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The Effectiveness of Organic Materials as Bioremediation Agents in Promoting Freshwater Crayfish Growth (*Cherax quadricarinatus*)

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Abstract

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Freshwater crayfish (*Cherax quadricarinatus*) are among the most economically valuable aquaculture species, with strong market potential in both domestic and international markets. However, one of the major challenges in freshwater crayfish farming is the deterioration of water quality caused by the accumulation of uneaten feed and metabolic waste. This study aimed to evaluate the effects of combinations of organic materials used as bioremediation agents on the absolute weight growth and specific length growth rate of freshwater crayfish. The experiment was conducted from August to September 2025 at CV. Cova Aquaculture using a Completely Randomized Design (CRD) consisting of four treatments with three replicates each (12 experimental units) over a 30-day culture period. The treatments consisted of powdered dried Indian almond leaves (*Terminalia catappa*) applied alone as the control (Treatment A), combined with spinach powder (Treatment B), pumpkin powder (Treatment C), or bean sprout powder (Treatment D). The organic materials were applied every three days and after each water exchange. Data were analyzed using one-way analysis of variance (ANOVA). Water quality parameters, including temperature, dissolved oxygen, and pH, were monitored daily. The results showed that the highest absolute weight growth and specific length growth rate were obtained in Treatment C, with an absolute weight gain of 0.78 g and a specific length growth rate of 2.16% day^{-1}. The combination of dried Indian almond leaf powder and pumpkin powder may function not only as an indirect itional supplement but also as an environmentally friendly bioremediation strategy that helps maintain water quality, thereby supporting the sustainable culture of freshwater crayfish.

Keywords: bioremediation, freshwater crayfish (*Cherax quadricarinatus*), growth, Indian almond leaves (*Terminalia catappa*), organic materials.

INTRODUCTION

Freshwater crayfish (*Cherax quadricarinatus*) have attracted increasing attention as an aquaculture species due to their rapid growth rate and promising economic value in intensive farming systems. However, one of the major challenges in freshwater crayfish culture is the deterioration of water quality caused by the accumulation of uneaten feed and metabolic waste (feces), which can induce physiological stress, inhibit growth, and even lead to mortality (Ruscoe *et al.,* 2005).

Poor water quality, particularly elevated concentrations of ammonia and nitrite, can adversely affect the health and productivity of freshwater crayfish, highlighting the need for effective management strategies (Yong-chun *et al.,* 2022). Water quality plays a crucial role in freshwater crayfish farming because these organisms require a stable aquatic environment with suitable physicochemical characteristics. The optimal water quality conditions for freshwater crayfish culture include a temperature of 24–31°C, a pH ranging from 6 to 8, ammonia concentrations below 1.2 ppm, and turbidity between 30 and 40 NTU (Tumembouw, 2011). Maintaining these parameters within the optimal range is essential to support crayfish growth and survival, and one promising approach is the application of organic materials.

Bioremediation is a process that utilizes beneficial microorganisms to treat contaminated water or waste by removing, reducing, or transforming pollutants through biological mechanisms (Jasmin *et al.,* 2020). In aquaculture, bioremediation focuses on the synergistic interaction between organic materials, microorganisms, and cultured organisms to stabilize water quality. The addition of specific organic materials can provide carbon sources and substrates for nitrifying and denitrifying bacteria, thereby facilitating the removal of ammonia, nitrite, and nitrate from the culture medium (Hou *et al.,* 2022).

Organic materials such as Indian almond leaves (*Terminalia catappa*), pumpkin, bean sprouts, and spinach have considerable potential as bioremediation agents while simultaneously serving as alternative nutrient sources rich in proteins, vitamins, and minerals. Their nutritional composition can promote the growth and health of freshwater crayfish while reducing dependence on relatively expensive commercial feeds (Chakraborty *et al.,* 2019). In addition, these organic materials may function as natural probiotics by enhancing the digestive system and improving nutrient utilization in freshwater crayfish (Wang *et al.,* 2024).

