



The effectiveness of eugenol in mitigating water quality, biochemical, and histopathological alterations during transportation of giant freshwater prawn juveniles

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Abstract

The giant freshwater prawn (*Macrobrachium rosenbergii*) is a high-value aquaculture commodity with increasing global demand. Adequate transportation of high-quality prawn juveniles is critical to ensuring sustainable production. Eugenol is widely known for its anesthetic characteristics. It can diminish metabolic rates and alleviate stress responses in aquatic organisms during handling and transportation. This study assessed the efficacy of eugenol, a natural anesthetic, in enhancing juvenile transport conditions by evaluating water quality, hemolymph biochemistry, histopathological alterations, and production performance. A randomized experimental design was employed with five eugenol treatments (0, 0.75, 1.87, 3.75, 5.63 mg/L) and control, combined with activated carbon and zeolite to maintain water quality during a 24-hour transport simulation. The results showed that the treatment with 1.87 mg/L eugenol yielded the best outcomes. This treatment maintained better water quality parameters, encompassing lower total ammonia nitrogen (TAN) and elevated dissolved oxygen (DO) levels compared to controls. Biochemical analysis revealed reduced stress response in juveniles, as evidenced by increased total protein, hemolymph glucose levels, high-density lipoprotein (HDL), and pH stability. Histopathological evaluation revealed minimal gill tissue hyperplasia and swift recovery post-transport from eugenol treatment. Treatment with 1.87 mg/L eugenol also achieved the highest juvenile survival rate (75.33%) at post-transportation (24 hours) and 10-day rearing. In contrast, the specific growth rate of all treatments was not significantly different. These findings underscore the potential of eugenol in enhancing the sustainability and transport efficacy of *Macrobrachium rosenbergii* juveniles.

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Introduction

The giant freshwater prawn (*Macrobrachium rosenbergii*) represents one of the high-value aquaculture products with perpetually escalating market demand. The global output of giant freshwater prawns in 2021 reached over 300 thousand metric tons, with a valuation surpassing \$2 billion. China is the foremost producer, producing

171,000 metric tons, constituting 54% of global production (1). A principal obstacle in facilitating the advancement of giant freshwater prawn aquaculture is the provision of high-quality seed (2). The quality of juveniles is heavily reliant on effective transportation methods, which is a crucial phase in guaranteeing the sustainability of *Macrobrachium rosenbergii* production (3). The transportation of giant freshwater prawn juveniles is a significant determinant in

facilitating the distribution of seeds to extensive cultivation site locations, even at considerable distances from seed production sites (2). Nonetheless, during transportation, there are frequent alterations in environmental conditions, which influence the physiological response (4). Modifications in ecological conditions to suboptimal levels during seed transportation will adversely impact the viability and efficacy of seed production; thus, efficient transportation methodologies are requisite to mitigate seed stress and mortality rates (3). Specific hematological biochemical parameters that can be discerned to evaluate stress response include glucose, total protein, pH, cholesterol, triglycerides, and HDL (5-10). Excessive excretion of ammonia-nitrogen ($\text{NH}_3\text{-N}$), CO_2 , and oxygen respiration by stressed fish can markedly diminish water quality (11-13). Stress attributable to variations in environmental conditions during transportation, such as temperature, and influencing fish biochemistry, can also impact gill histomorphology, resulting in epithelial hypertrophy and mucus secretion (14). Furthermore, elevated ammonia-nitrogen concentration in water during transport incites oxidative stress and apoptosis in fish hepatic tissues (11). In addition, if the stress condition occurs over an extended timeframe, it will detrimentally influence the fish's survivability and immunological response (15). Specific endeavors to establish conducive environmental conditions for *Macrobrachium rosenbergii* juveniles during transportation involve the utilization of additives such as eugenol, zeolite, and activated carbon (4,16,17). Eugenol serves in the capacity of a botanical anesthetic acquired from *Ocimum sanctum*, a range of Cinnamomum varieties, and *Myristica fragrans*; however, the *Syzygium aromaticum* species exhibits the highest output for eugenol extraction (18). Eugenol is advantageous for transporting live fish due to its capacity to enhance fish welfare and survival rates. Several studies indicate that eugenol effectively induces anesthesia in various fish taxa, including *Oreochromis niloticus* and *Seriola dumerili*. Eugenol alleviates stress levels during transportation by stabilizing physiological parameters such as plasma glucose while enhancing metabolic and antioxidant capacities in hepatic and gill tissues (19,20). Moreover, eugenol has been demonstrated to preserve water quality by elevating dissolved oxygen concentrations critical for fish health during transportation (20). Generally, eugenol functions as a promising alternative anesthetic, enhancing fish welfare during transportation (21). Zeolites operate as absorbers of ammonia and deleterious ions generated from the metabolic processes of organisms, thereby sustaining water quality (22). Concurrently, activated carbon is an adsorbent for organic substances that can compromise water quality (23). Combining these three constituents is anticipated to establish optimal environmental conditions for transporting giant freshwater prawn juveniles.

