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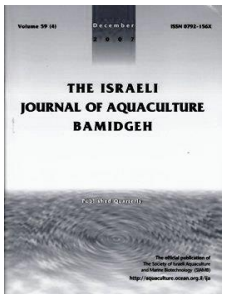

Effect of different levels of *Spirulina platensis* dietary supplementation on the growth, body color, digestion, and immunity of *Trachinotus ovatus*

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Keywords: *Spirulina platensis*; *Trachinotus ovatus;* growth performance; body color; digestion; immunity

Abstract

The effect of different levels of *Spirulina platensis* supplementation (0%, 1%, 2%, 3%, 4%, and 5%) on *Trachinotus ovatus* (initial weight of 5.8±0.04 g) growth, body color, digestion, and immunity were investigated. The experiment, comprising 3 replications of all supplementation treatments, was performed in 18 cages, 20 fish per cage. The fish were fed twice a day (08:00 and 17:00) to satiation over an experimental period of 8 weeks. There was no significant effect of *S. platensis* dietary supplementation on growth, body composition, and feed utilization in *T. ovatus* (P>0.05). Abdominal yellow color values markedly increased when feed was supplemented with 3%-5% *S. platensis*. Dietary *S. platensis* supplementation had no significant effect on intestinal amylase and lipase activity, while protease activity increased to some extent but was not statistically significant (P>0.05). As the dietary *S. platensis* levels increased, the content of alkaline phosphatase in the blood also increased, while blood glucose and malondialdehyde (MDA) content in the liver decreased (P<0.05). The MDA content was significantly lower with 3%-5% supplementation than with 0%-2% (P<0.05). *S. platensis* dietary supplementation can therefore improve body color and immunity in *T. ovatus*.

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Introduction

Trachinotus ovatus, known as golden pomfret or yellow wax pomfret, of the family Carangidae (Zhu et al., 1962; Zheng et al., 2014; Sun et al., 2014) was the test species in this trial. Its fast growth rate, simple diet, high immunity, and fresh meat source, make *T. ovatus* an ideal species for high-quality aquaculture in China. In 2014, domestic aquaculture production exceeded 150,000 tons. In natural water, *T. ovatus* is brightly colored as it feeds on a large number of carotenoid-rich macro and micro algae. In artificial culture, carotenoid content in the formula feed is lower, and other substances in the feed and aquaculture environment can affect carotenoid content. As a result, body color is duller than wild fish or fish fed fresh bait which affects commercial value.

In aquaculture production, *Spirulina platensis* has been widely used as biological feed or feed supplement due to its balanced nutrition (Yang 2011). Research shows that *S. platensis* contains many polysaccharides, carotenoids, unsaturated fatty acids, phycobiliproteins, superoxide dismutase, a variety of vitamins, and other biologically active substances that can improve the body color of fish, and promote growth and immunity to disease (Leng and Li, 2006). In this study, *S. platensis* supplementation at varying levels was added to formula feeds of *T. ovatus* to investigate the impact on growth, feed utilization, body color, activity of digestive enzymes, and immunity. Our study provided the theoretical basis and technical support for addressing the problem of body color change in aquaculture production of *T. ovatus*, without affecting growth and normal physiological function.

Materials and Methods

Experimental diets. Fish meal, peanut meal, soy protein concentrate, soybean meal, wheat middlings, squid viscera powder, bear yeast, fish oil, soy lecithin, mineral premix, vitamin premix, choline chloride, and *S. platensis* comprised the basic raw feed ingredients. Six experimental feeds (T1, T2, T3, T4, T5 and T6) were prepared by crushing all solid materials with a pulverizer and passing them through a 600 μm sieve. Each raw feed ingredient was weighed according to the feed formulation and ingredients were thoroughly mixed for 10 min. Fish oil was then added to the other ingredients, grated carefully and sieved through a 425 μm sieve. The ingredients containing fish oil were mixed for another 10 min then distilled water was added and stirred for 5 min. The mixture was then layered and pelleted into 2.0 mm and 2.5 mm diameter pieces. Pellets were kept at 16ºC for drying, packaged in a sealed bag, labeled, and stored at -20ºC.