Indian almond leaves contain 15–25% tannins on a dry-weight basis. These polyphenolic compounds possess antimicrobial properties, bind heavy metals, and have been reported to reduce populations of pathogenic bacteria such as *Aeromonas hydrophila* by 40–60% in aquaculture systems. Tannins can also chelate toxic nitrogen compounds, including ammonia (NH_3) and nitrite (NO_2^-), while reducing oxidative stress in crustaceans (Scabra *et al.,* 2021). Spinach contains approximately 25–30% crude protein (dry matter) and essential amino acids such as lysine and methionine, which play important roles in chitin synthesis. Dietary supplementation with 10% spinach has been shown to increase molting rates in crustaceans by up to 15%, while its high protein content supports muscle synthesis and enhances daily growth rates by approximately 0.8–1.2% (Agustami *et al.,* 2024). Pumpkin contains bioactive compounds, including omega-3 fatty acids at concentrations of approximately 0.5–1%, which have been reported to stimulate feed intake in aquatic organisms by up to 40% (Glencross, 2020). Meanwhile, bean sprouts contain digestive enzymes such as amylase (120–150 U g^{-1}) and protease (80–100 U g^{-1}), which improve feed digestibility. Supplementation with 15% bean sprouts has been reported to reduce the feed conversion ratio (FCR) in crustaceans from 2.1 to 1.7. Bean sprouts also provide essential amino acids, including lysine (4–6%) and leucine (3–5%), which contribute to improved exoskeleton quality and increase carapace thickness by approximately 1–15% (Li *et al.,* 2021).

Research on freshwater crayfish culture remains essential because this species is highly sensitive to fluctuations in water quality, which directly affect growth performance and survival. The present study proposes the use of inexpensive and environmentally sustainable organic materials as bioremediation agents to improve water quality, thereby minimizing stress and mortality while enhancing crayfish growth. Accordingly, this study aims to determine whether combinations of selected organic materials are effective in promoting the growth of freshwater crayfish (*Cherax quadricarinatus*).

MATERIALS AND METHODS

Research Duration and Location

This study was conducted over a period of 30 days at CV. Cova Aquaculture, located in Lembang Permatasari Residential Area, Tande Timur Village, Banggae Timur District, Majene Regency, West Sulawesi, Indonesia.

Research Equipment and Materials

The equipment and materials used in this study are presented in Tables 1 and 2, respectively.

Table 1. Equipment used in the study

No	Equipment	Function
1	Aquarium	Experimental tank for freshwater crayfish
2	Aerator	Supplies dissolved oxygen
3	Digital caliper	Measures crayfish length
4	pH meter	Measures pH
5	Thermometer	Measures water temperature
6	DO meter	Measures dissolved oxygen
7	Ammonia test kit	Measures ammonia concentration
8	Digital balance	Weighs freshwater crayfish
9	Blender	Grinds organic materials
10	Sieve	Filters ground materials
11	Hose	Assists with water exchange
12	Plastic basin	Mixes materials
13	Oven	Dries materials
14	Jar	Stores materials
15	Mortar and pestle	Crushes and mixes materials
16	Knife	Cuts materials
17	Sample bottle	Collects water samples
18	Camera	Documentation
19	Stationery	Records research data

Table 2. Materials used in the study

No	Material	Function
1	Freshwater crayfish (*Cherax quadricarinatus*)	Experimental animal
2	Freshwater	Culture medium
3	Indian almond leaves (*Terminalia catappa*)	Test material
4	Spinach	Test material
5	Pumpkin	Test material
6	Bean sprouts	Test material
7	Fengli feed	Crayfish feed

Research Procedure

The study consisted of three main stages: preparation of the experimental units and test animals, preparation of the test materials, and the rearing of freshwater crayfish. The first stage involved preparing the experimental tanks and test animals. All equipment was thoroughly washed with detergent, rinsed with clean water, wiped, and air-dried before use. The experimental animals were juvenile freshwater crayfish (*Cherax quadricarinatus*) approximately 2 months of age with an average body length of approximately 4 cm. The crayfish were obtained from a local hatchery.

The second stage involved preparing the test materials. The organic materials used in this study consisted of dried Indian almond leaves (*Terminalia catappa*), spinach leaves, pumpkin, and bean sprouts. All materials were washed thoroughly before use and then dried. The dried Indian almond leaves were ground into a fine powder using a blender. After grinding, the organic materials were mixed according to the treatment dosages specified in the experimental design.

