



Potential of *Artemia* biomass cultivated from white leg shrimp wastewater as a supplemental daily feed: Effects on shrimp growth performance, survival, and feed efficiency

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Abstract. Sustainable aquaculture requires innovative solutions to minimize environmental pollution and optimize resource utilization. This study investigated the potential of using wastewater from white leg shrimp (*Litopenaeus vannamei*) ponds to cultivate *Artemia*, and then using *Artemia* as a dietary supplement for shrimp, in order to develop a sustainable recirculating aquaculture model. The experiment consisted of five treatments, supplementing *Artemia* biomass (wet weight basis) at levels of 0% (control, 0% SA - Supplemented Artemia), 10% (10% SA), 20% (20% SA), 30% (30% SA), and 40% (40% SA) of the daily commercial feed, with four replicates per treatment. Shrimp were cultured in a recirculating biofilter system (200 L tank⁻¹) at a density of 60 ind tank⁻¹ for 40 days. The effects of dietary *Artemia* supplementation on growth performance, survival rate, and feed efficiency of white leg shrimp were evaluated. The results demonstrated that *Artemia* could be successfully cultured using shrimp pond wastewater, achieving high biomass productivity with a nutritional composition suitable for shrimp dietary supplementation. Supplementing *Artemia* into the diet significantly improved weight gain, biomass, survival rate, and feed efficiency of white leg shrimp. The supplementation levels of 30% SA and 40% SA improved weight gain by 2.94-7.18%, biomass by 14.54-14.89%, survival rate by 7.14-11.69%, and feed efficiency by 4.14-12.68% compared to the control ($p < 0.05$). These findings provide scientific evidence for the potential application of a recirculating aquaculture model that reduces environmental pollution and enhances resource use efficiency. The study suggests that a supplementation level of 30% SA is effective for enhancing the culture efficiency of white leg shrimp in intensive farming systems. However, further research is needed to optimize the *Artemia* production process using shrimp pond wastewater and determine the optimal supplementation levels for different cultured species and farming systems. The outcomes of this research contribute to the advancement of sustainable aquaculture practices, aiming at improving the productivity, quality, and value addition for aquaculture products, while reducing environmental impacts.

Key Words: *Litopenaeus vannamei*, nutrient recycling, recirculating aquaculture.

Introduction. Aquaculture, particularly white leg shrimp (*Litopenaeus vannamei*) farming, has become a globally significant industry, contributing substantially to food security and economic development. The global production of farmed shrimp has increased from 3.2 million tons in 2010 to 5.6 million tons in 2021, and is expected to continue growing in the coming years (FAO 2024). However, the rapid expansion of intensive shrimp farming has been accompanied by environmental challenges, such as increased wastewater generation and high feed costs. To produce one kg of marketable shrimp, the estimated water consumption in major shrimp-producing countries like Ecuador, India, China, Vietnam, and Thailand ranges from 14 to 76.8 m³ (Boyd et al 2021). Furthermore, shrimp can only assimilate 19.1-23.6% of the nitrogen from feed, while the remaining is discharged into the aquatic environment as organic matter (25.3-39.5%) and dissolved inorganic salts (NH₄⁺, NO₂⁻, NO₃⁻) accounting for 41.4-51.1% (Chen et al 2018). Wastewater from shrimp farming has negatively impacted the environment due to the accumulation of waste causing water eutrophication, disease

outbreaks, and biodiversity loss, consequently compromising the sustainability of shrimp farming (Iber & Kasan 2021; Tom et al 2021).