Determination of feed nutrients including content of feed moisture, crude protein, crude fat, and crude ash were determined according to national standards, i.e., GB/T6435-1986, GB/T6432-1994, GB/T6433-1994, and GB/T6438-1992. Feed formulation and biochemical composition are shown in Table 1.

Table 1. Basal diet and nutrition levels.

Items	*T1*	*T2*	*T3*	*T4*	*T5*	*T6*
Ingredients						
Fish meal	30	30	30	30	30	30
Peanut meal	13	13	13	13	13	13
Soy protein concentrate	12	11	10	9	8	7
Soybean meal	13	13	13	13	13	13
Wheat flour	17	17	17	17	17	17
Squid viscera powder	3	3	3	3	3	3
Brewers yeast	2	2	2	2	2	2
Fish oil	6	6	6	6	6	6
lecithin	1.5	1.5	1.5	1.5	1.5	1.5
Mineral premix [1]	1	1	1	1	1	1
Vitamin premix[2]	1	1	1	1	1	1
Choline chloride	0.5	0.5	0.5	0.5	0.5	0.5
S. platensis	0.0	1.0	2.0	3.0	4.0	5.0
Total	100	100	100	100	100	100
Proximate composition (% dry matter)						
Crude protein	50.27	50.52	49.83	49.84	49.61	50.00
Crude lipid	8.35	8.18	8.97	8.60	8.61	8.21
Ash	11.14	11.03	11.17	11.12	11.17	11.34
Moisture	9.69	10.00	9.13	9.64	9.48	9.67

Mineral premix 1(mg/kg): FeSO4·6H2O 800，GSO4·7H2O 180, NaCl 100, CuSO4·5H2O 10, ZnSO4·H2O 40, CoCl2·6H2O 28, MnSO4·H2O 60, The rest is zeolite powder;
Vitamin premix 2 (mg/kg): VA 40, VB1 45, VB2 22, VB3 168, VB5 102, VB6 40, VB7 0.4, VB8 450, VB9 10, VB12 0.04, VD3 0.1, VE 80, VK3 15, the rest is microcrystalline cellulose.

Feeding management. The experiment was conducted on fish rafts in the Shenzhen test base of the South China Sea Fisheries Research Institute. Fish used in the experiment were *T. ovatus* from the same incubation batch with initial weight of 5.8±0.04 g. After 1 week of acclimation the fish were divided into six treatment groups, one group per diet. Three replicates per treatment were set-up, 18 cages in total (1.0 m×1.0 m×1.5 m), each containing 20 fish. During the 8 week experiment fish were fed twice a day (08:00 h and 17:00 h) to satiation. Formula feed of 2.0 mm diameter was used first, and later substituted by 2.5 mm pellets. Water temperature in the cages ranged between 28-31ºC, salinity ranged between 15-17‰, pH was 7.8-8.5, and dissolved oxygen was >5 mg/L. In each experimental group, water quality, food intake, and mortality, were recorded daily. Samples were collected for analysis after the experimental 8 week period.

Determining effects on growth and feed utilization. At the end of the experimental period, fish were starved for 24 hrs and samples were collected. The fish were anesthetized with 10 mg/L eugenol, the total weight of each group was measured, and the fish counted. Six fish were randomly selected from each cage, and their weight and body length were measured. After dissection, total viscera weight and liver weight were determined. Calculation formulas for the effects on growth and feed utilization were as follows:

Survival rate (%) = number of fish after the experiment/number of fish at start of experiment x 100

Weight gain (%) = (final average weight–initial average weight)/initial average weight x 100

Specific growth rate (%/d) = [(ln final average weight–ln initial average weight)/experimental days

Feed conversion ratio = feed intake/weight gain in fish

Hepatosomatic index (%) = liver weight/fish body weight x 100

Viserosomatic index (%) = viscera weight/fish body weight x 100

Condition factor (g/cm^3) = (fish body weight/fish body length^3) x 100

Protein efficiency ratio (%) = (terminal average weight–initial average weight)/protein intake x 100.

