

**REPLACEMENT OF FISH MEAL BY JOJOBA MEAL
(*SIMMONDSIA CHINENSIS*) IN THE DIETS FOR RED TILAPIA,
OREOCHROMIS MOSSAMBICUS X *O. NILOTICUS* FRY**

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ABSTRACT: *The study was directed to estimate the effect of jojoba meal *Simmondsia chinensis* protein as alternative protein source of fish meal protein with replacing levels of 0, 25, 50,75 and 100 % on the performance of red tilapia, *Oreochromis mossambicus* x *O. niloticus* fry. One hundred and fifty fries (average weight 0.97 ± 0.08 g/fry) were randomly distributed into 15 aquaria, ten fish /aquaria fish were fed experimental diets in triplicate aquaria for 98 days. The inclusion of jojoba meal in the experimental diets did affect significantly the growth performance of red tilapia among all treatments. Significant differences were observed in terms of feed conversion ratio (FCR) and/or protein efficiency ratio (PER) by increasing jojoba meal protein level in the diets. Fish fed diet 2 which contained 25% jojoba meal protein had significantly the best growth performance parameters. Increasing jojoba meal protein significantly reduced the growth performance parameters. Fish fed D5 with 100% jojoba meal exhibited the highest value of crude protein. Fish fed D5 with 100% jojoba meal exhibited the highest value of crude protein. Also, the contents of carcass lipid and ash were increased significantly ($P < 0.05$) by increasing jojoba meal replacement in the experimental diets. No histopathological effect was observed among all treatments. It could be concluded that jojoba meal protein can be replaced in the diets for red tilapia fries up to 25% of fish meal protein without affecting the performance of red tilapia fries and/or histopathological indices.*

Key words: *Jojoba meal, red tilapia, growth performance, nutrition, histology.*

INTRODUCTION

Increasing the success of the aquaculture sector is related with decreasing the production cost. The fish meal protein and plant protein concentrates are the most expensive ingredients of fish diets (Naylor *et al.* 2009). Searching for alternative protein sources of fish meal protein was studied by several authors; soybean meal (El-Sayed 2004), cottonseed meal (El-Saidy and Gaber 2003), faba bean (El-Saidy *et al.* 2006), pea protein concentrate (Penn *et al.* 2011). Some of unconventional protein sources were studied in fish diets such as torula yeast (Olvera-Novoa *et al.* 2002), single cell proteins (Lim *et al.* 2005), and algae meal proteins (Nandeeshha *et al.* 1998, Hussein *et al.* 2013). Jojoba is a natural oil seed plant promising new crop that cultivates naturally in the desert or new lands. After the oil

extraction the left over contains from 26 to 33% crude protein (Nasser 2009) as well as carbohydrate and fiber (Elangovan and Shim 2000). El-Halawany (2004) stated that jojoba oil is not a vegetable oil but it is a liquid wax contained unsaturated acids (eicosenoic and oleic) and unsaturated alcohols (docosenol and eicosenol). It has been found that Jojoba meal contains four anti-nutritional compounds represent 11-15% of the meal which caused unpalatable and antagonistic effects on animals (Vermauti *et al.* 1998) which caused the inhibition and appetite devastation to animals (Lievens *et al.* 2003). The waste of jojoba seeds squeeze, as a by-product is a likely feedstuff after being detoxified (Motawe 2006). Further studies are required to increase the profitable value of jojoba meal. Consequently, the present study aims

to estimate the effect of jojoba meal as alternative protein source of fish meal protein in the formulated diets for red tilapia *Oreochromis mossambicus* x *O. niloticus* fry and its effect on the growth performance, carcass composition and fish histology.

MATERIALS AND METHODS

Fish and feeding trial

The feeding trial was conducted at the fish research laboratory (Faculty of Agriculture, Menoufia University). Red tilapia, *Oreochromis mossambicus* x *O. niloticus* fries were achieved from a governmental fish hatchery (21 kilo Marriott, Alexandria Governorate, Egypt). Fish were adapted to laboratory conditions for one week in fiber glass tank 1000 L. Ten fish averaged 0.97 ± 0.08 g/fish were distributed randomly into twelve 80-L glass aquaria. The formulated diets were fed to triplicate groups of fish three times at feeding rate of 6% of body weight at the beginning and decreased into 5% to the end of 14 weeks feeding period. The feeding rates were adjusted according to fish periodical live body weights every two weeks.

