



The Effect of Using Papaya (*Carica papaya* L) Leaves on Blood Glucose and Growth Rate of Asian Seabass (*Lates calcarifer*)

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ABSTRACT

This research aims to determine the effect of using papaya (*Carica papaya* L) leaves on blood glucose and growth rate of *Lates calcarifer*. This research used a completely randomized design (CRD) with 4 treatments and 3 replications. Treatment A with the addition of papaya leaves powder (*C. papaya*) of 3.25 g/L, B of 3.75 g/L, C of 4,25 g/L and D as a Control or without the addition of papaya leaves. The parameters measured were blood glucose, absolute growth and survival. The best treatment is treatment A (*C. papaya* 3.25 g/L) which produced fish blood glucose on the 60th day of 90 mg/dL, 100% survival rate, absolute weight growth and length of 6.43 g and 6.20 cm.

INTRODUCTION

Asian seabass (*Lates calcarifer*) grows quickly and adapts easily to the cultivation environment. Asian seabass has a high economic value, to meet domestic and foreign needs. As an export product, this type of fish is in great demand in foreign markets. However, this fish is classified as a carnivorous fish that can eat (Hasanah *et al.*, 2019). Generally, this cannibalism phenomenon is seen at the fry stage and begins to decline after the fry size reaches 7-10 cm. In general, cannibalism is related to genetics, lifestyle and differences in size in a group because genetic variation is the main factor (Jaya & Isnaini, 2013). Asian seabass (*L. calcarifer*) is one type of marine fish that has economic value in Indonesia. Asian seabass (*L. calcarifer*) cultivation has become a commercial activity, both in ponds and floating nets. In efforts to breed Asian seabass (*L. calcarifer*), the main obstacles are the low survival rate (SR) and susceptibility to disease. The low survival rate in asian seabass cultivation is influenced by several factors, including poor feeding, both in terms of quantity and quality of feed, and also fish experiencing stress.

With the increasing production of asian seabass, there are also many obstacles faced, one of which is stress due to disease attacks that inhibit fish growth and even this disease attack can cause fish death during breeding. This condition causes low appetite and low immunity of fish, so that growth is slow, fish are more susceptible to disease and eventually die. The main problem faced in asian seabass cultivation is that small fry are susceptible to stress (Wijaya *et al.*, 2024).

Efforts to control fish stress can be seen in the blood glucose profile of asian seabass. In this study, researchers used papaya leaves powder (*Carica papaya* L) to determine the effect on stress levels. This is based on research conducted by Mahasri *et al.* (2022) which showed that giving papaya leaves extract as an antiparasitic affects behavioral changes (stress) and blood glucose levels during administration of *Zeylanicobdella* sea leeches. In the marine environment, fish experience changes in physiological responses including primary and secondary responses. The first response occurs with an increase in hormones such as catecholamines and cortisol, while the second response that occurs is an increase in glucose. According to Mahasri *et al.* (2022) papaya leaves extract added to the aquarium container will increase blood glucose in asian seabass. Blood glucose levels are influenced by many factors, including food, liver glycogen storage capacity, and growth phase.

LITERATURE REVIEW

The classification of asian seabass is as follows based on (Kusumanti *et al.*, 2022).

Phylum	: Chordata
Phylum	: Vebrata
Class	: Pisces
Ordo	: Percomorphi
Family	: Centropomidae
Genus	: <i>Lates</i>
Species	: <i>Lates calcarife</i>

Asian seabass has an elongated, flat body shape, and has a pointed head with a concave upper part, in front of the convex dorsal fin and a wide caudal fin stem. The mouth is quite wide as a characteristic of a predatory fish and is accompanied by fine teeth, and the lower part of the operculum has strong spines. Small spines are found on the operculum, on the linea lateralis there are serrated lobes.

Asian seabass is a catadromous fish species, a fish that is euryhaline. The habitat of asian seabass can be found in lakes, lagoons, and river estuaries with a water level of between 10 and 15 ppt. newly hatched larvae (15-20 days or more). When they grow into 1 cm larvae, they can be found in freshwater environments such as rice fields and lakes. Larvae measuring 0.4-0.7 cm are found on the coast or river estuaries. In the Western Pacific and Indian Ocean, snapper is found in tropical and subtropical areas (Islamy *et al.*, 2023).