Determination of body composition. Three fish were randomly selected from each cage for body composition analysis. Moisture was determined by oven drying at 105°C until a constant weight was obtained, crude protein was measured by the Kjeldahl method (FOSS 2300), crude fat was determined by the Soxhlet extraction method (with petroleum ether as extraction agent; SoxtecTM 205, FOSS), and ash content was determined by the weight-loss method of burning in muffle furnace at 550ºC (Yatmato FO610C).

Measurement of skin color. The body color of *T. ovatus* was expressed using the color spaces L*, a*, and b* as recommend by the International Commission on Illumination, wherein L* indicates luminance value, –b* indicates a blue value, +b* indicates a yellow value, –a* indicates a green value, and +a* indicates a red value. After the experiment, the color value was measured using a color difference meter that was previously calibrated against a white board. Skin color values of the dorsal and abdominal regions of five fish from each cage were determined. The dorsal measurement comprised the region between the front edge of the dorsal fin and the lateral line, whereas the measurement region on the abdomen comprised the region between the pelvic fin and anal fin. Measurements were taken at night due to the potential effect of exogenous light on *T. ovatus* body color.

Determination of digestive enzymes indexes. Three fish from each cage were dissected after drawing blood from the caudal vein. The intestine and liver were

separated. A commercial kit (Nanjing Jiancheng Bioengineering Institute) was used to detect protease, lipase, and amylase activity in the intestine of *T. ovatus*.

Determination of immune indexes. Blood samples were centrifuged at 4℃, 3000 r/min for 10 min for the collection of upper serum. Liver homogenates were centrifuged for 10 min at 3000 r/min for the collection of the supernatant. Serum cholesterol (CHOL), triglyceride (TG), alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), serum glucose (GLU), and total protein were analyzed using a Hitachi 7180 automatic biochemical analyzer (882, Ichige, Hitachinaka-shi, Ibaraki-ken,Japan). Liver malondialdehyde (MDA) content was measured using a kit (Nanjing Jiancheng Bioengineering Institute, China).

Statistical analysis. Mean±SD was calculated using Microsoft Software Excel 2007 (Seattle, USA). Correlation tests were performed using SPSS 17.0 (SPSS Inc., Chicago, IL, USA). Differences among experimental groups were analyzed by one-way analysis of variance. If the difference between groups was significant (P<0.05), Duncan's multiple comparison analysis was used.

Results

Effect of different dietary S. platensis levels on growth performance and feed utilization in T. ovatus. Growth indexes and feed utilization in *T. ovatus* at 8 weeks, after administering experimental feeds with different levels of supplementation are shown in Table 2.

Table 2. Effects of dietary *S. platensis* levels on growth performance and feed utilization in *T. ovatus*

Treatments	T1	T2	T3	T4	T5	T6
Survival (%)	85.00±10.00	83.33±17.56	95.00±8.66	85.00±8.66	80.00±18.03	78.33±12.58
WG(%)	859.82±15.84	679.82±19.83	705.81±14.18	678.99±18.56	751.22±11.45	756.72±9.94
SGR (%/d)	3.77±0.11	3.42±0.09	3.47±0.19	3.40±0.31	3.55±0.28	3.58±0.06
FCR	1.13±0.09	1.21±0.02	1.32±0.10	1.24±0.27	1.19±0.28	1.34±0.05
HSI (%)	0.80±0.18	0.67±0.09	0.71±0.10	0.69±0.15	0.69±0.12	0.74±0.06
VSI (%)	5.25±0.36 ^{a}	4.72±0.23^{b}	5.09±0.50 ^{ab}	4.83±0.49^{b}	4.81±0.36^{b}	5.04±0.16 ^{ab}
CF (%)	3.63±0.21 ^{a}	3.64±0.22 ^{a}	3.53±0.23 ^{ab}	3.42±0.17 ^{b}	3.59±0.21 ^{ab}	3.63±0.17 ^{a}
PER (%)	1.96±0.16	1.82±0.03	1.68±0.12	1.84±0.38	1.95±0.52	1.65±0.07