The third stage involved rearing the freshwater crayfish. The prepared organic materials were applied to the culture water before stocking and subsequently every seven days following water exchange at a dosage of 3 g per experimental tank. The crayfish were fed twice daily, in the morning and afternoon, with a commercial diet at a feeding rate of 5% of total body weight per day. The rearing period lasted 30 days.

Experimental Design

This study employed a Completely Randomized Design (CRD) consisting of four treatments, each with three replicates. The experimental treatments were as follows:

Treatment A (Control) : 100% Indian almond leaf (*Terminalia catappa*) powder.
Treatment B : 50% Indian almond leaf powder + 50% spinach powder.
Treatment C : 50% Indian almond leaf powder + 50% pumpkin powder.
Treatment D : 50% Indian almond leaf powder + 50% bean sprout powder.

Measured Parameters
The parameters evaluated in this study included absolute weight growth, specific length growth rate, and water quality of the culture medium. Absolute weight growth was calculated using the equation proposed by Suseno (1983):

$$AWG = Wt - W0$$

Where:
AWG = Absolute weight growth (g)
W_t = Mean body weight of freshwater crayfish at the end of the experimental period (g)
W_0 = Mean body weight of freshwater crayfish at the beginning of the experimental period (g)

The specific length growth rate (SLGR) was calculated according to (Zonneveld et al., 1991) :

$$SLGR = \frac{Lt - L0}{t} x\ 100\ \%$$

Where:
SLGR = Specific length growth rate (% day^{-1})
Lt = Mean body length at the end of the experimental period (cm)
L_0 = Mean body length at the beginning of the experimental period (cm)
t = Duration of the experimental period (days)

As supporting data, water quality parameters were monitored twice daily, at 08:00 and 17:00 (Central Indonesia Time, WITA), including water temperature, pH, and dissolved oxygen (DO).

Data Analysis
The data obtained were analyzed statistically using one-way analysis of variance (ANOVA) at a 95% confidence level ($\alpha = 0.05$). When significant differences among treatments were detected, Tukey's Honestly Significant Difference (HSD) post hoc test was performed to identify differences between treatment means. All statistical analyses were conducted using IBM SPSS Statistics version 22. Water quality parameters were analyzed descriptively and compared with the recommended water quality standards for the culture of *Cherax quadricarinatus*.

RESULTS AND DISCUSSIONS
1. Absolute Weight Growth

Absolute weight growth is defined as the difference between the final body weight and the initial body weight of an organism after a specified rearing period. This parameter measures the overall increase in biomass without considering the initial body weight or the growth rate per unit of waktu (Effendie, 1997). time (Effendie, 1997). The results of the absolute weight growth of freshwater crayfish are presented in Figure 1**.**

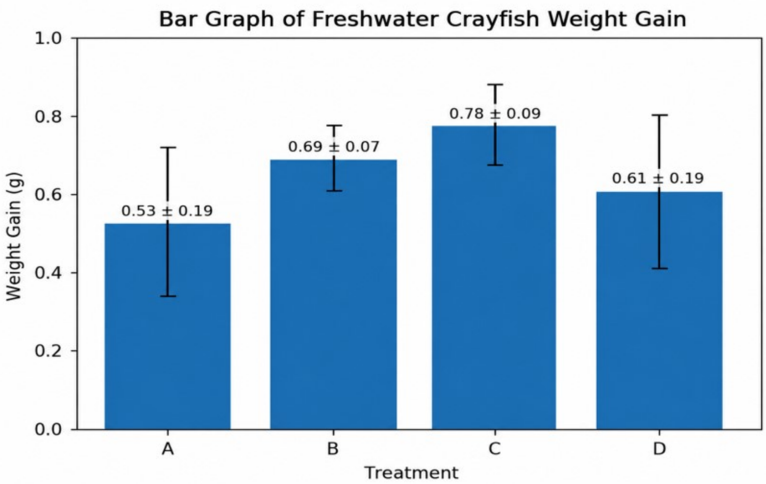

Figure 1. Graph of Absolute Weight Growth

The results showed that the highest absolute weight growth was observed in Treatment C, which consisted of a combination of dried Indian almond leaves (*Terminalia catappa*) and pumpkin, with a mean weight gain of 0.78 g. However, one-way analysis of variance (ANOVA) indicated that the treatments had no statistically significant effect on absolute weight growth ($p > 0.05$). Nevertheless, the value obtained in the present study was higher than that reported by Safir et al. (2023), who found a maximum absolute weight gain of 0.49 g.

