

## **Original Research Article**

# **The Impact of Addition of Soybean Oil to Increase the Growth, EPA and HSI in Biofloc Catfish**

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### **ABSTRACT**

This study aimed to determine the level of increasing of omega-3 fatty acids EPA, Hepatosomatic Index (HSI), Specific Growth Rates (SGR), and Average Daily Growth (ADG) with the addition of soybean oil to feed as supplement. This research method uses a Complete Randomized Design (CRD) consisting of 4 treatments and 4 tests. The different concentration of soybean oil in the feed were considered as treatment, A (0%); B (2%); C (4%); and D (6%). The test fish used was catfish (*Clarias sp*) from Bandung Regency, West Java. Feed as much as 2.5% of the biomass weight is given 2 times a day, namely at 07 a.m and 16 p.m for 30 days in a biofloc tank. The conclusion of this study was that the addition of soybean oil showed the best result is in the addition of 4% soybean oil to the feed. The dose showed that the addition of soybean oil as much as 4% to the feed was able to have the best influence on the increase in Omega-3 eicosapentaenoic acid/EPA fatty acids by 25 mg/100g with the highest Hepatosomatic Index (HSI) value of  $1.52 \pm 0.11$ , the best Specific Growth rate (SGR) value of 1.1% and Average Daily Growth (ADG) value of 0.27%.

*Keywords: Soybean Oil, Catfish, Eicosapentaenoic Acid, Specific Growth Rates, Average Daily Growth*

### **1. INTRODUCTION**

Soybeans are a type of legume plant that has high nutrition. Every 100 g of soy dry matter contains 35 g of protein, 35 g of carbohydrates, 18-20 g of fat as well as other nutritional content. Soybeans are known as oilseeds that contain several beneficial nutrients including proteins, carbohydrates, vitamins, and minerals. Dried soybeans contain 36% protein, 19% oil, 35% carbohydrates (17% of which are dietary fiber), 5% minerals, 0.25% calcium, 0.28% magnesium, 0.58% total posfor and several other components including vitamins [1-2]. Soybeans are usually processed as the basic ingredients of various products, one of which is soybean oil. The virtue of soybean oil is that it contains high protein levels compared to other types of nuts and contains high levels of unsaturated fatty acids and contains natural antioxidants [3]. Soybean protein is one of the cheapest sources of dietary protein [4]. Soybean protein was considered a good substituent for animal protein [5], and its nutritional profile except sulfur amino acids (methionine and cysteine) is almost similar to animal protein because soybean protein contains most of the essential amino acids needed for animals and human nutrition. Most of the fatty acid content of soy consists of essential fatty acids that are needed by the body. The main components contained in soybean oil are unsaturated fatty acids (85%) and saturated fatty acids (15%) [6]. According to [7] soybean

oil contains 49.9% linoleic acid and 9.1% linolenic acid. With this high content of fatty acids, soybean oil has the opportunity to increase the content of omega-3 fatty acids through catfish feeding.

Catfish (*Clarias* sp.) is one of the most popular types of fish commodities in Indonesia that can be cultivated on limited land and water sources with high stocking density, the cultivation technology used is relatively easy to master by the community, with low business capital, easy marketing, and short cultivation time [8]. According to [9] catfish have quite good prospects for development. Another advantage of catfish is that it contains omega-3 fatty acids which are relatively higher compared to other types of freshwater fish but lower when compared to seawater fish [10]. The age and weight of the fish greatly affect the content of unsaturated fatty acids and omega-3 fatty acids of catfish. According to [11] the content of unsaturated fatty acids in catfish with a weight size of 140-170 gr/fish is the highest is 60.725%. While the lowest saturated fatty acid content is 34.47%. The highest EPA content is 0.96% at a weight of 200-250 gr/fish and the lowest is 0.84% at a weight of 100 g/fish.

Omega-3 belongs to a type of polyunsaturated fatty acid that has multiple bonds. The first bond lies in the third carbon atom of the methyl omega group, the next bond lies in the third carbon atomic number of the previous double bond. Natural fatty acids include omega-3 fatty acids, one of which is *eicosapentaenoic acid* (C20: 5) or EPA [12].

