

## Effects of Replacing Soybean Oil with Palm Oil on Growth Performance, Muscle Composition, and Serum Biochemical Indices of Juvenile GIFT Tilapia: Postprint

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### Abstract

This experiment aimed to investigate the effects of replacing soybean oil with palm oil on growth performance, muscle composition, and serum biochemical indices of juvenile GIFT tilapia. Six hundred juvenile GIFT tilapia with an initial body weight of 8.80 g were selected and randomly divided into 5 groups with 3 replicates per group and 40 fish per replicate. The five groups of experimental fish were fed five isonitrogenous (31.60%) and isolipidic (9.30%) diets in which palm oil replaced 0 (control), 25%, 50%, 75%, and 100% of soybean oil in the basal diet, designated as G0, G25, G50, G75, and G100, respectively. The experimental period lasted 56 days. The results showed: 1) Replacement of soybean oil with palm oil at different proportions had no significant effects on final body weight, weight gain rate, specific growth rate, feed conversion ratio, and survival rate of juvenile tilapia ( $P > 0.05$ ). 2) Compared with the G0 group, there were no significant changes in condition factor, viscera-somatic index, hepatosomatic index, or muscle moisture, crude protein, crude lipid, and crude ash contents in the palm oil groups (G25, G50, G75, G100) ( $P > 0.05$ ); muscle crude lipid content in the G50 group was significantly higher than that in the G100 group ( $P < 0.05$ ). 3) Replacement of soybean oil with palm oil at different proportions had no significant effect on serum cholesterol, triglyceride, low-density lipoprotein cholesterol, and total protein contents in juvenile tilapia ( $P > 0.05$ ). Compared with the G0 group, serum high-density lipoprotein cholesterol content in the G50 and G100 groups, serum alanine aminotransferase activity in the G25 and G50 groups, and serum aspartate aminotransferase activity in the G50 and G75 groups were significantly decreased ( $P < 0.05$ ). It was concluded that, based on growth performance as the evaluation criterion, soybean oil in juvenile GIFT tilapia diets could be completely replaced by palm

oil; when the replacement proportion of palm oil for soybean oil exceeded 50%, it significantly affected muscle crude lipid content and some serum biochemical indices of juvenile GIFT tilapia.

## Full Text

### Effects of Soybean Oil Replacement by Palm Oil on Growth Performance, Muscle Composition and Serum Biochemical Indexes of Juvenile Genetically Improved Farmed Tilapia (*Oreochromis niloticus*)

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#### Abstract

This experiment was conducted to investigate the effects of replacing soybean oil with palm oil on growth performance, muscle composition, and serum biochemical indexes in juvenile genetically improved farmed tilapia (GIFT). Six hundred juvenile GIFT with an initial body weight of 8.80 g were randomly divided into five groups with three replicates per group and 40 fish per replicate. The five groups were fed five isonitrogenous (31.60% crude protein) and isolipidic (9.30% crude fat) diets in which palm oil replaced 0% (control), 25%, 50%, 75%, and 100% of the soybean oil in the basal diet, designated as G0, G25, G50, G75, and G100, respectively. The experiment lasted for 56 days. The results showed: (1) No significant differences were observed in final body weight, weight gain rate, specific growth rate, feed conversion ratio, or survival rate among all groups ( $P > 0.05$ ). (2) Compared with the G0 group, no significant changes were found in condition factor, viscerosomatic index, hepatosomatic index, or muscle moisture, crude protein, ether extract, and ash contents in the palm oil groups (G25, G50, G75, and G100) ( $P > 0.05$ ), though the muscle ether extract content in the G50 group was significantly higher than in the G100 group ( $P < 0.05$ ). (3) Replacement of soybean oil with palm oil at different proportions had no significant effect on serum cholesterol, triglyceride, low-density lipoprotein cholesterol, or total protein contents ( $P > 0.05$ ). However, serum high-density lipoprotein cholesterol content in the G50 and G100 groups, serum alanine aminotransferase activity in the G25 and G50 groups, and serum aspartate aminotransferase activity in the G50 and G75 groups were significantly reduced compared with the G0 group ( $P < 0.05$ ). These results indicate that, based on growth performance as the evaluation criterion, soybean oil can be completely replaced by palm oil in juvenile GIFT diets, though replacement proportions exceeding 50% significantly affect muscle ether extract content and some serum biochemical indexes.