Although no statistically significant differences were detected among treatments ($p > 0.05$), the tendency toward greater weight gain in crayfish receiving combinations of organic materials suggests the potential application of natural organic matter as a bioremediation strategy in freshwater crayfish culture. This approach is preventive, environmentally sustainable, and eco-friendly because it relies on natural biological processes to improve water quality while simultaneously supporting the growth performance of cultured organisms.

Indian almond leaves (*Terminalia catappa*) release bioactive compounds, including tannins, flavonoids, and saponins, which exhibit antibacterial and antifungal properties (Rizal *et al.,* 2021)**.** These compounds can suppress the abundance of pathogenic bacteria in the culture water, thereby reducing the risk of disease and physiological stress in freshwater crayfish. A healthier microbial environment allows crayfish to allocate more energy toward growth rather than immune responses and stress adaptation.

Pumpkin contains readily degradable carbohydrates that serve as an excellent carbon source for beneficial bacteria and other microorganisms, promoting their proliferation and the formation of nutrient-rich biofilms. Freshwater crayfish are omnivorous and detritivorous organisms that naturally consume decomposing organic matter, including microbial biofilms, as supplementary food. Consequently, the enhanced biofilm production may provide an additional natural food source rich in nutrients, while bioactive compounds such as carotenoids may improve health status and feed utilization efficiency. Furthermore, pumpkin powder has been suggested to stimulate microbial communities involved in nitrification and denitrification processes, thereby contributing to improved water quality (Hastuti *et al.,* 2022).

Maintaining stable populations of nitrifying and denitrifying bacteria in aquaculture systems can be achieved through the supplementation of specific nutrients that stimulate microbial growth and activity. Essential nutrients, particularly nitrogen and phosphorus, are required to sustain bacterial metabolism and community stability (Effendi *et al.,* 2015). In bioremediation technology, this process is referred to as biostimulation, in which nutrients are added to the environment to enhance the growth and metabolic efficiency of indigenous microorganisms responsible for degrading contaminants (Jekti, 2018).

2. Specific Length Growth Rate

The results of the one-way analysis of variance (ANOVA) indicated that the treatments had no statistically significant effect on the specific length growth rate of freshwater crayfish ($p > 0.05$), as shown in Figure 2.

