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Research Article

Buton Forest Onion Extract (*Eleutherine bulbosa* (Mill.) Potential on Growth Performance of Vannamei Shrimp (*Litopenaeus vannamei*)

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Abstract

Background and Objective: Vannamei shrimp *Litopenaeus vannamei* is an economically valuable aquaculture commodity but still faces problems due to disease and growth performance. Buton forest onion extract *Eleutherine bulbosa* (Mill.) Urbhas been shown can protect the shrimp from disease. This study aimed to evaluate the potential of the Buton forest onion extract on the growth of vannamei shrimp in tarpaulin pond. **Materials and Methods:** The experiments were conducted for 90 days by adding Buton forest onion extract in feed at a dosage of 40 g kg⁻¹. The Buton forest onion bulbs was extracted by maceration method using 96% ethanol. The test feed was made through the re-pelleting method. Treatment include supplementation of Buton forest onion extract diet supplemented with Buton forest onion extract diet at twice a week (treatment A) and control was without Buton forest onion extract (treatment K). Parameter measurement include final weight, average daily growth (ADG), feed conversion ratio (FCR), survival rate and water quality (temperature, salinity, turbidity and pH). **Results:** This study was demonstrated the addition of Buton forest onion extract on feed with a frequency twice a week in significantly improve the growth performance of vannamei shrimp by improving the final weight and average daily growth at 65 and 90 days of maintenance. Water quality during the study was still within the optimum range for *L. vannamei* shrimp growth. **Conclusion:** This study showed that supplementation of Buton forest onion extract was able to improve the growth performance of shrimp in tarpaulin ponds.

Key words: Buton forest onion extract, growth performance, tarpaulin pond, vannamei shrimp

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Vannamei shrimp *Litopenaeus vannamei* is one of the leading export aquaculture commodities for fisheries in Indonesia. The high demand for vannamei shrimp has encouraged various centers of cultivation to increase their production through intensification. Intensive cultivation of vannamei shrimp that is not supported by a good system and technology could cause various obstacles, especially those related to diseases. One type of disease that is often found in vannamei shrimp is vibriosis caused by the bacterium *Vibrio harveyi*¹. The *V. harveyi* is an opportunistic pathogen and is classified as a marine Gram-negative luminous organism². This bacteria is a natural micro-florain seawater which can cause mortality in shrimp larvae in hatcheries³ and nauplius, zoea, mysis and post larvae stadia to adult shrimp in enlargement ponds⁴.

Many efforts have been made to prevent *V. harveyi* infection in vannamei shrimp cultivation such as the use of herbal medicines. Utilization of herbal medicinal in aquaculture has increased in popularity because the extraction process is simple, inexpensive and the products are safe for animals and the aquatic environment. The use of herbal medicinal plants can prevent bacterial infections because they contain antibacterial compounds and materials to improve the immune system in organisms⁵. One of the herbal plants that can be used to prevent *V. harveyi* infection in shrimp is the Buton forest onion extract (*E. bulbosa*). Buton forest onion extract can inhibit the growth of *V. harveyi* because it contains flavonoid, tannin, saponin, quinone, steroid and triterpenoid compounds which can inhibit the metabolism of *V. harveyi*⁶. This phytochemical content is able to stimulate the immune system⁷. Onions have been known to also have antibacterial and antioxidant effects⁸. Various nutrient-rich medicinal plants could increase the growth of cultivated organisms⁹. Several studies such as the use of garlic and shallots on sea bass (*Dicentrarchus labrax*) showed an increase in growth, feed utilization and fish protein content¹⁰.

Our recent study showed that Buton forest onion extract has capability to improve the growth performance and the immunity responses of the shrimp and suppress *V. harveyi* cells growth in laboratory scale research. Based on these reasons, this study was expected to provide information on the use of Buton forest onion extract to improve the growth performance of vaname shrimp in tarpaulin ponds.

MATERIALS AND METHODS

The study was conducted between April and August, 2018 in Lasalimu Village, South Lasalimu district, Buton Regency, South East Sulawesi.

Extraction: The Buton forest onion bulbs was obtained from Kanawa Village, South Lasalimu district, Buton Regency, South East Sulawesi. The Buton forest onion extract was produced by maceration method⁶. Buton forest onion bulbs that had flowered or were 3-4 months old were cleaned and then sliced thinly and dried in an oven for 48 h at 60°C. The dried onion slices were then ground and sifted to produce a flour/powder. The Buton forest onion powder was extracted with 96% ethanol as the solvent at a ratio of 1:4 (w/v). Then the maceration process was conducted at room temperature for 24 h using a magnetic stirrer. The first maceration results were filtered using Whatman® No. 41 filter paper. The maceration process was conducted twice. The first, second and third maceration results were combined and then concentrated with a vacuum evaporator at 40°C to produce a Buton forest onion extract.

