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Effects of Spirulina Meal Supplementation on Growth and Survival Rate of Royal Whiptail Catfish Fry (*Sturisoma panamense*)

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Abstract

Royal Whiptail Catfish (*Sturisoma panamense*) have high-stress levels in the early stages of their life. This is a significant concern in the field of fish farming. One of the crucial factors in the cultivation of royal whiptail catfish is the addition of vegetable or algae protein to the feed according to the habits of herbivorous fish. This study aimed to determine the best dose of spirulina that can enhance the growth and survival rates of royal whiptail catfish fry. There were four treatment groups, i.e. feed without spirulina (K), a dose of 3% (SP-3%), a dose of 6% (SP-6%), and a dose of 9% (SP9%). The study was conducted at the Tetra Aquaria Company in Sukabumi City in April - May 2024. The spirulina culture was dried in an oven at 60 °C for 24 hours and ground using a mortar. The powdered feed was mixed with spirulina according to the treatment dose. The fish larvae used were newly hatched larvae, and the maintenance period during the treatment was 21 days. The maintenance container uses a tray measuring 40 cm x 30 cm x 15 cm. The study results showed that the best group was SP-6% with an average survival rate of 87% ± 4.62, significantly different compared to the K, SP-3%, and SP-9% treatments. The daily growth rate value of SP-9% showed the best significant difference (P <0.05) from the control treatment and 3% dose, however not significantly different (P> 0.05) with SP-6%.

INTRODUCTION

The royal whiptail catfish (*Sturisoma panamense*) is known as panamense. This fish is native to Central and South America (Ren *et al.* 2019). Panamense is a calm fish and has a slender and elongated body shape. It is brown or yellow with a small black pattern along the sides so that it becomes its attraction. Most of them are herbivores that feed on algae, vegetables, and biofilms (Burbano 2022). This commodity has high prospects for cultivation because it is unique and has a high selling price (Akbar 2021). However, panamense cultivation is still difficult because many fail in larval stadia.

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Panamense larvae have a high level of stress if their needs are not met, one of which needs to be considered is feed.

Aquadiction World (2023) states that to accelerate the growth of *panamese* larvae, a high-protein diet is needed, and this fish needs vegetables or algae when it is still in the stadia larvae. This study will examine the composition of algae for panamense fish larval feed. The algae used is a type of *Spirulina platensis*. Spirulina has a high protein content of between 59-63%, fatty acids of around 7-10%, carbohydrates 13.6%, multivitamins, especially vitamin B12, and minerals (Grosshagauer *et al*. 2020, Shekharam *et al.* 1987). Spirulina has been shown to increase protein in fish feed and has reportedly shown promising results in several species, such as tilapia *Oreochromis niloticus* (Lu & Takeuchi 2004), guppies *Poecilia reticulata* (Dernekbasi *et al*. 2010), goldfish *Carassius auratus* (Vasudhevan & James 2011), barred knifejaw *Oplegnathus fasciatus* (Kim *et al*. 2013), catfish *Clarias batrachus* (Dar *et al.* 2014), rainbow trout *Oncorhynchus mykiss* (Yeganeh *et al*. 2015), olive flounder *Paralichthys olivaceus* (Kim *et al*. 2015), beluga *Huso huso* (Adel *et al*. 2016), and oscar *Astronotus ocellatus* (Mohammadiazarm 2021).

The dose of spirulina that will be used for panamense has not been studied in depth, so it needs to be researched. The dosage of spirulina refers to the research of Nazhiroh (2019) and Agung et al. (2021), who provide spirulina for carp and zebrafish feed starting from doses of 3%, 6%, and 9%. This dose has a good effect on the growth performance and survival of chef fish and zebras. Thus, the addition of spirulina with the right dose to the commercial feed of panamense fish larvae needs to be researched to get optimal growth and increase survival.