Values are means ± SD of three replications. Means in the same row with different superscripts are significantly different (P<0.05). WG, weight gain; SGR, specific growth rate; FCR, feed conversion ratio; HSI, hepatosomatic index; VSI, viserosomatic index; CF, condition factor; PER, protein efficiency ratio

The survival rate among groups varied, with the highest in the T3 treatment group and the lowest in T6, but there was no statistical difference among the groups (P>0.05). Weight gain, feed conversion ratio, and hepatosomatic index in the T1 control group, without *S. platensis* supplementation, were the highest, lowest, and highest, respectively, among all treatment groups; however, there were no significant differences between groups (P>0.05). Differences in viserosomatic index were not significant among T2-T6 groups which received *S. platensis* supplementation (P>0.05), and were lower than in the T1 control group. Condition factor was the highest in T2 and lowest in T4. Protein efficiency was not significantly different between groups (P>0.05).

Effects of dietary S. platensis levels on T. ovatus body composition. There was no significant difference in body composition between groups (P>0.05) in all treatments (Table 3). Compared with other groups, T4 had the highest crude protein content and the lowest crude fat content, but these differences were not significant (P>0.05).

Table 3. Effects of dietary *S. platensis* levels on body composition in *T. ovatus*

Treatments	*T1*	*T2*	*T3*	*T4*	*T5*	*T6*
Protein	58.80±0.47	56.18±2.60	58.24±0.54	59.29±0.86	57.89±2.18	58.12±1.64
Lipid	27.77±1.54	27.75±0.91	27.32±2.89	24.93±2.67	28.62±1.84	26.82±1.94
Ash	12.59±0.56	13.16±1.12	13.29±0.21	13.46±0.16	12.76±0.59	13.28±1.21
Moisture	67.28±1.04	67.00±0.60	68.12±1.26	68.46±0.82	67.14±1.08	68.02±1.44

Values are means ± SD of three replications. There was no significant difference between any of the treatments.

Effects of dietary S. platensis levels T. ovatus body color. With the increase of dietary *S. platensis* levels, skin color changed markedly; the skin color of fish in groups T3-T6 was a substantially brighter golden yellow than in the T1 and T2 groups; the greatest color change occurred in T4 group (Figure 1).

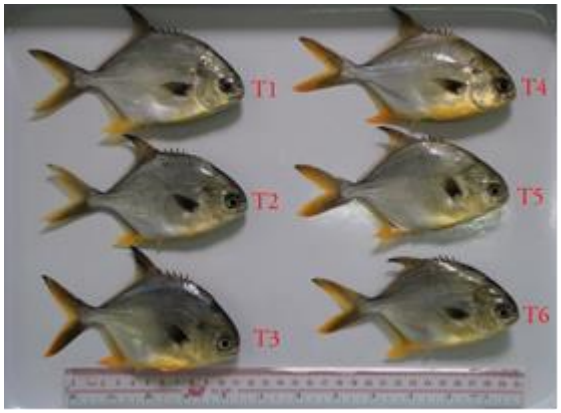

Fig.1 Effects of dietary *S. platensis* levels on the body color in *T. ovatus*

There were no significant changes in dorsal skin color value (L*, a*, and b* values) among experimental groups (P>0.05), the same applied to abdominal skin luminance (L*) and red/green values (a*) (P>0.05) (Table 4).

Table 4. Effects of dietary *S. platensis* levels on skin color values of L*. a*. b* in *T. ovatus*

Treatments		*T1*	*T2*	*T3*	*T4*	*T5*	*T6*
Dorsum	L*	74.63±3.23	71.47±4.69	72.05±4.53	74.44±3.46	75.52±4.72	72.37±4.70
	a*	-1.29±0.33	-0.54±0.69	-0.61±1.00	-0.81±0.74	-1.17±0.74	-0.61±0.75
	b*	7.71±3.05	8.42±3.01	8.54±2.92	8.65±2.82	8.54±2.48	9.86±1.48
Belly	L*	94.76±2.35	92.68±5.65	93.76±1.40	91.70±2.72	93.04±2.87	90.63±3.28
	a*	-2.08±0.58	-2.11±0.96	-2.44±0.96	-1.96±1.00	-2.55±0.73	-2.66±0.89
	b*	11.31±5.16 [a]	10.83±9.16 [a]	20.39±11.30 [b]	28.10±11.93 [b]	25.78±8.10 [b]	27.15±13.87 [b]

Values are means ± SD of three replications. Means in the same row with different superscripts are significantly different (P<0.05).

