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Impact of Partial Replacement of Fishmeal with Pumpkin Meal on Growth Performance, Feed Utilization, and Body Composition of the Nile Tilapia Fingerling (*Oreochromis niloticus*).

Khattab H. M[1], Ragaa A. Ahmed[2,*] , Ayah M. Desoky[3] and A. M. Singer[3]
1. Dept.of Anim. Prod. Fac. Of Agric. Ain Shams Univ. Cairo, Egypt
2. Dept. of Aquaculture Fac. of Fish and Fisheries Technology, Aswan Univ. Egypt
3. Dept. Of Animal and poultry production, Fac. of Agric. and Natural Resources, Aswan Univ. Egypt.
[*]**Corresponding Author: ragaa10@yahoo.com**

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ABSTRACT

Twelve ponds were used in this experiment to study the effect of replacing fishmeal with pumpkin meal in the diet of Nile tilapia using four levels of soybean replacement (C=0, T1=50, T2=75, and T3=100) of the total fish meal of the control diet C. Each one was replicated in three ponds for 60 days. Nile tilapia fingers (2.41g) were transferred to the experimental ponds at a rate of 50 fingerlings/hap. At the end of the experimental period, there were no statistically significant differences in the survival rates between the fish fed on experimental diets. The highest significant (P>0.05) final body weight (FBW) and weight gain (WG) were achieved with the group fed on T2 followed by the group fed on T1. The group fed on T1 and T2 achieved the highest significant Specific Growth Rate (SGR) followed by the T3 group. Treatment 2 recorded the best PER and FCR followed by T3. Otherwise, the group fed the control diet gave the lowest FCR and PER. The highest (P>.05) PPV value was obtained for fish fed T2 with no significant differences among All experimental groups. The fish fed on the control diet was significantly higher(P>. O5) in the body moisture and crude protein content than the rest of the treatment fish was significantly higher (P > 0.05) in the body moisture content and less crude protein content than the other treatment.

INTRODUCTION

In conjunction with the global demand for aquatic species, aquaculture has increased dramatically (Daoud *et al*., 2020). In 1961, per capita fish consumption6 was increased from 9.0 kg to 20.2 kg in 2020, and is expected to increase to 21.4 kg in 2030 and the world population is expected to reach 10 billion (FAO, 2020), At the global level, seafood makes up 15% of animal protein consumed, averaging 20.2 kg per person in 2020 (Boyd *et al*., 2022). With the population increase, the demand for seafood increases .Coupled with the decreasing availability of wild fisheries due to climate change and related issues, it has led to a mismatch between the demand for and supply of fish products, which has led to the need to find other methods of fish production (Tran *et al*., 2019). During the past two decades aquaculture has been the fastest-growing food production system (Muhala *et al*., 2021; Maulu *et al*., 2021; Ragasa *et al*., 2022).Aquaculture contributes substantially to global food and nutrition security,and it is a major producer of aquatic foods.(Edwards *et al*., 2019). One of the most widely farmed fish species is the Nile tilapia (*Oreochromis niloticus*) and contributes to food security, providing affordable fish for the resource poor. (Maulu *et al*., 2021, Eissa *et al*., 2022, Kord *et al*., 2022),and is currently cultured in more than 140 countries (Zhang *et al*., 2020). Because of the high fertility,rapid growth, good performance under suboptimal nutritional conditions,and ability to resist poor water quality, Nile tilapia is an ideal fish species for aquaculture. (Hassanien *et al*., 2011, Opiyo *et al*., 2018, Tibihika *et al*., 2020). Access to affordable and sustainable feed ingredients that maintain fish growth and well-being is a requirement for improving profits in fish farming (Allam *et al*., 2020, Khalil *et al*., 2022). Currently , pumpkin (Cucurbita spp.) is receiving much attention for its nutritional and health protective properties. (Montesano *et al*., 2018), and clinical application (Tanaska *et al*., 2020). The objective of this study was to investigate the effects of different dietary concentrations of pumpkin (T. occidentalis) meal on growth performance , and body composition, of Nile tilapia (*Oreochromis niloticus*) fingerlings .

MATERIALS AND METHODS

Experimental fish, and diet preparation

Nile tilapia fingerlings were obtained from General Authority for Fish Resources Development (GAFRD), - Sahari region, Aswan city

Four isonitrogenous diets were formulated from commercial ingredients The composition and chemical analysis of the experimental diets are shown in Table (1). The dry ingredients were passed through a sieve (aperture of 0.3 mm in diameter) before being mixed into the diet. Emulsified oil was added with equal quantity of water with 0.7% phosphatidylcholine (lecithin) according to (El- Dahhar and El- Shazly, 1993), to the experimental rations. Then the boiling water was mixed with the mixtures at a rate of 50% for pelleting.