The formulated diets and feeding

Five experimental diets were formulated to be isonitrogenous and isocaloric in terms of crude protein (30.3%) and gross energy (4.1 Kcal/ g). Five experimental diets were formulated including the control as basal diet (D1) without jojoba meal, followed by four diets. FM was replaced by jojoba meal at 25, 50, 75 and 100 % (D2, D3, D4 and D5, respectively). The ingredients of the experimental diets are showed in Table 1. All ingredients were mixed adding water about 40% moisture. The experimental diets were passed through a mincer with die into 1-mm diameter spaghetti-like strands, sun dried and stored in airtight containers. Proximate compositions of the experimental diets were determined according to AOAC methods (2000).

Water quality

Water temperature and dissolved oxygen were measured every other day using an oxygen meter. pH was monitored twice weekly using pH meter. During the 14-week feeding trial, water temperature was 25.6 ± 0.9 °C, the dissolved oxygen was 6.5 ± 0.5 mg⁻¹ and pH was 8.5 ± 0.2 .

Sample analysis

The fish body composition consisted of crude protein, moisture, and ash were performed by standard procedures (AOAC 2000). After 14-week feeding period all fish were counted and weighed to calculate percent weight gain (PWG; $[(BW - \text{initial BW}) \times 100 / \text{initial BW}]$), feed conversion ratio (FCR; dry feed consumed/WG), feed efficiency ratio (FER; WG/ dry feed consumed), protein efficiency ratio (PER; WG/protein intake), specific growth rate (SGR; $[(\ln \text{ final BW} - \ln \text{ initial BW}) \times 100 / \text{days}]$), and survival ($[(\text{no. of fish at the end of the experiment} / \text{no. of fish at the beginning of the experiment}) \times 100]$). The initial sample of 15 fish was frozen at -18 °C for analysis of whole body composition. At the end of the feeding trial, six fish from each tank were sampled for biochemical analysis. Fish were homogenized individually for whole body composition and frozen at -18 °C for proximate chemical analysis at the laboratory of the faculty of Agriculture at Menoufia University.

Histological analysis

By the end of the feeding trial, six fish were randomly sacrificed from each treatment. The viscera were dissected and preserved in 10% neutral buffered formalin for 48 h. The liver and intestine were separately dissected and examined; further processing according to standard histological techniques was followed (Banchfort *et al.* 1996). Slides, stained routinely with hematoxylin and eosin (H&E) stain for examination through the light electric microscope.

Partial and complete replacement of fish meal by jojoba meal.

Table 1. Ingredients composition of the formulated diets for red tilapia

Replacement level %	Experimental diets				
	0	25	50	75	100
Ingredients (%)					
Fish meal (60 % CP)	20.0	15.0	10.0	5.0	0.0
Jojoba meal (24% CP)	0.0	12.5	25.0	37.5	50.0
Soybean meal (44% CP)	40.0	40.0	40.0	40.0	40.0
Corn starch	31.4	23.9	16.4	8.9	1.4
Vegetable oil	4.0	4.0	4.0	4.0	4.0
Vitamins and minerals premix ¹	2.0	2.0	2.0	2.0	2.0
Vitamin C	0.1	0.1	0.1	0.1	0.1
Molasses	2.0	2.0	2.0	2.0	2.0
Methionine	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100
Chemical analysis (% DM)					
Dry Matter	88.36	88.43	88.50	88.50	88.64
Crude protein	30.3	30.0	30.4	30.6	30.2
Ether extract	5.41	5.53	5.66	5.66	5.90
Crude fiber	3.91	4.48	5.05	5.05	6.18
Nitrogen Free Extract	40.51	39.55	38.58	38.58	36.67
Crude ash	8.23	8.57	8.91	8.91	9.59
Calculated energy value					
GE (Kcal/Kg) ²	4110.07	4104.78	4099.5	4098.29	4088.57
DE (Kcal/Kg)	3082.55	3078.59	3074.62	3073.71	3066.43

¹Vitamins and minerals mixture (mg or IU if mentioned kg⁻¹diet): vitamin A, 8000 IU; vitamin D3, 4000 IU; vitamin E 50 IU; vitamin K3, 19IU; vitamin B2, 25mg; vitamin B3, 69mg; nicotinic acid, 125mg; thiamine, 10mg; folic acid, 7 mg; biotin, 7mg; vitamin B12, 75mg; choline, 400mg and vitamin C, 200 mg. 300 mg I, 100mg Co, 100mg Si, 50000mg Zn, 70000mg Mn, 30000mg Fe, 4000 Cu, and CaCo3 even 1 KG.