Papaya plants contain sap, which is a substance called papain. Papain is a sulfhydryl protease from papaya sap. This papain enzyme is usually found in the stems, leaves and fruit of the papaya itself. This enzyme can work optimally at temperatures ranging from 38-80 °C. Papain enzyme can be used to help digest protein. The use of papain enzyme as a source of protease enzyme can improve the quality of low crude protein feed. Papaya leaves also contain alkaloids, flavonoids, and saponins. Papain is one of the most powerful enzymes produced by all parts of the papaya plant, except the seeds and roots. The part of the plant that produces the most sap is found in the papaya fruit (Aravind *et al.*, 2013).

In addition to the papain enzyme, there are several compounds that can be proven through phytochemical tests. This phytochemical test aims to determine the presence or absence of bioactive components in a test sample. From the results of phytochemical tests conducted by (Waruwu *et al.*, 2021) papaya leaves contain positive alkaloids, triterpenoids, steroids, flavonoids, saponins and tannins. These compounds are believed to be able to kill insect pests. Based on the results of phytochemical analysis tests also from the Spice and Medicinal Plants Research Center, Cimanggu Bogor, it shows that papaya leaves quantitatively contain 1.5% flavonoids and the normal level of flavonoids in papaya leaves is 0-2%.

METHODOLOGY

This research was conducted for two months (60 days), namely in June - August 2024 at the Tablolong Beach Fish Fry Center, West Kupang District, Kupang Regency. Department of Marine and Fisheries of East Nusa Tenggara Province.

The fish used were asian seabass with a size of 6-7 cm with a stocking density of 10 per treatment. The use of papaya leaves in powder form is done naturally, namely by drying using sunlight and then ground. Papaya leaves powder is given to fish every two weeks according to the treatment with the soaking method. The soaking process is carried out by giving powder to the fish's living medium according to the treatment and soaking for 20 minutes. After that the fish are returned to the maintenance container.

This study used a completely randomized design (CRD) with 4 treatments and 3 replications, namely:

- Treatment A : Papaya leaves powder (*C. papaya*) 3.25 g/L
- Treatment B : Papaya leaves powder (*C. papaya*) 3.75 g/L
- Treatment C : Papaya leaves powder (*C. papaya*) 4.25 g/L
- Treatment D : Control or Without Papaya leaves powder (*C. papaya*)

Test Parameters

Glucose (mg/dL)

Blood glucose measurement in asian seabass using the *Glucosure Star* tool. For measurement using the *Glucosure Star* tool, the fish blood sample obtained is then dripped onto a glucose test strip, then the test strip is inserted into the glucose meter so that the blood glucose results contained are read. This measurement was measured before being given papaya leaves powder and after being given papaya leaves in each treatment to determine changes in glucose in asian seabass.

Absolute Weight Growth (g)

Measurement The calculation of absolute weight is calculated using the formula of Effendy *et al.* (1997) as follows:

$$Wm = Wt - Wo$$

Description:

- Wm : Absolute weight growth (grams)
- Wt : Final average weight (grams)
- Wo : Initial average weight (grams)

Absolute Length Growth (cm)

The calculation of absolute length growth uses the formula of Effendy *et al.* (1997) as follows:

$$Pm = Lt - Lo$$

Description:

- Pm : Length Growth
- Lt : Final length of the study at week t
- Lo : Initial length of the study

Survival Rate (%)

Observation The survival rate (SR) was obtained based on that proposed by Effendy *et al.*, (1997) as follows:

$$SR = Nt / No \times 100\%$$

Description:

- SR : Survival Rate (%)
- Nt : Number of Fish at the end of the study
- No : Number of Fish at the beginning of the study

RESEARCH RESULT

Blood Glucose of Asian seabass (Lates calcarifer)

Blood glucose in the fish's body is usually used to determine the level of stress in the fish. Blood glucose functions as the main supply source of energy. The blood glucose of asian seabass treated with the addition of papaya leaves flour can be seen in Figure 1.

