

THE USE OF CLOVE OIL (*Syzygium aromaticum*) AS AN ANESTHETIC TO THE SURVIVAL RATE OF VANAME (*Litopenaeus vannamei*) POSTLARVAE IN TRANSPORTATION

Penggunaan Minyak Cengkeh (*Syzygium aromaticum*) Sebagai Anestesi Untuk Meningkatkan Tingkat Kelangsungan Hidup Larva Vaname (*Litopenaeus vannamei*) Dalam Transportasi

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ABSTRACT

Closed transportation of vannamei post larvae at hatchery facilities is still considered relatively ineffective. This is evidenced by several problems encountered in the field, such as post transport survival rates that have not yet reached a high category. This study aimed to determine the survival rate of vannamei post larvae transported under a closed system with the application of clove oil. The study employed a Completely Randomized Design (CRD) consisting of four treatments with three (3×) replications, resulting in a total of twelve (12) experimental units : P1 (0%), P2 (0.10%), P3 (0.20%), and P4 (0.30%). The application of different clove oil dosages in closed transportation showed significant differences among treatments in terms of postlarval survival ($P < 0.05$). The survival rates obtained were 53.1% for treatment P1, 87.9% for P2, 88.7% for P3, and 62.4% for P4. Based on measurements of water quality parameters during the study, the conditions within the plastic bag containers after 24 hours of transportation remained within tolerance limits suitable for vannamei postlarvae maintenance. Water quality values after transportation were recorded as follows: temperature 29.38-29.41 °C, dissolved oxygen 4.70-4.75 ppm, pH 7.30-7.39, and ammonia 0.02-0.05 ppm.

Keywords : Survivale Rate *Litopenaeus vannamei*, *Syzygium aromaticum*, Transportation

ABSTRAK

Pengangkutan larva udang vannamei secara tertutup di fasilitas penetasan masih dianggap relatif tidak efektif. Hal ini dibuktikan dengan beberapa masalah yang ditemui di lapangan, seperti tingkat kelangsungan hidup pasca pengangkutan yang belum mencapai kategori tinggi. Penelitian ini bertujuan untuk menentukan tingkat kelangsungan hidup larva udang vannamei yang diangkut dalam sistem tertutup dengan aplikasi minyak cengkeh. Penelitian ini menggunakan Rancangan Acak Lengkap (CRD) yang terdiri dari empat perlakuan dengan tiga (3×) ulangan, sehingga menghasilkan total dua belas (12) unit percobaan: P1 (0%), P2 (0,10%), P3 (0,20%), dan P4 (0,30%). Aplikasi berbagai dosis minyak cengkeh dalam pengangkutan

tertutup menunjukkan perbedaan signifikan antar perlakuan dalam hal kelangsungan hidup pasca larva ($P < 0,05$). Tingkat kelangsungan hidup yang diperoleh adalah 53,1% untuk perlakuan P1, 87,9% untuk P2, 88,7% untuk P3, dan 62,4% untuk P4. Berdasarkan pengukuran parameter kualitas air selama penelitian, kondisi di dalam wadah kantong plastik setelah 24 jam pengangkutan tetap berada dalam batas toleransi yang sesuai untuk pemeliharaan larva udang vannamei. Nilai kualitas air setelah pengangkutan tercatat sebagai berikut: suhu 29,38-29,41 °C, oksigen terlarut 4,70-4,75 ppm, pH 7,30-7,39, dan amonia 0,02-0,05 ppm.

Kata Kunci: Tingkat Kelangsungan Hidup *Litopenaeus vannamei*, *Syzygium aromaticum*, Transportasi

INTRODUCTION

Vannamei (*Litopenaeus vannamei*) is a flagship product of the fisheries sector and has currently become one of Indonesia's leading commodities in enhancing export competitiveness in the global market. Moreover, this species possesses several advantages, including rapid growth, resistance to diseases, and high economic value (Latifah *et al.*, 2025). Global demand for shrimp continues to increase, resulting in a growing need for the availability of high quality and sustainable shrimp seed. The quality of the seed used is a crucial component in the success of shrimp aquaculture. The transportation process from hatcheries to grow out sites greatly influences this quality. This condition motivates shrimp farmers to continuously carry out grow out activities in a sustainable manner. Such grow-out operations must be consistently supported by hatchery activities to minimize the risk of shortages in vannamei post larvae supply at the production level. According to (Jannah *et al.*, 2024) hatchery operations represent a critical stage in aquaculture activities, as they determine subsequent phases, particularly the grow out stage. In other words, hatchery activities are processes aimed at producing seed, which serves as the primary input component in grow out operations.