Twelve ponds were used in this experiment to study the effect of replacing fishmeal by pumpkin meal in the diet of Nile tilapia using four levels of soybean replacement (0, 50, 75, and 100) of the total fish meal of the control diet C. Each one was replicated in three ponds. Nile tilapia fingers (2.41g) were transferred to the experimental ponds at a rate of 50 fingerlings / hap. Each pond was supported by continuous artificial ventilation. The replacement ratios in the experimental diets were as follows:

C: zero Pumpkin (control diet).

T1 : 50 % Pumpkin.

T2: 75 % Pumpkin.

T3: 100 % Pumpkin.

Growth performance and feed utilization:

Fish was fed with treatments diets twice daily and feed amounts are adjusted every two weeks based on weight change. At the end of the experimental period the fish in each pond were counted and the weight of the group was taken to evaluate the absolute and relative average weight gain (WG), specific growth rate (SGR), food conversion ratio (FCR), protein productive value (PPV), and protein efficiency ratio (PER) using the following formulae as described by **Brown (1957), Winberg (1956), Castell and Teiws (1980), and Miller and Bender (1955)**, respectively.

WG = final average weight (g)−initial average weight (g).

SGR (% d–1) = 100 × (ln Wt − ln W0)/t

Where Wt and W0 represent final and initial body weights of fish, respectively, and t represents the duration of the feeding trial.

FCR = dry weight of feed (g) / wet weight gain by fish (g); and PER = wet weight gain by fish (g) / protein intake (g).

Where protein intake (g) = protein (%) in feed × total weight (g) of diet consumed / 100.

PPV % = 100 * (Pt –P0) / protein intake g.

Where:

P0: Protein content in fish carcass at the start.

Pt: Protein content in fish carcass at the end.

Water temperature, pH and dissolved oxygen concentration were routinely monitored in all ponds. At the beginning and end of the feeding trial, samples of fingerlings were analyzed for body chemical composition.

Table 1: Composition and chemical analysis of the diets with the four levels of Pumpkin meal replacement used to feed Nile tilapia fingerlings.

ITEMS (g/kg)	C	T1	T2	T3
Wheat flour	100	100	100	100
Pumpkin meal	0	90	60	120
Fishmeal	120	30	60	0
Wheat bran	150	150	150	150
Soybean meal	346	346	346	346
Corn meal	200	200	200	200
Corn oil	30	30	30	30
Sun flour oil	20	20	20	20
Vit.&min.mix*	30	30	30	30
Ascorbic acid	4	4	4	4
Proximate analysis on dry matter basis				
Dry mater	90.22	90.11	90.03	90.02
Moisture	9.78	9.89	9.97	9.98
Crude protein (N × 6.25)	33.22	32.94	31.92	31.46
Crude fat	7.24	7.45	7.40	7.20
Crude fiber	6.36	6.29	6.48	6.13
Ash	6.10	6.53	6.88	6.94
Carbohydrate (NFE)^{u8}	47.08	46.79	47.32	48.27
Gross energy (GE) kcal/100g3 **	408.14	405.15	401.12	400.48

**GE (kcal/100 g DM) = CP x 5.64 + EE x 9.44 + NFE x 4.11 calculated according to (McDonald *et al*., 1973).

*Each 100 gram of vitamin and mineral contained: Mineral: Zn, 2.50 mg; Mn, 16.00 mg; Fe, 31.50 mg; Cu, 5.50; I, 0.55 mg; Ca, 1.15 gm and P, 450 mg. Vitamins: A, 7500000 IU; Bi, 100 mg; B3, 500 mg; B6, 150 mg; B12, 2.5 mg; E, 100 mg; K, 100 mg; Pantothenic acid, 275 mg; Folic acid, 100 mg and vit. D3, 7500 IU.

Fish and feed analyses:

Feed samples were taken to determine the proximate composition analyses including moisture, protein, lipid, and ash contents according to AOAC.**(2012)**. NFE was calculated by the difference according to the following equation: NFE = 100 - [Moisture % + Ash % + % lipid + % protein + Fiber %]**.** GE content of the diets was estimated according to the following factors (5.64, 9.44 and 4.11 kcal/100g) for protein, lipid and NFE, respectively **(McDonald *et al.*, 1973).**

Statistical analysis:

Analysis of variance (ANOVA) was used to test for significant differences (p < .05) between the various treatment means obtained for the growth, feed utilization, and body composition. All statistical analyses were conducted using a IBM-SPSS v. 28 software.