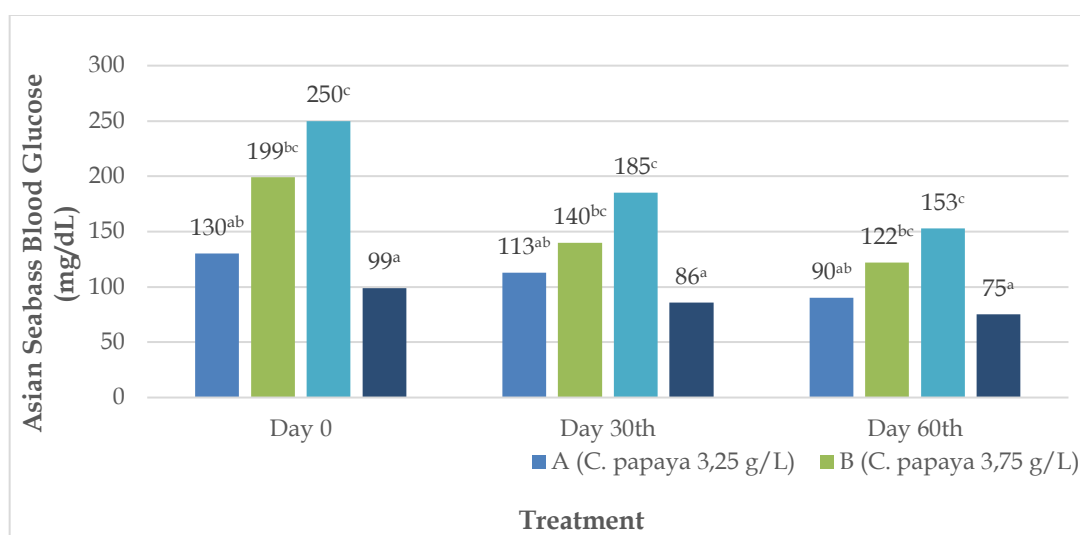


Figure 1. Blood Glucose of Asian Seabass (*Lates calcarifer*)

The blood glucose levels of white sea bass produced vary. On day 0, the highest blood glucose level was in treatment C (*C. papaya* 4.25 mg/dL) at 250 mg/dL and the lowest was in treatment D (Control) at 99 mg/dL. On day 30, the highest glucose level was in treatment C (*C. papaya* 4.25g/L) at 185 mg/dL and the lowest was in treatment D (Control) at 86 mg/dL day. On the 60th day, the highest glucose levels were found in treatment C (*C. papaya* 4.25g/L) at 153 mg/dL and the lowest were found in treatment D (Control) at 75 mg/dL. The glucose levels of snapper fish for each treatment from day 0 to day 60 experienced a decrease. A significant decrease occurred in the treatment with the use of papaya leaves.

Absolute Weight Gain of Asian seabass (Lates calcarifer)

The absolute weight gain in asian seabass increased during the 60-day maintenance period with papaya leaves flour treatment. The absolute weight growth of asian seabass fish can be seen in Figure 2.