To address the wastewater and waste issues in shrimp farming, several advanced shrimp farming technologies have been researched and applied, such as biofloc technology, recirculating aquaculture systems (RAS), aquaponics in RAS, aquamimicry, microalgae-assisted aquaculture, and integrated multi-trophic aquaculture (IMTA) (Choo & Caipang 2015; Tom et al 2021; Lothmann & Sewilam 2023). These technologies share the common feature of utilizing dissolved nutrients and water reuse based on the integration of autotrophic (microalgae) and heterotrophic (mollusks, copepods) organisms to convert waste into useful biomass, thereby reducing negative environmental impacts (Magnotti et al 2016; Kumar et al 2017; Tom et al 2021; Lothmann & Sewilam 2023). However, these technologies or farming models also have certain limitations, such as high production costs, long culture periods, management difficulties, large space requirements, and low economic efficiency of associated/secondary products (Tom et al 2021; Lothmann & Sewilam 2023). Consequently, their practical application in production has not been truly effective, and water exchange-based shrimp farming remains the most widely adopted solution in many countries, especially in Vietnam (Pongthanapanich et al 2019).

Artemia, a small crustacean, is widely used in aquaculture as a nutritious food source for shrimp and fish larvae (Sorgeloos et al 2001). It has diverse sizes, high nutritional value, and is convenient to use and store. It can be used in various forms, such as decapsulated cysts, fresh biomass, live, frozen, and dried. To date, numerous studies have applied *Artemia* as a supplementary feed for aquaculture species like black tiger shrimp (*Penaeus monodon*), white leg shrimp, and mud crab (*Scylla paramamosain*), yielding positive results (Dhert et al 1993; Anh et al 2011; Bengtson et al 2018). Another significant advantage of *Artemia* is its water filtration ability and application potential in aquaculture wastewater treatment. Studies have shown that an adult *Artemia* can filter up to 64 mL of water per hour and can filter food particles smaller than 50 μm (Ogburn et al 2023; Li et al 2024). Some studies have used aquaculture wastewater to cultivate *Artemia* biomass to recover nutrients from the wastewater and convert them into the biomass of this zooplankton (Gharibi et al 2021; Magnotti et al 2016; Yao et al 2018). However, this *Artemia* source is mainly reused in dried form, replacing fishmeal in the production of aquafeed (Anh et al 2023).

This study aimed to evaluate the effects of supplementing fresh *Artemia* biomass, cultivated from shrimp pond wastewater, on the diet of *L. vannamei* in an intensive farming system. The study hypothesizes that *Artemia* supplementation will improve shrimp growth, survival, and feed utilization efficiency while contributing to mitigating the environmental impact of pond wastewater. Four *Artemia* supplementation regimes (10, 20, 30, and 40% of the total feed) were tested to determine the appropriate supplementation level, thereby contributing to the effective utilization of wastewater and enhancing the growth of farmed shrimp. The results of this study will provide crucial information for developing more sustainable and efficient shrimp farming strategies, addressing both the environmental and economic challenges faced by the industry.

Material and Method

Artemia biomass culture. *Artemia franciscana* strain originated from Vinh Chau, Soc Trang, Vietnam, was used for biomass production. Wastewater from intensive shrimp ponds with suitable environmental parameters and high microalgae content was filtered through a 50 μm mesh bag and stored in 8 m^3 tanks under aeration and shading conditions for 3-5 days to promote microalgae growth before being used for *Artemia* culture. *Artemia* was cultured in 3 m^3 composite tanks placed under a roof with continuous aeration. The initial stocking density was 1,500 ind L^{-1} . For the first 5 days, *Artemia* was fed *Chaetoceros muelleri* microalgae according to the feeding protocol described by Coutteau et al (1992). From day 6 onwards, the prepared wastewater was supplemented into the culture tanks 3 times day^{-1} , with gradually increasing volumes from 5-40% per tank, each time, depending on the age and biomass of *Artemia*. Waste

was removed daily by siphoning, and 70% of the water was regularly exchanged every 3 days with wastewater directly from shrimp ponds through a 50 µm filter bag. After 15 days of culture, *Artemia* was harvested, and biological parameters and biomass were determined. Samples were soaked in freshwater, drained, divided into small portions and stored at -20°C for use in experiments. The approximate biochemical composition of *Artemia* biomass was also analyzed according to AOAC methods (AOAC 2006).

