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Effect of Indian Pluchea Leaf (*Pluchea indica*) addition on feed-on growth performance and survival rate of *Litopenaeus vannamei*

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Abstract: This study aims to determine the effect and the best dose of Indian Pluchea leaf extract on artificial feed on the growth performance and survival rate of Vanname. The sample of this study was shrimp aged PL 21 obtained from shrimp farmers in Pekalongan. The study employed an experimental method with a completely randomized design (CRD) with 4 (four) treatments with 3 (three) replications, A (Indian Pluchea leaf extract of 0 ml/kg feed), B (Indian Pluchea leaf extract 5 ml/kg feed), C (Indian Pluchea leaf extract 10 ml/kg feed), D (Indian Pluchea leaf extract 15 ml/kg feed). The addition of Indian Pluchea leaf extracts to feed had a very significant effect on the growth of vanname. The best dose was in treatment D with a growth of 5.61 grams, FCR (feed conversion ratio) around 1.17 and SR (survival rate) 100 %. The water quality parameters were found during the observation: temperature of 29-32°C, the salinity of 20-25 ppt, and pH of 7.8 - 8.2.

Keywords: Growth, *Indian Pluchea*, Feed, Vanname

1. Introduction

There are numerous changes happening in the aquaculture industry right now, both in terms of the technologies used for production and the kinds of products that are being grown. Vannamei shrimp is one of the commodities that are on the rise and is developed until now due to its huge potential. Vannamei shrimp is a type of shrimp that has a high economic value of IDR 60 thousand/kg[1] and is in great demand on the world market.

Accordingly, the high stocking density of vannamei shrimp aquaculture requires a large amount of feed availability since they have to increase the dose or frequency of feeding. Feed is one of the important factors in vannamei shrimp cultivation activities. Moreover, the financing for feed in shrimp cultivation reaches 40-70% of the variable production cost[2]. Under these conditions, it is necessary to take any action to increase the growth rate of vannamei shrimp in order to reduce production costs, especially feed.

One of the preventive measures is feeding using the addition of natural ingredients that can be obtained easily and cheaply to increase growth and support the survival rate of vannamei shrimp. Natural ingredients that can be used include Indian Pluchea leaves (*Pluchea indica*). Indian Pluchea tree is a type of plant that grows a lot in the environment. Indian Pluchea leaves contain compounds

that are useful for the body namely phytochemicals which are antioxidants that can inhibit the work of free radicals to improve growth performance and stimulate the immune system. The compounds contained in Indian Pluchea leaves are alkaloids, tannin, essential oils, vit A, vit C, and flavonoids[3]. The percentage values of the phytochemical content of Indian Pluchea leaves were flavonoids of 4.18%, tannins of 2.351%, essential oils of 1.88%, and alkaloids of 0.316%. As the aforementioned explanation, it can be seen that the content of Indian Pluchea leaves contains flavonoids, vitamins, and secondary metabolites that can stimulate growth and are anti-microbial.

Previous studies on the use of Indian Pluchea leaves to prove that Indian Pluchea leaves affect the growth of catfish fry [4] and are effective in inhibiting the growth of *Pseudomonas fluorescent* bacteria in carp[5]. However, currently, there is no literature on the use of Indian Pluchea leaves in shrimp growth. It is necessary to conduct a study on the effect of Indian Pluchea leaf extract on the growth of vannamei shrimp.

2. Materials and Methods

This study was done at the Slamaran Aquaculture Laboratory, Universitas Pekalongan. The study was conducted based on randomized design (CRD) with 4 treatments and 3 replications. The treatments applied were varied in the addition of Indian Pluchea leaf extract to the feed as follows:

1. Treatment A : Indian Pluchea leaf extract of 0 ml/kg of feed
2. Treatment B : Indian Pluchea leaf extract of 5 ml/kg of feed
3. Treatment C : Indian Pluchea leaf extract of 10 ml/kg of feed
4. Treatment D : Indian Pluchea leaf extract of 15 ml/kg of feed

Vannamei shrimp PL 21 were obtained as samples from shrimp farming in the Degayu Pekalongan area. 120 shrimps were divided into 10 shrimps per container. Each container was a 25-liter aquarium equipped with an aerator.