RESULTS AND DISCUSSION

Based on what has been reported in previous studies on the benefits of pumpkin seed proteins and their use on a large scale, and due to its positive medical effects as antioxidants, cancer diseases, and liver diseases (Caili *et al*., 2006), it is expected to be applied in the formulations of feed for animals and aquatic animals as protein substitutes. In the current study all experimental diets were accepted by the fish and there were no pathological signs or symptoms were detected during the experiment. The reason could be that pumpkin protein is characterized by its high content of essential amino acids and high functional features (Vinayashree and Vasu, 2021). The culture environment was favorable as the water temperature was within the range of tilapia culture and the pond were aerated to increase the dissolved oxygen level.

Growth performance

Fig (2) shows survival, final body weight (FBW), weight Gain (WG), and specific growth rate (SGR) of Nile tilapia fingerlings fed varied levels of pumpkin. Fuadi *et al*. (2021) used the survival rate as an indicator of the ability of fish to survive, as it is calculated as the survival rate of animals after exceeding a certain period. (Nasyrah *et al*., 2019) estimated several factors affecting the ability of fish to adapt to various environmental conditions and thus their ability to survive and resist food shortages, handling stress, competition, high densities, heat stress, and other abiotic environmental conditions. In this study, the replacement of fishmeal meal with different levels of pumpkin meal did not affect the sustenance rates of Nile tilapia, and there were no statistically significant differences in the survival rates between the fish fed on experimental diets.

In a study presented by Jun *et al*. (2006) it was reported that the content of Pumpkin Cucurbita mochata is large and varied, as it contains a group of vitamins (A, C, and E) and minerals (with manganese, magnesium, and potassium). In addition, cucurbita mochata is rich in carotene and mineral salts.

The highest significant (P<0.05) FBW and WG were achieved with the group fed on T2 followed by the group fed on T1 while the group fed on control diet (C) and T3 achieved the lowest (P<0.05) FBW and WG. The group fed on T1 and T2 achieved the highest significant SGR followed by T3 group, while the lowest significant SGR was achieved with the group fed on C diet of Nile tilapia (*O. niloticus*).

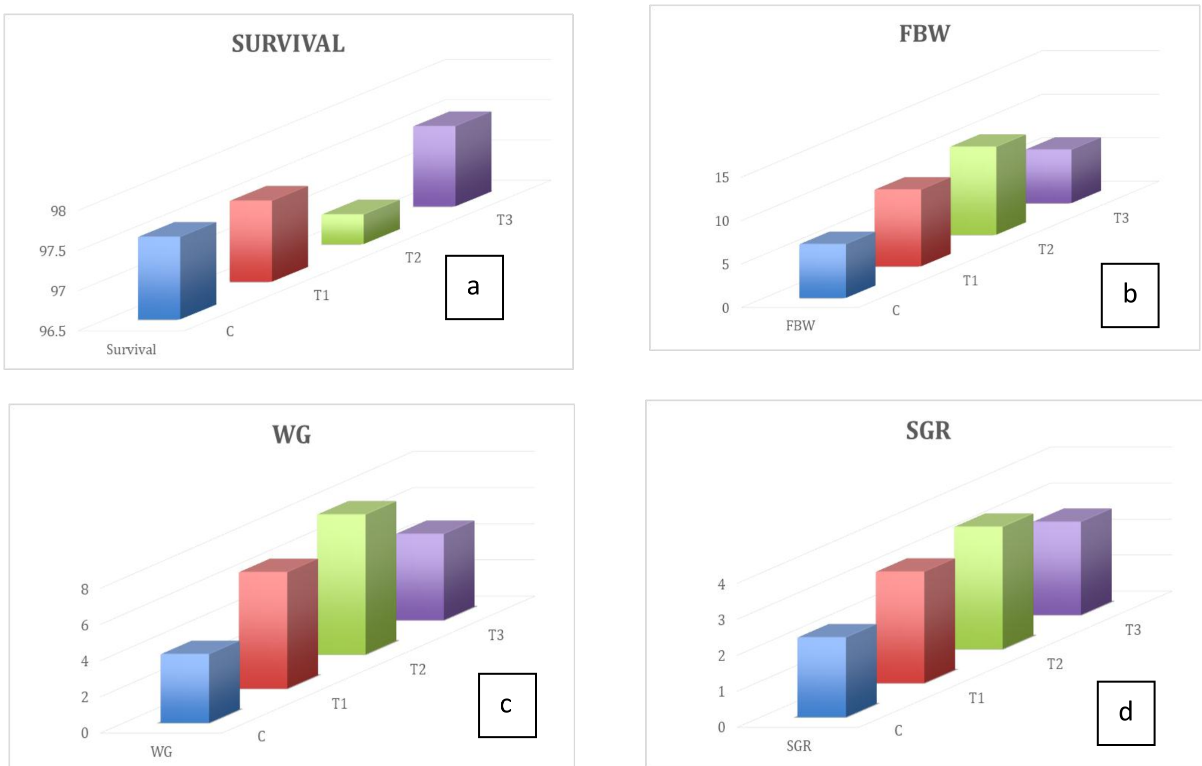

Fig (1): Effect of four levels of pumpkin meal (0, 50, 75 and 100 %) replaced fish meal in the basal on **a:** Survival **b:** Final Body weight (FBW) **c:** Waight gain (WG) and **d:** specific growth rate (SGR) of Nile tilapia (Oreochromis niloticus), 2.41g initial BW.

