Artemia sp. enrichment with vitamin C and taurine to support growth and survival rate of vaname (Litopenaeus vannamei) larvae: early study

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Abstract. The aim of the study was to determine the effect of Artemia sp enrichment with vitamin C and taurine on the growth and survival rate of Vaname (Litopenaeus vannamei) post larvae. The data was collected using randomized design with four treatment groups, which are control (group A), Artemia sp enriched with 50 mg vit CL1 media (group B), Artemia sp enriched with 50 mg taurine L1 media (group C), and Artemia sp enriched with 25 mg vit C and 25 mg taurine L1 media (group D). Data were analyzed statistically using ANOVA followed by LSD/Tukey at 5% level, using the growth and survival rate of the vaname post larvae and water quality as the data variable.

1. Introduction
Vaname shrimps (Litopenaeus vannamei), known as Pacific white shrimps, has been formally declared as the most valuable commodity for aquaculture by the Ministry of Marine Affairs and Fishery of Indonesia in 2001. Since then, aquaculture of vaname shrimps has developed rapidly in many coastal areas in Indonesia. Strategies of providing availability and good quality of shrimp farming assets were proposed to increase the national competitiveness of shrimp production and shrimp business player [1]. It is essential to conduct a study related to shrimp seeds or shrimp larva to provide good quality of shrimp seeds.

Shrimp seeds production from the shrimp hatcheries is expected to produce the best quality of shrimp larvae. However, most of these hatcheries encountered high mortality and low growth rate and development of larvae/juvenile shrimps. Balanced and good nutrition is needed as energy sources for the larvae/juvenile shrimps growth and development [2]. Artemia is known as the best natural diet source of nutrition for these juveniles.

In many aquaculture productions, water quality and nutrition are monitored for the high performance of the juveniles. Nutrition is usually stored in the fish meal, which is the main protein source in the diets for aquaculture species [3], [4]. As the aquaculture production increases, the demand on fish meals will also be high and expand the fish meal industry, leading to a higher fish meal price. Research shows that adding vitamin C and taurine into juveniles diets can improve their performance or quality. Adding vitamin C in diets proven to be successfully improve the survival rate and body protection of juvenile tiger shrimp, Penaeus monodon [5].

Taurine, as one of free amino acids containing sulfur, is widely distributed in many different animal tissues [6]. Taurine with other amino acids also involve in osmoregulation of shrimp. Increases in seawater salinity will boost the concentration of these three amino acids including taurine [7].
Therefore, this study aimed to determine the effects of dietary addition of vitamin C and taurine in natural diet, *Artemia*, on the growth performance and survival rate of white or vaname shrimp (*L. vannamei*).

2. Material and Methods

2.1. Time and Location

This research was conducted in September 2020 at the CV Hatchery, Manunggal 23, Anyer, Banten.

2.2. Diet Preparation

Frozen *Artemia* was weighed and toed and put in media enriched with vitamin C and taurine as treatments for 60 minutes. After 60 minutes, they were filtered and rinsed with seawater prior to given to juvenile white shrimp. Small aquariums with a volume of 1.5 L were used for this enrichment of *Artemia* and seawater was added to the aquarium as much as 600 ml. Enrichment to *Artemia* was given twice a day and the study was conducted for 8-days of observation.

2.3. Juvenile White Shrimp Preparation

Juvenile white shrimp were collected from CV Manunggal 23 – Karang Suraga of Anyer District – Banten Province. Juvenile white shrimp used in this study was in post-larvae stadia (PL-1). Twenty small aquariums with 5 L capacity were used and filled with 2 L of filtered seawater. Each aquarium consisted of 100 larvae L⁻¹. They were fed with enrichment *Artemia* with/without vitamin C and/or taurine. After eight days of observations, the growth and survival ratio were determined.

2.4. Experimental Design

This study was conducted in a complete randomized design with four treatments and five replications. Group A used as control and was given *Artemia* without enrichment; Group B was given *Artemia* enriched with vitamin C for 50 mg L⁻¹; Group C was given *Artemia* enriched with taurine for 50 mg L⁻¹; Group D was given *Artemia* enriched with vitamin C for 25 mg L⁻¹ and taurine for 25 mg L⁻¹.

2.5. Water Management

During the study, seawater was not changed but added during the feeding period. Parameter of water quality was determined at PL-1, PL-4 and PL-8 (which can be seen in Table 3).

2.6. Data Collection and Analysis

The growth of the white shrimp larvae (PL) was determined in early study prior to the given treatment and after 8-days of observation by measuring the body’s length from rostrum to the tail (in mm). While the survival rate was calculated by the number of larvae prior to and after treatments. Survival ratio (%) = 100 x final number/initial number. Results were expressed as mean ±SEM. Analysis data was conducted using ANOVA followed with LSD (P<0.05). All statistical analyses were performed using the Minitab18 (USA).

