



Impact of Fed Containing Different Levels of Diets Supplementation *Spirulina platensis* on Growth, Haematological, Body Composition and Biochemical Parameters, of Gurami (*Osphronemus gouramy*)

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Abstract

Spirulina platensis (*S. platensis*) is a blue-green algae, shaped like a spiral, contains high nutritional value including proteins, vitamins, minerals and essential amino acids. The objective of this study is to evaluate the effects of supplementation *S. platensis* in diets and to get the level of supplementation *S. platensis* in diets gurami fish, *Osphronemus gouramy* best promote growth, hematological, body composition and biochemical blood parameters. The fish are used to measure the average weight 2.52 ± 0.17 g. The treatments tested was 5 groups, replicated 3 times for each group. The dietary levels *S. platensis* of five treatment group were: C (control group), P1, P2, P3, P4, respectively supplementation *S. platensis* 0, 2, 3, 4, and 6 g/kg of diet, the trial period is 56 days. The results showed that the level difference *S. platensis* supplementation can increase growth, hematological and biochemical blood parameters gurami. Best growth, body composition, hematological and biochemical blood parameters of gurami is obtained at the level of supplementation *S. platensis* 6 g/kg of diet. Findings from this study indicate that supplementation *S. platensis* 6 g/kg of diet in aquaculture gurami increasing growth, body composition and immunity body fish.

Keywords: immunity, *Osphronemus gouramy*, *Spirulina platensis*, supplementation

Introduction

Spirulina platensis is a blue-green algae, filamentous, spiral shape with high nutritional value content. These algae have a composition the content of nutrients, for example, 60-70% protein, essential amino acids, chlorophyll, glycogen, vitamins in a concentration balanced and Gamma Linolenic Acid (GLA). *Spirulina platensis* contain bioactive compounds with antioxidant activity that could be used to supplement food (Bermejo, Pinero, & Villar, 2008), high in protein so it can be used as a source of micronutrients (Phang, Miah, Yeoh, & Hashim, 2000). Nutrient content of *S. platensis* is very complete, vitamins B12, β -carotene, phosphorus, iron, calcium (Gutierrez-Salmean, Fabila-Castillo, & Chamorro-Cevallos, 2015). Blue-green algae, also containing phycocyanin (Jaime-Ceballos, Hernandez-Llamas, Garcia-Galano, & Villarreal, 2006), C-phycocyanin showed antioxidant activity and may as a substitute for heparin because it serves as an anticoagulant (Kamble, Gaikar, Padalia, & Shinde, 2013). These algae can protect the environment through waste recycling and energy conservation (Saranraj & Sivasakthi, 2014). Plant cell walls is generally covered by the cellulose, however, *S. platensis* cell wall composed of complex sugars that are easily digested by fish (Belay, 2002).



Simanjuntak, Dana, Wirawidjaja and Supriyadi (2002; 2003) reported that supplementation of *S. platensis* 4 g/kg diet suppressed the mortality of fish, boost immunity views of histological liver, spleen and kidneys. Even the fish tested challenged with pathogenic bacteria *Aeromonas hydrophila* recovery occurs, the wound caused by bacterial infection covered by the new network. *Spirulina* is a good source of energy that can be used as an essential component in animal feed (Kim, Rahimnejad, Kim, & Lee, 2013).

Gurami, *Osphronemus gouramy* (*O. gouramy*) is a freshwater fish, including into familia Anabantidae, a fish that is very popular in Indonesia. These fish can weigh up to 5 kg, a thick meat and savory taste. These fish can be kept in water without the circulatory system because it has a labyrinth plate. Therefore, gurami can be maintained in ponds with dirty water. However, the growth of gourami is relatively slow, to reach the size of 500 g takes 18 months (Sitanggang & Sarwono, 2001). Gurami fish are also susceptible to the disease, especially in winter or heavy rainfall, even mortality in eggs and larvae stage can reach 100%. This will cause a great loss to the farmers gurami.