METHODOLOGY

Ethical Approval

All procedures carried out in this study are under the standard procedures and the fish were properly kept. The animals used are harmless and do not cause environmental pollution.

Place and Time

The research was carried out in March - April 2024. Spirulina culture and simplicia manufacturing were carried out at the Laboratory of the Fish Hatchery Technology and Management Study Program, IPB Vocational School, Sukabumi Campus. Meanwhile, the treatment of fish is carried out at the Tetra Aquaria Company located at Situ Endah, Lembursitu District, Sukabumi City, West Java.

Research Materials

The tools used in this study were 2 L jars, 15 L mineral water gallons, rectangular trays measuring 40 cm x 30 cm x 15 cm, calipers (Tricle Brand, Indonesia), round mortars, aeration hoses and aeration stones, aluminum foil, and plankton. The materials used in this study were panamense larvae, spirulina seeds, Walne fertilizer, natrium thiosulfate, and chlorine.

Research Design

The research design used a Complete Randomized Design (RAL), which has 4 treatments and 3 replicates. The treatment group included test feed not given control spirulina supplementation (C), test feed with 3% spirulina supplementation (SP-3%), test feed with 6% spirulina supplementation (SP-6%), and test feed with 9% spirulina supplementation (SP-9%).

Work Procedure

Container Preparation

The container used for spirulina culture uses a 2 L jar and a gallon of mineral water measuring 15 L. Then the fish rearing container uses a rectangular tray with a size of 40 cm x 30 cm x 15 cm as many as 12 trays. Spirulina culture containers and fish rearing were sterilized using chlorine at a dose of 0.01 g/L and then left for 24 hours. The aeration hose and aeration stone to be used are also put into a sterilization

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container containing a chlorine solution. After 24 hours, all equipment is rinsed with fresh water and then dried. The maintenance medium before use is sterilized with chlorine at a dose of 0.05 mL/L for 24 hours. Then, the maintenance medium is neutralized with Na-thiosulfate at a dose of 0.025 ml/L and waited 24 hours before it could be used.

Spirulina Culture

The spirulina used is a type of *Spirulina plantesis*. Spirulina culture uses the Buwono & Nurhasanah (2018) procedure. Spirulina is cultured on a laboratory scale through two stages. The first is a 1-liter scale using a 2-liter container, and the second stage is a 10-liter culture with a 15-liter container. The container to be used is washed first and then dried. The first stage of the 1-liter scale culture begins with a jar filled with 800 ml of water and 1 ml of Walne fertilizer (dose 1 ml/L) added. Spirulina seeds are added as much as 200 mL (20% of the total volume) and aerated. After 7 days, spirulina can be used for the second stage of culture. The second stage of the 10 L scale begins with a container filled with 8 L of water and 2 L of spirulina seeds added, then 10 ml of Walne fertilizer and aeration. After 14 days, spirulina is harvested by filtering using planktonet.

Spirulina Powder Preparation

Yields in 10 L cultures produce wet-weight spirulina weighing 225 g. The process of making spirulina powder is carried out by wet spirulina spread evenly on a heat-resistant container and covered with aluminum foil. Drying is done by heating it in the oven at 60°C for 24 hours. The dried spirulina is put into the mortar and then ground until smooth and powdered. The powder is stored in a closed, airtight container.

Test Feed

An amount of 100 g of commercial powder feed mixed with 3 g, 6 g, and 9 g of spirulina is stirred until well mixed, the characteristics of the feed have been mixed well, namely, the color of the powder pellets becomes darker than before mixing spirulina. As a binder, 5 g of gelatin is needed to be mixed with 100 ml of water, then the gelatin solution is cooked over low heat until it boils and a gel-like texture is obtained. The gelatin that is already in the form of a gel is mixed with spirulina feed and stirred until smooth until the texture of the feed becomes like paste. The test feed is analyzed proximate before it is carried out before fish rearing. The proximate analysis tested was fat, carbohydrate, protein, moisture content, and ash content.