Experimental shrimp. *L. vannamei* used in the experiment had a size range of 6.1-6.7 cm and 1.3-1.5 g ind⁻¹, exhibiting a good ability to utilize *Artemia* biomass. The initial average size of experimental shrimp was 6.41±0.31 cm and 1.42±0.13 g ind⁻¹.

Experimental design and system. The experiment was conducted at the Cam Ranh Centre for Tropical Marine Research and Aquaculture, Aquaculture Institute, Nha Trang University, Vietnam. The system consisted of 20 composite tanks (200 L tank⁻¹) with an appropriately designed water supply and drainage system. Wastewater from the culture tanks was treated by a recirculating biological filtration system before being pumped back into the system. The culture tanks were equipped with air stones for oxygen supply and waste aggregation. A commercial shrimp feed with a nutritional composition meeting the requirements of shrimp (Protein>36%, lipid>4%) was used. Frozen *Artemia* biomass was thawed, drained, and the feeding amount was determined according to the experimental treatments. The experiment included 5 treatments corresponding to 5 levels of *Artemia* supplementation in the diet: 0 (control, 0% SA - Supplemented *Artemia*), 10% (1% SA), 20% (20% SA), 30% (30% SA), and 40% (40% SA), calculated based on the weight of commercial feed used per day. Shrimp were fed commercial feed 4 times day⁻¹ and supplemented with *Artemia* biomass once day⁻¹ (except for the control). Shrimp were cultured at a density of 60 ind tank⁻¹. Each treatment was replicated 4 times over a period of 40 days.

Management and monitoring. *L. vannamei* were fed commercial feed at a rate of 8% body weight day⁻¹, divided equally into 4 meals. The feed amount was adjusted based on the shrimp's feeding response and the amount of leftover feed. Culture tanks were siphoned regularly 4 times day⁻¹, 2 hours after feeding. Leftover feed was separately stored for each tank and air-dried to calculate feed utilization efficiency at the end of the experiment. Daily, 7-10% of the total system water was exchanged. Water quality parameters were maintained within optimal ranges for shrimp culture: temperature 28-31°C, salinity 30-35‰, dissolved oxygen ≥5 mg O₂ L⁻¹, pH 8.1-8.4, alkalinity 120-160 mg CaCO₃ L⁻¹, TAN<1 mg L⁻¹, and NO₂⁻<5 mg L⁻¹. After 40 days of culture, shrimp were harvested to determine growth performance, survival rate, and feed utilization efficiency.

Data collection and evaluation. Water quality parameters: dissolved oxygen content, temperature, and pH were measured using Milwaukee (MW600) and ENZO 7011 handheld meters. Alkalinity, TAN, and NO₂⁻ were determined using Sera test kits. Measurement frequency was once every two days.

Artemia biomass parameters. The total length of *Artemia* was determined by randomly measuring 30 individuals using a stereo microscope (Olympus SZ61) with a calibrated eyepiece micrometer. The wet weight of individual *Artemia* was determined by randomly measuring three samples, each consisting of 200 drained *Artemia* individuals, using an electronic balance (accuracy: 0.001 g). The wet biomass of *Artemia* in the tank was determined by weighing the total harvested *Artemia*.

Shrimp growth performance. The total length (TL, cm) and body weight (BW, g) of shrimp at the beginning and end of the experiment were determined by randomly measuring 30 individuals per tank. Length was measured from the rostrum tip to the telson end using a caliper (accuracy: 0.1 mm), while weight was measured using an electronic balance (Viet Nhat, 0.01 g). Growth parameters and survival rate were calculated as follows:

Specific growth rate in length:

$$\text{SGR}_L (\% \text{ day}^{-1}) = 100 \times [\ln(L_2) - \ln(L_1)] / t$$

Specific growth rate in weight:

$$\text{SGR}_W (\% \text{ day}^{-1}) = 100 \times [\ln(W_2) - \ln(W_1)] / t$$

Coefficient of variation in length:

$$\text{CV}_L (\%) = 100 \times \text{SD}_{L_2} / L_2$$

Coefficient of variation in weight:

$$\text{CV}_W (\%) = 100 \times \text{SD}_{W_2} / W_2$$

Condition factor:

$$\text{CF} (\text{g cm}^{-3}) = 100 \times W_2 / (L_2)^3$$

Survival rate:

$$\text{SR} (\%) = 100 \times N_2 / N_1$$

Feed utilization efficiency. Feed utilization efficiency was evaluated based on the amount of feed consumed, uneaten feed, and shrimp growth performance. The following indicators were calculated:

Feed intake:

$$\text{FI} (\text{g ind}^{-1}) = \text{Total feed consumed} / \text{Final number of individuals}$$

Daily feeding rate:

$$\text{FR} (\% \text{BW day}^{-1}) = 100 \times \text{FI} / [(W_1 + W_2) / 2 \times t]$$

Feed conversion ratio:

$$\text{FCR} = \text{FI} / (W_2 - W_1)$$

Protein efficiency ratio:

$$\text{PER} = (W_2 - W_1) / \text{PI}$$

Where:

L_1 , L_2 - the initial and final total lengths (cm);

W_1 , W_2 - the initial and final body weights (g) of shrimp;

t - the experimental duration (40 days);

SD - the standard deviation;

N_1 , N_2 - the initial and final numbers of shrimps;

PI - the amount of protein intake.

Statistical analysis. Data were analyzed using IBM SPSS Statistics version 26.0. Normality and homogeneity of variances were tested prior to applying one-way ANOVA to assess differences among treatments. When significant differences were detected, Duncan's Multiple Range Test was used for pairwise comparisons at a significance level of 5% ($p < 0.05$). Results are reported as mean values \pm standard error (SE).

Results

Artemia biomass production. The results demonstrated the successful cultivation of *Artemia* biomass using wastewater from white leg shrimp farming (Table 1). The average wet and dry biomass yields were $56.43 \pm 3.66 \text{ g m}^{-3}$ and $4.93 \pm 0.26 \text{ g m}^{-3}$ wastewater, respectively. Biochemical composition analysis revealed that the *Artemia* biomass contained $51.60 \pm 0.82\%$ crude protein and $12.33 \pm 0.40\%$ crude lipid on a dry weight basis. These findings highlight the high nutritional value of *Artemia* cultured using white leg shrimp wastewater and its potential as a supplementary feed for shrimp aquaculture.

Table 1

Results of *Artemia* culture using wastewater as a supplementary feed for *Litopenaeus vannamei* in the experiment

No.	Parameters	Values
	Size and biomass of <i>Artemia</i>	6.72±0.84
1	Length (mm)	6.72±0.84
2	Wet weight (mg ind ⁻¹)	5.94±0.61
3	Survival rate (%)	42.13±2.41
4	Wet biomass (g m ⁻³ wastewater)	56.43±3.66
5	Dry biomass (g m ⁻³ wastewater)	4.93±0.26
Proximate biochemical composition		
1	Moisture (% wet weight)	91.13±0.79
2	Crude protein (% dry weight)	51.60±0.82
3	Crude lipid (% dry weight)	12.33±0.40
4	Carbohydrate (% dry weight)	17.13±0.25
5	Ash (% dry weight)	18.23±0.60

Data are presented as mean±SD (n=3).

Water quality parameters. The water quality parameters in the experimental system remained within acceptable ranges for the growth and development of white leg shrimp (Table 2). The average temperature, salinity, dissolved oxygen, pH, and alkalinity were 28.07–29.02°C, 31.4–33.1‰, 5.81–6.20 mg O₂ L⁻¹, 8.08–8.32, and 139.7–152.5 mg CaCO₃ L⁻¹, respectively. Although total ammonia nitrogen (TAN) and nitrite (NO₂⁻) concentrations showed a slight increasing trend over the experimental period, with ranges of 0.53–0.97 mg L⁻¹ and 0.85–1.87 mg L⁻¹, respectively, they remained within the recommended limits for shrimp aquaculture (Boyd & Tucker 2012; Van Wyk & Scarpa 1999).