Gurami cultivation and aquaculture conditions also need to be improved to achieve accelerated growth and immunity of fish. The increase in production is largely determined by the composition of the feed material, a high protein content is necessary for the parent to spawn so that the eggs produced are of good quality (Djajasewaka, 2004). One solution that can be done is by feeding management of high nutritional value which can promote the growth as well as to boost immunity, namely the provision of *S. platensis* in feed as a dietary supplement.

The use of *S. platensis* as a food supplement has been done by several researchers. Ungsethaphand, Peerapornpisal, Whangchai and Sardud (2010) of the results of the research say that the carcass proximate composition of red hybrid tilapia fed *S. platensis* showed no significant difference compared with the control. Supplementation soybean meal with *Spirulina* at different levels showed significant differences compared with the control. In the cultivation of Rainbow trout, can use *Spirulina* as a substitute protein source of soybean meal (Ahmadzade-Nia et al., 2011). Simanjuntak, Moeljopawiro, Artama and Wahyuono (2011b) proved that the extract of *S. platensis* can improve IgG and IgM mice Balb C were challenged with tachyzoite, *Toxoplasma gondii*.

Research on different levels of *S. platensis* on diet supplementation on growth, hematological, body composition and biochemical gurami has not been done. Therefore, this research needs to be done in order to investigate the effects of supplementation of *S. platensis* in gurami feed. The success of supplementation *S. platensis* on gurami will save the cost of feed, increasing growth and immunity body fish.

The objective of this study is to evaluate the effects of supplementation *S. platensis* in diets and to get the level of supplementation *S. platensis* in diets gurami fish, *Osphronemus gouramy* best promote growth (weigh and body length), hematological (red cell count, leukocyte count, hematocrit and hemoglobin level), body composition and biochemical blood parameters (total protein, albumin, globulin levels and the ratio of albumin: globulin).

Materials and Methods

Experimental Fish

Gurami (*Osphronemus gouramy*) (Figure 1.), as many as forty five, 3 months old obtained from spawning a pair of parent. Parents gurami comes from the village of Singasari, Banyumas regency, Central Java Province, Indonesia. Gurami was placed on the aquarium fiber measuring 40 cm x 60 cm x 60 cm as much as 15 pieces and was given oxygen using electric water pump and temperature maintained by heaters. Fishes were kept for one week to acclimate to the laboratory conditions. Fish were divided into five groups.

Experimental Diets

Supplementation *S. platensis* on feed

Five experimental diets were formulated to contain 0, 2, 3, 4, 6 g/kg of commercial pellets, each treatment was replicated three times. The treatments were tested, namely: C = control (commercial feed, without supplementation *S. platensis*); P1 = supplementation of *S. platensis* 2 g/kg of feed; P2 = supplementation of *S. platensis* 3 g/kg of feed; P3 = supplementation of *S. platensis* 4 g/kg of feed; and P4 = supplementation of *S. platensis* 6 g/kg of feed. *Spirulina platensis* supplementation on feed made with the following procedure: 100 ml of distilled water were placed in a spray (sprayer), then added 2 g of *S. platensis* powder is then shaken until homogeneous. Commercial pellets (1 kg) placed in the tray, *S. platensis* solution is sprayed on the feed, stirring gently and evenly so. Food that has been supplemented with *S. platensis* dried under the sun to dry, after a dry cooled at room temperature. The feed is placed on a clean container and sealed and ready tested on gurami. Supplementation of *S. platensis* level 3, 4, and 6 g/kg of feed made in accordance with the procedure described above. Feed given twice daily (08.00 and 16.00) as much as 5% of the total weight of fish per aquarium, for 56 days. The proximate composition of feed before being tried out, previously in the analysis. For feed composition analysis methods described in Faculty of Animal Husbandry, Jenderal Soedirman University, Purwokerto. The result are shown in Table 1.

Data Measurement

Growth measurements gurami (weight and body length) is done every two weeks. Body length measurements performed using millimeter blocks paper while the body weight measurement is done using a digital scale with a precision of 0.01 g.