Experimental design: The experiments were conducted for 90 days by adding Buton forest onion extract in feed at a dosage of 40 g kg⁻¹. The test feed was made through the re-pelleting method. Commercial pellets were mixed with the Buton forest onion extract and then carboxil methyl cellulose 3% and water were added. The treatments in this study consisted of two treatments: Treatment A (supplementation of Buton forest onion extract diet at twice a week) and K (control without supplementation of Buton forest onion extract diet). Then the mixture was made into pellets again and was dried in an oven at 60°C.

Proximate analysis: The proximate analysis on feed samples were calculated triplicate following standard procedures¹¹. Crude protein was determined by Kjeldahl method (N 9 6.25) Auto Kjeldahl System (Kjeltec TM 2300 Foss Tecator, Sweden). Crude lipid was calculated gravimetrically after extratction with ether extraction (Soxtec System HT6, Tecator AB, Sweden). Moisture content by drying in an oven at 105°C for 24 h. Ash content was determined gravimetrical by incineration in a muffle furnace at 600°C for 6 h. Crude fiber was estimated after acid and alkali digestion and loss in mass by combustion at 600°C for 3 h. Carbohydrate content was

calculated based on difference calculation. Nitrogen-free extract (NFE) was determined from 100-(crude protein + crude lipid+crude fiber+total ash).

Water quality measurement: Water quality parameters measurement were temperature, salinity, turbidity and pH. Temperature was measured twice everyday by using thermometer. Salinity, pH and turbidity were measured every week by using refractometer, pH meter and turbidimeter.

Maintenance of shrimp: The shrimp was obtained from PT. Central Protein a Prima Hatchery, Takalar district, Indonesia. As many as 300 shrimp were reared in tarpaulin ponds sized $3 \times 2 \times 1.2 \text{ m}^3$ for 90 days. The shrimp were diet the commercial feed twice a day and supplementation of Buton forest onion extract diet at twice a week. Shrimps were sampled every week after 65 days the age of shrimp maintenance until 90 days. At 65 and 90 days, 30 shrimp per pond were collected as samples for monitoring their growth. The final weight was the weight of the shrimp at the end of the maintenance period¹². Average daily growth of shrimp (ADG, g/day) during the experimental period was calculated according to the formula:

$$\text{ADG} = \frac{W_f - W_i}{t}$$

where, W_f and W_i are final and initial body weight and t is duration of experimental period. Feed conversion ratio was calculated according to formula:

$$\text{FCR} = \frac{F}{W_f - W_i}$$

where, F is weight of food supplied to fish during study, W_f and W_i are final and initial body weight. Survival rate was calculated according to formula:

$$\text{SR} = \frac{\text{Remained shrimp number}}{\text{Stocked shrimp number}} \times 100$$

Statistical analysis: All the data collected for the parameters were analyzed using the IBM SPSS Statistic for Windows, version 16.00 (IBM Corp, Armonk, NY, USA) at a significance level of 0.05. Significant differences between treatments were determined using a *post hoc* Duncan test.

RESULTS

The results of the feed proximate analysis showed that the treatment feed and control feed were not significantly different.

Treatment A: Treatment A feed contained 36.72% protein, 7.08% lipid, 32.24% nitrogen-free extract material, 2.34% coarse fiber, 13.51% moisture and 8.11% ash.

Control K: Control K feed contained 41.28% protein, 7.53% lipid, 23.73% nitrogen-free extract material, 2.21% coarse fiber, 14.08% moisture and 11.17% ash.

The results of the study revealed that after a maintenance period of 90 days, the shrimp in treatment A (supplementation of Buton forest onion extract diet at twice a week) reached the final weight of 22.72 ± 0.02 g/shrimp while the control shrimp were only 16.62 ± 0.02 g/shrimp as shown in Fig. 1.

The ADG in treatment A (supplementation of Buton forest onion extract diet) was higher than in the control treatment both after 65 and 90 days of maintenance (Fig. 2). The ADG in treatment A was 0.27 ± 0.002 g/day after 65 days of maintenance and 0.31 ± 0.001 g/day after 90 days of maintenance. The ADG in the control was 0.22 ± 0.001 g/day after 65 days of maintenance and 0.22 ± 0.002 g/day after 90 days of maintenance as shown in Fig. 2.