EPA fatty acids are abundant in fish and foods that contain high fatty acids such as soybean oil. This fatty acid belongs to the LCPUFA (*Long Chain Polyunsaturated Fatty Acid*) group which has an important role in visual function, and brain development helps in the formation of blood cells and the heart, nourishes the blood, and also helps good blood circulation [13]. EPA fatty acids are precursors of prostaglandin-3 (which inhibit platelet aggregation), thromboxane-3, and leukotin-5 groups (all eicosanoids). EPA fatty acids also serve as builders of most parts of the cerebral cortex of the brain and the normal growth of other organs [14]. In addition, according to [15], an important role of omega-3 fatty acids is that they can help improve memory for people with Alzheimer's.

In research [11], the addition of soybean oil to feed was able to produce a relatively high EPA of 5.05%. Meanwhile, in study [16], a mixture of 9% squid oil and 3% soybean oil was able to produce an EPA of 5.10%.

The use of biofloc systems is considered effective and able to increase productivity. Because of a narrow biofloc tank, it can produce more fish. Biofloc technology is carried out by adding organic carbohydrates to the maintenance medium to increase the C/N ratio and stimulate the growth of heterotrophic bacteria that can assimilate inorganic nitrogen into bacterial biomass. Heterotrophic bacteria are believed to be able to convert ammonia - nitrogen discharge from a cultivation system into bacterial biomass that has the potential to be used as a source of feed for fish [17]. Thus, production costs can be reduced, especially in terms of feed. In addition, the time required is relatively shorter than using conventional cultivation systems [18]. Based on research [19], the growth of fish with a biofloc system has increased faster than non-biofloc. The 45th day of catfish growth with a biofloc system continued to increase higher with an average of 45.61gr, while non had an average of 43.51gr. The biofloc system also increases the HSI value of 0.71 compared to non-biofloc which is 0.60 [20].

Catfish with a biofloc system are able to be superior to other types of fish because they had an arborescent that can support the growth of fish in high stocking density and sufficient water quality. Therefore, the purpose of addition soybean oil to catfish feed is to suspect the most appropriate dosage to increase the omega-3 fatty acid EPA and promote the growth of catfish (*Clarias* sp). The results of the study are expected to be able to provide information for farmers and the public about the important role of fat in feed to improve the quality of catfish by using biofloc systems.

The purpose of this study is to determine the effect of the addition of soybean oil in feed on the *eicosapentaenoic acid*/EPA fatty acid content and catfish growth (*Clarias sp.*). the results of the study are expected to be able to provide information for farmers and the public about the important role of fat in feed to improve catfish quality by using biofloc systems.

## 2. METHODS

This research was conducted in August – September 2021 at the Inland Fisheries Area Laboratory, Ciparanje, Padjadjaran University. The test fish used was an enlarged catfish (*Clarias sp.*) obtained from resident catfish farmers in the village of Cileunyi Kulon, Cileunyi District, Bandung Regency, West Java Province. The feed used to perform the study. The test feed contains different fats in each treatment. Feeding was carried out 2 times a day during the maintenance period of 30 days in a fiber tub tank with a size of 68x68 cm. Other adjuvants include probiotics, molasses, and adhesives.

The stage of making feed was mixed feed and soybean oil with the addition of progol as an adhesive, then stir until homogeneous and the feed is dried by aerating.

The study was conducted experimentally with a Complete Randomized Design (CRD) consisting of four treatments and four replays. The treatment used was the addition of soybean oil to the feed with the following concentrations.

A: Control (without the addition of soybean oil)

B: Feed with the addition of soybean oil 2%

C: Feed with the addition of soybean oil 4%

D: Feed with the addition of soybean oil 6%

Acclimatization in this study consists of acclimatization of cultivation media and test feed given during the maintenance period to assist in good growth and be able to survive during the maintenance process. The water medium was carried out to precipitate with the addition of aerator to removed particles or substances that can inhibit the sustainability of fish. During the acclimatization process, the fish were given commercially pellet from Hi-Pro Vite 781 (31-33% protein, 4-6% fat, 3-5% fiber and 9-10% water). Mixed feed with soybean oil little by little with the aim that the fish would be able to receive the feed given during the study.