Extraction is done by crude extraction method by smoothing the leaves with a blender and then macerating. After maceration, it is evaporated for several hours to obtain the result. The feed given in the process of rearing vannamei shrimp is commercial feed mixed with Indian Pluchea leaf extract. The feed was mixed with Indian Pluchea leaf extract by spraying under the predetermined treatment dose, then dried by aerating. After the feed was dried, it can be given to the shrimp. The aerator was turned off 15 minutes prior to the feeding. The feed was spread evenly. Then, the aerator was turned on 15 minutes after feeding. The feed was stored in a protected, dry, and sterile place. Parameters observed in this study were absolute weight growth, Feed Conversion Ratio, Survival Rate, and water quality. The calculations used were as follows:

2.1. Absolute Growth

The absolute weight growth was calculated to get the results of the weight gain of vannamei shrimp that had been reared, using a certain formula

$$AG = W_2 - W_1[6]$$

Where:

- AG : Absolute Growth (g)
 W₂ : Final weight (g)
 W₁ : Initial weight (g)

2.2. Feed Conversion Ratio (FCR)

Calculation of the feed conversion ratio was carried out at the end of the study to determine the amount of feed eaten during the rearing period, namely:

$$FCR = F / W_t - W_0 + D[7]$$

Description:

- F : Amount of Feed Consumed (g)
 W_t : Final Body Weight (g)
 W : Initial Body weight (g)

D : Weight of death fish (g)

2.3. Survival Rate (SR)

The survival rate is the number of life levels of vannamei shrimp from initial stocking to the end of the rearing period, calculated using a certain formula;

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

Where:

N_t : Number of live fish at the end of the study

N_o : Number of fish at the beginning of the study

2.4. Water Quality Parameters

The water quality parameters observed included temperature, pH, and salinity. The temperature was measured daily using a thermometer; pH was measured using a pH meter; salinity was measured using a refractometer.

The effect of treatment on the growth of vannamei shrimp was determined by collecting the data to be compared and analyzed statistically for variance. The normality of the data was tested using the Liliefors test while the homogeneity was tested using the Bartlett test. If the data are normal and homogeneous then the data are analyzed using one-way Analysis of Variance (ANOVA). Furthermore, water quality data were analyzed descriptively.

3. Results and Discussion

3.1. Absolute Growth

Vannamei shrimp biomass growth increased in each treatment. The growth with the highest yield was shown in treatment D with an average value of 5.61 grams. Then, it was followed by treatment C of 5.03 grams, treatment B of 4.49 grams, and treatment A as a control with an average weight of 3.41 grams as a control (Table 1). This study found that the addition of Indian Pluchea leaf extracts to feed had an effect on the growth of vannamei shrimp.

Before further tested, biomass growth data was statistically tested for normality and homogeneity of data variance as attached in Appendix 2 and 3. The results of the normality test were $L_{max} (0.1138) < L_{table} 5\% (0.275)$ and $1\% (0.242)$, indicating that the data were normally distributed. After that, the homogeneity test presents the results of $X^2 (2.7296) < X^2 5\% (7.81)$ and $X^2 1\% (11.34)$, indicating that the data were homogeneous. Analysis of variance (ANOVA) was carried out and presents a very significant effect of different feeding treatments on the growth of vannamei shrimp seed biomass.

Table 1. Average growth of vannamei shrimp

Treatment	Average Growth (g)
A	3.41±0.33 ^c
B	4.49±0.26 ^b
C	5.02±0.09 ^{ab}
D	5.61±0.24 ^a

Note: A : Indian Pluchea leaf extract of 0 ml/kg of feed
 B : Indian Pluchea leaf extract of 5 ml/kg of feed
 C : Indian Pluchea leaf extract of 10 ml/kg of feed

D : Indian Pluchea leaf extract of 15 ml/kg of feed

(Different letter notation in one column shows a significant difference $P < 0.05$)

It is suspected that the feed given with sufficient composition and with the addition of Indian Pluchea leaf extract into the feed has had a good impact on shrimp growth. Indian Pluchea leaf extract contains vitamin C, amino acids, tannin, saponin, and flavonoids that are able to maximize the digestive process and absorption of nutrients, especially protein. Flavonoids derived from soybeans can improve growth performance and disease resistance in fish. Increasing flavonoids also improve feed quality so as to increase protein content in the feed[8]. This is supported by the statement that the presence of active compounds from plants in a way can suppress the presence of anti-nutrients in the feed so as to increase protein sources[9]. Vitamin C in Indian Pluchea leaves has a very important function in the metabolism of the shrimp body. The presence of vit C in the feed could increase the secretion of digestive enzymes[10] so that the feed will be more easily digested by the body. In addition, the role of vit c is also very important for the molting process in shrimp and formatting fish bone. It helps increase the content of important minerals such as phosphorus, iron, calcium, and other minerals. The mineral content in the body of prawns increases along with the higher dosage of Vit C content[11]. Other compounds such as tannins and saponins have a role as anti-oxidants that help maintain body immunity. In line with Quideau et al[12] that tannins have the ability to stimulate the body's defense system. Good immune conditions will increase growth. Furthermore, the positive effects of essential oils are their ability to increase appetite, and growth, as an immunomodulator, and also anti-stress[13]. Prolonged stress conditions in animals will have a negative impact on the quality of life of these organisms such as adverse effects on growth, endurance, and even resistance to disease attacks. Prolonged exposure to stress will produce negative effects on life functions such as growth, development, behavior, reproduction, disease resistance, and even death in fish[14]. The antibacterial content in flavonoids could stabilize intestinal conditions to increase growth[15]. In treatments B and D, the growth was lower than in treatment D due to the lower dose of Indian Pluchea leaf extract on the feed.