Larval Maintenance

The fish used are newly hatched larvae that are 1 day old and kept until 21 days. One container is filled with 25 fish larvae. Fish are fed with ad satiation treatment with a frequency of feeding 2 times daily at 08.00 and 15.00. Spraying is carried out every morning, and the water changes by as much as 80% daily. During maintenance, water quality is maintained to meet SNI 6484.4 (2014) standards, namely temperatures ranging from 25-30°C; Ph 6,5-8; DO >3 mg/L; ammonia 0-0.5 mg/L; nitrite 0.1-0.5 mg/L. and nitrate <20 mg/L. Fish length data was collected every 7 days by measuring the total body length using a caliper.

Data Collection

At the end of the rearing, all the fish were measured for the total length and the total live fish. The growth parameters to be analyzed include length growth parameters (Zonneveld et al., 1991), average daily length growth (Sari et al., 2017), and survival rate (Effendie, 2002) using the following formula:

Length growth (cm) = final body length – initial body length

$$\text{Average daily length growth (cm/day)} = \frac{\text{final body length} - \text{initial body length}}{\text{duration of experiment}}$$

$$\text{Survival rate}(\%) = \frac{\text{Final number of fish}}{\text{initial number of fish}} \times 100$$

Feed proximate was analyzed with the standard methods (AOAC 1995). Moisture

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content analysis was carried out by drying at 105 °C. Crude protein, lipid, and ash content were determined using the Kjeldahl method (Kjeltec 2100), the ether extraction method (SER 148/3 Solvent Extractor, Velp, Italia), and muffle furnace at 550 °C for 8 hours.

Water quality parameters consist of water physics measured daily such as temperature, pH, and dissolved oxygen (DO) using a thermometer, pH meter, and DO meter. Chemical water was measured weekly using ammonia using a water test kit.

Data Analysis

The field data obtained were processed and analyzed, then tested for normality and homogeneity before conducting a variety analysis (ANOVA). The Smallest Real Difference follow-up test with a confidence interval of < 95% using the Duncan test.

RESULTS AND DISCUSSIONS

Water quality parameters

Water quality parameters during 21 days of maintenance are presented in Table 1. The water quality parameters including temperature, pH, DO, and ammonia, were not remarkably ($p > 0.05$) impacted by different levels of Spirulina dose. The level of ammonia in the dose 9% was higher compared to the 3% and 6% doses.

Table 1. Growth and survival in royal whiptail catfish *Sturisoma panamense* fish fed without the addition of spirulina (C), fed with spirulina dose 3% (SP-3%), dose 6% (SP-6%), and dose 9% (SP-9%).

Parameters	Dose of Spirulina			
	C	SP-3%	SP-6%	SP-9%
Temperature (°C)	27,29 ± 0,48	27,26 ± 0,48	27,22 ± 0,44	27,21 ± 0,45
pH	7,42 ± 0,21	7,45 ± 0,22	7,47 ± 0,26	7,45 ± 0,30
DO (ppm)	6,84 ± 0,65	6,24 ± 0,20	6,31 ± 0,24	6,44 ± 0,58
NH_3 (ppm)	0,25 ± 0,02	0,35 ± 0,03	0,42 ± 0,04	0,55 ± 0,02

DO: Dissolved oxygen.

In this research, feed optimized with US soybean meal showed positive results compared to control/commercial feed. This can be seen in Figure 2. The average weight of the treatment using feed optimized with US soy is 1262.4 g, while the control feed is 1220 g. This shows that feed with a US soy formulation has good nutritional content for fish growth from fingerlings to market size. However, the final average of tilapia in all treatment diets was above 1200 g, and there were no significant differences ($P > 0.05$). Statistical analyses revealed no significant differences in the treatment, specifically between the control and trial feeds regarding the ABW ($P>0.05$). The ABW average calculated for the control feed was 523.68 436.60, whereas that trial feed (optimized with US soybean meal) was 547.45434.16. Previous studies by Ehsani *et al*. (2014) replaced fishmeal with fermented soybean meal on the average final weight of juvenile yellowfin showed no significance compared to the control group ($P > 0.05$). The final average weight from the control was 6.88 g, and the average final weight replaced with 10% fermented soybean meal was 6.87 g.