A natural material that can be used as an alternative to reduce mortality rates during long-distance transportation of fish seed is clove oil (Mikhsalmina *et al.*, 2017). Clove oil contains major active compounds, namely eugenol, eugenyl acetate, and β -caryophyllene, which function as anesthetic, antiemetic, carminative, stimulant, antispasmodic, and antiseptic agents (Wimadani & Fitri, 2024). Clove oil functions as an anesthetic, which in this context serves to sedate fish. When fish are sedated, their activity levels are reduced, thereby suppressing metabolic processes and consequently decreasing the production of carbon dioxide, which readily dissolves in water. One effective approach to minimizing fish stress during transportation and maintaining high survival rates is the application of anesthesia at low temperatures. This method is considered effective because it does not leave harmful residues (Hayati *et al.*, 2022). Fish subjected to anesthesia exhibit reduced metabolic activity, resulting in lower oxygen requirements and minimal production of metabolic byproducts such as ammonia and carbon dioxide.

Closed transportation of vannamei post larvae at hatchery facilities is still considered relatively ineffective, as evidenced by the emergence of several practical issues in the field, such as post transport survival rates that have not yet reached a high category (Anggoro *et al.*, 2022). This condition encourages hatchery operators to continuously innovate in order to ensure that closed transportation of vannamei postlarvae can be conducted effectively and achieve high survival rates. In general, shrimp hatcheries are not located in close proximity to grow out sites, and transportation duration varies; consequently, declines in water quality particularly dissolved oxygen levels may occur, leading to physiological stress and reduced survival of vannamei seed (Zaidy & Eliyani, 2021). Based on this background, it is necessary to conduct research on the use of clove oil for the transportation of vannamei post larvae. The

objective of this study is to determine the optimal dosage of clove oil to maximize the survival of shrimp seed transported under closed conditions for 24 hours.

RESEARCH METHODOLOGY

Time and Place

The study was conducted in October 2025 at the Shrimp Hatchery Teaching Factory, Field Practice Installation, Technical University of Fisheries, Ministry of Marine Affairs and Fisheries, Republic of Indonesia, Serang Campus, located on Jl. STP Raya Banten, Kasemen District, Serang City, Banten Province.

Working Method

This study employed a Completely Randomized Design (CRD) with four treatments and three replicates, resulting in a total of twelve (12) experimental units, as follows: P1: 0%, P2: 0.10%, P3: 0.20%, and P4: 0.30%.

Test Seed

The shrimp seed used in this study were postlarvae at stage PL-9, with a density of 1,600 individuals per liter of water. The vannamei postlarvae selected as test animals were healthy, free of deformities, uniform in size, and actively swimming. Prior to transportation, the postlarvae underwent a one day fasting period to reduce metabolic activity. The post larvae were then placed in plastic bags, which were inserted into styrofoam containers with ice, and subsequently loaded into a vehicle for 24 hour transportation. After transportation, the styrofoam containers were opened simultaneously, and the number of surviving post larvae in each plastic bag was counted. Following transportation, the shrimp seed were maintained for seven (7) days in aquaculture containers specifically, 75 liter buckets equipped with aeration. During this period, feeding was conducted regularly, and survival rates were recorded at the end of the maintenance period.

Media Preparation

The initial step involves preparing 30 × 60 cm plastic bags, rubber bands, styrofoam containers, ice, adhesive tape, oxygen cylinders, and clove oil. One liter of water is added to each plastic bag, followed by the addition of clove oil at concentrations of 0%, 0.10%, 0.20%, and 0.30%. Subsequently, 1,600 post larvae are introduced into each bag, and oxygen is supplied at a ratio of 1:3 (oxygen:water) (Anggoro *et al.*, 2022) The plastic bags were then securely tied and placed into styrofoam containers pre filled with ice. The containers were sealed with adhesive tape to maintain a stable internal temperature. Next, the styrofoam containers were loaded into a vehicle for a 24 hour transportation period. Each treatment was repeated three times (3×) to ensure replication.

Treatment

Survival Rate

To assess the success of shrimp seed transportation, the measured parameter was the post transport survival rate of the vannamei post larvae. After transportation, the post larvae were maintained for seven (7) days in 75 liter buckets equipped with aeration to evaluate the effect of clove oil use on closed system post larvae transportation. The survival rate of vannamei post larvae was calculated using the formula proposed by Effendie (1977), as cited in (Zaidy *et al.*, 2021).

$$SR = \frac{N_t}{N_o} \times 100\%$$

Description :

SR : Survival rate (%).

Nt : Number of vannamei post larvae alive at the end of the experiment.