Feed utilization

Pumpkin's outstanding content of water-soluble vitamins, polysaccharides, antioxidants, tocopherols, minerals, and amino acids has been reported in previous studies **(Alibas, 2007; Stevenson *et al*., 2007; Guiné *et al*., 2011)**. Containing all these ingredients may be a major reason for improving growth performance and utilization of food. With the diversity of the form in which it can be used, pumpkin can be recommended as a future ingredient in aquaculture nutrition **(Dada and Abiodun, 2014; Murray *et al*., 2014; Lovatto *et al*., 2017).**

The present findings (Table 2) clearly indicated that T2 (75% pumpkin meal) had the best significant feed conversion ratio (FCR), being 1.28 followed by T3 (50% pumpkin meal), being 1.65. There was no significant differences was detected between the group fed on T1 (50% pumpkin meal) and the control group in feed conversion ratio (FCR). Results of protein efficiency ratio (PER) showed a statistically significant difference ($P < .05$) among dietary treatments. Fish fed diet with T2 gave the best value of PER (2.65) followed by T3. No significant differences were detected between groups fed on T3 and that fed on T1 (1.95 and 2.32). Otherwise, group fed control diet gave the lowest PER (1.39). Likewise, the protein productive value (PPV) was significantly (P<.05) affected by pumpkin addition. The highest (P<.05) PPV value were obtained for fish fed T2 (160.39) followed by T3, T1 and then control being 153.18, 111.98, 100.34 respectively, with no significant differences among them. **Hashemi (2013)** stressed that pumpkin contains omega-6 and omega-3, in addition to vitamin E with a very high concentration **(Ryan *et al*., 2007**), and these components are mainly crucial for improving growth and utilization of feed for aquatic animals.

Table (2): Feed conversion ratio (FCR), protein efficiency ratio (PER), and protein productive value (PPV) of Nile tilapia (*O. niloticus*), fed at four levels of Pumpkin meal replaced fish meal in a basal diet.

	C	**T1**	**T2**	**T3**
FCR	2.39 ± 0.07^{a}	2.12 ± 0.08^{a}	1.28 ± 0.08^{c}	1.65 ± 0.11^{b}
PER	1.39 ± 0.05^{c}	1.95 ± 0.28^{b}	2.65 ± 0.16^{a}	2.32 ± 0.17^{ab}
PPV	100.34 ± 9.44^{a}	111.98 ± 24.15^{a}	160.39 ± 28.79^{a}	153.18 ± 13.65^{a}

a, b, c, within the same row without a common superscript are significantly different (p <0.05).

Body Composition

Determining the content of protein, fat, ash, and moisture in the fish body can be considered a good indicator of the fish's health and physiological balance **(Saliu *et al*., 2007)**.

In the current study (Table 3), the body moisture content were decreased significantly when feeding on the experimental diet. The fish fed the control meal achieved higher crude protein content than the rest of the meals. It is clear from the results that there is an inverse relationship between the fish body moisture content and the protein content. This is consistent with the report of **Valente *et al*. (2006) and FAO (1999)** that body moisture content is inversely related to protein content. There were no differences in the body ash content with different experimental diets.

Table (3) Chemical composition of Nile tilapia (*O. niloticus*), fed at four levels of Pumpkin meal replaced fish meal in a basal diet.

	C	**T1**	**T2**	**T3**
Moisture	77.95 ± 0.18^{a}	76.45 ± 0.19^{b}	76.29 ± 0.22^{b}	76.32 ± 0.21^{b}
Crude protein	66.25 ± 0.21^{a}	65.44 ± 0.17^{b}	65.31 ± 0.15^{b}	64.75 ± 0.12^{b}
Crude Fat	18.81 ± 0.14^{b}	19.25 ± 0.18^{a}	19.38 ± 0.18^{a}	19.42 ± 0.24^{a}
Ash	14.01 ± 0.22^{a}	14.11 ± 0.21^{a}	14.18 ± 0.23^{a}	14.46 ± 0.22^{a}

a, b, c, within the same row without a common superscript are significantly different (p <0.05).

CONCLUSION

The results of the current study indicated that the replacement of fish meal in the diet of Nile tilapia fingerlings up to a level of 50% had a higher positive effect than the control diet on growth performance, nutritional efficiency, and body composition.

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