The data observed in this study are the content of omega-3 fatty acids EPA, Hepatosomatic Index (HSI), Specific Growth Rates (SGR), and Average Daily Growth (ADG).

### 2.1 Omega-3 EPA

EPA's omega-3 fatty acids were tested using the gas chromatography method at the Saraswanti Indo Genetech Laboratory and analyzed descriptively.

### 2.2 Hepatosomatic Index (HSI)

According to [21] , the value of the *hepatosomatic index* (HSI) can be calculated using the following formula.

$$HSI = \frac{W_h}{W_t - W_h} \times 100$$

Information:

HSI = Hepato Somatic Index

W<sub>h</sub> = Heavy heart (g)

W<sub>t</sub> = Weight (g)

## 2.3 Specific Growth Rates (SGR)

To see the growth of fish specifically, the formula used to calculate *specific growth rates* (SGR) according to [22] is.

$$SGR = \frac{\ln Wt - \ln Wo}{t} \times 100\%$$

Information:

Wt = Average weight of fish at the end of maintenance (g)

Wo = Average weight of fish at the beginning of maintenance (g)

t = Length of maintenance time

## 2.4 Average Daily Growth (ADG)

The daily growth rate using the formula according to [23] is.

$$ADG = \frac{ABW_2 - ABW_1}{T}$$

Information:

ABW<sub>2</sub> = Average weight of the second or subsequent sampling (g/fish)

ABW<sub>1</sub> = Average weight of the first or previous sampling (g/fish)

T = Sampling time distance (days)

## 3. RESULTS AND DISCUSSION

### 3.1 Omega-3 EPA

Based on the results of studies that have been carried out, a level of 4% is the most effective dose in growth, hepatosomatics index, specific growth rate and average daily growth. The level was thought to have a higher chance of the presence of fatty acids contained in catfish. According to [24], the large HSI content will be suspected to be directly proportional to the amount of fatty acids contained in the fish.

Result of omega-3 eicosapentaenoic acid/EPA analysis is shown in table below.

**Table 1. EPA fatty acid in catfish**

Treatment	EPA fatty acid (mg/100g)
Without adding soybean oil	16.4
With addition of 4% soybean oil	25

Note: The result of chromatography gas test by Saraswanti Indo Genetech [25]

Table 1 above shows that feed with the addition of 4% soybean oil and without addition produces different omega-3 levels due to the addition of different soybean oil to the feed. The treatment without the addition of soybean oil only obtained omega-3 fatty acidS EPA 16.4 mg / 100g, while the addition of soybean oil as much as 4% was able to produce a higher value of omega-3 fatty acids EPA of 25 mg / 100g. The yield of EPA fatty acid content in the addition of soybean oil is 4% increased when compared to without the addition of soybean oil. This is in line with the research of [11] showing that the application of soybean oil to feed produces a relatively high EPA of 5.05% increased from a concentration of 5% to 10%. Meanwhile, the research [16] on the addition of feed with 9% squid oil and 3% soybean oil was able to produce a proportion of EPA fatty acids of 5.10%.

The difference in the EPA fatty acid content of catfish is thought to be due to the composition of fats and fatty acids in fish depending on the type of food consumed by fish [26]. The nutritional balance of catfish feed is very important in supporting the growth and quality of catfish. Fatty acid nutrients from soybean oil are used as additives that play a role

in growth. Lack or deficiency of fatty acids in fish can lead to disruption of growth function, feed efficiency till death [27] .

Increased levels of omega-3 fatty acids EPA catfish can provide benefits for those who consume catfish. These omega-3s play a role in vision function, connecting most of the cerebral cortex of the brain and other normal growth [28] as well as helping in the formation of blood cells and the heart, nourishing the blood, and also helping with good blood circulation [13]. Lack of omega-3 fatty acids can inhibit the process of growth, brain development, nerve cell problems, and decrease vision function [29].

### 3.2 Hepatosomatic Index (HSI)

The results in table 2 showed that the treatment given to soybean oil in catfish feed showed a noticeable difference ( $P < 0.05$ ) with the highest HSI value of  $1.52 \pm 0.11$  in the C treatment (4%). This is following [30] on the addition of soybean substitutions, which influences HSI values ranging from  $1.06 \pm 0.01$  to  $1.44 \pm 0.01$ .