²GE (Gross energy) was calculated according to NRC (1993) by using factors of 5.65, 9.45 and 4.22 Kcal per gram of protein, lipid and carbohydrate, respectively.

Statistical analysis

Differences among dietary treatments were tested by one-way ANOVA. The percentage data of weight gain and specific growth rate were arcsine transformed before the ANOVA analysis. Differences were considered significant at the $P < 0.05$. The differences among means were determined

using Duncan 's multiple range test (Duncan 1955).

Results

Growth performance and survival rate (%)

The effect of jojoba meal protein as replacement of fish meal protein on the

growth performance of red tilapia *Oreochromis mossambicus* x *O. niloticus* fries fed the formulated diets are shown in Table 2. Significant differences ($P < 0.05$) in terms of final weight, weight gain and weight gain % were observed among all treatments. Fish fed control diet D1 exhibited the higher final weight and weight gain (8.95 and 8.05 g/fish) and followed by fish fed D2 (8.15 and 7.20 g/fish) diet. Additionally, increasing jojoba meal protein in the diets decreased the final weight and weight gain. Also, the results of specific growth rate (SGR % day⁻¹) showed the lowest value (1.62) with fish fed D5 and followed by fish fed D4 diet. There was no effect of jojoba meal protein in the formulated diets on the survival rate (%) of red tilapia.

Feed utilization

The effect of replacement FM protein with jojoba meal protein on feed utilization of red tilapia fries fed the experimental diets illustrated in Table 2. Feed intake differed significantly ($P < 0.05$) between fish fed the experimental diets. The feed consumed reduced significantly ($P < 0.05$) with

increasing jojoba meal replacement level in the diets to 100%. Feed conversion ratio (FCR) were significantly ($P < 0.05$) differed between all treatments. The best value (2.24) was observed with fish fed D1 without jojoba meal and the worst value (2.83) was observed with fish fed D5. Protein efficiency ratio (PER) was not differ significantly ($P > 0.05$) between fish fed D1, D2 and D3 and differed with that fed D4 and D5. Increasing jojoba meal replacement level decreased PER significantly ($P < 0.05$).

Fish body analysis

The contents of fish body including moisture, protein, lipid and ash were observed in Table 3. It was observed that moisture and protein content increased with increasing jojoba meal replacement level above 50%. A gradual increment in lipid contents was observed as the level of jojoba meal inclusion increase in the experimental diets and differed significantly ($P < 0.05$) among all treatments. It was observed that ash content increased significantly ($P < 0.05$) with increasing jojoba meal replacement level.

Table 2. The performance of Red tilapia *Oreochromis mossambicus* x *O. niloticus* fry (average 0.97±0.08 g/fish) fed the experimental diets with different levels of jojoba meal for 14 weeks. Values are mean ± SD of triplicate groups.

Values*	D1	D2	D3	D4	D5
IBW (g fish ⁻¹)	0.90±0.0	0.95±0.05	0.96±0.05	1.1±0.0	0.95±0.05
FBW (g fish ⁻¹)	8.95±0.15 ^e	8.15±0.15 ^d	7.25±0.35 ^c	5.45±0.15 ^b	4.65±0.25 ^a
TWG (g fish ⁻¹)	8.05±0.15 ^e	7.20±0.10 ^d	6.30±0.40 ^c	4.35±0.15 ^b	3.70±0.20 ^a
WG (%)	894.4 ±16.7 ^d	758.9±29.5 ^c	665.4±77.3 ^b	395.5±13.6 ^a	389.5±0.56 ^a
SGR (% day ⁻¹)	2.34±0.02 ^d	2.19±0.04 ^c	2.07±0.11 ^b	1.63±0.3 ^a	1.62±0.0 ^a
FI (g fish ⁻¹)	18.04±0.56 ^e	17.07±0.29 ^d	14.78±0.92 ^c	12.13±0.26 ^b	10.44±0.19 ^a
FCR	2.24±0.03 ^a	2.37±0.01 ^a	2.35±0.01 ^a	2.79±0.15 ^b	2.83±0.21 ^b
FER	0.45±0.01 ^b	0.42±0.0 ^b	0.43±0.0 ^b	0.36±0.02 ^a	0.35±0.25 ^a
PI (g)	5.5±0.18 ^e	5.17±0.09 ^d	4.48±0.28 ^c	3.68±0.07 ^b	3.16±0.06 ^a
PER	1.47±0.02 ^b	1.39±0.01 ^b	1.41±0.01 ^b	1.18±0.06 ^a	1.17±0.09 ^a
Survival %	100	100	100	100	100

Means given in the same row having different superscript letters were significantly different at ($P < 0.05$).