Feed conversion ratio is done every two weeks with the calculation formula:

$FCR = \text{feed consumed by fish} / \text{weight gain by fish}$

The hematological data of gurami (number of erythrocyte, number of leukocyte, hemoglobin concentration and hematocrit value) calculation at the end of the study. Blood was drawn from the heart using a 1 ml syringe that has been moistened with anti-coagulant (EDTA). Calculation of the total number of erythrocytes and leukocytes using haemocytometer "Improved Double Naubauer's", measurement of hemoglobin levels using haemometer "Assistant" and measurement of haematocrit value used microhematocrit "Hawkskey haematocrit reader" (Chairlan & Lestari, 2011).

For body composition analysis methods (analysis proximate) described in Faculty of Animal Husbandry, Jenderal Soedirman University, Purwokerto. The result are shown in Table 3.

For biochemical parameters, at the end of the experimental period, six fish per group were collected. Blood was collected from all the alleged differences in levels of supplementation of *S. platensis*. Blood was drawn from the heart using a syringe 1 mL of 0.5 mL and placed in microtubes. Blood left in the refrigerator for 10 minutes, then centrifuged at a speed of 4000 rpm for 20 minutes. The supernatant was put in a new microtube and the sediment was removed. Serum was separated from each blood sample for the biochemical examination. Total protein levels in serum was determined According to photometric test According to the biuret method. Serum albumin concentration was measured as described by photometric test using Bromocresol green. Blood serum globulin was calculated by subtracting the concentration of albumin from that of the total protein and albumin/globulin ratio (A/G ratio) was calculated by dividing the albumin concentration over that of globulin.

Statistics Analysis

Data were presented as mean \pm standard deviation (SD). The data were analyzed by analysis of variance (ANOVA) to identify the significantly different groups at ($P < 0.05$) by one way ANOVA with post hoc LSD multiple comparison test using SPSS software statistical program (SPSS for windows ver. 21.00, USA).

Results

Feed composition (analysis proximate) with different levels supplementation *S. platensis* (experimental diets) are shown in Table 1.

The body weight and body length of gurami (*O. gouramy*) at various levels of diet supplementation *S. platensis* were measured every two weeks for 56 days. Data as shown in Figure 2 and Figure 3.

The feed conversion ratio at various levels of supplementation *S. platensis* in the gourami diet was measured every two weeks for 56 days. The data as shown in Table 2.

The data on the calculation of hematological gurami were measured at the end of the study (56 days) include: the number of erythrocyte (RBCs), leukocyte count (WBCs), hemoglobin concentration and hematocrit values as shown in Table 3.

Results of the proximate analysis of body composition showed that there were significant increases in body composition gurami along with increased levels of *S. platensis* in the feed given (Table 4).

Total protein serum, albumin, globulin levels, albumin/globulin (A/G) ratio of gurami as shown in Table 5.

Discussion

Growth Gurami Feed with Experimental Diets

The results of measurements of weight and body length growth of gurami showed that increasing levels of supplementation of *S. platensis* in the diet followed by increased growth of body length and weight of gurami. Further test results with the Latin Square Design (LSD) for different levels of supplementation showed that the control was significantly different from all levels of supplementation *S. platensis* (2, 3, 4 and 6 g/kg) ($P < 0.05$). Further test results with LSD for different of the time indicates that the difference in duration of *S. platensis* a significant increase in weight and body length growth of gurami ($P < 0.05$). Increased growth of weight and body length of gurami followed with increasing treatment time. Best growth is obtained at the level of supplementation of *S. platensis* 6 g/kg of feed, for *S. platensis* contains high nutritive value.