Compared with the control group T1, there was no significant difference in the abdominal yellow value (b*) in T2 (P>0.05) and b* was significantly higher in T3–T5 groups (P<0.05); it was the highest in T4 group, and then stabilized. These values were consistent with the observations (Figure 2).

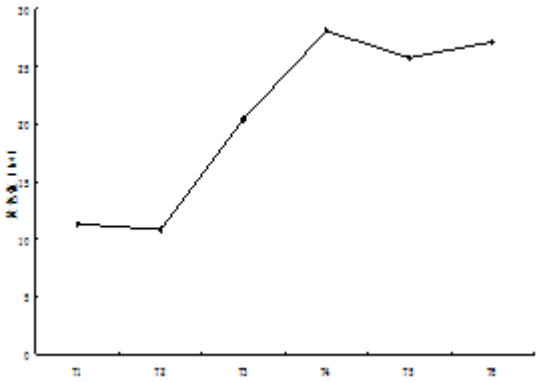

Fig.2 The relationship between dietary *S. platensis* levels and abdominal skin yellow values (b*) in *T. ovatus.*

Effects of dietary S. platensis levels on digestive enzyme activity in T. ovatus. There were no significant differences in intestinal amylase and lipase activity among the different treatment groups (P>0.05). Protease activity in T5 was highest, and was significantly higher than in groups T1-T4 (P<0.05), but was not significantly different (P>0.05) from T6 (Table 5).

Table 5. Effects of dietary *S. platensis* levels on the activity of digestive enzymes in *T. ovatus*

Treatments	*T1*	*T2*	*T3*	*T4*	*T5*	*T6*
Amylase (U/mg)	0.28±0.04	0.29±0.07	0.26±0.01	0.28±0.01	0.23±0.03	0.28±0.08
Lipase (U/g)	4.10±0.46	4.45±1.30	3.80±0.65	5.44±1.68	4.53±1.31	4.99±1.27
Protease (U/mg)	0.58±0.01^{a}	0.61±0.09^{a}	0.58±0.08^{a}	0.62±0.08^{a}	0.77±0.15^{b}	0.66±0.02^{ab}

Values are means ± SD of three replications. Means in the same row with different superscripts are significantly different (P<0.05).

Effects of dietary S. platensis levels on immunity indexes in T. ovatus. For the serum biochemical indexes, TG, ALT, AST, total protein content did not significantly differ among treatment groups. Group T6 had significantly higher CHOL than the other groups (P<0.05), while the differences between groups T1-T5 were not significant (P>0.05). When dietary *S. platensis* levels increased, ALP content in the blood became elevated, whereas GLU content decreased. With regard to the liver immunity index, MDA showed a decreasing trend and was considerably lower in groups T4-T6 than in T3 (Table 6).

Table 6. Effects of dietary *S. platensis* levels on the activity of digestive enzymes in *T. ovatus*

Treatment	*T1*	*T2*	*T3*	*T4*	*T5*	*T6*
CHOL	5.10±0.26^{a}	4.81±0.25^{a}	4.84±0.45^{a}	4.97±0.48^{a}	4.48±0.18^{a}	5.71±0.29^{b}
TG	1.21±0.22	1.15±0.06	1.55±0.31	1.48±0.18	1.45±0.17	1.39±0.21
ALT	1.67±0.58	2.33±0.58	1.67±1.15	1.00±1.00	2.00±2.00	3.33±2.52
AST	27.67±9.29	23.33±3.21	24.00±12.29	33.00±6.56	27.33±10.02	32.00±12.12
ALP	47.67±3.21^{a}	45.00±4.36^{a}	44.67±1.53^{a}	49.33±3.51^{ab}	50.33±9.50^{ab}	58.67±6.03^{b}
GLU	8.95±2.53^{ab}	8.82±2.65^{ab}	10.07±2.58^{b}	4.91±2.29^{ac}	3.15±0.36^{c}	5.27±1.72^{ac}
TP	32.73±0.76	30.03±1.17	31.43±3.77	31.73±2.57	32.23±3.76	34.00±0.10
MDA	7.07±0.62^{ab}	9.47±5.88^{b}	9.70±4.27^{b}	2.30±0.55^{a}	2.01±0.28^{a}	2.18±0.15^{a}

Values are means ± SD of three replications. Means in the same row with different superscripts are significantly different (P<0.05).