Further studies are essential to formulate transport methodologies capable of mitigating stress levels and

enhancing production effectiveness. In addition, experiments on an expanded scale are needed to ascertain their efficacy and applicability within the aquaculture sector. The objective of this research was to assess the efficacy of eugenol in optimizing juvenile transport conditions of *Macrobrachium rosenbergii*, with a focus on water quality, hemolymph biochemistry, histopathological alteration, and production effectiveness to bolster aquaculture sustainability. The outcomes of this investigation are anticipated to furnish pragmatic solutions to sustainably enhance the successful transportation of giant freshwater prawn juveniles.

Materials and methods

Ethical approve

This study was conducted at the Environmental Laboratory, Department of Aquaculture, Faculty of Fisheries and Marine Science, IPB University, following Ethical Approval No. 204/KH.06/SKE/07/2024, issued on 1st July 2024.

Experimental design

The research methodology employed a wholly randomized framework, encompassing five interventions of eugenol concentrations predicated on the previous 96-hour LC_{50} (7.5 mg/L) assay result multiplied by 10%, 25%, 50%, 75%, and 0%, namely, eugenol concentrations of 0.75 mg/L (A); 1.87 mg/L (B); 3.75 mg/L (C); 5.63 mg/L (D) and 0 mg/L (E) along with a single intervention utilizing solely water devoid of supplementary zeolite, activated carbon, and eugenol (F), with all interventions replicated threefold. The granulation of activated carbon implemented was $-40/\pm 60$ mesh with a Cation Exchange Capacity (CEC) of 3.91 me/100 g. The zeolite was $-40/\pm 60$ mesh with a CEC of 82.39 me/100 g. Activated carbon and zeolite doses used in treatments A, B, C, D, and E are 10 g/L and 20 g/L, respectively (17). The prawn juvenile employed is a giant freshwater prawn juvenile of *Macrobrachium rosenbergii* exhibiting a length range of 3.67 - 4.61 cm and an average mass of 0.412 ± 0.007 grams.

Transportation simulation procedure

The container for simulated shrimp transportation was a polyethylene (PE) bag filled with 1.5 Liters of freshwater augmented with substance for treatment A, B, C, D, E, and F, subsequently enriched with pure oxygen at a ratio of 2:1 relative to water volume. The PE bag also incorporates shelter in the form of nets measuring 20 x 20 cm². The density of juveniles is 100 individuals/L. The PE bag is stored in a Styrofoam container filled with ice cubes and subsequently sealed. Transportation simulation is executed for 24 hours, and the Styrofoam container is lightly shaken for 5 minutes every 1 hour.

Rearing procedure

Upon the conclusion of the transportation simulation, the juveniles from each treatment and replication were transferred into a glass aquarium and reared for 10 days. The dimensions of the aquarium utilized are 50x50x40 cm, which has been filled with fresh water to a depth of 40 cm and aerated, subsequently equipped with 30 units of PVC pipe shelters and 3 units of net shelters measuring 20 x 20 cm², with a larval density of 50 individuals per aquarium.

Water quality and production performance measurement

Water quality parameters such as temperature, DO, pH, CO₂, alkalinity, TAN, and nitrite were assessed utilizing the APHA methodology (24) and recorded at temporal intervals of 0, 4, 8-, 14-, 20-, and 24-hours during transport simulations. The survival rate (SR) and Specific Growth Rate (SGR) indicators were calculated employing the technique of Setijaningsih (25); the assessment of these indicators was executed after the transport simulation as well as at the termination of the rearing period in the aquarium.

Hemolymph analysis

Examination of physiological parameters of hemolymph in the form of glucose concentration utilizing the methodology of Li (26), quantification of total protein by Lowry (27), and evaluation of cholesterol levels employing the technique of Mercier (28). Measurement of HDL levels by the approach of Ruiz-Verdugo (29), pH assessed by (pH meter, HORIBA LaQuatwin), and hemolymph triglycerides assay utilizing the method of Barham and Trinder (30). Physiological parameters were evaluated at the commencement of the transport simulation (0 hours), after the simulation duration (24 hours), and subsequently during juveniles rearing in the aquarium at 27, 36, 48, 96, 192, and 264 hours (10th day).

Gill analysis procedure

Gills tissue was observed at the end of the transportation simulation (24 hours) and 264 hours. Gill sample analysis was performed at the Pathology Laboratory, Faculty of Veterinary Medicine, IPB University, following the method outlined by Jin (31). Samples fixed in a paraformaldehyde solution for 24 hours were subsequently rinsed with water, dehydrated using a graded ethanol series 70-95%, and cleared with xylene. Following paraffin embedding, the gill samples were sectioned into 4-5 μm thick slices using a microtome. The prepared sections were stained with haematoxylin and eosin, mounted with neutral resin, and examined under an optical microscope for imaging and analysis (32-36).