No : Number of vannamei post larvae at the beginning of the experiment.

Water Quality

During the study, water quality parameters in the transportation containers were measured both before and after the transportation process. The parameters and measurement methods for water quality are presented in Table 1.

Table 1. Water Quality Parameters and Measurement Methods

| Parameter | Unit | Measuring Instrument | Measurement Method |
|------------------|------|----------------------|--------------------|
| Temperature | °C | Termometer | In situ |
| Dissolved Oxygen | ppm | DO meter | In situ |
| pH | - | pH meter | In situ |
| Ammonia | ppm | Test kit | In situ |

Data Analysis

The post transport survival data of vannamei post larvae were calculated for each treatment and subsequently analyzed using Analysis of Variance (ANOVA) at a 95% confidence level with SPSS version 26. If significant differences were detected, further analysis was conducted using Duncan's multiple range test. Water quality measurements were analyzed descriptively.

RESULTS AND DISCUSSION

Survival Rate

The results of the treatment using clove oil in closed transportation on the survival of vannamei post larvae are presented in Table 2.

Table 2. Survival rate of vannamei post larvae

| Treatment | Survival Rate (%) |
|-----------|-------------------|
| P1 | 53,1 ^a |
| P2 | 87,9 ^c |
| P3 | 88,7 ^c |
| P4 | 62,4 ^b |

Description :

P1 : Clove oil dose 0%

P2 : Clove oil dose 0.10%

P3 : Clove oil dose 0.20%

P4 : Clove oil dose 0.30%

Based on the table above, the use of clove oil in shrimp post larvae transportation showed significant differences in survival rates among treatment groups ($P < 0.05$). The survival rates observed were 53.1% for P1, 87.9% for P2, 88.7% for P3, and 62.4% for P4. Analysis of clove oil usage across treatments indicates that higher doses tended to result in lower survival rates, whereas lower doses provided the highest survival. This suggests that clove oil is effective as a natural anesthetic agent for live fish transportation. The eugenol content induces a sedative

effect on vannamei post larvae, resulting in reduced swimming activity and eventual sedation. The study results indicate that the highest survival rate, 88.7%, was achieved in treatment P3, suggesting that this represents the optimal clove oil dosage for shrimp post larvae transportation. These findings are consistent with previous research (Fardhani *et al.*, 2025) which stated that the administration of different clove oil doses had a significant effect on the highest survival rate of Nile tilapia, reaching 95%. Other studies have shown similar results (Wimadani & Fitri, 2024) also suggested that post-transport survival of fish seed reached 100% with the use of clove oil. The active component of clove oil, eugenol, acts as an anesthetic, reducing the physiological activity of the fish, which is consistent with the statement (Rozik *et al.*, 2025) that eugenol can reduce central nervous system activity, decrease oxygen consumption, motor activity, and stress responses in fish. When metabolism decreases, oxygen consumption during transportation is reduced, carbon dioxide levels decline, and ammonia accumulation in the plastic bags remains low, which is supported by the statement (Anggoro *et al.*, 2022) which stated that the metabolism of shrimp post-larvae, producing feces during transportation, can lead to a decline in water quality. The effect of clove oil use has been successfully investigated in several studies; in some cases, it had a positive impact on fish survival, while in others, it did not significantly affect survival. These findings are in line with the research (Mikhsalmina *et al.*, 2017) which reported that the post transport survival rate of milkfish (*Chanos chanos*) seed was 67.3%.

Water Quality

The results of water quality parameter measurements during the study are presented in Table 3.

Table 3. Water Quality Parameter Measurements During the Study

| Treatment | Temperature (°C) | | Dissolved Oxygen (ppm) | | pH | | Ammonia (ppm) | |
|-----------|------------------|-------|------------------------|-------|--------|-------|---------------|-------|
| | Before | After | Before | After | Before | After | Before | After |
| P1 | 26,10 | 29,38 | 5,12 | 4,70 | 7,01 | 7,39 | 0,02 | 0,05 |
| P2 | 26,12 | 29,40 | 5,13 | 4,74 | 7,03 | 7,35 | 0,02 | 0,04 |
| P3 | 26,10 | 29,37 | 5,14 | 4,75 | 7,02 | 7,36 | 0,02 | 0,03 |
| P4 | 26,11 | 29,41 | 5,12 | 4,74 | 7,01 | 7,30 | 0,02 | 0,05 |