The FCR in treatment A (fed the Buton forest onion extract) was not significantly lower than the control at 65 days (1.10 ± 0.007) but it was significantly lower than control at

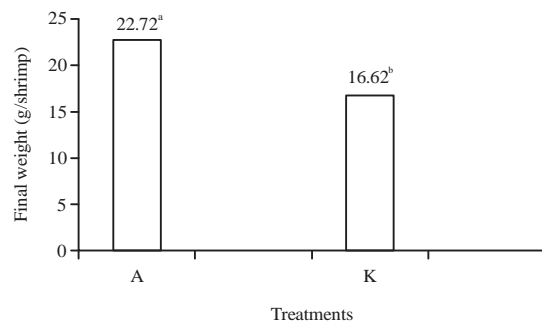


Fig. 1: Final weight of vaname shrimp. Treatment A (supplementation of Buton forest onion extract diet at twice a week) and K (control or supplementation without Buton forest onion extract diet)

Different letters over each treatment bar in same day (Mean \pm SD) indicated significant difference (Duncan, $p < 0.05$)

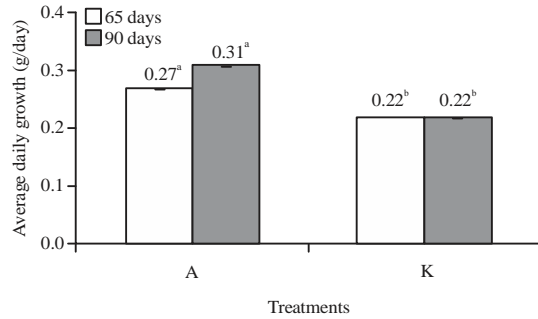


Fig. 2: Average daily growth rate of vaname shrimp. Treatment A (supplementation of Buton forest onion extract diet at twice a week) and K (control or supplementation without Buton forest onion extract diet) Different letters over each treatment bar in same day (Mean±SD) indicated significant difference (Duncan, $p<0.05$)

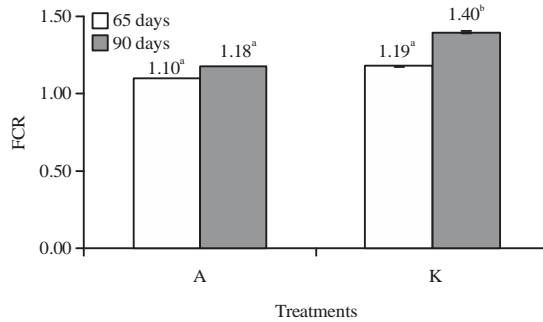


Fig. 3: Feed conversion ratio in vaname shrimp. Treatment A (supplementation of Buton forest onion extract diet at twice a week) and K (control or supplementation without Buton forest onion extract diet) Different letters over each treatment bar in same day (Mean±SD) indicated significant difference (Duncan, $p<0.05$)

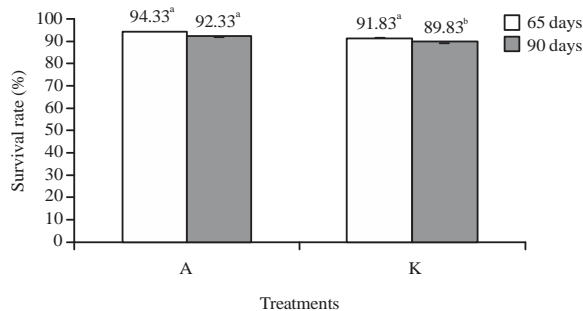


Fig. 4: Survival rate of vaname shrimp. Treatment A (supplementation of Buton forest onion extract diet at twice a week) and K (control or supplementation without Buton forest onion extract diet) Different letters over each treatment bar in same day (Mean±SD) indicated significant difference (Duncan, $p<0.05$)

Table 1: Water quality during the study

Treatments	Temperature (°C)	Salinity (g L ⁻¹)	Turbidity (cm)	pH
A	29-30	28-30	30-40	7.5-8.2
K	29-30	28-30	25-37	7.4-8.2

90 days (1.18 ± 0.008) of maintenance. The FCR in the control treatment was 1.19 ± 0.004 and 1.40 ± 0.013 (Fig. 3).

The survival rate in treatment A was not significantly higher than in the control at 65 days ($94.33 \pm 0.33\%$) but it was significantly higher than in the control at 90 days of maintenance ($92 \pm 33\%$) (Fig. 4). The SR in the control was 91.83 ± 0.17 and $89.83 \pm 0.50\%$, respectively. Water quality (Table 1) included temperature, salinity, turbidity and pH during the study was still within the optimum range for *L. vannamei* shrimp growth.