Partial and complete replacement of fish meal by jojoba meal.

Table 3. Body composition of red tilapia *Oreochromis mossambicus* x *O. niloticus* fry (average 0.97 ± 0.08 g/fish) fed the experimental diets with different levels of jojoba meal for 14 weeks. Values are mean \pm S D of triplicate groups.

Treatments	Moisture	Protein	Lipid	Ash
D1, Control	68.7 \pm 1.1 ^c	51.2 \pm 1.2 ^a	15.1 \pm 0.2 ^a	7.2 \pm 0.1 ^b
D2, 25% diet	66.3 \pm 0.4 ^a	55.2 \pm 0.0 ^b	17.3 \pm 0.2 ^b	6.5 \pm 0.2 ^a
D3, 50% diet	67.0 \pm 0.4 ^b	51.5 \pm 1.4 ^a	18.7 \pm 0.2 ^d	7.8 \pm 0.1 ^c
D4, 75% diet	68.2 \pm 0.1 ^c	55.6 \pm 0.0 ^b	18.1 \pm 0.1 ^c	10.2 \pm 0.3 ^d
D5, 100%	69.3 \pm 0.0 ^d	59.3 \pm 1.2 ^c	19.5 \pm 0.3 ^e	10.6 \pm 0.0 ^e

Means given in the same column having different letters were significantly different at ($P < 0.05$).

Histology analysis

Liver histology from fish fed control and jojoba meal replacement diets with different levels (25, 50, 75 and 100 %) for 14 weeks is concisely showed in Fig. 1. The normal structure liver was observed in the fish of control group (Fig. 1A). The hepatic parenchyma of fish fed diet supplemented with jojoba meal replacement at 25% showed no alterations but dilatation in the central vein (Fig.1 B). Also, Focal hemorrhage was detected in the hepatic parenchyma associated with congestion in the hepatic sinusoids of fish fed diet supplemented with jojoba meal at 50% (Fig.1 C). Additionally, congestion was detected in the central vein and sinusoids, hepatocytes contained larger deposits and the nuclei of the hepatocytes were pushed to the cell wall of fish fed diet supplemented with jojoba meal at 75 and 100 % for 14 weeks (Fig. 1D, E). The normal structure intestine of fish fed the control diet was observed (Fig. 2 A). However, the intestine of fish fed diets supplemented with jojoba meal at different levels (25, 50, 75 and 100%) showed diffuse goblet cells formation in the lining mucosal epithelium accompanying with inflammatory cells infiltration in the underlying lamina propria (Fig. 2 B, C, D and E).

Discussion

Fish performance

The results showed that replacement of jojoba meal at a level of 25% (D2) instead of fish meal in the experimental diets enhanced the growth performance of red tilapia (Table 2). Also, increasing jojoba meal replacement level in the experimental diets significantly decreased ($P < 0.05$) the growth of red tilapia. The same results were observed with Labib *et al.* 2012 in the diets for Nile tilapia fingerlings. Khalil *et al.* 2009 attributed the adverse effects of increasing the inclusion level of jojoba meal in the experimental diets on growth performance of red tilapia to increase the level of simmondsin existing in jojoba meal. Moreover, the same authors reported that increasing the inclusion level of jojoba level decrease in growth performance led to affect significantly all organs indices and liver functions enzymes.

Feed utilization

The inclusion of non-extracted jojoba meal in the experimental diets did affect significantly ($P < 0.05$, Table 2) the feed consumed for red tilapia fry. The opposite results were observed with Motawe 2006 who found no significant differences in feed intake and feed conversion ratio ($P > 0.05$) of tilapia fed (0, 25, 50, 75 and 100%) of non- extracted jojoba meal replacement

instead of fish meal. Also, Nabil *et al.* 2010 concluded that the inclusion of non-extracted jojoba meal in replacement to soybean protein at 25 and 50% levels had insignificant effects on tilapia feed consumed. Lievens *et al.* 2003 suggested that the toxic material (simmondsine) found in the jojoba meal caused the inhibition of food intake and appetite suppression to animals and chicken. The same results were

found with Cokelaere *et al.* 1995. Finally, it has been concluded that simmondsine should be removed from the waste of jojoba cake after oil extraction (Fernanda *et al.* 2007). In the current study, PER decreased significantly with increasing the jojoba level in the diets. These results are in contrast with Norhan and Toutou 2015 who found improvement in PER values of sea bream fed jojoba meal replacement diets.