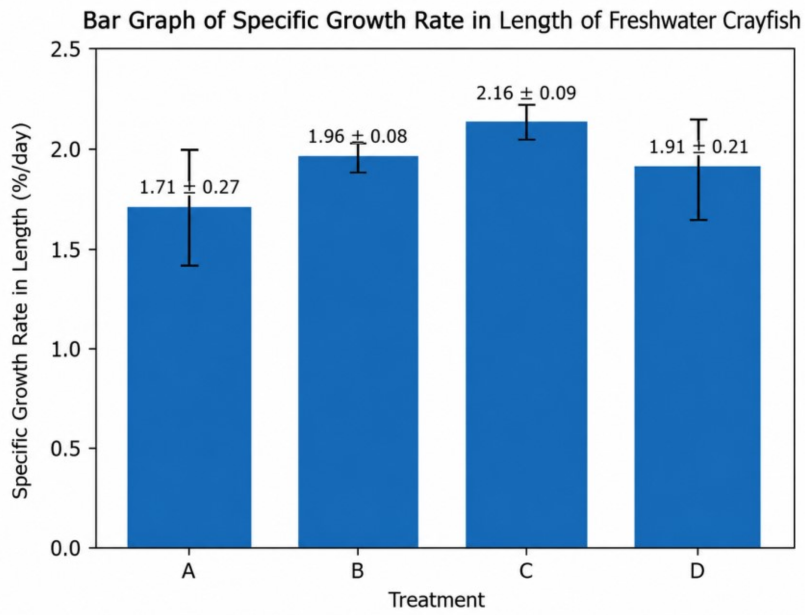

Figure 2. Graph of the Specific Length Growth Rate

The results showed that the highest specific length growth rate was obtained in Treatment C, consisting of a combination of dried Indian almond leaves (*Terminalia catappa*) and pumpkin, with a value of 2.16% day^{-1}. This value was higher than those reported by Handajani et al. (2025), who observed specific length growth rates ranging from 0.25 to 1.10% day^{-1}, and Siburian *et al.,* (2019) Siburian et al. (2019), who reported a maximum value of 1.15 ± 0.08% day^{-1}.

In crustaceans, length growth is closely associated with the molting process, which is highly sensitive to the accumulation of toxic compounds such as ammonia (NH_3) and nitrite (NO_2^-). The addition of pumpkin powder in Treatment C may have served as a readily degradable organic carbon source, thereby promoting the biostimulation of beneficial aquatic microorganisms. In bioremediation systems, the availability of organic carbon provides an energy substrate for heterotrophic and nitrifying bacteria, which contribute to the stabilization of water quality by facilitating nitrogen transformation processes.

The tannins present in Indian almond leaves are also presumed to exhibit antibacterial and antifungal activities, thereby reducing stress and the risk of infection in juvenile freshwater crayfish. Furthermore, water conditioned with Indian almond leaves more closely resembles the natural habitat of freshwater crayfish, creating a more favorable environment that reduces physiological stress and allows a greater proportion of metabolic energy to be allocated to growth. Pumpkin powder dispersed in the culture water may also release carotenoids and vitamins, which can be absorbed indirectly through natural feeding activities or by consuming nutrient-rich detritus. These compounds may function as supplementary nutrients while simultaneously acting as prebiotics that stimulate beneficial microbial communities, thereby enhancing the natural food web within the culture system.

The brownish coloration of the water resulting from Indian almond leaves creates a shaded and relatively dark environment that is more compatible with the nocturnal behavior of freshwater crayfish. Such environmental conditions may reduce stress and improve overall welfare. In addition, the antimicrobial properties of tannins and flavonoids suppress the proliferation of pathogenic bacteria and fungi in the culture water and on the crayfish exoskeleton. By reducing disease pressure, more energy can be allocated to somatic growth rather than immune defense (Yusanti & Mulyani, 2024).

The high fiber content of pumpkin powder gradually decomposes in the culture water, providing a prebiotic substrate for beneficial microorganisms inhabiting both the aquatic environment and the digestive tract of freshwater crayfish. A healthier microbial community can indirectly improve digestive efficiency and nutrient utilization. Furthermore, the decomposition of pumpkin powder may stimulate the growth of natural food organisms, including phytoplankton, zooplankton, and microbial biofilms, which serve as supplementary food sources for freshwater crayfish. The increased availability of these natural food resources may contribute to improved growth performance (Dahniar & Wamnebo, 2023).

3. Water Quality

The experimental treatments may have indirectly influenced the growth and physiological performance of freshwater crayfish throughout the culture period by affecting water quality conditions. Water quality is one of the most important environmental factors determining the survival, health, and growth performance of cultured freshwater crayfish. The water quality parameters measured during the experimental period are presented in Table 3.

Table 3. Water Quality Parameters During the Experimental Period

	Treatment				Water Quality Standard (SNI 01-7246-2006)
	A	B	C	D	
Temperature (°C)	26.8–28.9	27.0–29.5	26.8–29.2	27.0–29.0	28–33
pH	7.87–8.09	7.89–8.07	7.86–8.01	7.83–8.07	6–7
Dissolved oxygen (mg L^{-1})	5.08–6.68	4.54–5.94	4.69–5.91	4.92–5.91	> 4
Ammonia (mg L^{-1})	0.00–0.11	0.00–0.35	0.00–0.12	0.00–0.15	≤ 0.10