Discussion

Effects of S. platensis on growth, feed utilization, and body composition in T. ovatus. In recent years, research on aquatic diets supplemented with *S. platensis* has focused mainly on effects on growth indexes. Feed supplemented with an appropriate amount of *S. platensis* can significantly improve fish growth performance and feed utilization. This has been reported in Pengze crucian carp, koi, Jian carp, allogynogenetic crucian carp, and other cyprinids (James et al. 2009; Liu et al., 2004; Yang 2011; Luo *2006*; Dong et al., 2008). *S. platensis* has been applied predominately to fry culture in shellfish and crustaceans as well as an antibacterial agent (Bhuvaneswari et al, 2009). The growth performance and survival rate of fry significantly improved and fry immunity was promoted using feed supplemented with *S. platensis* (Zhong et al., 2011). In the current study, *T. ovatus* was fed on feed supplemented with 1%-5% *S. platensis*. There were no significant effects of *S. platensis* supplementation on growth performance, body composition, and feed utilization. The reason for this might be due to the fact that *S. platensis* supplementation level was low and the culture period was short, therefore a promotional effect on growth performance cannot be positively determined. Feed supplemented with *S. platensis* did, however, reduce the hepatosomatic and viserosomatic indexes as well as the condition factor, increase the proportion of fish muscle, and improve its commercial value.

Effects of S. platensis on body color of T. ovatus. Body color is not only an important characteristic for fish classification, but also a measurement of health status; however, it does not directly affect the value of commercial fish. Fish body color is mainly determined by carotenoids that are not synthesized by fish; they can only be obtained through absorption, precipitation, and transformation from food (Goodwin 1985). *S. platensis* is a rich source of carotenoids (0.2%–0.4%), mainly comprising lutein and β-carotene. These pigments have a relatively high rate of absorption and conversion in aquatic animals, and can be metabolized into astaxanthin and cantharidin zeaxanthin that brighten the color of aquatic animals, as well as improve their ornamental and commercial value (Liu 2002).

In aquaculture, *S. platensis* was initially used as body color toner for koi, golden fish, and other ornamental fish, and had an obvious hyperchromic effect (James et al., 2009; He and Zhang 1999; Xu 1999). Previous studies have shown that feed supplemented with *S. platensis* can increase body color in the blood parrot cichlid, and the color gradually deepened with an increase in *S. platensis* supplementation or feeding time

(Zhang et al., 2009). Shape and body color of eels have also undergone significant changes with 1%-3% *S. platensis* supplementation (Qiu and Hong 1992). When feed was supplemented with 3%-6% *S. platensis*, the body color of *Plecoglossus altivelis* considerably improved after 10 weeks (Wei D et al., 2009). The body color of *Pseudocaranx dentex* was also obviously improved after feed supplementation with 5%–10% *S. platensis* powder (Okada et al., 1991). *Trachinotus ovatus*, also known as the golden pomfret, has a vivid color and a golden abdomen in the wild; however, when cultured in an artificial environment, its body color turned white and its abdomen became dull due to the lack of algae and relevant pigments in the feed. In the present study, when the feed was supplemented with 1%-5% *S. platensis*, the body color was obviously improved and the abdominal yellow value (b*) markedly increased after feeding for 8 weeks. During the culturing of *T. ovatus*, feed supplemented with nutrient-rich *S. platensis* had significant hyperchromic effects.