**Keywords:** GIFT (*Oreochromis niloticus*); soybean oil; palm oil; growth performance; muscle composition; serum biochemical indexes

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Soybean oil is a high-quality lipid source commonly used in aquatic commercial feeds, particularly in freshwater fish diets. With the rapid development of aquaculture and the feed industry, soybean oil consumption has increased dramatically, leading to substantial price increases that have gradually limited its use in aquatic feeds. Identifying more economical and high-quality lipid alternatives to soybean oil has become a key research focus in the aquatic feed industry. Palm oil represents an important component of the global oils and fats market, accounting for over 30% of total world production. Since the 1970s, palm oil production has grown faster than any other vegetable oil, with global output exceeding 62 million tons in 2012. Palm oil primarily contains palmitic acid, oleic acid, and linoleic acid, with approximately 50% saturated fatty acids, 40% monounsaturated fatty acids, and 10% polyunsaturated fatty acids. The monounsaturated fatty acids are mainly oleic acid, while polyunsaturated fatty acids are primarily linoleic acid. Rich in vitamin E, -tocotrienol, -carotene, -carotene, and lutein, palm oil serves as a suitable alternative to fish oil and soybean oil.

Previous studies have demonstrated that replacing a certain proportion of fish oil with palm oil does not significantly affect growth performance or feed utilization in spotted sea bass (*Lateolabrax maculatus*), African catfish (*Clarias gariepinus*), Atlantic salmon (*Salmo salar* L.), rainbow trout (*Oncorhynchus mykiss*), or European sea bass (*Dicentrarchus labrax* L.). Cheng Minjie reported that when dietary lipid content was 10% in half-smooth tongue sole (*Cynoglossus semilaevis*) feeds, replacing 32–60% of fish oil with palm oil benefited growth and health while reducing feed conversion ratio and improving muscle nutritional composition.

Tilapia (*Oreochromis niloticus*) is a subtropical fish species, and genetically improved farmed tilapia (GIFT) represents an excellent strain developed through selective breeding in recent years. Characterized by rapid growth, plump and tender flesh, and high fillet yield, GIFT has become one of the main cultured species in South China and an important export product with significant market value. Although numerous studies have investigated nutritional requirements and immune health in tilapia, research on soybean oil alternatives in tilapia feeds remains unreported. Therefore, this study examined the effects of different palm oil replacement proportions on growth performance, muscle nutritional composition, and serum biochemical indexes in juvenile GIFT to provide a theoretical basis for palm oil application in tilapia feeds.

## 1. Materials and Methods

**1.1 Palm Oil** The experimental palm oil was purchased from Yihai (Guangzhou) Cereals & Oils Industry Co., Ltd., originating from Indonesia

with a melting point of 24°C. Fatty acid composition was analyzed using gas chromatography-mass spectrometry (Agilent 6890N-5975, USA) with an Agilent DB-23 column (30 mm × 0.25 mm × 0.25 m) under chromatographic and mass spectrometric conditions following the method of Liu Suihua et al. The contents of C14:0, C16:0, C18:0, C18:1, C18:2, C18:3, C20:0, and C20:1 in the experimental palm oil were 0.94%, 38.45%, 3.78%, 44.34%, 11.22%, 0.22%, 0.35%, and 0.19%, respectively, expressed as percentages of total fatty acids.

**1.2 Experimental Diets** A basal diet (G0, control) was formulated using soybean meal, rapeseed meal, and corn gluten meal as primary protein sources, soybean oil as the main lipid source, and strong wheat flour as the primary carbohydrate source. Four additional diets were prepared by replacing 25% (G25), 50% (G50), 75% (G75), and 100% (G100) of the soybean oil in the basal diet with palm oil on an equal-weight basis. All five experimental diets were isonitrogenous (31.60% crude protein) and isolipidic (9.30% crude fat). Dietary composition and nutrient levels are presented in Table 1, and fatty acid composition is shown in Table 2. After thorough mixing, the experimental diets were processed into 1.50 mm diameter extruded pellets using a BUHLER-125 twin-screw extruder (Switzerland) and stored at room temperature.