**Table 2. Hepatosomatic Index (%)**

Component	HSI			
	A (0%)	B (2%)	C (4%)	D (6%)
HSI	$1.15 \pm 0.15^a$	$1.42 \pm 0.21^{\text{from}}$	$1.52 \pm 0.11^b$	$1.41 \pm 0.20^{\text{from}}$

Note: a.b = similar letter notation means that there is no noticeable difference in the level of the test Duncan has a value of 5%.

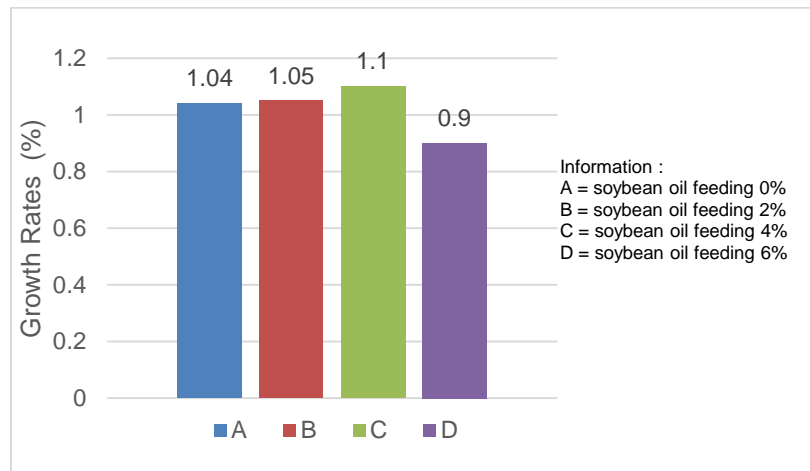
An increase in hepatosomatic values shows that there **was** glycogen and fat storage in the liver and muscles as a source of energy. High hepatosomatic values indicate the amount of fat content and fatty acids that can be used as a source of energy to support their growth. Low hepatosomatic values indicate the ability of fish to utilize poor feed, and low nutrient balance so that protein is used as an energy source and causes the growth rate to be slow [31].

According to [32], an increase in HSI values indicates an increase in the amount of nutrients absorbed and causes the amount of accumulated nutrients in the liver to increase as well. According to [33], the liver is one of the places where fat is stored. The liver also has a big role in the occurrence of metabolism and fat synthesis. Fat stored in the liver will cause an increase in size in the liver. The amount of fat content in the fish liver is thought to be directly proportional to the amount of fatty acids contained in the fish.

One of the benefits of using a biofloc system is that it was able to increased the hsi value compared to a non-biofloc system. Female catfish with a biofloc system produced a value of 0.71, and the non-biofloc system is 0.60. while male catfish with a biofloc system produced a value of 1.2 and a non-biofloc system was 1.0 [20]. Similarly, research [34] using the biofloc system in tilapia produced the highest value of 3.35 and the control treatment was 1.80.

### 3.3 Spesific Growth Rates (SGR)

Growth is defined as a change in fish in units of weight, size, or volume over time. The specific growth rate is calculated to find out the weekly growth of fish expressed in percentages (%).



**Fig 1. Specific Growth Rates (SGR) in Catfish**

In figure 1 above, it shows that the application of 4% soybean oil to catfish feed shows the highest growth rate value of 1.1%, and the application of 2% soybean oil shows a growth rate value of 1.05%, without giving soybean oil or 0% shows a growth rate value of 1.04% and giving soybean oil 6% shows a growth rate value of 0.9%. The decrease in the growth rate value in the D treatment (6%) is suspected to be because the added soybean oil causes a decrease in the quality value of the feed and the nutritional content becomes unbalanced for catfish.

In contrast to [35] using 4% fat supplementation consisting of a mixture of 2% coconut oil and 2% pecan oil in tilapia fry, produced a higher growth rate value of 2.09%, as well as [30] study on the addition of soybean substitution in barley fish feed, showed the highest growth rate value of 2.60%.