According to this study, Ahmadzade-Nia et al. (2011) found that the body weight of rainbow trout by different levels of soybean meal with *Spirulina* on all treatments was higher than the control. *Spirulina platensis* can be used as a substitute for fish meal in rainbow trout feed with respect to the high protein content and does not result in a negative effect on the performance of fish (Teimouri, Amirkolaie, & Yeganeh, 2013). This is because *S. platensis* contains high nutritional value, such as Gamma Linoleic Acid (GLA) and digestive enzymes (Demir & Tukul, 2010), vitamins (especially vitamin A and B12) and polyunsaturated fatty acids (PUFAS) (Wang, Pan, Sheng, Xu, & Hu, 2007), high protein (60-70 %) and minerals (Saranraj & Sivasakthi, 2014), essential fatty acids, vitamins, minerals, pigments (Ali & Saleh, 2012) that is needed for the growth of fish.

Similar results were also reported by Dernekbası, Una, Karayucel and Aral (2010), supplementation *S. platensis* in feed provide better growth compared with commercial feed on a diet of guppy. Moreira, Ribeiro, Duarte, de



Morais and Souza-Soares (2013) said that the *S. platensis* strain LEB-18 can be used as an alternative essential minerals and nutrients for the human body because it contains high protein and minerals (iron).

Spirulina platensis plays an important role in animal nutrition and can be used as a complement feed of fish, shrimp and poultry (Saranraj & Sivasakthi, 2014). Phycocyanin content of blue-green algae can be used as a functional food (Prasanna, Sood, Suresh, Nayak, & Kaushik, 2007; Abd El Baky, El Baroty, & Ibrahim, 2015; Liu, Huang, Zhang, Cai, & Cai, 2016). *Spirulina platensis* has been used as a dietary supplement in animals (Vonshak & Tomaselli, 2002). This finding is consistent with research Zeinab, Aly, Faiza and Fatma (2015), the provision of *S. platensis* on Nile Tilapia feed is very effective stimulating growth. Supplementation of *S. platensis* in fish feed significant increase the performance of growth compare to control (Jana, Saroch, & Borana, 2014; Simanjuntak, Wibowo, & Indarmawan, 2015; Abu-Elala, Galal, Abd-Elsalam, Mohey-Elsaeed, & Ragaa, 2016), potential is used as a natural food supplement to increase fish growth and no negative effect (Teimouri et al., 2013) and can be used as a raw material for feed (Simanjuntak, Soedibya, & Wibowo, 2014). However, at low temperatures, supplementation 3-7% *Spirulina* in juvenile olive flounder may not improve growth performance (Kim, Rahimnejad, Kim, Lee, & Lee, 2013).

Thus it can be said that the increase in the growth of gurami is due to supplementation of *S. platensis* in feed. Increased levels of *S. platensis* supplementation in the diet actually increases body weight and length growth of gurami. This suggests that *S. platensis* can be digested by the gurami. In accordance with the statement Beresto (2001) that *S. platensis* cell walls do not contain cellulose. The cell walls consist of mucopolimer murein so easily digested by digestive enzymes secreted by the fish. Research results show that feed supplementation *S. platensis* 7.5 and 10% did not change growth related parameters in rainbow trout compared to the other treatment. Supplementation of *S. platensis* 10 g/kg feed on *O. niloticus* improve growth performance (Ibrahim, Mohamed, & Ibrahim, 2013), use of 1% *Spirulina* in Tilapia fingerlings diets improves fish growth (Belal, Khalafalla, & El-Hais, 2012) and supplementation 10% to their feed of rainbow trout increase growth performance (Sirakov, Velichkova, & Nikolov, 2012).

Results of the proximate analysis showed that the increased level of diet supplementation *S. platensis* also increase the protein level of feed. Protein is one of the nutrients that play an important role for the growth of animals including fish. *Spirulina platensis* also contains vitamins and minerals that are very important for fish health. Accordingly, the results of this study indicate that the growth in weight and body length obtained at the highest gurami fish treated supplementation *S. platensis* 6 g/kg diet (highest level).

The result of the feed conversion ratio indicates that the higher level of supplementation *S. platensis* in the gurami diet, the smaller the obtained FCR value. The lower the FCR value indicates that the diet is getting better and more efficient and is used for growth. This is probably caused by the nutritional value of feed given higher along with the increasing level of supplementation *S. platensis*. If the nutrient value of feed given is low, it will affect the digestive system of gurami fish. Thus, in the cultivation of gurami, protein content of feed or feed composition that will be given to consider, that by adding *S. platensis* in the diet as a food supplement to the nutritional needs of the fish can be met.