**1.3 Experimental Fish and Culture Management** The experiment was conducted in an indoor recirculating aquaculture system at the Guangzhou National Agricultural Science and Technology Park of Guangdong Academy of Agricultural Sciences. Juvenile GIFT were purchased from Foshan Nanhai Tongwei Aquatic Technology Co., Ltd. and temporarily reared in outdoor net cages (3.0 m × 3.0 m × 3.0 m) at the Guangzhou National Agricultural Science and Technology Park for two weeks of acclimation, during which they were fed to satiation twice daily (08:30 and 17:00) with Guangdong Tongwei 151 high-grade extruded fish feed. The experimental system consisted of 24 cylindrical fiberglass tanks (80 cm diameter × 70 cm height) with a water volume of approximately 300 L each. At the start of the experiment, 600 healthy juvenile GIFT with uniform size, averaging 8.80 g, were selected and stocked into 15 culture tanks at 40 fish per tank. The 15 tanks were randomly assigned to five groups with three replicates each, corresponding to the five dietary treatments (G0, G25, G50, G75, and G100). Fish were hand-fed to apparent satiation twice daily (08:30 and 17:00) at 2-3% of body weight, with feeding amounts adjusted based on consumption and growth. Daily records were maintained for feed intake, water temperature, and mortality. Throughout the experiment, continuous aeration maintained dissolved oxygen concentration above 8.0 mg/L under natural light conditions. Mean water temperature was 28.8°C, ammonia nitrogen concentration was below 0.06 mg/L, nitrite concentration was below 0.05 mg/L, and pH ranged from 7.6 to 8.0. The experimental period lasted 56 days.

**1.4 Sample Collection and Analysis** At the start of the experiment, 20 fish with body weights similar to the initial experimental fish were sampled and stored at  $-20^{\circ}\text{C}$  for subsequent analysis. At the end of the feeding trial, fish were fasted for 24 h before final sampling. Fish from each tank were counted and weighed collectively. Five fish per tank were randomly selected for blood collection via caudal vein puncture; blood samples were pooled, centrifuged at  $4^{\circ}\text{C}$  and 4,000 r/min for 10 min to obtain serum, which was stored at  $-80^{\circ}\text{C}$  for serum biochemical analysis. Another five fish per tank were randomly selected for morphometric measurements. Additionally, five fish per tank were randomly sampled, and dorsal muscle tissue from the same side was collected, pooled, and stored at  $-20^{\circ}\text{C}$  for routine muscle composition analysis.

Moisture content in feed and muscle samples was determined by oven drying at  $105^{\circ}\text{C}$  (GB/T 6435-1986). Crude protein content was measured using the Kjeldahl method (GB/T 6432-1994). Ether extract content was determined by ether extraction (GB/T 6432-1994). Ash content was measured by incineration at  $550^{\circ}\text{C}$  (GB/T 6438-1992). Calcium content was determined by ethylenediaminetetraacetic acid (EDTA) titration (GB/T 6436-2002). Total phosphorus content was measured by molybdenum yellow colorimetry (GB/T 6437-2002). Dietary crude fiber content was determined by acid-alkali digestion and filtration (GB/T 5009.10-2002).

Serum cholesterol (CHOL), triglyceride (TG), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and total protein (TP) contents, as well as alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities, were measured using an automated Roche biochemical analyzer.

**1.5 Index Calculations** Weight gain rate (WGR, %) =  $100 \times (\text{final body weight} + \text{weight of dead fish} - \text{initial body weight}) / \text{initial body weight}$   
Specific growth rate (SGR, %/d) =  $100 \times (\ln \text{ final body weight} - \ln \text{ initial body weight}) / \text{feeding days}$   
Feed intake (FI, g/fish) =  $\text{total feed intake} / [(\text{initial fish number} + \text{final fish number}) / 2]$   
Feed conversion ratio (FCR) =  $\text{total feed intake} / (\text{final body weight} + \text{weight of dead fish} - \text{initial body weight})$   
Survival rate (SR, %) =  $100 \times \text{final fish number} / \text{initial fish number}$   
Condition factor (CF,  $\text{g}/\text{cm}^3$ ) =  $100 \times \text{body weight} / \text{body length}^3$   
Viscerosomatic index (VSI, %) =  $100 \times \text{viscera weight} / \text{body weight}$   
Hepatosomatic index (HSI, %) =  $100 \times \text{liver weight} / \text{body weight}$

**1.6 Statistical Analysis** Experimental data are expressed as mean  $\pm$  standard deviation. Statistical analysis was performed using SPSS 19.0 software. Homogeneity of variance was first tested; if the assumption was met, one-way ANOVA was conducted, followed by Tukey's multiple comparison test for significant differences. If variance homogeneity was not satisfied, Dunnett's T3

test was used for multiple comparisons. Significance was set at  $P < 0.05$ .

## 2. Results

**2.1 Effects of Soybean Oil Replacement by Palm Oil on Growth Performance of Juvenile GIFT** As shown in Table 3, replacement of soybean oil with palm oil at different proportions had no significant effect on final body weight, WGR, SGR, FI, FCR, or SR of juvenile GIFT ( $P > 0.05$ ). Survival rates in all groups exceeded 96.60%, with the G0 and G75 groups showing the highest final body weight, WGR, and SGR, while the G25 group exhibited the lowest FCR.