Data analysis

The quality of water and biochemical parameter data were subjected to statistical examination utilizing variance analysis (ANOVA) with the F-test at a 95% confidence

interval employing Minitab Statistical Software version 16.1.1. to ascertain the influence of treatments on the observed parameters. Fisher's subsequent test is used to determine significant disparities among treatments. The histological parameters were subjected to descriptive analysis.

Results

Water Quality

Based on figure 1, there was a decrease in water quality during the transport simulation, encompassing a reduction in DO and pH and an augmentation in TAN, nitrite, alkalinity, and CO₂ levels. Treatments not subjected to eugenol (E and F) exhibited a trend for inferior water quality compared to treatments administered with eugenol. The treatment of E and F during transport has a proclivity to diminish DO and pH values alongside elevated TAN and CO₂ values. Water quality across all treatments recorded during a 10-day rearing met the requisite aquaculture standard criteria (Table 1).

Table 1: Water quality parameters during juvenile rearing compared with the reference water quality requirements

Parameters	Rearing period	Reference sources
DO	(4.95 – 5.93) mg/L	(>4.48) mg/L (32)
pH	7.47 – 7.89	7 – 8.50 (33)
TAN	(0.21 -0.34) mg/L	<0.90 mg/L (34)
CO ₂	(1.99 – 3.42) mg/L	<5.90 mg/L (35)
NO ₂	0.01-1.53 mg/L	<5.00 mg/L (5)
Alkalinity	109.21-134.44 mg/L	100.0-150.0 mg/L (36)

Haemolymphs biochemical and production performance

Based on figure 2, treatment B exhibited better stress levels during the transportation simulation (24 h) than other treatments. Furthermore, this can be evidenced by the biochemical quantification values of hemolymph (glucose, total protein, HDL, triglycerides), which tend to converge with the values before transport (0 h). At the 27th to 264th hours after transport, biochemical alterations that transpired in all treatments failed to demonstrate significant distinct patterns among treatments. The highest survival rate during the transportation and rearing period was attained by the B treatment, which was 75.33±4.04 and 89.33±5.03%, while the lowest in the F treatment were 39.67±10.97 and 60.00±3.46%. The application of eugenol during transport exhibited no discernible effect (p>0.05) on SGR during juvenile rearing (Table 2).

Gill histopathology

Based on figure 3, after the transport simulation (24th hour), the gill tissue in each intervention exhibited hyperplasia. However, after the rearing of juvenile shrimp for 10 days, hyperplasia of the gill tissue in treatments A, B, C, D, and E was no longer observed.