Hematological Gurami Feed with Experimental Diets

Hematological examination needs to be done to determine the health status of the fish. Fish infected with the disease will lose their appetite, so that a low hematocrit value. Hemoglobin is the blood pigment that serves to



bind oxygen, low hemoglobin level indicates the binding of oxygen is low. Hematological parameters is influenced by several factors, such as: nutrition (Simanjuntak, Yuwono, & Rachmawati, 2006), gender (Acharya & Mohanty, 2012), disease (Simanjuntak, Moeljopawiro, Artama, & Wahyuono, 2011a). Hematological parameters will show the effects of nutritional supplementation *S. platensis* in the diet (Simpore, Pignatelli, & Musumeei, 2007) and can be used to determine the immune status of an animal. Simanjuntak et al. (2006) on fish Nile (*Osteochilus hasselti* C.V.) reported that supplementation *S. platensis* in the diet can improve hematological. Simanjuntak et al. (2011a) proved that the extract of *S. platensis* can improve hematological mice Balb C were challenged with tachyzoite, *Toxoplasma gondii*. Increased hematological parameters indicated an immune enhancement of gurami.

Fish blood is composed of a variety of cells, where each cell has a different function. However, white blood cells, hemoglobin and hematocrit plays an important role in the immune response of fish. Hemoglobin levels low will cause the individual to experience anemia, hematocrit value low will result in individuals susceptible to various diseases. The results showed that increasing levels of supplementation of *S. platensis* was followed by an increase in hematological gurami, supplementation treatment is higher than the control. Increased hematological also indicated an increase in the survival rates fish. In contrast to the results of research Ungsethaphand et al. (2010) found that the fish fed supplemented with different concentrations of *S. platensis* not provide a significant difference in the survival rates of fish. Feeding *S. platensis* of hybrid red tilapia (*O. niloticus* x *O. mossambicus*) also did not provide a significant difference in all treatment groups compared with the control (El-Sheekh, El-Shourbagy, Salaby, & Hosny, 2014). On the contrary, Jana et al. (2014) recorded that feeding *Spirulina* to fish improved survival rates, the highest obtained in fishes fed with 5% *Spirulina*.

Spirulina platensis contains vitamins, which one function of vitamin is in erythropoiesis. Zeinab et al. (2015) reported that administration of *S. platensis* on Nile Tilapia feed increases hemoglobin compared to control; suplementasi 10% *S. platensis* increase hematocrit value (Ibrahem et al., 2013). *Spirulina platensis* contains phycocyanin, detection routine examination revealed that the blood parameters of red blood cells, platelets and white blood cells in the normal range for animals treated phycocyanin (Liu et al., 2016). Research Moreira, Behling, Rodrigues, Costa and Souza-Soares (2013), in which malnourished rats supplemented with *S. platensis* LEB-18 for 30 days to get that increased hematological.

The results showed that different levels of supplementation of *S. platensis* on feed provides hematological differences gurami. Hematological increases with increasing levels of supplementation *S. platensis*, the highest obtained at supplementation *S. platensis* 6 g/kg of feed. This research is in accordance with Promya and Chitmant (2011) who reported that received feed fingerlings roomates 5% *A. platensis* showed an increase red and white blood cell counts and immunity stimulating capacity. *Spirulina platensis* containing C-phycocyanin and antioxidant compound that can help built the immunity (Abd El Baky et al., 2015). *Spirulina platensis* pigments contain carotenes and other antioxidants that have activity (Wang et al., 2007), the use of *Spirulina* can improve the immunity capacity of the animals consume it roomates (Abu-Elala et al., 2016; Bermejo et al., 2008).

Body Composition of Gurami Feed with Experimental Diets.