Table 2

Water quality parameters during the experimental period

Days	Parameters						
	Temperature (°C)	Salinity (‰)	Dissolved oxygen (mg L ⁻¹)	pH	Alkalinity (mg CaCO ₃ L ⁻¹)	TAN (mg L ⁻¹)	NO ₂ -N (mg L ⁻¹)
10	28.2±0.32	31.4±0.51	6.20±0.12	8.08±0.10	146.8±11.21	0.53±0.16	0.85±0.25
20	28.1±0.50	32.3±0.62	5.81±0.11	8.10±0.11	139.7±12.23	0.60±0.21	1.21±0.31
30	28.5±0.69	32.8±0.66	6.12±0.13	8.17±0.15	142.8±10.91	0.82±0.33	1.41±0.34
40	29.0±0.47	33.1±0.71	5.91±0.18	8.32±0.21	152.5±16.18	0.97±0.41	1.87±0.45

Data are presented as mean±SD (n=5).

Growth performance. Dietary supplementation of *Artemia* significantly influenced the growth performance of white leg shrimp (Table 3). Shrimp fed a diet with 40% SA exhibited significantly higher weight growth parameters, including final body weight (W₂) and specific growth rate for weight (SGR_w), compared to those in the 10% SA and control (0% SA) groups (p<0.05). However, no significant differences in weight growth parameters were observed among the 20% SA, 30% SA, and other experimental groups (p>0.05). Interestingly, *Artemia* supplementation did not significantly affect the length growth parameters, length coefficient of variation (CV_L), weight coefficient of variation (CV_w), or condition factor (CF) of the shrimp (p>0.05).

Shrimp biomass production. The dietary inclusion of *Artemia* significantly enhanced the harvested white leg shrimp biomass (Table 3). The shrimp biomass in the *Artemia* supplementation groups (10% SA–40% SA: 3.05–3.24 g L⁻¹) was significantly higher than that in the control (2.82 g L⁻¹) (p<0.05). The supplementation of *Artemia* resulted in a substantial increase in shrimp biomass, ranging from 8.16 to 14.89%, compared to the control, highlighting the effectiveness of this feeding strategy in improving white leg shrimp culture yield.

Table 3

Growth performance and survival rate of *Litopenaeus vannamei* cultured under different *Artemia* supplementation treatments

Parameters	Treatments				
	0% SA	10% SA	20% SA	30% SA	40% SA
L ₁ (cm)	6.41±0.31	6.41±0.31	6.41±0.31	6.41±0.31	6.41±0.31
W ₁ (g)	1.42±0.13	1.42±0.13	1.42±0.13	1.42±0.13	1.42±0.13
L ₂ (cm)	13.21±0.08	13.18±0.31	13.41±0.14	13.35±0.13	13.67±0.17
W ₂ (g)	14.63±0.11 ^a	14.57±0.15 ^a	15.16±0.31 ^{ab}	15.06±0.16 ^{ab}	15.68±0.28 ^b
SGR _L (% day ⁻¹)	1.81±0.02	1.80±0.06	1.84±0.03	1.84±0.03	1.89±0.03
SGR _w (% day ⁻¹)	5.83±0.02 ^a	5.82±0.03 ^a	5.92±0.05 ^{ab}	5.91±0.03 ^{ab}	6.00±0.05 ^b
CV _L (%)	2.94±0.40	3.23±0.57	3.95±0.95	3.51±0.91	3.72±0.76
CV _w (%)	7.92±0.80	8.08±0.87	8.59±0.64	8.16±0.58	9.45±0.63
CF (g cm ⁻³)	0.64±0.01	0.64±0.04	0.63±0.02	0.63±0.01	0.62±0.02
BM (g L ⁻¹)	2.82±0.06 ^a	3.10±0.01 ^b	3.05±0.07 ^b	3.24±0.09 ^b	3.23±0.10 ^b
SR (%)	64.17±1.08 ^a	70.83±1.08 ^b	67.08±1.25 ^{ab}	71.67±2.04 ^b	68.75±1.85 ^{ab}

Data are presented as mean±SE (n=4). Mean values with different superscript letters in the same row indicate significant differences (p<0.05). SA, supplemented *Artemia*; L₁, initial length; W₁, initial weight; L₂, final length; W₂, final weight; SGR_L, specific growth rate for length; SGR_w, specific growth rate for weight; CV_L, coefficient of variation for length; CV_w, coefficient of variation for weight; CF, condition factor; BM, biomass; SR, survival rate.