Based on the results of water quality parameter measurements during the study, the conditions in the plastic bag containers after 24 hours of transportation remained within the tolerance limits suitable for vannamei post larvae maintenance. The temperature values before transportation ranged from 26.10 to 26.12°C, and after transportation ranged from 29.38 to 29.41°C. These values are still within acceptable standards for vannamei seed transportation, which is consistent with previous research (Anggoro *et al.*, 2024). The water temperature during transportation remained relatively stable and within a range suitable for the survival of post larvae, with temperatures during 48-hour transport ranging from 29.13 to 29.33°C. This is consistent with (SNI, 2010), which states that for transport durations of less than 6 hours, the water temperature should be 26–28°C, whereas for 18–24 hours it should be 20–22°C. In closed-system transportation, temperature plays a critical role in reducing metabolic activity; lower temperatures decrease post larvae activity, thereby reducing oxygen demand and the production of metabolic wastes such as ammonia and carbon dioxide (CO₂). As a result, the post larvae become more resilient during transportation.

The dissolved oxygen (DO) levels before shrimp post-larvae transportation ranged from 5.12 to 5.14 ppm, and decreased to 4.70–4.75 ppm after 48 hours of transport. This indicates a decline in DO levels during transportation; however, these values are still within a suitable

range for shrimp seed maintenance. This is consistent with (Madyowati *et al.*, 2021), who reported that dissolved oxygen levels in post transport fish ranged from 4.78 to 4.9 ppm. Dissolved oxygen is essential as the primary source of oxygen for vannamei post larvae to respire and carry out metabolic processes, thereby supporting their survival during transportation. This aligns with (Wahyuni *et al.*, 2022), who stated that dissolved oxygen transports nutrients via the shrimp's blood to the entire body, producing energy necessary for physiological activities. Similarly, (Albab *et al.*, 2025) emphasized that dissolved oxygen is a vital element for shrimp respiration and metabolic processes, and low DO levels can lead to breathing difficulties and physiological stress.

The pH values before transportation ranged from 7.01 to 7.03, and after post transportation, they ranged from 7.30 to 7.39, indicating that the water remained within a suitable range for the survival of vannamei post larvae. This is consistent with (Mikhsalmina *et al.*, 2017), who reported that the final pH of fish seed after closed system transportation ranged from 6.5 to 7. The role of pH in closed transportation is to maintain the physiological balance of the fish; stable pH supports osmoregulation and metabolic processes, whereas excessively low or high pH can cause stress and respiratory disturbances in the post larvae. This aligns with the statement by William & Robert (1992), cited in (Nasmi *et al.*, 2017), that pH decline during transportation is caused by increased carbon dioxide concentration, which reacts with water to form carbonic acid (H_2CO_3). Stable pH reduces stress and mortality risk, while low pH can suppress microbial activity, helping to limit the growth of bacteria that deplete oxygen and produce harmful metabolites. This is supported by (Wahyuningsih & Gitarama, 2020), who stated that under slightly alkaline conditions (pH 7–8.5), nitrifying bacteria grow optimally; however, at $\text{pH} \geq 8.5$, the growth of *Nitrobacter* is inhibited compared to *Nitrosomonas*. Both *Nitrosomonas* and *Nitrobacter* require sufficient oxygen, and if oxygen levels are inadequate, nitrification is hindered, leading to increased ammonia and nitrite concentrations.

The ammonia concentration before and after transportation ranged from 0.02 to 0.05 ppm, indicating that the ammonia levels in the plastic containers remained within a suitable range for the survival of vannamei post larvae. This is consistent with (Anggoro *et al.*, 2024), who reported post transport ammonia levels of 0.03–0.09 mg/L in post larvae transported using pure oxygen. These findings suggest that the use of clove oil during transportation is highly effective, as it restricts post larvae movement due to the anesthetic effect of eugenol, thereby reducing metabolic waste production during transport. This is further supported by the study of (Wimadani & Fitri, 2024), which reported that water quality tests during Nile tilapia transportation using clove oil as an anesthetic showed ammonia levels of 0.06 mg/L. Ammonia is utilized in photosynthesis during the daytime, while in the morning, evening, and night, it is not processed through photosynthesis.

CONCLUSION

Based on the results of this study, it can be concluded that the use of clove oil at different doses significantly affected the survival of vannamei post larvae during closed system transportation. The treatments P1 (0%), P2 (0.10%), P3 (0.20%), and P4 (0.30%) showed significant differences in survival rates among groups ($P < 0.05$). The highest survival rate was observed in treatment P3 at 88.7%, while the lowest was in the control group P1 at 53.1%. Water quality parameters during post transportation were as follows: temperature 29.38–29.41°C, dissolved oxygen (DO) 4.70–4.75 ppm, pH 7.30–7.39, and ammonia 0.02–0.05 ppm.

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