The percentage of protein in body composition gurami highest obtained at treatment P4 that supplementation of *S. platensis* 6 g/kg diet (53.14%), followed by treatment of P3 and P2 (49.92%), P1 (47.91%) and the lowest was obtained in control (47.45%). Increasing the percentage of protein feed is followed by increase the percentage in protein body composition gurami. Percentage of fat body composition gurami obtained highest in treatment P4



(29.73%) followed by P1, C, P3 and P2. The increasing percentage of protein feed (highest P4) followed by increase the percentage in protein body composition gurami, but not so with the percentage of fat. The highest percentage of dietary fat obtained at P2, whereas the lowest body fat percentage composition in P2, but the highest obtained at P4. Thus, it can be said that the increase in the percentage of body composition protein followed by increased body fat percentage composition of gurami.

Protein, when mixed with non protein and starch material, it can enrich the nutrient composition of feed (Patocka, Cervenkova, Narine, & Jelen, 2006; Pogaku, Seng, Boonbeng, & Kallu, 2007). Moreira et al. (2013) suggested that *S. platensis* strain LEB-18 very well applied in feed to overcome nutritional deficiencies because they contain a variety of minerals and other nutritional components (carbohydrates, fat and fiber, protein and ash). Changes fat and protein content in body fish associated with altered synthesis and deposition levels in the muscle (Abdel-Tawab & Ahmed, 2009; Karakatsouli, 2012).

The content of nutrients contained in *S. platensis* can improve body composition of gurami. High levels of protein, vitamins, minerals, amino acids and essential fatty acids, play an important role in improving body composition. Research Blavan, Devi, Shanti, Radhakrishnan and Poongodi (2010) on *Macrobrachium rosenbergii* PL fed nauplii Artemia enriched with *Spirulina* report that significantly improve the biochemical composition of the body. Feeding *Spirulina* 20% significantly improve body composition of larvae *Cyprinus carpio*. This can be seen in the protein composition and body fat (Abdulrahman & Ameen, 2014).

Improved feed composition was also followed by an increase in body composition of gurami. The results of this study reported that the supplementation of *S. platensis* in feeding gurami can improve body composition of gurami fish.

Biochemical Parameters Findings in Blood Serum

Serum protein composition (biochemical parameters) and level of each component (total protein, albumin and globulin) depending on the type of fish, age, and sexual maturity life cycle, diet, health and environmental factors. Fish albumin levels play a role in the metabolism of plastic and plays an important role in the functioning of colloidal osmotic pressure of the blood and the transportation of chemicals exogenous and endogenous metabolites (Baker, 2002). Globulin serves as a carrier/transport and the role of this protein is essential for living things (Preeti & Seema, 2014). Biochemical parameters of fish, especially protein can be used to determine the health status of fish and holding the key role in the immune response (De Pedro, Guijarro, Lopez-Patino, Marines-Alvares, & Delgado, 2005). Thus the determination of total protein, albumin and globulin in the serum of fish is a diagnostic tool that is fairly reflecting the health of fish, liver, metabolic status and stress conditions.

Research Mutlu, Aydin, and Kutlu (2015) reported that the total protein and albumin Nile tilapia, *O. niloticus* decreases because exposed to copper sulfate, fish that are exposed to subacute intoxication deltamethrin also showed a decrease in total protein and albumin (Abdelkhalek, Ghazy, & Abdel-Daim, 2015).

The results showed that increasing levels of supplementation *S. platensis* increases total protein, albumin and globulin compared to controls. However, in contrast to the ratio of albumin/globulin (A/G) decreased in fish fed treatment compared with controls. Biochemical parameters of gurami obtained the highest supplementation *S. platensis* 6 g/kg diet.

This is consistent with research Zeinab et al. (2015), increased levels of *S. platensis* also increase serum total protein and globulin in all groups compared to controls whereas, albumin level was not a significant difference,



indicating that *S. platensis* can improve immunity and health of *O. niloticus*. Biochemical analysis provides important information about the health of fish (De Pedro et al., 2005). Exogenous factors, such as management of fish farming (Rehulka, Minarik, & Rehulkova, 2004; Svobodova et al., 2008), stress (Cnaani, Tinman, Avidar, Ron, & Hulata, 2004) and diseases (Chen, Jin, & Wang, 2005), determining the composition of blood. Knowledge of serum albumin is necessary to know the status of the physiological and biochemical of fish blood.