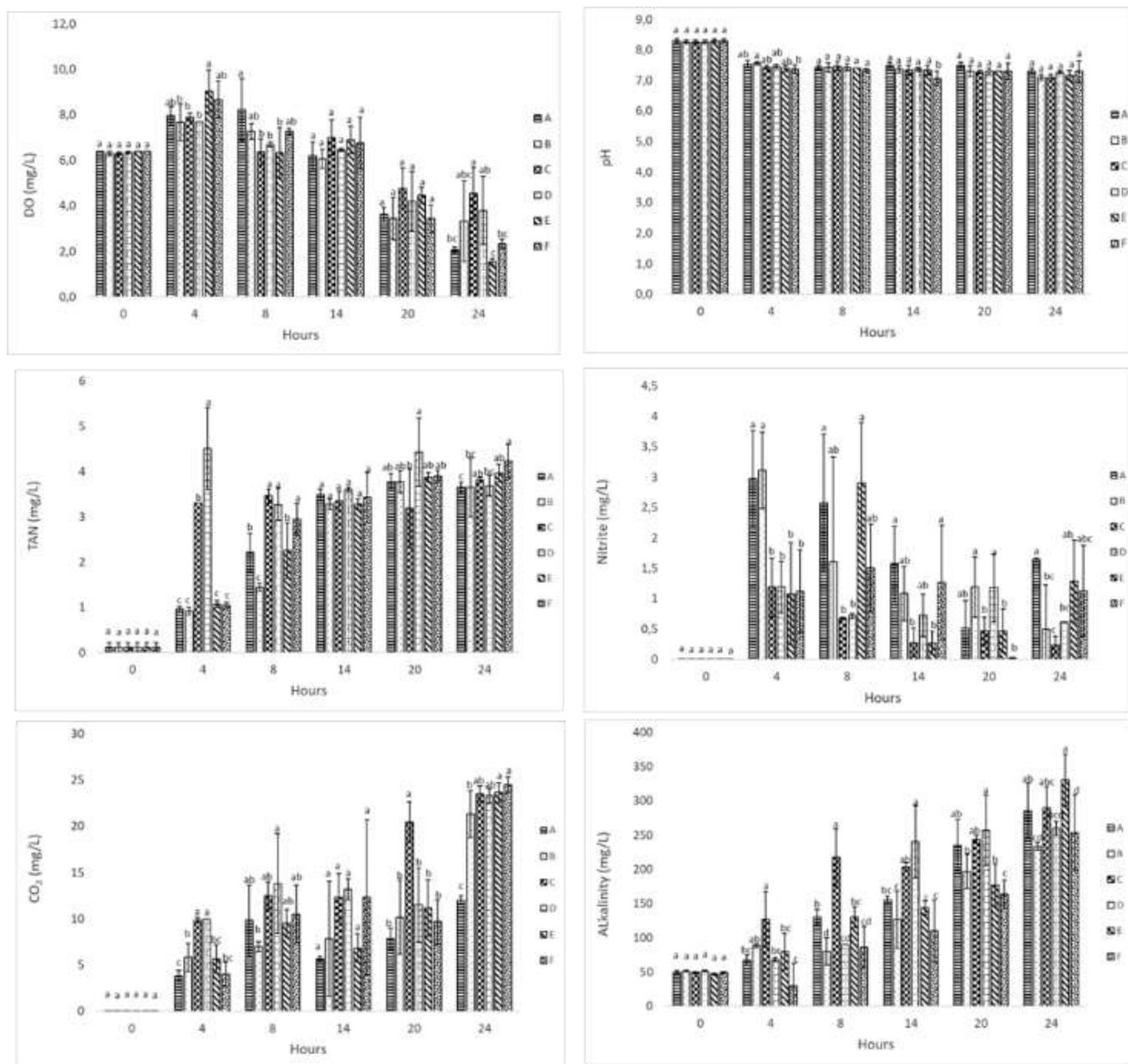


Figure 1: Water quality data was recorded during transportation simulation. A: 0.75 mg/L eugenol; B: 1.87 mg/L eugenol; C: 3.75 mg/L eugenol; D: 5.63 mg/L eugenol; E: 0 mg/L eugenol; F: freshwater without eugenol, zeolite, and activated carbon. Different lowercase letters in the graph indicate significant differences (P<0.05).

Table 2: *Macrobrachium rosenbergii* Juvenile production performance during the study

Parameters	A	B	C	D	E	F
SR transport (%)	61.33±6.03 ^{bc}	75.33±4.04 ^a	68.00±5.00 ^{ab}	66.33±4.73 ^{ab}	49.67±10.12 ^{cd}	39.67±10.97 ^d
SR rearing (%)	73.33±3.06 ^c	89.33±5.03 ^a	80.00±4.00 ^b	74.00±2.00 ^c	64.67±1.15 ^d	60.00±3.46 ^d
SGR (%/day)	2.18±0.12 ^a	2.30±0.11 ^a	2.23±0.07 ^a	2.29±0.12 ^a	2.11±0.09 ^a	2.09±0.11 ^a

Values in rows with different letters differ significantly (P < 0.05).