Previous studies (El-Saidy and Gaber, 2002) conducted a study that analyzed a commercial tilapia diet that contained 30% soybean meal and 20% fish meal with diets that had all of the protein originating from soybean meal, and the result after feeding for ten weeks, feed which containing 55% soybean meal outperformed the commercial tilapia diet concerning weight gain, final weight, protein efficiency ratio, feed conversion, and feed intake.

Average Daily Growth (ADG)

The results of this study found that the overall average daily growth of Nile tilapia with feed control and feed with US soybean meal can be seen in Figure 3.

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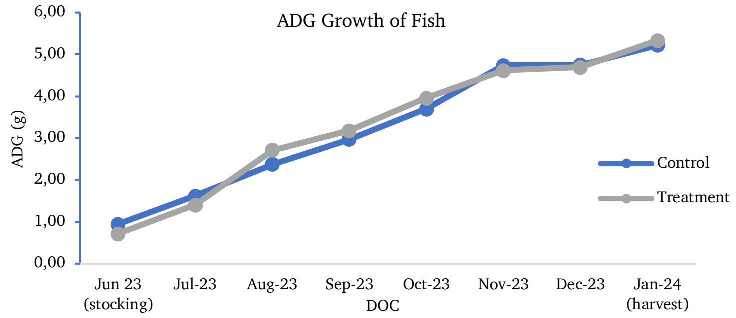

Figure 3. Comparability of overall ADG between control feed and feed optimized with the US soybean meal.

The growth performance can be seen in Figure 3, where tilapia fish given control/commercial feed shows a significant increase in ADG with a value of 5.21 g, while compared to fish given feed with US soybean meal formulation had better ADG than control, which is 5.34 g. However, statistical analyses using the T-test revealed no significant differences between this treatment of both the control feed and trial feed (P >0.05). The ADG average calculated for the control feed was 3.34 1.47, whereas that trial feed (optimized with US soybean meal) was 3.421.46. Based on the previous study by Ehsani *et al.* (2014), the ADG from the control feed was 4.40, while from feed replaced with 10% fermented soybean meal was 4.36.

Additionally, based on the result from this study proved that US soybean meal has a positive effect on tilapia growth due to treatment better than control. Soy product is utilized as livestock feed as they provide a benefit in growth performance, but this depends on the dosage that farmers use for feed manufacture (Sinha *et al.*, 2013). To identify a purpose for this by-product for freshwater aquaculture production, several studies have been conducted recently.

Biomass

Based on the results of the study, it was found that the overall biomass of Nile tilapia with feed control and feed with US soybean meal can be seen in Figure 4.

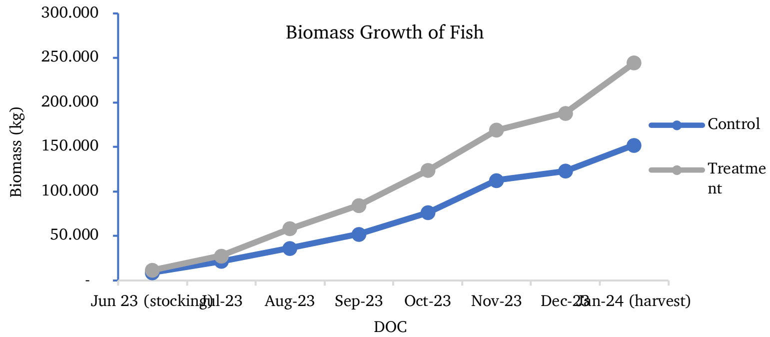

Figure 4. Liver abnormalities: degeneration (blue), congestion (green), vacuolization (red), and necrosis (black circle).