Survival rate. The survival rate of *L. vannamei* was significantly influenced by the dietary supplementation of *Artemia* (Table 3). Shrimp fed diets with 10% SA and 30% SA had significantly higher survival rates than those in the control, with increases of 10.38% and 11.69%, respectively (p<0.05). However, no significant differences in survival rates were detected among the 20% SA, 40% SA, and other experimental groups (p>0.05).

Feed utilization efficiency. The feed utilization parameters of white leg shrimp varied with the level of *Artemia* supplementation (Table 4). Feed intake (FI, g ind⁻¹) and daily feeding rate (FR, % BW day⁻¹) were significantly higher in the 10% SA and 30% SA groups compared to the control (p<0.05). Similarly, the feed conversion ratio (FCR) in the 20% SA-40% SA groups was significantly better than in the control (p<0.05). However, the protein efficiency ratio (PER) was higher in the control compared to the 10% SA and 30% SA groups (p<0.05). These findings suggest that the supplementation of *Artemia* in the diet positively influenced the feeding behaviour and feed utilization efficiency of white leg shrimp, with the 30% SA level being identified as effective for enhancing the nutritional performance of white leg shrimp in this study.

Table 4

Feed utilization efficiency of *Litopenaeus vannamei* cultured under different *Artemia* supplementation treatments

Parameters	Treatments				
	0% SA	10% SA	20% SA	30% SA	40%SA
FI (g ind ⁻¹)	11.51±0.26 ^a	12.59±0.22 ^b	12.18±0.28 ^{ab}	12.97±0.34 ^b	12.86±0.34 ^b
FR (% BW day ⁻¹)	3.59±0.06 ^a	3.94±0.10 ^b	3.67±0.07 ^{ab}	3.94±0.11 ^b	3.76±0.10 ^{ab}
FCR	1.45±0.01 ^b	1.42±0.02 ^{ab}	1.39±0.02 ^a	1.39±0.01 ^a	1.38±0.01 ^a
PER	3.19±0.05 ^b	2.91±0.08 ^a	3.14±0.06 ^{ab}	2.93±0.08 ^a	3.09±0.08 ^{ab}

Data are presented as mean±SE (n=4). Mean values with different superscript letters in the same row indicate significant differences (p<0.05). SA, supplemented *Artemia*; FI, feed intake; FR, feeding rate; BW, body weight; FCR, feed conversion ratio, and PER, protein efficiency ratio.

Discussion. *Artemia* is recognized as an essential feed in the nursing of many aquatic animal groups due to its nutritional advantages, size diversity, convenience in use and storage, and as an ideal subject for supplementing essential nutrients through enrichment techniques (Bengtson et al 2018; Cahyadi et al 2020; Madkour et al 2022). Therefore, the application of various forms of *Artemia* (live, fresh, frozen, dried, etc.) in

the nursing of crustacean species (shrimp, crab, lobster) and marine fish has been implemented and has yielded positive results in terms of growth, survival rate, feed utilization efficiency, as well as in general health indicators (Sorgeloos et al 2001; Anh et al 2011; Madkour et al 2022). However, most of these studies used *Artemia* in the form of hatched cysts, nauplii, or meta-nauplii to supplement the early stages of shrimp culture (postlarvae aged 10-25 days) or fish fry (transitional stage from rotifers to artificial feed) (Huy et al 2023; Madkour et al 2022). The current study is one of the pioneering studies using *Artemia* biomass as a supplementary feed for shrimp in the second phase of a three-phase shrimp farming process, corresponding to an age of approximately 20-60 days. At this stage, shrimp reach a size large enough to effectively utilize *Artemia* biomass, which is much larger in size compared to the previous stages. To ensure uniform quality throughout the experiment, we used frozen *Artemia* biomass as a supplementary feed for shrimp. The research results have shown the potential effectiveness of using *Artemia* biomass as a supplement for shrimp in intensive shrimp farming models.