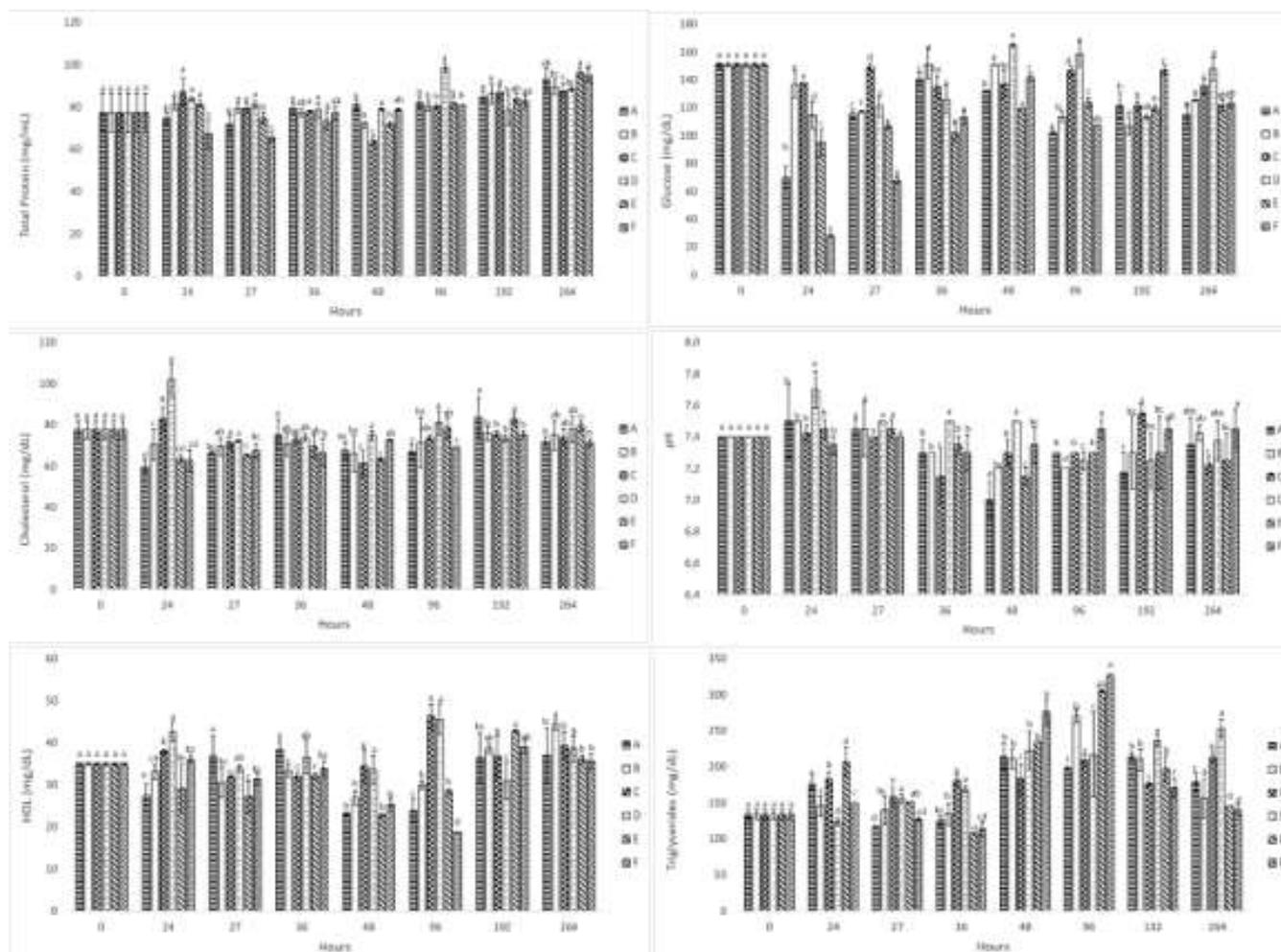


Figure 2: Haemolymph biochemical data were recorded during the transportation simulation (0 and 24 hours) and rearing period (27, 36, 48, 96, 192 and 264 hours). A: 0.75 mg/L eugenol; B: 1.87 mg/L eugenol; C: 3.75 mg/L eugenol; D: 5.63 mg/L eugenol; E: 0 mg/L eugenol; F: freshwater without eugenol, zeolite, and activated carbon. Different lowercase letters in the graph indicate significant differences ($P < 0.05$).

Discussion

Modifications in water quality that occur while transporting live fish can be connected to a range of intertwined factors, such as the physiological tension endured by the fish, the increase of metabolic residues, and limiting environmental conditions (9). Based on figure 1, it is observable that during transportation simulation, there is a deterioration in water quality, which encompasses a reduction in dissolved oxygen and pH values alongside an escalation in total ammonia nitrogen, alkalinity, and carbon dioxide values. Throughout transportation, fish undergo heightened respiratory rates as a reaction to stress, which culminates in augmented oxygen consumption, contributing to diminished DO levels in the water (12,37). Elevated carbon dioxide concentrations resulting from fish respiration further influence the reduction in DO. As CO_2 concentrations

rise, the pH of the water may decline, which consequently can impact the fish's capacity to assimilate oxygen (13).

The result of augmented fish metabolism during the transportation interval is heightened excretion of nitrogenous byproducts, notably ammonia. The elevated nitrogen waste discharge during transportation is a principal factor contributing to the increased ammonia concentrations in water, constituting one of the fish's primary stressors (37). The accumulation of carbon dioxide from respiratory activity can precipitate a decline in pH, which subsequently influences the equilibrium between unionized ammonia (NH_3) and ionized ammonium (NH_4^+), frequently culminating in elevated total ammonia measurements in water (38). The ion-exchange characteristics of zeolite facilitate the substitution of ammonium ions with alternative cations in water. This mechanism can induce an augmentation in the concentration of bicarbonate ions, a

crucial element of alkalinity. The interchange of ammonium ions with bicarbonate ions can effectively enhance the alkalinity of water (39). Furthermore, heightened carbon

dioxide concentrations promote more excellent solubility of calcium carbonate and calcium silicate in water, thereby elevating alkalinity parameters (40).