The total biomass of tilapia after post-stocking until harvest can be seen in Figure 4. The graph shows a significant difference in the increase of tilapia biomass every month. Both treatments showed positive results where the biomass increased significantly with the increase of day of culture (DOC) cultivation. However, there is a large difference in the total biomass at the end of the cultivation period. The fish

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fed with the treatment by optimizing the US soybean meal formulation had a greater biomass mass compared to the control/commercial feed, which was 244.144 kg, while the control feed was only 152.247 kg. From these data, it can be concluded that the effect of feed-given formulations using US soy has a positive impact on the amount of biomass in Tilapia fish. Similar to Nile tilapia in this study, omnivorous fish showed a high degree of acceptability towards the replacement of soybean meal for fishmeal.

However, favorable growth outcomes were preserved when fishmeal was replaced by 50% or more. Some authors suggest the soybean meal's production process may affect the digestibility performance for commercial fish like Nile tilapia (Tangendjaja, 2015).

Survival Rate (SR)

The results of the study show that the overall survival rate (SR) of Nile tilapia with feed control and feed with US soybean meal can be seen in Figure 5.

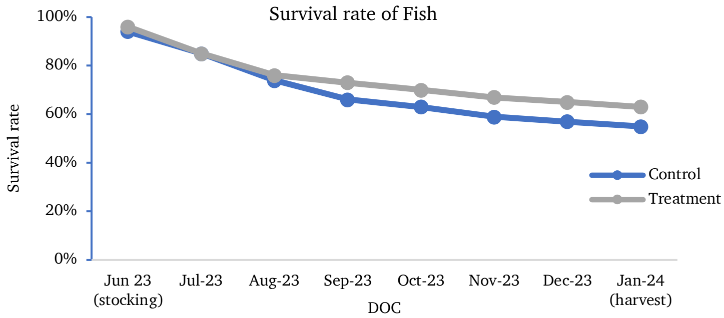

Figure 5. Comparability of overall survival rate between control feed and feed optimized with the US soybean meal.

SR (Survival Rate) is a determining factor in the success of fish farming activities. Based on the results of the study, the mortality rate was caused by several factors, such as water quality, fish health, weather conditions, etc. The main cause of reducing SR in this study was weather conditions, where in the rainy season it influences the water quality, especially in water temperature (decreased until 2°C), though the temperature is still safe for tilapia farming. However, sudden temperature changes caused the fish to become stressed and susceptible to diseases, which influenced the survival rate due to the high mortality occurrence at that time. The survival rate of tilapia in all treatment diets was above 50%, and there were no significant differences (P> 0.05). Previous studies by Ehsani *et al.* (2014) replaced fishmeal with fermented soybean meal on the survival rate of juvenile yellowfin showed no significance compared to the control group (P> 0.05). The survival rate from the control was 95%, and the survival rate by replacing with 10% fermented soybean meal was also 95%.

However, based on the result from this study, fish fed using the formulation with the US. Soy showed a percentage of the survival rate value until the end of the culture period of 64.2% while using the control feed of 55.1%. This showed that the fish given the treatment feed had a better survival rate than fish given control/commercial feed. It can be concluded that the content of nutrients in the feed can affect the health and survival of fish. According to a study by Kader *et al.* (2012), fermented soybean products can enhance blood biochemical parameters; nevertheless, determining the inclusion is necessary for improved fish health and growth.

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Feed Conversion Ratio (FCR)

The results of the study showed that the overall feed conversion ratio (FCR) of Nile tilapia with feed control and feed with US soybean meal can be seen in Figure 6.