This study has also demonstrated that culturing *Artemia* using wastewater from intensive shrimp farming activities is feasible and yields high biomass productivity, with suitable crude protein and lipid contents of 51.6% and 12.3% dry weight, respectively, which meets the needs of white leg shrimp well. This result is consistent with previous studies on the utilization of aquaculture wastewater to culture *Artemia* biomass, with the nutritional composition of the obtained *Artemia* samples having crude protein and crude lipid contents ranging from 50-60% and 10-18% dry weight, respectively (Léger et al 1987; Anh 2009; Huy et al 2020; Bengtson et al 2018). Due to this nutritional advantage, *Artemia* biomass has been used as a direct supplementary feed or as a replacement for fishmeal in the production of feed for aquaculture species, yielding positive results (Castro et al 2009; Anh 2009; Anh et al 2023). Our study contributes further evidence on the ability to directly use *Artemia* cultured from shrimp farming wastewater to supplement the diet of white leg shrimp. This approach also opens up prospects for applying recirculating/integrated models in aquaculture, helping to minimize environmental pollution, improve resource use efficiency, and simultaneously create a nutritious and safe feed ingredient for aquaculture (Huy et al 2020; Anh et al 2023). The utilization of filter-feeding species such as seaweed/algae, molluscs, and fish that consume organic debris/vegetation to treat water and recirculate it into the farming system is not new (Granada et al 2016). However, these groups of species often have limitations such as requiring large areas, long culture periods, and generating low-value products, making it difficult to immediately use them as feed for shrimp to close the process. Therefore, the approach of using *Artemia* biomass has many more advantages; however, further research is needed to optimize the efficiency of this model before applying it to commercial production.

Supplementing *Artemia* in the diet of white leg shrimp has brought many benefits in the current study. Shrimp fed a diet supplemented with 30-40% *Artemia* achieved significantly higher growth rates and biomass compared to the control, with increases ranging from 2.92-7.18% and 14.54-14.89%, respectively. Simultaneously, feed utilization efficiency was also considerably enhanced, with increases of 10.38-12.68% in feed consumption and 4.14-4.83% in feed conversion efficiency. This result is consistent with many previous reports related to the use of *Artemia* as a supplementary feed for aquatic species such as black tiger shrimp, freshwater prawn, marine crab, and many fish species (Anh 2011; Madkour et al 2022). The improved growth of shrimp can be explained by the high protein content, abundance of essential amino acids, and polyunsaturated fatty acids (DHA, EPA, etc.) found in *Artemia* (Castro et al 2009; Léger et al 1987). These essential factors have been proven to play a crucial role in promoting growth and enhancing feed utilization efficiency in many aquatic species, especially crustaceans and marine fish (Bengtson et al 2018; Madkour et al 2022). Furthermore, in the juvenile stage, exogenous digestive enzymes from live feeds such as *Artemia* have been demonstrated to play a significant role in the formation, development, and functional maturation of the digestive system (enzymes, gut structure), thereby enhancing nutrient absorption capacity, which in turn stimulates the growth and

development of the cultured species (Madkour et al 2022; Prusińska et al 2020; Kamaszewski et al 2014). Additionally, in shrimp and fish nursing, the combined feeding of live and formulated feeds is also believed to enhance prey capture response and support the digestion of formulated feeds, thereby improving feed utilization efficiency (Anh 2009; Anh 2011).