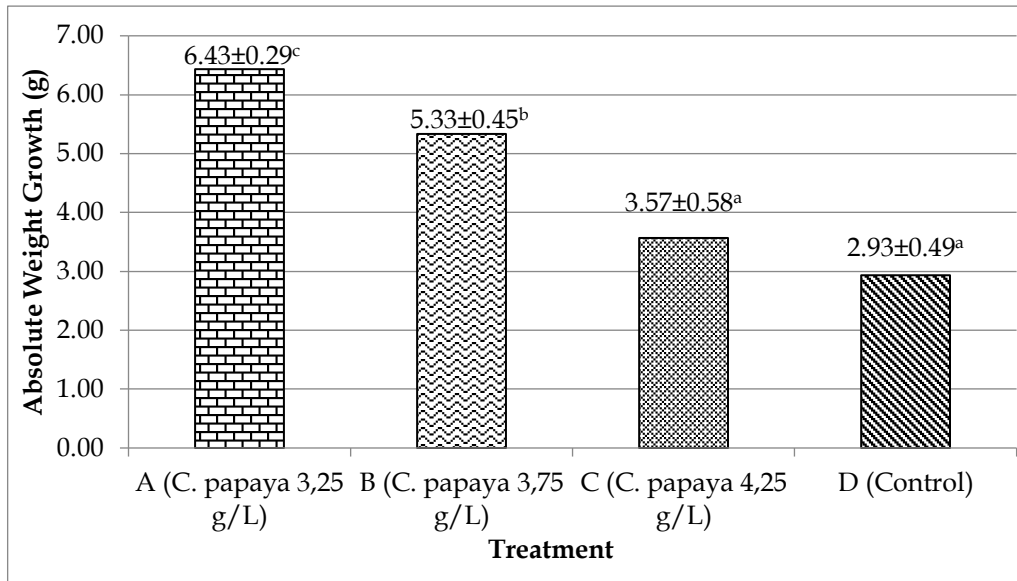


Figure 2. Absolute Weight Growth of Asian Seabass (*Lates calcarifer*)

The results of the study in Figure 2. show the difference in absolute weight growth of asian seabass fish (*L. calcarifer*) with different combinations of papaya leaves flour. The highest absolute weight growth of fish was found in in treatment A (*C. papaya* 3.25 g/L) with a value of 6.43±0.29 g, followed by treatment B (*C. papaya* 3.75 g/L) of 5.33 ±0.45 g, then treatment C (*C. papaya* 3 ,75 g/L) of 3.57 ±0.58 g, while the lowest absolute weight of fish was in treatment D (0 g/L) of 2.93 ±0.49 g. The ANOVA test data on this parameter showed that the administration of papaya leaves flour had a significant effect. on absolute weight growth so that further tests were carried out to determine the differences between each treatment, where all treatments were significantly different from each other.

Absolute Length Growth of Asian seabass (Lates calcarifer)

The absolute length growth of asian seabass is influenced by the level of stress so that it affects growth. The growth in length of asian seabass can be seen in Figure 3.

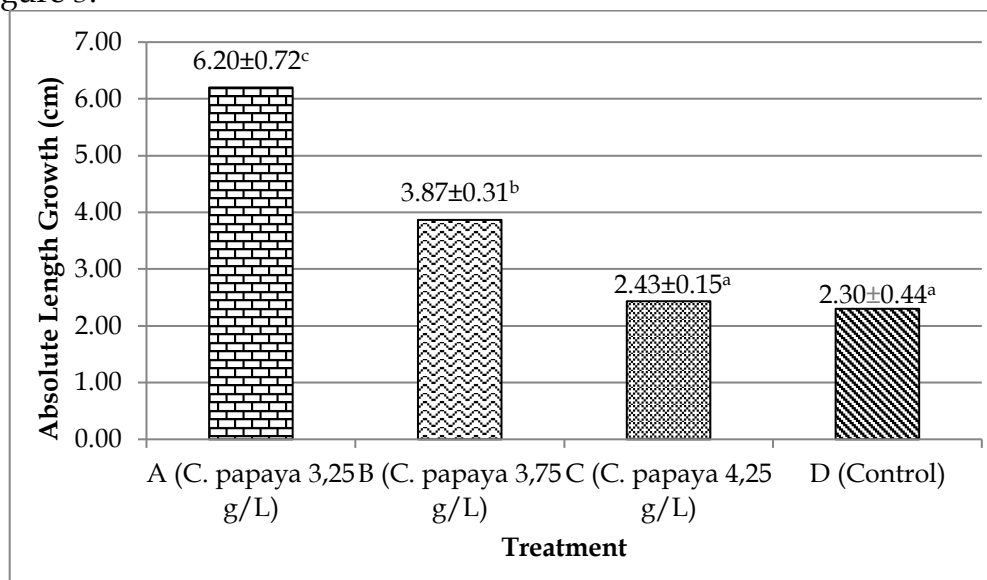


Figure 3. Absolute Length Growth of Asian Seabass (*Lates calcarifer*)