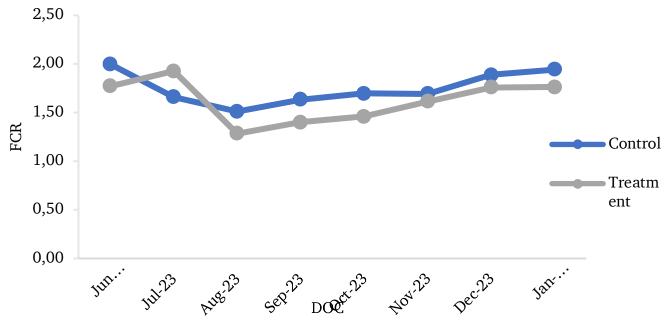

Figure 6. Comparability of overall FCR between control feed and feed optimized with the US soybean meal.

In this study, the FCR value can be seen in Figure 5. Fish given feed with US soybean meal formulations have a better FCR of 1.76 than fish given control/commercial feed of 1.94. The feed conversion ratio (FCR) was calculated to evaluate the feed utilization given to the fish. If the FCR value is lower, then the feed quality is good and effective for the growth of fish body weight. However, statistical analyses revealed no significant differences in the treatment, specifically between the control feed and trial feed regarding the FCR (P>0.05). The FCR average calculated for the control feed was 1.75 0.17, whereas that trial feed (optimized with US soybean meal) was 1.62 0.22. Based on the result from this study showed fluctuating results of FCR during the culture period, this case was caused by several factors such as weather and mortality rate, especially after 85 days post-stocking. When bad weather and high mortality occurred, most fish got a low fish appetite, which influenced the amount of feed per day and impacted the total of feed in the sampling period as well as the total biomass at the sampling time.

According to Wijayanti and Pebriani (2021), differences in FCR results depend on the protein contained in each feed ingredient, and environmental, quality, and quantity of feed affect the growth and conversion of the feed. Conversion of the feed. The lower the conversion value, the more efficiently the feed is converted into meat. The main components of soybean products include protein, lipids, fiber, and minerals. According to some studies, soybeans offer several functional qualities, including the capacity to serve as an antioxidant, the ability to prevent cardiovascular disease, the ability to reduce calorie consumption, and the potential to maintain a healthy hepatic lipid profile (Kim *et al*., 2016).

Water Quality

Water temperature is one of the most influencing environmental factors affecting the growth of fish. The better the water quality during the study, the better the growth and survival will be. The results from the observations show that the value of water quality is in optimal condition on Lake Toba. An average temperature value of 25 °C – 27 °C is in the good range for the growth and survival of Nile tilapia. This results from the following statement from Leonard and Skov (2022) that an optimal rearing temperature for Nile Tilapia of 25-30°C. Furthermore, the DO (dissolved oxygen) value shows the optimal DO for tilapia culture was in the optimal range of 5-7 mg/L. According to Indriati and Hafiludin

(2022), dissolved oxygen (DO) concentration the more it is, the better it is for aquaculture, but the best is between 5 and 7 ppm. Low DO levels occur due to the pollution of toxic ammonia compounds from feces and metabolite waste of fish and feed residues.

CONCLUSION

Based on the growth parameters in this research, the comparison between tilapia fish fed with US soybean meal optimized has better growth in average body weight (ABW) with the better value of 1262.4 g, average daily growth (ADG) with the better value of 5.34 g, biomass with the better value of 244.144 kg, survival rate (SR) with the better percentage of 64.2%, and feed conversion ratio (FCR) with the better value of 1.76. However, there was no significant effect ($P > 0.05$) on the SR (Survival rate), Feed Conversion Ratio (FCR), ADG (Average Daily Growth), and final ABW (Average Body Weight) in both the experimental diets.

CONFLICT OF INTEREST

None of the authors had any conflicts of interest upon writing and publishing this manuscript article.

AUTHOR CONTRIBUTION

Juanda: author correspondence, collecting data, researcher, sampling, Friska Setiawani Saragih: contributed to article writing and performed statistical, Hasim Djamil: contributed data analysis, Apryani Susanti: manuscript writing and analysis.

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