrynevych N., 2023 Assessment of energy and biological value of *Cherax quadricarinatus* meat after feeding crayfish with different types of feed. *Tehnologîa Virobnictva i Pererobki Produktiv Tvarinnictva* 1(2):12-21.
- Zubaidah A., Prabowo A., Andriawan S., 2022 *Oreochromis niloticus* immune responses examination: a case study of environmental toxicology. *Aquaculture, Aquarium, Conservation & Legislation* 15(5):2363-2373.
- Zubaidah A., Samsundari S., Jaelani M. A. Q., 2021 [Effectiveness of adding papaya leaf extract (*Carica papaya* L) for preventing saprolegniasis in puntung carp eggs (*Cyprinus carpio*)]. *Jurnal Akuakultur Rawa Indonesia* 9(2):109-117. [in Indonesian]
- \*\*\* AOAC, 2012 Official method of analysis. Association of Official Analytical Chemists (19<sup>th</sup> Edition), Washington D.C., USA, 771 p.

Received: 17 May 2025. Accepted: 15 August 2025. Published online: 30 August 2025.

Authors:

Hany Handajani, Department of Aquaculture, Faculty of Agriculture and Animal Science, University of Muhammadiyah Malang, Malang, East Java, 65144, Indonesia, e-mail: handajani@umm.ac.id

Fathimatuz Zahroh, Department of Aquaculture, Faculty of Agriculture and Animal Science, University of Muhammadiyah Malang, Malang, East Java, 65144, Indonesia, e-mail: fathimatuz@gmail.com

Dony Prasetyo, Department of Aquaculture, Faculty of Agriculture and Animal Science, University of Muhammadiyah Malang, Malang, East Java, 65144, Indonesia, e-mail: donyprasetyo@umm.ac.id

Soni Andriawan, Biotechnology Development Center, University of Muhammadiyah Malang, Malang, East Java, 65144, Indonesia, e-mail: soniandriawan@umm.ac.id

This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

How to cite this article:

Handajani H., Zahroh F., Prasetyo D., Andriawan S., 2025 Optimizing growth and shell pigmentation in *Cherax quadricarinatus* juveniles using purple sweet potato extract. *AAFL Bioflux* 18(4):1970-1983.