Effects of S. platensis on digestion and immunity in T. ovatus. S. platensis can influence growth promotion and appetite in a variety of aquatic animals, and feed supplemented with *S. platensis* can increase digestive enzyme activity in aquatic animals. Protease activity in the intestine, liver, and pancreas were substantially elevated in allogynogenetic crucian carp (*Carassius auratus gibelio*) when feed was supplemented with *S. platensis*, and since its activity was much higher than amylase, it could be concluded that *S. platensis* mainly affects protease activity (Huang et al., 2009; Dong et al., 2008). Dietary *S. platensis* supplementation in this study had little impact on the activity of intestinal amylase and lipase in *T. ovatus*, but increased intestinal protease activity to some extent, similar to the results from the studies on allogynogenetic crucian carp (*Carassius auratus gibelio*). There may be two reasons for these *S. platensis* effects on endogenous enzyme activity. First, *S. platensis* contains 60%-70% protein; once the dietary protein increases, protease would be stimulated and correspondingly increase. Second, *S. platensis* contains an abundance of lactic acid bacteria and other beneficial bacteria that can improve intestinal microenvironment, stimulate the secretion of digestive enzyme, and improve intestinal digestive enzyme activity.

Health status, physiological condition, metabolism, and immunity can be reflected in fish blood biochemical indexes (Zhou et al., 2001). Among the serum biochemical indexes in each treatment group of *T. ovatus*, TG, ALT, AST, and TP did not significantly differ, indicating that no adverse effect on fish health status and metabolic ability occurred as a result of *S. platensis* supplementation. Blood GLU, the primary energy supply substance, is relatively constant under normal conditions, playing an important role in maintaining the normal physiological function of the body. The GLU concentration of fish can be influenced by hunger, density, nutritional factors, and health status, among other variables. When a fish is in a harmful environment, basal metabolism increases, and a large amount of glycogen is decomposed and enters the blood. Since a higher metabolism requires a larger amount of GLU, levels can increase quickly over a short period (Wang 2007). GLU concentrations in silver carp (*Hypophthalmichthys molitrix*) with septicemia and in tilapia (*Oreochromis niloticus*) infected with *Edwardsiella tarda* have been shown to be significantly elevated (Mi et al., 1993; Benli et al., 2004). In the current experiment, with an increase of dietary *S. platensis* supplementation, GLU level in blood of *T. ovatus* showed a tendency to be lowered; however, it was still within the normal range (2.78-12.72 mmol/L) indicating normal metabolic level and immunity in each group of *T. ovatus*. ALP, a non-specific phosphate hydrolase, is mainly involved in fish metabolic regulation, directly participates in transfer and metabolic processes of phosphate groups, and plays an important role in non-specific immune response in organisms (Huang et al., 2005). In our experiment, ALP activity in the blood of *T. ovatus* showed an increasing tendency, indicating that feed with high *S. platensis* supplementation can enhance fish immune function; however, this requires further investigation.

Lipid peroxidation is a complex and disruptive chemical reaction process. It can not only reduce biological membrane mobility and cell activity in animals, but can also produce a large amount of free radicals, MDA, and other harmful substances (Sargent et al., 1999).

Polyunsaturated fatty acids (PUFA) can improve the antioxidant capacity of the liver and pancreas in fish; however, high doses of PUFA may increase the MDA levels in the liver and pancreas, thereby affecting fish health (Pan et al., 2014). *S. platensis*, which is rich in PUFA and other substances important in biological activity, can increase antioxidant enzyme activity, capture free radicals, and consequently enhance antioxidant capacity (Wang et al., 2009). MDA is the final product of the lipid peroxidation reaction and its content reflects the extent of lipid peroxidation. In this experiment, a decreasing trend of MDA content was observed in the liver of *T. ovatus*; MDA in the fish group with 3%-5% *S. platensis* supplementation was substantially lower than in the group with 0%-2%. This indicates that the accumulation of lipid peroxides could be effectively reduced by *S. platensis*, resulting in a reduced degree of free radical injury on liver cells, similar to the results of *Cranoglanis bouderius multiradiatus* (Lv et al., 2015). Feed supplemented with *S. platensis* can considerably increase the antioxidant capacity of *T. ovatus*.

Conclusion

Feed supplemented with 3%–5% *S. platensis* can significantly improve body color and immunity in *T. ovatus*.

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