Besides the significant improvement in growth and feed utilization efficiency, the survival rate of shrimp in the *Artemia*-supplemented groups was also higher compared to the control group, especially in the 10 and 30% supplementation groups, with increases ranging from 10.38-11.69%. This result is similar to findings from previous studies that also used different forms of *Artemia* as supplements in the nursing of various shrimp and fish species (Madkour et al 2022; Anh et al 2011). *Artemia* is used as a feed for the nursery stage in most marine fish species, and supplementing *Artemia* helps reduce fish mortality, cannibalism, and uneven growth (Sorgeloos et al 2001). Some studies have shown that, in addition to essential amino acids and polyunsaturated fatty acids, *Artemia* also contains many other substances with high biological activity such as β -glucan, peptidoglycan, vitamin C, and carotenoids (Léger et al 1987; Sorgeloos et al 2001). These nutrients play a vital role in enhancing resistance, immunity, and stress tolerance in many cultured shrimp and fish species (Ciji & Akhtar 2021). Notably, salinity or formalin shock tests in black tiger shrimp (*Penaeus monodon*) postlarvae after being fed a diet supplemented with *Artemia* in the form of live or dried feed replacing a portion of the protein in the formulated feed showed better tolerance compared to the diet without *Artemia* supplementation (Dhert et al 1993; Anh et al 2023). Furthermore, feeding live fresh *Artemia* significantly reduced the rate of cannibalism in crab larvae, decreasing by 1.6-3.5 times compared to frozen *Artemia*, fresh shrimp, and dried *Artemia* (Anh 2011). Several studies also confirm that providing suitable live feeds significantly reduces cannibalism in many crustacean species, especially during the molting stage (Romano & Zeng 2017). A complete and balanced nutritional composition may play an essential role in satisfying nutritional needs, thereby contributing to reducing cannibalism and increasing the survival rate of crustaceans (Anh 2011; Romano & Zeng 2017). The higher survival rate in the supplemented groups compared to the control, especially in the 10 and 30% groups in the current study, on the one hand, affirms the positive role of supplementing *Artemia* in the diet of white leg shrimp; however, this result also suggests that further research is needed to determine the optimal supplementation level for shrimp survival while minimizing the amount of *Artemia* used.

This study demonstrates the potential of using *Artemia* biomass cultivated in shrimp farming wastewater as a feed for the same species. The research opens up the possibility of an approach that holds significance for the current shrimp farming industry, in terms of economic, environmental, and sustainable development aspects. However, as an initial assessment, our study still has some limitations that need to be addressed. Firstly, we have not determined the detailed nutritional composition of the supplemented *Artemia*, particularly the essential amino acids, polyunsaturated fatty acids, exogenous digestive enzymes, and other important nutrients, or the relationship between this supplementary nutritional composition and the nutritional composition of the shrimp at harvest. Secondly, although there were no abnormal manifestations related to diseases throughout the experimental period, further studies on the biosafety of the *Artemia* source used in this approach are required. Additionally, many technical indicators need to be researched, supplemented, and refined before they can be applied to commercial-scale production practices. Addressing the aforementioned limitations is crucial for enhancing the effectiveness and feasibility of cultivating *Artemia* using wastewater from shrimp farming activities and using it as a feed for this species, thereby closing the recycling process and improving the economic and environmental efficiency of shrimp farming in the current challenging context.

Conclusions. The research findings indicate that *Artemia* can be successfully cultivated using wastewater from shrimp ponds, and supplementing *Artemia* biomass into the diet at a level of 30% of daily commercial feed leads to significant improvements in growth performance, survival rate, and feed efficiency of shrimp. This result suggests that

Artemia biomass can substantially replace commercial feed, potentially reducing production costs and environmental impacts of shrimp farming. These findings highlight the potential of this approach for efficiently recycling nutrients, reducing environmental pollution, and enhancing the sustainability of shrimp farming activities. However, before widespread application in production practices, more in-depth studies are needed on the relationship between the nutritional composition of supplemented *Artemia* and cultured shrimp, and the biosafety of using this feed source. Further research is also necessary in order to improve the process on a larger scale, to optimize efficiency and to ensure economic feasibility.

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