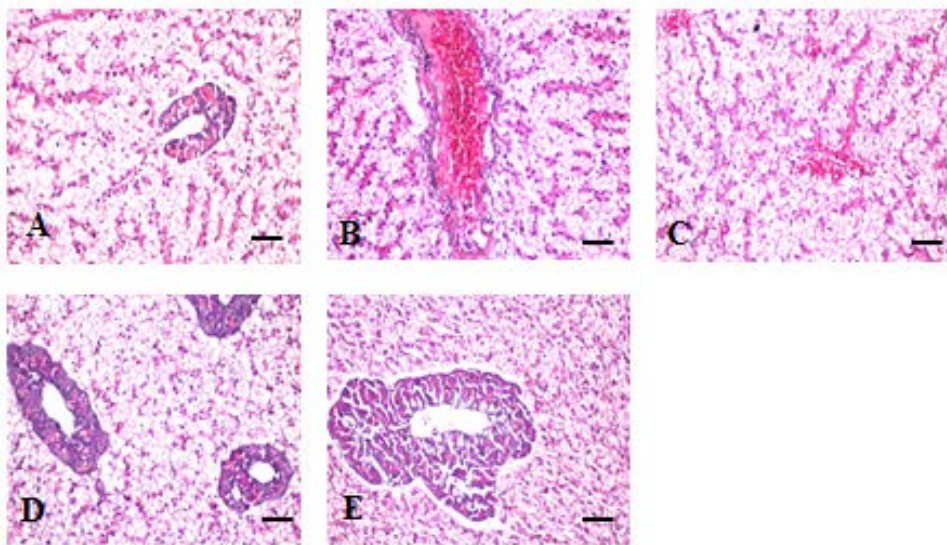


Figure (1): Histopathological changes in the liver of Red tilapia fed the experimental diets.

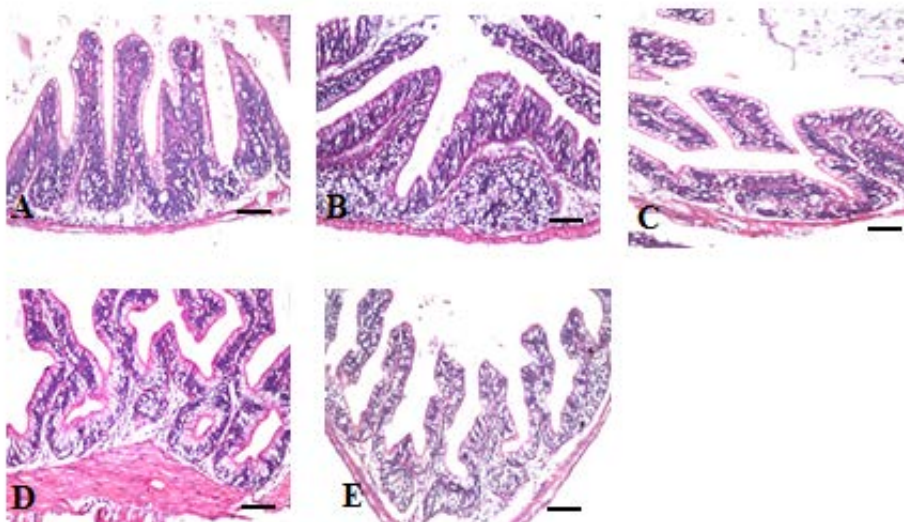


Figure (2): Histopathological changes in the intestine of Red tilapia fed the experimental diets.

Carcass composition

The body composition of fish fed the experimental diets differed significantly ($P < 0.05$, Table 3) among all treatments in terms of moisture, protein, lipids and ash contents. These results are in contrast with Khalil *et al.* 2009 who found that the body composition of mono-sex *O. niloticus* did not differ significantly ($P \geq 0.05$) with increasing non- extracted jojoba meal replacement level up to 100% in the experimental diets. Fish fed D5 diet exhibited the higher values of lipids and ash contents. Moreover, fish fed D1 diet exhibited the lowest value. These results performed the opposite trend by Elangovan and Shim (2000) with tin foil barb.