3. Results and Discussion

The survival rate and growth of the vaname shrimp larvae (PL-1 to PL-8) after 8 days of observation can be seen in following tables.

The highest survival percentage of vaname shrimp larvae is in the aquarium enriched with vitamin C and taurine. This condition seemed to be driven by the effect of vitamin C and taurine within *Artemia*. Diets supplemented with vitamin C could improve the growth by gaining weight and decreased the feed conversion of prawns, but the survival rate and molting frequency roughly constant [8]. However, in the aquarium with *Artemia* enriched taurine alone (C) had significantly more survival ratio compared to the control group (A). The study shows that the increased survival ratio is
presumably affected by taurine. Nevertheless, deficiency in vitamin C could lower food conversion and growth, incomplete molting, lower stress resistance [9], and immune response in juvenile marine fish [10]. Enrichment of *Artemia* with vitamin C in the diet also increases the survival ratio and growth of catfish *Pangasianodon* sp. larvae [11].

In our study, the use of vitamin C of 50 mg/L was very low compared to other study by [12], which used 0.9 g L\(^{-1}\). This concentration is the optimum level of vitamin C in *Artemia*. Therefore, the treatment with only vitamin C (group B) did not show any difference in survival ratio compared to the control group (A).

Meanwhile, the treatment of taurine only to *Artemia* (group C) increased the survival ratio significantly compared to those in the control group (A). We could say that taurine might enhance the ability of the larvae of vaname to survive. Carnivorous biota (such as fish and shrimp) could accumulate more taurine than plant-eating fish [13]. Some studies also indicate that taurine supplementation significantly improved growth and feed efficiency than taurine-free control diet [14]. The use of taurine in the vaname larvae body could be responsible for the anti-oxidative and protective effect, as seen in other fish, such as zebrafish [15]. Taurine was also known to be the essential amino acid in diets for most fish [16].

In contrast, even though the enrichment of taurine in *Artemia* significantly differed compared to the control group by looking only at body length, taurine enrichment in *Artemia* did not exceed the growth of the vaname larvae compared to the treatment of only vitamin C to enriched *Artemia* (Table 2).

Based on the results of the analysis of the stomach of Danio albolineatus eating insects (100%). Nevertheless, all the treatment groups significantly increased the growth of the vaname larvae compared to the control group by looking at the body length. These results indicate that vitamin C or taurine alone enhances body length growth. Simultaneously, the combination of both gave the highest positive impact on both body length growth and survival ratio.

*Vaname* larvae/juveniles used in this study were in Post Larvae (PL-1) stadium and underwent PL-8. At the PL-1, the juveniles were actively moving for feeding [17]. This activity needs the nervous system to work properly. The taurine obtaining from *Artemia* enrichment presumably worked in their nerve system, helping them move to find their diets. Another study using taurine enrichment in rotifer also indicated significant improvement in survival and development of larval vaname [18]). However, determining of the amount of taurine concentration in juvenile shrimp body should be undergone.

### Table 1. The Survival ratio (%) of Vaname shrimp larvae in different treatments

| Treatment Group | X ± SEM (%) |
|-----------------|-------------|
| A               | 62.80 ± 2.65\(^c\) |
| B               | 69.00 ± 2.17\(^b,c\) |
| C               | 74.80 ± 1.16\(^b\) |
| D               | 84.00 ± 1.05\(^a\) |

\(^{a,b,c}\)Statistical significant with LSD at 5%

### Table 2. The body length growth of Vaname shrimp larvae in different treatments

| Treatment Group | X ± SEM (mm) |
|-----------------|-------------|
| A               | 2.96 ± 0.05\(^d\) |
| B               | 3.40 ± 0.05\(^b\) |
| C               | 3.22 ± 0.04\(^c\) |
| D               | 3.66 ± 0.03\(^a\) |

\(^{a,b,c}\)Statistical significant with LSD at 5%
Water quality is another essential thing needed in shrimp culture. We picked only four water quality parameters because there was no water changing of the media in this study. However, all the four of water quality parameters were given in ranges values indicated by the Indonesian National Standard (SNI) for vaname production (Table 3).

Table 3. Water quality of the treatment media during 8-days observation

| Parameter | Media  | SNI |
|-----------|-------|-----|
| Temperature (°C) | 30 – 31 | 28.5 – 31.2 |
| DO (mg L⁻¹) | 5.6 – 5.7 | 5.0 – 6.7 |
| pH | 7.80 | 7.9 – 8.2 |
| Salinity % | 32 | 25 – 35 |

4. Conclusion
In this early study, enrichment of Artemia as a natural diet with vitamin C and taurine with a 25 mgL⁻¹ concentration in each media increased the survival ratio and the growth in body length of the juvenile vaname shrimp in post larvae stadia.

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