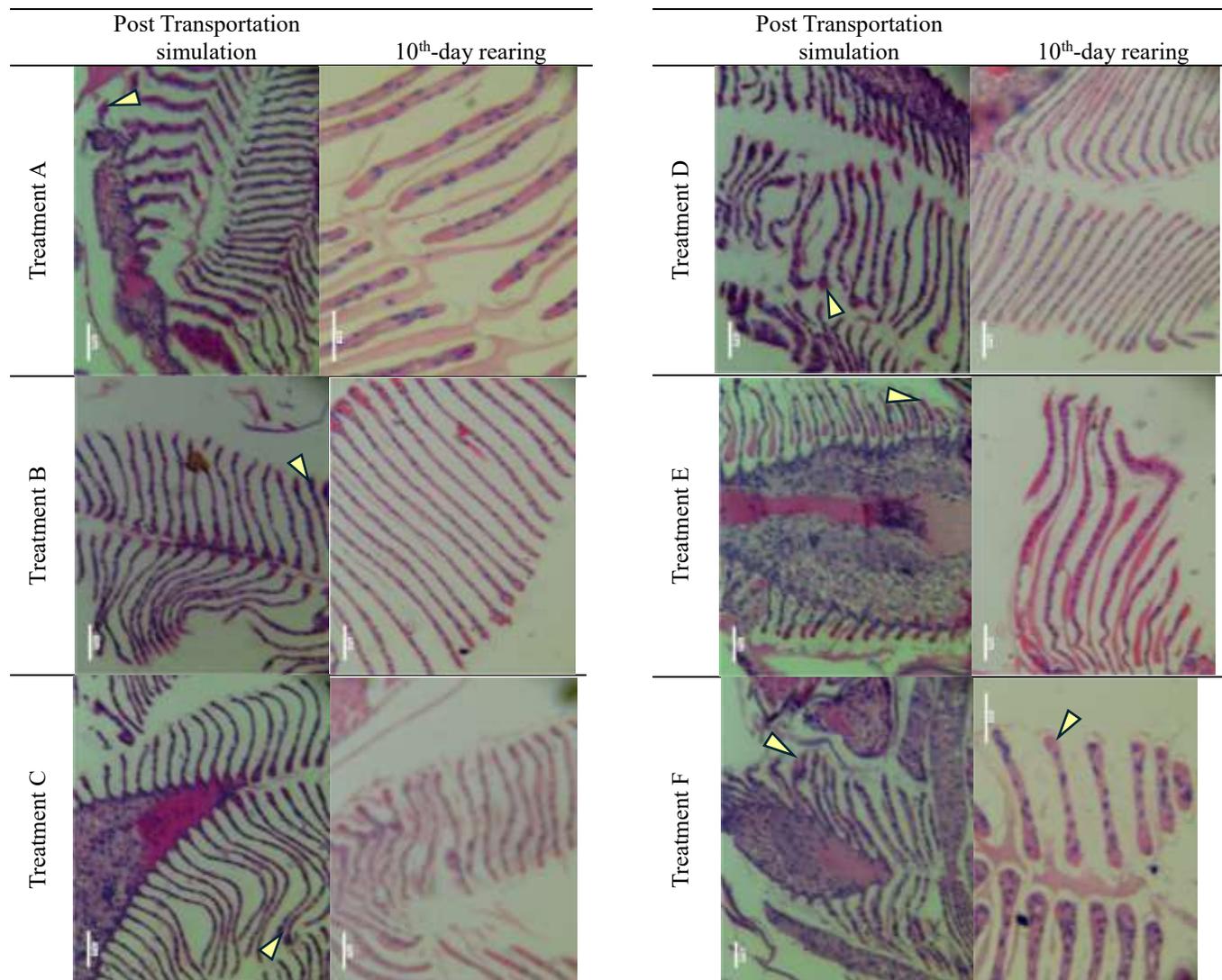


Figure 3: Micrograph of giant freshwater prawn (*Macrobrachium rosenbergii*) juvenile gills at post-transportation simulation and after rearing for 10 days. All treatments exhibited gill hyperplasia (yellow arrowhead) immediately after transportation simulation. A: 0.75 mg/L eugenol; B: 1.87 mg/L eugenol; C: 3.75 mg/L eugenol; D: 5.63 mg/L eugenol; E: 0 mg/L eugenol; F: freshwater without eugenol, zeolite, and activated carbon. Haematoxylin and eosin staining. Bar: 50 and 70 μm .

Figure 1 shows that applying eugenol in giant freshwater prawn juveniles' transportation exhibited a better water quality response during the study. Compared with the controls, this manifests from the enhanced DO, TAN, nitrite, and CO_2 values. Eugenol functions as a proficient anesthetic to diminish the metabolic activity of fish. Eugenol decreases overall fish stress levels, consequently reducing oxygen consumption and minimizing the generation of metabolic byproducts such as ammonia and carbon dioxide (41-43).