Total protein, albumin and globulin can be increased by feeding high nutritious. Feeding high-nutrition will replenish the nutritional value of an animal, so the animals are not malnourished. Supplemented *S. platensis* LEB-18 for 30 days increased the biochemical profile of mice blood (Moreira et al., 2013). Results of the proximate analysis of feed showed that the higher the level of supplementation of *S. platensis*, the higher the percentage of protein and fat.

Conclusion

Different levels of supplementation of *S. platensis* in the diet can increase growth, hematological, body composition and blood biochemical parameters of gurami fish. Supplementation of *S. platensis* 6 g/kg of diet give the best results. Thus, for the successful cultivation of gurami fish, *Osphronemus gouramy*, need to add *S. platensis* in the diet in order to obtain increasing growth and immunity.

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Table 1. The proximate composition of diets.

Parameters (%)	Treatment				
	C	P1	P2	P3	P4
Water	8.61	8.62	7.15	15.67	10.71
Protein	29.26	29.32	31.22	38.79	39.22
Lipid	5.33	6.35	7.24	5.07	6.04
Fiber	6.51	6.74	7.95	7.97	7.14
Ash	9.31	9.32	9.26	8.58	8.94
BETN	49.59	48.26	44.33	39.94	38.66

Table 2. Feed Conversion Ratio (FCR)

Feed Conversion Ratio (FCR)	Treatment				
	C	P1	P2	P3	P4
Day 14	1.322	1.300	1.284	1.258	1.262
Day 28	1.275	1.145	1.088	1.062	1.055
Day 42	2.457	2.382	2.432	1.988	1.477
Day 56	4.235	2.398	2.310	1.997	1.725

Table 3. Hematological of fish fed with experimental diets.

Parameters	Treatment				
	C	P1	P2	P3	P4
RBCs (cells/mm ³)x 10 ⁶	3.21 ± 0.02	3.46 ± 0.08	3.66 ± 0.08	3.77 ± 0.05	3.94 ± 0.11
WBCs (cells/mm ³)x10 ⁵	2.12 ± 0.08	2.31 ± 0.04	2.51 ± 0.03	2.22 ± 0.04	2.86 ± 0.13
Hemoglobin (g/dl)	6-7	7-8	7-8	7-8	7.8-8.4
Hematocrit (%)	30-35	34-37	37-39	42-45	42-47

Table 4. Body composition of gurami fed with experimental diets

Parameters (%)	Treatments				
	C	P1	P2	P3	P4
Water	4.03	3.99	3.83	2.60	2.33
Protein	47.91	48.38	49.92	49.92	53.14
Lipid	27.99	28.10	23.95	26.91	29.73
Fiber	1.20	1.27	2.70	1.37	1.70
Ash	6.61	6.84	12.45	8.12	4.77
BETN	16.29	15.41	10.99	13.68	10.66

Table 5. Blood biochemical of fish fed with experimental diets.

Treatments	Parameters (g/dL)			
	Total Protein	Albumin	Globulin	A : G ratio
C	3.64 ± 0.03	1.83 ± 0.06	1.81 ± 0.07	1.01 ± 0.07
P1	4.06 ± 0.04	1.96 ± 0.02	2.10 ± 0.03	0.93 ± 0.02
P2	4.22 ± 0.08	2.07 ± 0.03	2.19 ± 0.02	0.94 ± 0.02
P3	4.41 ± 0.09	2.16 ± 0.03	2.25 ± 0.14	0.96 ± 0.07
P4	4.84 ± 0.18	2.30 ± 0.02	2.54 ± 0.15	0.91 ± 0.05