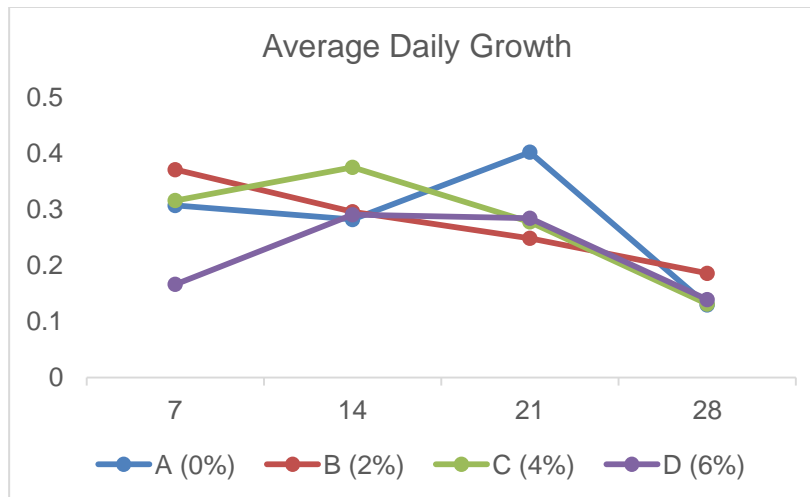
According to [36], The use of oil as a source of fat in fish feed is very important to support fish growth. The use of oil in feed can act as a substitute for protein as an energy source so that energy use can be saved and protein can be absorbed to be optimally utilized in growth. The high-fat content due to the addition of soybean oil causes the activity of lipogenic enzymes to decrease, thus inhibiting the synthesis of fatty acids [37] and can increase the chances of fat peroxidase and affecting the artibut sensors in muscles [38-39].

Fat peroxidation is thought to reduce the efficiency value of feed and HSI (treatment D). This proves that catfish are able to take advantage of the addition of fat from soybean oil up to the limit of 4%. Existence of this decrease is suspected that the feed given has an unbalanced nutritional value. According to [40], the composition of feed with high-fat content will affect the activity of the enzymes amylase, lipase, and protease which can cause an increase in the occurrence of fat peroxidase resulting in a low growth rate.

### 3.3 Average Daily Growth (ADG)

Based on the ADG chart (Figure 2) catfish growth during maintenance fluctuates at each treatment. This is thought to be due to competition for feed which causes fish growth to increase and decrease significantly. The average daily growth ranges from 0.21 to 0.27%. The highest value was found in the C (4%) treatment of 0.27% and the smallest value was found in the D (6%) treatment of 0.21%. This is different from the research of [41] shows an ADG value of 0.9% for catfish in biofloc tanks.

There is a decrease in the average daily growth due to the use of protein feed in catfish magnification size allocated for metabolic processes, breeding, energy sources, growth, reproduction, and activity [42].



**Fig 2. Average Daily Growth in Catfish**

The use of feed with the addition of soybean oil can increase growth at a dosage of up to 4%. Catfish need fat with a content ranging from 4-5% of the weight of the feed. The use of excess fat can reduce daily feed consumption and reduce fish growth [43]. The maximum use of fat in the feed can encourage fish to grow faster and produce a **protein-sparing effect**. The **protein sparing effect** can balance the use of fats and carbohydrates for metabolic activities so that the protein contained in fish can be used for growth [44].

The application of biofloc technology was one of the alternatives to solving the problem of aquaculture waste, aquaculture waste would be used by heterotroph bacteria to become additional feed for fish. Nitrogen waste produced by aquaculture organisms is converted into protein bacterial biomass that is good for growth [45]. Sangkuriang catfish (*Clarias gariepinus*) farming performed with a biofloc system is able to produce an ADG value of 1.4 gr/fish/day [41].

#### 4. CONCLUSION

Based on this study, it can be concluded that the addition of soybean oil of various levels (0%,2%,4%,6%), the best level is that the addition of 4% soybean oil to the feed was able to increase growth and HSI. The level is thought to have the highest EPA omega-3 fatty acid content compared to other levels. The observations showed that the value of omega-3 fatty acid *eicosapentaenoic acid*/EPA was 25 mg/100g, the highest HSI value was  $1.52 \pm 0.11$ , the best SGR value was 1.1% and the ADG value was 0.27%.

#### 5. COMPETING INTERESTS:

Authors have declared that no competing interests exist.



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