**2.2 Effects of Soybean Oil Replacement by Palm Oil on Physique Indexes of Juvenile GIFT** Table 4 shows that replacement of soybean oil with palm oil at different proportions had no significant effect on CF, VSI, or HSI of juvenile GIFT ( $P > 0.05$ ).

**2.3 Effects of Soybean Oil Replacement by Palm Oil on Muscle Nutritional Composition of Juvenile GIFT** As presented in Table 5, no significant differences were observed in muscle moisture, crude protein, or ash contents among all groups ( $P > 0.05$ ). Although muscle ether extract content in the replacement groups did not differ significantly from the G0 group ( $P > 0.05$ ), the G50 group exhibited significantly higher muscle ether extract content than the G100 group ( $P < 0.05$ ).

**2.3 Effects of Soybean Oil Replacement by Palm Oil on Serum Biochemical Indexes of Juvenile GIFT** Table 6 indicates that replacement of soybean oil with palm oil at different proportions had no significant effect on serum CHOL, TG, LDL-C, or TP contents ( $P > 0.05$ ). Compared with the G0 group, serum HDL-C content in the G50 and G100 groups was significantly reduced ( $P < 0.05$ ); additionally, HDL-C content in the G75 group was significantly higher than in the G50 and G100 groups ( $P < 0.05$ ). Serum ALT activity in the G25 and G50 groups and AST activity in the G50 and G75 groups were significantly lower than in the G0 group ( $P < 0.05$ ), while other replacement groups showed no significant differences ( $P > 0.05$ ).

## 3. Discussion

**3.1 Effects of Soybean Oil Replacement by Palm Oil on Growth Performance of Juvenile GIFT** The present results demonstrate that replacing soybean oil with palm oil at different proportions had no significant effect on WGR, FCR, or SR of juvenile GIFT, with the G0 and G75 groups showing the highest WGR and SGR, and the G25 group exhibiting the lowest FCR. These findings are consistent with previous studies on palm oil replacement of fish oil in catfish, Atlantic salmon, and rainbow trout. Song Yizhen et al. reported that feeding juvenile GIFT (initial weight 11.19 g) diets containing 4.00% chicken fat,

fish oil, palm oil, peanut oil, or lecithin oil for 60 days resulted in significantly lower SGR in the palm oil group compared with chicken fat, peanut oil, and lecithin oil groups, but no significant difference compared with the fish oil group. Cheng Minjie found that when dietary lipid content was 10.00% in half-smooth tongue sole feeds, replacing 60% of fish oil with palm oil significantly improved WGR compared with the all-fish-oil group. Bahurmiz et al. completely replaced 8.00% fish oil in diets with crude palm oil, palm olein, or refined palm oil and observed no significant effects on final body weight, SGR, feed efficiency, SR, physique indexes, or hematocrit in red hybrid tilapia (*Oreochromis sp.*) raised from stocking to marketable size over 20 weeks. Lim et al. reported that dietary crude or refined palm oil at 8.00% in African catfish feeds significantly improved growth performance, protein retention, and muscle vitamin E concentration.

Palm oil is rich in natural antioxidants such as vitamin E, and dietary vitamin E content increases progressively with higher palm oil inclusion levels. In this experiment, WGR, FCR, and SR in all replacement groups did not differ significantly from the control (G0), while the G75 group showed superior FBW, WGR, and SGR compared with other replacement groups, possibly due to increasing dietary vitamin E content with higher replacement proportions. Li Zhihua et al. fed juvenile tilapia (initial weight 3.25 g) diets supplemented with 0, 25, 250, or 2,500 mg/kg vitamin E for 56 days and found that the 250 mg/kg group exhibited significantly higher WGR and SGR than other groups, suggesting that appropriate vitamin E levels enhance tilapia growth and antioxidant capacity, while excessive vitamin E inhibits growth. Gao et al. reported that increasing palm oil replacement of fish oil in spotted sea bass diets progressively increased dietary vitamin E content, with 60% replacement showing no significant effects on final body weight, WGR, SGR, or FCR. Ng et al. found that replacing 0, 25%, 50%, 75%, or 100% of fish oil with palm fatty acid distillate in African catfish feeds containing 10% crude fat increased dietary vitamin E from 78.68 to 304.81 g/g, with 25% replacement significantly improving final body weight and WGR compared with the all-fish-oil group.