Figure legends

Figure 1. Histopathological changes in liver of Red tilapia fed different diets (A, Control) without any supplementation, followed by three diets with jojoba meal at 25, 50, 75 and 100 % (B, C, D, E, respectively). (A) Showing normal histological structure. (B) No alterations but dilatation in the central vein. Focal hemorrhage was detected in the hepatic parenchyma associated with congestion in the hepatic sinusoids of fish fed diet supplemented with jojoba meal at 50% (C). Congestion was detected in the central vein and sinusoids, hepatocytes contained larger deposits and the nuclei of the hepatocytes were pushed to the cell wall of fish fed diet supplemented with jojoba meal at 75 and 100% (D, E respectively). (H&E staining); scale bars = 40 μm .

Figure 2. Histopathological changes in the intestine of Red tilapia fed different diets (A, Control) without any supplementation, followed by four diets with jojoba meal at 25, 50, 75 and 100 % (B, C, D, E, respectively). No specific pathological changes were observed in the intestine of fish fed the control diet without any supplementation (Fig.2 A). The intestine of fish fed diets

supplemented with jojoba meal at different levels (25, 50, 75 and 100%) showed diffuse goblet cells formation in the lining mucosal epithelium associated with inflammatory cells infiltration in the underlying lamina propria (Fig. 2 B, C, D and E). (H&E staining); scale bars = 40 μm .

Conclusion

From the obtained results, it is recommended to replace fish meal protein with jojoba meal up to 25% without any negative effects on the performance, feed utilization and fish histology of the red tilapia, *Oreochromis mossambicus* x *O. niloticus* fries. Further studies are needed to maximize the commercial benefits from jojoba meal by detoxification methods and other fish species.

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إحلال بروتين مسحوق الجوجوبا كبديل لبروتين مسحوق السمك في إعداد علائق أسماك البلطي الأحمر

دياب محمد سعد الصعدي ، إبتهاال السيد محمد حسين

قسم إنتاج الدواجن والأسماك - كلية الزراعة - جامعة المنوفية

الملخص العربي

أجريت هذه الدراسة بهدف تقييم تأثير مسحوق الجوجوبا كبديل بروتيني لمسحوق السمك علي نمو وأداء زريعة أسماك البلطي الأحمر. استخدم في هذه الدراسة عدد 150 زريعة بلطي أحمر بمتوسط وزن ابتدائي 0.97 ± 0.08 جم/ سمكة ووزعت الأسماك عشوائياً على 15 حوض زجاجي بمعدل 10 سمكات بكل منها وتم تغذية الأسماك على العلائق التجريبية في ثلاث مكررات لكل عليقة واستمرت التجربة لمدة 98 يوم. أظهرت النتائج أن إضافة مسحوق الجوجوبا إلى العلائق التجريبية كان لها تأثيراً معنوياً علي نمو وأداء زريعة البلطي الأحمر بين كل المعاملات. زيادة مسحوق الجوجوبا في العلائق التجريبية كان له تأثيراً معنوياً علي معدل التحول الغذائي و الكفاءة البروتينية بين كل المعاملات. وجد أن أفضل نمو وأداء ظهر في مجاميع الأسماك المغذاة علي العليقة التي تحتوي علي 25% بروتين من مسحوق الجوجوبا. أيضاً وجد أن زيادة مسحوق الجوجوبا في العلائق التجريبية بمعدل 100% أدت إلي زيادة معنوية في محتوى جسم الأسماك من الدهن والرماد. لم يظهر أي تأثير هستوباثولوجي لمسحوق الجوجوبا علي الحالة الصحية للأسماك بين كل المعاملات. من النتائج المتحصل عليها يمكن أن نوصي بإستخدام مسحوق الجوجوبا كبديل بروتيني لمسحوق السمك في علائق زريعة أسماك البلطي الأحمر بمعدل 25% دون أي تأثيرات عكسية علي النمو والأداء والحالة الصحية للأسماك .

أسماء السادة المحكمين

كلية الزراعة بمشتهر - جامعة بنها

كلية الزراعة - جامعة المنوفية

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Partial and complete replacement of fish meal by jojoba meal.