**Figure 1.** Gurami Fish (*Osphronemus gouramy*)

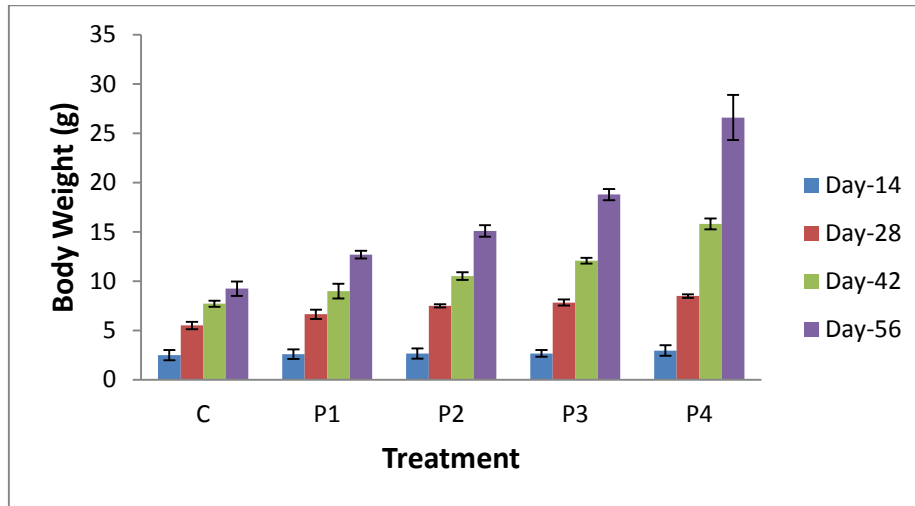


Figure 2. Average gurami body weight during the 56 days of maintenance.

Description: C : Control

P1 : Supplementation *S. platensis* 2 g/kg of diet

P2 : Supplementation *S. platensis* 3 g/kg of diet

P3 : Supplementation *S. platensis* 4 g/kg of diet

P4 : Supplementation *S. platensis* 6 g/kg of diet

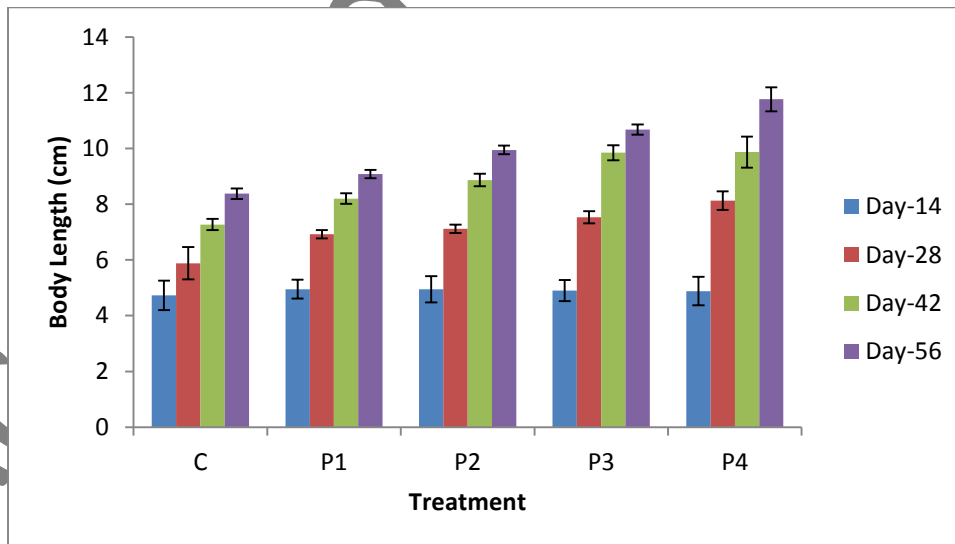


Figure 3. Average gurami body length during the 56 days of maintenance.

Description: C : Control

P1 : Supplementation *S. platensis* 2 g/kg of diet

P2 : Supplementation *S. platensis* 3 g/kg of diet

P3 : Supplementation *S. platensis* 4 g/kg of diet

P4 : Supplementation *S. platensis* 6 g/kg of diet