Applying activated carbon and zeolite in the treatment yielded a better water quality response than the controls. The concentration of TAN in the E treatment was diminished compared to the F treatment (control). Zeolites and activated carbon are efficacious in eliminating ammonia from water. Zeolite operates through ion exchange, wherein it sequesters ammonium ions from the water, thereby decreasing the concentration of ammonia, which is deleterious. Combining zeolite and activated carbon in a closed transportation system

effectively upholds water quality during the transport process and enhances fish survival (44).

According to Figure 2, it can be observed that glucose concentrations in treatment A, D, E, and F at the end of transportation simulation (24 h) encounter hypoglycemia, characterized by glucose levels significantly beneath normative glucose levels (preceding transportation simulation). Hypoglycemia may transpire owing to the juvenile undergoing considerable stress throughout the transport procedure. When fish are subjected to stress, there is a substantial cortisol secretion. Cortisol is a stress-associated hormone that exerts a crucial influence on glucose metabolism. Cortisol initiates glycogenolysis and gluconeogenesis, which typically elevate blood glucose levels in reaction to stress (45). Nonetheless, if stress is extended or acute, this may culminate in the exhaustion of glycogen reserves, leading to hypoglycemia (46).

This phenomenon is distinctly observable during the transportation of live fish exhibiting elevated density levels, wherein an augmentation in blood cortisol concentrations is not invariably succeeded by a corresponding elevation in blood glucose concentrations (47). Although cortisol concentrations escalate in response to stress, glucose concentrations may occasionally diminish or even decline should the stress persist over an extended duration (46). Overall, hemolymph glucose concentrations at the end of transport simulation and during juvenile rearing indicated that the application of eugenol in treatments B and C yielded better stress response rates than the other treatments. From the findings of this study, it is evident that eugenol possesses a proclivity to ameliorate stress in fish more effectively than the control group (47).

Some of the biochemical parameters evaluated also manifested a similar correlation with the trajectory of hemolymph glucose levels at the end of the transport simulation (24 hours). Based on Figure 2, total protein, cholesterol, triglycerides, HDL, and pH, particularly in treatments administered eugenol (B, C, and D), appear to be elevated compared to other treatments. When fish are subjected to stress, cortisol concentrations will be augmented. Elevated cortisol can activate protein synthesis in the liver, resulting in heightened levels of total protein in the circulatory system, such as increased globulin concentrations; this response implies a form of fish's adaptation to manage stress (48). Furthermore, under stress conditions, fishes will undergo alterations in their energetic metabolism, which culminates in augmented protein synthesis as the organism endeavors to attain homeostasis (49). A significant constituent of total protein (TP) is albumin. Albumin fulfills a crucial function in preserving oncotic pressure, which is essential for the equilibrium of fluids within the circulatory system (50). In periods of stress, heightened total protein levels can assist in stabilizing blood osmolarity and averting edema, thereby promoting homeostatic mechanisms (50). The propensity towards

hypoglycemia in treatments A, D, and F renders the fish incapable of effectively responding to the stress that arises, attributable to the incapacity to swiftly achieve homeostasis. This phenomenon can be evidenced by diminished cholesterol, triglycerides, HDL, and pH concentrations during the 24-hour transport period.

Elevated cholesterol and triglyceride levels are frequently associated with fulfilling energy demands for stress-induced homeostatic processes (51,52). When fish face sudden stress, there is a notable rise in triglycerides, HDL, and cholesterol levels, indicating that energy availability, exhibited through triglyceride levels, plays a crucial role in modulating stress reactions in fish (10). The primary hormone indicative of stress, cortisol, is instrumental in regulating lipid metabolism by facilitating triglyceride and cholesterol biosynthesis and mobilization from adipose tissue (53,54). As illustrated in Figure 2, it is revealed that throughout the study, the administration of eugenol resulted in an escalation of HDL concentration in the hemolymph. Acknowledged for its extensive range of physiological functions, HDL is substantial in cholesterol transport and exhibits anti-inflammatory properties, which stressful environments may influence. HDL facilitates mechanisms that counteract inflammation and protect cell damage, which could be beneficial in mitigating the adverse effects of stress (55,56). Furthermore, this suggests that eugenol application reduces stress response during transportation.

Eugenol administration also induces elevated hemolymph pH levels compared to controls at the end of transportation simulation (24 hours). Acute stress can be correlated with heightened synthesis of lactate and CO₂ in the bloodstream, thereby reducing blood pH (57). Overall, the findings of biochemical hemolymph analyses during the 27th hour of the rearing period (3 hours post-transportation) across all treatments already demonstrate a normative biochemical status of the hemolymph, analogous to the condition preceding transportation. Therefore, this indicates that 3 hours after juveniles' transportation, they can acclimate to revert to standard conditions before being transported. Throughout the rearing period until 264 hours, no statistically significant stress impact was observed among treatments. During the rearing period of 10 days, the juveniles were cultured under identical water quality parameters, ensuring no environmental discrepancies affected their physiological conditions.