3.2. Feed Conversion Ratio (FCR)

Vannamei shrimp feed conversion ratio during the best rearing was shown in treatment D with a value of 1.17, followed by treatment C with a value of 1.18, treatment B with a value of 1.29, and treatment A with a value of 1.41.

Table 2. Average value of Feed Conversion Ratio of vannamei shrimp

Treatment	FCR
A	1.41±0.015 ^c
B	1.29±0.023 ^b
C	1.18±0.012 ^a
D	1.17±0.032 ^a

Note: Different letter notation in one column shows a significant difference between treatments ($P < 0.05$)

A lower FCR value indicates that the feed consumed by fish is more efficiently used for growth, whereas a larger FCR value indicates that the feed consumed is less effective. Indian Pluchea leaf extract also contains essential oils and flavonoids, which are phytochemicals and have an important role in the digestive tract, thereby increasing fish digestibility of feed. The presence of essential oils stimulates the intestinal mucosa to secrete a certain amount of compounds that can induce positive changes in intestinal morphology, as well as provide anti-inflammatory, and antioxidant activity[16]. Essential oils can also affect the amount and type of secretions produced by the intestinal mucosa and change the physical and chemical properties of the intestinal environment so that the absorption process

is maximized. Flavonoids can improve feed efficiency and absorption due to their ability to increase metabolism. The flavonoids in cinnamon can stimulate increased metabolism in the body as well as antioxidants[17]. Increased metabolism will accelerate the breakdown of blood glucose, and fatty acids and improve fish health so that protein can be utilized for growth. The high feed conversion ratio in treatment A was due to the absence of Indian Pluchea leaf extract compared to treatments B, C, and D

3.3. Survival Rate (SR)

The addition of Indian Pluchea leaf extract to the feed did not affect the survival rate of vannamei shrimp. Treatments A, B, C, and D resulted in a 100% survival rate, indicating that there were no fish mortality in the study.

Table 3. The survival rate of vannamei shrimp

	Treatment			
	A	B	C	D
No	30	30	30	30
Nt	30	30	30	30
SR	100%	100%	100%	100%

The SR value in this study achieves 100 % for all treatments. The high survival rate is due to the active substances in Indian Pluchea leaves, including flavonoids, tannins, and vitamin C, which have the ability to boost the immune system and act as antioxidants. Antioxidants are electron-giving chemicals or reductants that can inhibit, prevent or slow down oxidation reactions[18]. The presence of antioxidants could protect body cells from harmful radicals substances. In addition to vitamin C, there are tannins in Indian Pluchea leaves that also make a positive contribution, namely protecting the body from possible pathogen attacks. Tannins have the potential as anti-microbials that are effective against pathogenic fish[19].

3.4. Water Quality

The results of the observation of the pH value in the vannamei shrimp rearing medium for 30 days ranged from 7.8 to 8.0. This can be said to be in good condition for the growth of the vannamei shrimp. The optimal pH for the growth of vannamei shrimp ranges from 6.5 to 9.0, while a low pH can reduce the growth rate of vannamei shrimp[20].

The results of observations of salinity in the maintenance of vannamei shrimp for 30 days ranged from 20-23 ppt. The increase and decrease in salinity that occurred were still within the optimal range and supported the growth and survival of vannamei shrimp. Good salinity for the growth of vannamei shrimp is in the range of 15-23 ppt[21]. Based on these data, it can be concluded that the salinity of the rearing medium water during the 30 days of the study was still within the normal range. Therefore, it was feasible to support the growth and survival rate of vannamei shrimp.

The temperature observed in vannamei shrimp rearing media for 30 days ranged from 28 - 30°C. The optimal temperature for rearing vannamei shrimp is 28 - 32°C[20]. Thus, the water temperature during the rearing in this study is in a condition that is suitable for the life of vannamei shrimp.

4. Conclusion

The addition of Indian Pluchea leaf which is abundant in nature to feed affects the growth of vannamei shrimp. The best dose of adding Indian Pluchea leaf extract to feed on the growth of vannamei shrimp was in treatment D (15 ml of Indian Pluchea leaf extract/kg of feed) with an average of 5.61 grams.

5. Acknowledgement

I declare that in this study all researchers followed all institutional and national guidelines for the care and use of laboratory animals.

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