The research results in Figure 3. show that the highest absolute length growth of asian seabass was found in treatment A (*C. papaya* 3.25 g/L) with a value of 6.20 ± 0.72 cm, followed by treatment B (*C. papaya* 3.75 g/L) was 3.87 ± 0.31 cm, then treatment C (*C. papaya* 3.75 g/L) was 2.43 ± 0.15 cm, while the lowest absolute length growth rate of fish was found when feeding D (0 g/L) at 2.30 ± 0.44 cm. The ANOVA test data on this parameter showed that the provision of leaves flour had a significant effect on the absolute length growth of asian seabass fish, so that further tests (LSD) were carried out to determine the differences between each treatment that could be, where all treatments were significantly different from each other.

Survival Rate of Asian seabass (*Lates calcarifer*)

Fish survival rate refers to the percentage of fish that survive in a population over a period of time. Fish survival rate is an important indicator in aquaculture, as it reflects the success of aquaculture management, fish health, and environmental quality. Fish Survival Rate asian seabass during the study is shown in Figure 4.

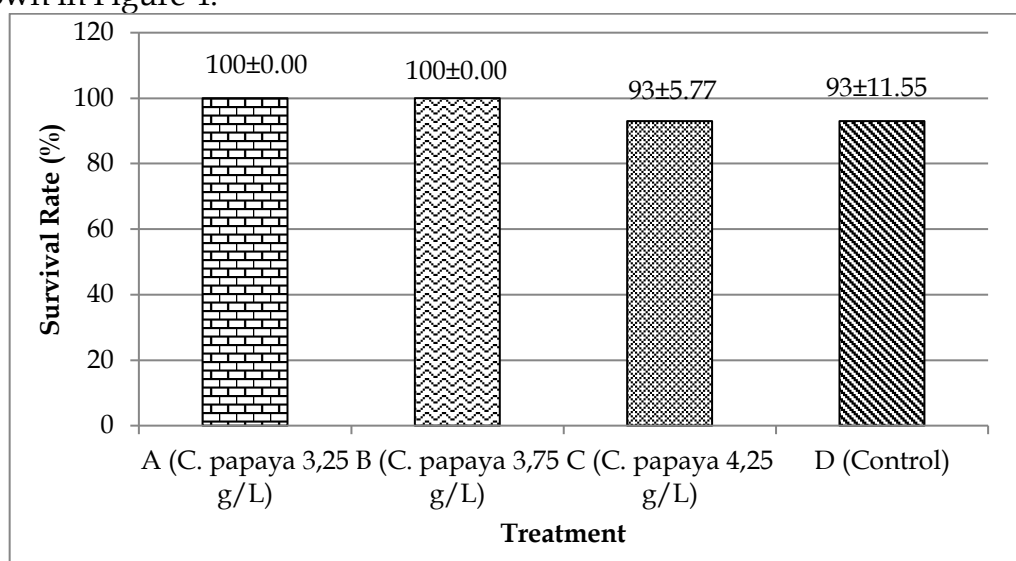


Figure 4. Survival Rate of Asian Seabass (*Lates calcarifer*)

The survival rate of asian seabass as a result of the different methods of giving papaya leaves flour in Figure 4 shows the survival rate. The highest survival value of asian seabass was found in treatments A and B of 100%, then followed by treatment C of 93%. The ANOVA test data on this parameter showed that the provision of papaya leaves flour treatment did not have a significant effect on the survival rate of asian seabass.

DISCUSSION

The use of papaya leaves powder during the maintenance period resulted in a decrease in fish glucose levels from day 0 to 60. This is thought to be due to the role of papaya leaves powder in reducing stress levels in asian seabass. The

increase in blood glucose at the beginning of the breeding season is caused by fish stress in a new environment. The stress experienced by fish causes hyperglycemia, which means a rapid increase in blood sugar over time (Shabrina *et al.*, 2018). The blood glucose levels of asian seabass in this study exceeded the normal range of normal fish blood sugar levels, which is 40-90 mg/dL (Shabrina *et al.*, 2018). Juharni *et al.*, (2022) stated that high blood sugar levels indicate that fish are adapting to the new environment and water quality.