The gill tissue exhibited histopathological alterations manifested as hyperplasia without any evidence of significant impairment. Hyperplasia is conventionally identified by primary lamellar hypertrophy; it is frequently associated with the proliferation of mucous-secreting cells as an adaptive response to environmental stressors, such as fluctuations in pH, accumulation of CO₂, ammonia, and other metabolic byproducts (34,58). Treatments D and C displayed the most pronounced hyperplasia at the onset of

the rearing period. In contrast, treatment F demonstrated the most protracted tissue recuperation. The findings of the histological examination of the gills on the tenth day of rearing, all treatments, except for treatment F, exhibited a return of gill tissue to baseline conditions. The primary lamellar morphology in treatment F retained a greater thickness than in other treatments, implying a diminished likelihood of recovery (59). In summary, gill tissue regeneration was apparent from the reduced thickening of hyperplasia across all treatments by the conclusion of the observation period.

The utilization of eugenol culminated in enhanced juvenile viability at the end of the transportation simulation (24 hours) and throughout a 10-day rearing period. Eugenol is a gentle anesthetic that soothes aquatic organisms during transportation and diminishes metabolic activity, cortisol concentrations, and stress-related gene expression, thereby fostering improved water quality to mitigate mortality during transportation (4). This assertion is substantiated by numerous studies indicating that the application of eugenol does not yield a significant long-term effect on fish growth and performance post-transport (43,60).

Conclusions

The results of this study showed that the anesthetic properties of eugenol during transport were instrumental in reducing the stress level of juvenile shrimp. Moreover, eugenol did not adversely affect juvenile shrimp development at the rearing phase's end. Therefore, this indicates that the dose of eugenol used in this study is deemed safe to apply during transport, as it does not interfere with the growth potential of juvenile shrimp over a long period. Based on the findings of this study, it can be concluded that the treatment with 1.87 mg/L eugenol was the optimal intervention, yielding better stress levels, water quality, and survival rate (75.33±4.04%) during simulated transport compared to other treatments.

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Conflict of interest

The authors declare no conflict of interest.

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فعالية الأوجينول في التخفيف من التغيرات في جودة المياه والتغيرات البيوكيميائية والنسجية المرضية أثناء نقل صغار الجمبري العملاق في المياه العذبة

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الخلاصة

يعتبر الجمبري العملاق (*Macrobrachium rosenbergii*) سلعة عالية القيمة للاستزراع المائي مع تزايد الطلب العالمي. يعد النقل المناسب لصغار الجمبري عالي الجودة أمراً بالغ الأهمية لضمان الإنتاج المستدام. يشتهر الأوجينول على نطاق واسع بخصائصه المخدرة. يمكن أن يقلل من معدلات الأيض ويخفف من استجابات الإجهاد في الكائنات المائية أثناء المناولة والنقل. قيمت هذه الدراسة فعالية الأوجينول، وهو مخدر طبيعي، في تحسين ظروف نقل صغار الجمبري من خلال تقييم جودة المياه، والكيمياء الحيوية للهيموليمف، والتغيرات النسيجية المرضية، وأداء الإنتاج. تم استخدام تصميم تجريبي عشوائي مع خمسة معالجات يوجينول (0، 0.75، 1.87، 3.75، 5.63 ملغم / لتر) ومجموعة سيطرة، جنباً إلى جنب مع الكربون المنشط والزيوليت للحفاظ على جودة المياه خلال محاكاة النقل على مدار 24 ساعة. أظهرت النتائج أن العلاج بـ 1.87 ملغم / لتر يوجينول أسفر عن أفضل النتائج. حافظت هذه المعالجة على معايير جودة مياه أفضل، بما في ذلك انخفاض نيتروجين الأمونيا الكلي (TAN) ومستويات الأكسجين المذاب المرتفعة (DO) مقارنة بمجموعة السيطرة. كشف التحليل الكيميائي الحيوي عن انخفاض استجابة الإجهاد لدى صغار الجمبري، كما يتضح من زيادة البروتين الكلي، ومستويات الجلوكوز الهيموليمف، والبروتين الدهني عالي الكثافة (HDL)، واستقرار الأس الهيدروجيني. كشف التقييم النسيجي المرضي عن الحد الأدنى من تضخم أنسجة الخياشيم والتعافي السريع بعد النقل من علاج الأوجينول. كما حققت المعالجة بـ 1.87 ملغم/لتر من الأوجينول أعلى معدل لبقاء الأحداث (75.33%) في مرحلة ما بعد النقل (24 ساعة) والتربية لمدة 10 أيام. في المقابل، لم يكن معدل النمو المحدد لجميع العلاجات مختلفاً بشكل كبير. تؤكد هذه النتائج على إمكانات الأوجينول في تعزيز استدامة وفعالية نقل صغار الجمبري.