Aquatic animal growth performance is primarily influenced by dietary fatty acid composition, with different fish species exhibiting varying digestive utilization of the same oil. Yong Wenyue reported that tilapia have higher requirements for linoleic acid than other fatty acids and that vegetable oil supplementation is more beneficial for tilapia growth. In this experiment, both soybean oil and palm oil are vegetable oils, and increasing palm oil replacement proportion gradually reduced dietary C18:3 and C18:2 contents (C18:3 decreased from 4.44% to 2.31%, C18:2 from 45.96% to 35.46%). The comparable growth performance across all replacement groups indicates that dietary C18:2 and C18:3 contents after complete soybean oil replacement by palm oil still meet the growth requirements of tilapia.

### **3.2 Effects of Soybean Oil Replacement by Palm Oil on Physique Indexes and Muscle Composition of Juvenile GIFT** The present re-

sults show that replacing soybean oil with palm oil at different proportions had no significant effect on CF, VSI, or HSI of juvenile GIFT, indicating that such replacement does not significantly affect morphological characteristics. No significant differences in muscle moisture, crude protein, or ash contents were observed between replacement groups and the control group, suggesting that palm oil replacement does not affect protein deposition in muscle. However, the G100 group exhibited significantly lower muscle ether extract content than the G50 group, possibly because increasing palm oil replacement proportion gradually reduced dietary C18:2 and C18:3 contents, leading to an imbalance in their ratio (C18:2:C18:3 was 11.19:1 in G50 and 15.35:1 in G100), which subsequently affected lipid deposition in muscle. Cao Junming et al. reported that increasing housefly maggot meal replacement of fish meal in diets caused polyunsaturated fatty acid deficiency, resulting in decreased muscle ether extract content in Pacific white shrimp (*Litopenaeus vannamei*). Zhang Lijian et al. reported that the main fatty acids in tilapia muscle are C16:0, C18:1, C18:2, and C22:6. In this experiment, the G50 diet contained higher C16:0 content (12.02%) than the control and other replacement groups, which may have contributed to higher muscle ether extract content in the G50 group.

**3.3 Effects of Soybean Oil Replacement by Palm Oil on Serum Biochemical Indexes of Juvenile GIFT** High-density lipoprotein cholesterol (HDL-C) is a blood lipoprotein that transports cholesterol, triglycerides, and other substances from peripheral tissues to the liver for catabolism, preventing cholesterol deposition in blood vessel walls. The present results show that serum HDL-C content in the G50 and G100 groups was significantly lower than in the control group, which differs from Cheng Minjie' s finding that replacing 80% of fish oil with palm oil significantly increased serum HDL-C content in half-smooth tongue sole, potentially benefiting lipid metabolism. This discrepancy may be attributed to differences in experimental fish species and the lipid source being replaced. Torstensen et al. found that complete replacement of fish oil with palm oil had no significant effect on plasma cholesterol, triglyceride, very low-density lipoprotein cholesterol (VLDL-C), LDL-C, or HDL-C contents in Atlantic salmon.

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are primarily located in hepatocytes and serve as markers of liver function. Under normal conditions, these enzyme activities are low and relatively stable in fish blood, but increase significantly when liver damage occurs. The present results show that serum ALT and AST activities in juvenile GIFT decreased initially and then increased with higher palm oil replacement proportions, with ALT activity in the G25 and G50 groups and AST activity in the G50 and G75 groups being significantly reduced, suggesting that palm oil at these replacement proportions may improve liver health status. This effect may be related to the progressive increase in natural antioxidants such as vitamin E and  $\beta$ -carotene, enhancing dietary antioxidant capacity with higher palm oil inclusion. Studies have shown that replacing fish oil with palm oil or palm fatty acid distillate in

African catfish feeds increases dietary vitamin E content and significantly improves antioxidant capacity in liver and muscle, protecting liver health. Gao et al. replaced 0, 40%, 60%, and 100% of oxidized fish oil with palm oil, increasing dietary vitamin E from 0 to 246 mg/kg, and found that palm oil replacement reduced lipid peroxidation and improved antioxidant capacity in spotted sea bass after 50 days. Han Yuzhe et al. reported that incorporating appropriate proportions of palm oil in spotted sea bass diets could alleviate liver damage caused by oxidized fish oil.

### Conclusion

Based on the experimental results, three main conclusions can be drawn. First, isocaloric replacement of 25-100% soybean oil with palm oil had no significant effect on growth performance, physique indexes, or routine muscle composition (except ether extract content) of juvenile GIFT. Second, isocaloric replacement of 50% soybean oil with palm oil significantly affected serum HDL-C content and ALT and AST activities. Third, using growth performance as the evaluation criterion, when dietary crude protein and ether extract contents are 31.60% and 9.30%, respectively, and soybean oil content is 4.00%, palm oil can completely replace soybean oil in juvenile GIFT diets.

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