Blood glucose levels will further affect decreased blood volume, decreased leukocytes, decreased liver glycogen, shrinking stomach diameter and thinning of the mucosal layer. However, in this study it can be seen that blood glucose levels in treatment A (*C. papaya* 4.25g/L) quickly decreased from 130 mg/dL to normal again at 90 mg/dL. Glucose is important to meet the high energy needs caused by stress, because stress diverts energy from normal metabolic processes to energy used to activate the body's systems in response to stress (Eka *et al.*, 2019). The decrease in blood sugar indicates that fish use sugar energy to respond and adapt to stress. The ability of fish to adapt to the environment, the duration and magnitude of the stress response depends on the species, intensity and duration of the response. In fish, especially carnivorous species, long-term increases in blood sugar (hyperglycemia) can be observed even after fish consume carbohydrate-rich foods (Syakirin *et al.*, 2024).

The growth of asian seabass is influenced by protein consumption. In treatment A, the highest growth was obtained because in this treatment the fish consumed more feed and the feed consumed was used for growth. According to research (Herlina, 2016) and (Arief *et al.*, 2009), protein also plays a role in various metabolic processes and body functions of fish. Papaya leaves contain alkaloid compounds carpaine, caricak santin, violak santi, papain, saponin flavonoids, and polytenol. In fact, good nutrition affects good growth responses (Hasanah *et al.*, 2019). Alkaloids, steroids, flavonoids, saponins, triterpenoids and essential oils to support the digestion of food protein (Yunus & Yushra, 2023). Papaya leaves flour contains the enzyme papain which produces proteolytic enzymes. Papain is a proteolytic enzyme from papaya flour, *Carica papaya*, which can hydrolyze protein into amino acids (Saleky, 2021). Hasina *et al.* (2012) reported that papaya flour contains protease enzymes (protein decomposers), namely papain and chymopapain. These two enzymes have the ability to break bonds in protein molecules so that proteins can be broken down into amino acids.

The high absolute length growth of the treatment (*C. papaya* 3.25 g/L) is due to the high level of fish appetite and also contains high protein so that it produces more energy in the fish according to (Wardoyo *et al.*, 2024). The content of active compounds and enzymes in papaya leaves helps stimulate appetite and accelerate the digestion process in fish (Syakirin *et al.*, 2023). Through the process of soaking papaya leaves, it helps accelerate the absorption of nutrients in the feed consumed by asian seabass.

The factor that causes the survival rate of asian seabass to reach 100% is that the nutrients contained in the feed can be optimally utilized for the metabolic process and increase the immune system. In addition, water quality supports the normal life process of fish. (Kumalasari, 2021) which states that feed and water

quality parameters play an important role in the survival of aquatic organisms. At harvest, the high presentation obtained indicates that the quality of feed and environmental quality are appropriate during the cultivation process (Rahmaningsih *et al.*, 2013). The success of aquaculture is determined by several factors, one of which is the survival rate. Aquaculture production data supported by high survival will affect the production and profits that will be generated, so this factor must be a primary concern in aquaculture (Khalil *et al.*, 2021).

CONCLUSIONS AND RECOMMENDATIONS

Papaya leaves flour with soaking process affects blood glucose, absolute weight growth and absolute length growth but does not affect the survival rate of asian seabass. The best treatment of papaya leaves flour dose in treatment A is (*C. papaya* 3.25 g/l) which produces fish blood glucose on day 60 of 90 mg/dL, absolute weight growth of 6.43 g, absolute length growth of 6.20 cm and survival of 100%. The suggestion that can be given is that further research is needed related to lower doses of papaya leaves flour mixed in the asian seabass maintenance container.

ADVANCED RESEARCH

The author hopes that in the future researchers can use the same method and apply it to different types of fish to see its effects.

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