DISCUSSION

Results showed that the supplementation of Buton forest onion extract diet at twice a week at a dose of 40 g kg^{-1} feed could improve shrimp growth. This can be seen from the final weight of the shrimp in treatment A which was significantly higher than that of the control after 90 days of maintenance ($p<0.05$). The average daily growth was significantly higher than in the control at 65 and 90 days of maintenance ($p<0.05$). These results indicated the ability of supplementation of Buton forest onion extract diet to improve the growth performance in shrimp. Some studies also found similar results. Supplementation garlic and onion to sea bass (*Dicentrarchus labrax*) could increase growth, feed utilization and fish protein content¹⁰. The administration of herbal ingredients in the form of adding ginger to fish feed could significantly improve fish's growth performance compared to fish that were not given herbal ingredients¹³. In addition, the use of herbal ingredients such as cinnamon (*Cinnamomum zeylanicum*) in the diet of tilapia could increase the specific growth rate, feed efficiency, protein efficiency and energy utilization¹⁴.

The improved growth performance in the shrimp supplementation of Buton forest onion extract diet was thought to be caused by the presence of ingredients that play a role in feed digestion in shrimp. Buton forest onion extract has phytochemicals in the form of flavonoid, tannin, saponin, quinone, steroid and triterpenoid compounds⁶. Some studies have found that ingredients contained in plants were not only anti-pathogenic bioactive compounds but also nutrients such as amino acids, carbohydrate, minerals and vitamins. All of these ingredients play a role in increasing appetite¹⁵, accelerating the metabolism¹⁶ and improving enzyme activity

in the digestive tract¹⁷ so that the digestive process becomes more effective which results in an increased growth¹⁸.

The effectiveness of the digestive performance can be measured using various parameters including the Feed Conversion Ratio (FCR). Results of this study indicated that the supplementation of Buton forest onion extract diet had a significantly lower FCR compared to the control treatment at 90 days of maintenance ($p < 0.05$). The FCR is the amount of feed needed to produce a certain shrimp weight. The smaller FCR value, the greater amount of feed is converted into flesh. The FCR value was not significantly different at 65 days of maintenance but it was significantly different at 90 days of maintenance. The FCR value of shrimp treated with Buton forest onion extract diet being lower than that of the control at 90 days is an indicator that the supplementation of Buton forest onion extract diet could increase the appetite and the digestive performance of shrimp after 65 days. The addition of herbal ingredients in feed acts as an anti-bacterial, antifungal and immunostimulant for the body and could also increase the appetite because it contains appetite-enhancing ingredients¹⁹. This was similar to the findings of other study²⁰, who discovered that the use of herbal ingredients such as cinnamon leaf extract (*Cinnamomum burmannii*) could increase protein digestibility in catfish, resulting in a higher body protein content. The high protein retention in the shrimp's body caused them to experience faster growth. However, an excessive administration of herbal ingredients in feed would actually disrupt the growth performance. Bio-active content in plants such as tannin have the ability to form protein complexes, therefore, they could inhibit microorganism growth or enzyme activity²¹.

Supplementation of Buton forest onion extract diet was also found to maintain a significantly higher shrimp survival rate compared to the control ($p < 0.05$) at 90 days maintenance, although at 65 days it was not significantly different. Buton forest onion extract contains phytochemicals such as phenolic, polyphenol, alkaloid, quinone, terpenoid, lectin and polypeptide compounds which are good for preventing pathogenic infections and maintaining shrimp health⁷. In addition to containing ingredients that can increase growth, the onion extract also contains various nutrients such as proteins that are good for maintaining shrimp health⁹.

CONCLUSION

The application of Buton forest onion extract on shrimp cultured in tarpaulin ponds was able to improve the growth performance by improving the final weight and average daily growth at 65 and 90 days of maintenance. Vannamei shrimp

that were given the Buton forest onion extract also had a lower feed conversion ratio and higher survival rate at 90 days of maintenance compared to those of the control.

SIGNIFICANCE STATEMENT

This study discovered the ability of Buton forest onion extract diet to improve the growth performance in shrimp in tarpaulin ponds which could be beneficial for fish farmers. This study could help researchers to uncover the critical areas of utilizing herbal extracts for improving the growth of shrimp have not been extensively studied. Thus a new theory on the application of Buton forest onion extract diet to improve the growth performance of shrimp may be arrived at.

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