The water temperature recorded throughout the experimental period remained within the range considered suitable for the growth and survival of freshwater crayfish. In aquaculture systems, water temperature is one of the most important environmental variables influencing the physiological performance of cultured organisms. Previous studies have reported that the optimal temperature for freshwater aquaculture ranges from 28 to 32°C (Astiyani et al., 2024). Specifically, experiments on juvenile *Cherax quadricarinatus* reared at 26, 28, 30, and 32°C demonstrated the highest daily growth rate and survival at 28°C, while quadratic regression analysis also identified 28°C as the optimal culture temperature. Additional studies evaluating feeding physiology and feed utilization at 23, 27, and 31°C, together with recent nutritional studies on redclaw crayfish, have suggested an optimal growth range of approximately 23 ± 1 to 31 ± 1°C, with 24–30°C recommended for intensive culture and juvenile production (Cheng *et al.,* 2025).

The dissolved oxygen (DO) concentrations recorded during the study remained above 4 mg L^{-1}, satisfying the minimum requirement for the optimal growth and development of freshwater crayfish. Adequate dissolved oxygen is particularly important during molting because oxygen demand increases substantially during this physiologically demanding process. If DO concentrations fall below 4 mg L^{-1} during periods of mass molting, the risk of unsuccessful molting increases considerably, as crayfish may become exhausted before completely shedding their exoskeleton, resulting in mortality (Handajani et al., 2025). Freshwater crayfish possess a unique physiological adaptation that allows them to survive temporarily outside water provided that their gills remain moist, enabling them to absorb oxygen directly from the atmosphere and survive under such conditions for extended periods (Effendi *et al.,* 2015). Nevertheless, maintaining adequate dissolved oxygen in the culture water remains essential for normal growth and physiological performance.

The pH values recorded during the experimental period were slightly higher than the range specified in the Indonesian National Standard (SNI 01-7246-2006). However, they remained within the range reported by previous studies to support normal physiological function in *C. quadricarinatus*. Several studies have demonstrated that a pH between 7.5 and 8.2 promotes physiological stability by maintaining osmotic balance and metabolic activity, thereby reducing stress and supporting growth (Handajani *et al.,* 2025)**.** Water pH is a critical factor in freshwater crayfish culture because highly acidic conditions (pH < 5) may result in mortality, whereas highly alkaline conditions (pH > 9) can suppress appetite, leading to reduced feed intake and slower growth (Rihardi *et al.,* 2013).

Among the treatments, Treatment B exhibited the highest ammonia concentration (0.35 mg L^{-1}), exceeding the value recommended by the Indonesian National Standard (SNI 01-7246-2006). Elevated ammonia concentrations may adversely affect crayfish survival and overall culture performance. Nevertheless, Novita *et al.,* (2024) **r**eported that ammonia concentrations of 0.44 mg L^{-1} were still associated with satisfactory growth and survival of freshwater crayfish, while (Sugiura, 2018) recommended maintaining ammonia concentrations below 1 mg L^{-1} in fish and crustacean aquaculture systems to minimize toxic effects.

Overall, the water quality observations suggest that the organic materials applied in this study functioned not only as indirect nutritional supplements but also as environmentally friendly bioremediation agents that contributed to maintaining favorable water quality conditions for freshwater crayfish culture. Their application may enhance microbial activity, support nutrient cycling, and improve the overall stability of the culture environment, thereby promoting healthier and more sustainable aquaculture production.

CONCLUSION

Based on the results of this study, the highest absolute weight growth and specific length growth rate of freshwater crayfish (*Cherax quadricarinatus*) were obtained in Treatment C, which consisted of a combination of dried Indian almond leaves (*Terminalia catappa*) and pumpkin. This treatment produced an absolute weight gain of 0.78 g and a specific length growth rate of 2.16% day^{-1}.

The water quality observations suggest that the organic materials used in this study may function not only as indirect nutritional supplements but also as environmentally friendly bioremediation agents that help maintain favorable water quality conditions in freshwater crayfish culture. Although the treatments did not produce statistically significant differences in growth ($p > 0.05$), the observed improvements indicate that the application of selected organic materials has potential as a sustainable strategy to support freshwater crayfish production. Further studies involving longer culture periods and a broader range of treatment dosages are recommended to validate these findings.

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