INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
ISSN Print: 1560–8530; ISSN Online: 1814–9596
10–234/IAZ/2011/13–2–239–244
http://www.fspublishers.org

Full Length Article

Effect of Feeding Olive Waste on Growth Performance and Muscle Composition of Nile Tilapia (*Oreochromis niloticus*)

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ABSTRACT

Olive waste was evaluated as a substitute ingredient for wheat bran in tilapia (*Oreochromis niloticus*) diets. Five experimental diets viz., 0 (control), 25, 50, 75 and 100% olive waste (OW) replaced with wheat bran were prepared and randomly allotted to triplicate groups with 10 fish in each group. Fish were reared in glass aquaria at a water temperature of 29 ± 1^0C with a dark/light cycle of 12 h. Fish were fed at 3% of body weight twice daily for a period of 70 days. The increasing level of olive waste in tilapia diets significantly ($P < 0.05$) reduced the growth performance and efficiency of feed utilization in fish. However, the replacement of wheat bran with 25% OW did not affect ($P > 0.05$) the feed intake (FI) weight gain (WG), specific growth rate (SGR), condition factor (k), feed conversion ratio (FCR) and protein efficiency ratio (PER) in fish as compared to fish fed control (100% wheat bran). The replacement of wheat bran with 50, 75 and 100% OW significantly ($P < 0.05$) reduced the growth performance and efficiency of feed utilization in Nile tilapia in terms of feed intake, body weight gain, SGR, condition factor, FCR and PER. The substitution of olive waste with wheat bran at all levels did not affect ($P > 0.05$) the moisture and crude protein contents of fish muscles. The fish fed diet containing 100% OW, however, showed significantly higher muscle lipid contents as compared to others. An irregular pattern was observed in the ash content of fish muscles fed various experimental diets. In conclusion, locally produced olive waste in Saudi Arabia can be used as a substitute ingredient for wheat bran only up to 25% level in tilapia diets without compromising their growth performance and efficiency of feed utilization.

Key Words: Nile tilapia; Olive waste; Specific growth rate; Feed conversion ratio; Muscle composition

INTRODUCTION

Aquaculture is an important source of human food and like all other agricultural and animal farming activities its productivity also totally depends on the provision and supply of nutrient inputs. Fish is considered as one of the best quality protein source and therefore, most of the developing countries including Kingdom of Saudi Arabia (KSA) are concerned about increasing their aquaculture production and to make it more and more cost effective. World's Tilapia production has increased drastically over the past few years. It was estimated to be 2.5 million tonnes in the year 2005 with a forecast of >3.5 million tonnes by the year 2010 (FAO, 2006; Josupeit, 2007). The demand for fish and fish products is continuously increasing in the Kingdom. The average per capita consumption of fish in the KSA during the year 1971 was only 2.40 kg/year (Al-Qunaibet *et al*., 1993) that went up to 7.9 kg/year in 2001 and is continuously on the increase (Ministry of Agriculture, 2003). The production of Nile tilapia in KSA has also increased in the last few years due to the introduction of appropriate latest aquaculture technologies including the development of high performance artificial compound aquafeeds required for efficient growth in intensive production systems.

Aquafeeds account for about 40 to 60% of the total cost of aquaculture operations. It is therefore necessary to look for new non-traditional low cost feedstuffs to decrease the overall cost of fish production. The use of alternate feed resources for the development of Aquafeeds has therefore gained significance as the traditional ingredients are either becoming costly or less available (Naylor *et al*., 2000; Ali & Salim, 2004; El-Sayed, 2007; Goddard *et al*., 2008). The production of wastes from the agri-food industry has increased considerably over the recent years for various economical as well as environmental reasons. These wastes include cereal by-products, catering wastes, brewery by-products, plant wastes and wastes from the pasta and other food processing industry (Chaubert, 1995; Um-e-Kalsoom *et al*., 2009). Appropriate strategies need to be developed for the optimum use of such waste products, in particular to recycle them in compound animal or aquafeeds.

To cite this paper: Al-Asgah, N.A., E.M. Younis, A.A. Abdel-Warith, A.A. El-Khaldy and A. Ali, 2011. Effect of Feeding olive waste on growth performance and muscle composition of Nile tilapia (*Oreochromis niloticus*). *Int. J. Agric. Biol.*, 13: 239–244

Nile tilapia is an opportunistic omnivorous fish species that can consume a variety of such industrial wastes, field wastes or unutilized plant residues. El-Gendy (2006) reported that replacing 25% of soybean meal protein with broad bean leaves, cucumber and squash in tilapia diets did not show any adverse effects on their growth and efficiency of feed utilization parameters. Hassanen *et al*. (2005a) studied the effect of incorporating unconventional energy sources (haulm of tomato, cantaloupe, cucumber & squash) also leaves of eggplant, pepper and olive pulp instead of wheat flour in the diets of hybrid tilapia. Al-Asgah (1988) demonstrated that date palm seeds can be substituted for wheat bran-barley mixture in carp diets up to 75%. It has also been shown that waste date meal (WDM) could be used up to 30% as an alternate ingredient for soybean meal in practical tilapia diets without compromising the growth (Azaza *et al*., 2009).

A few recently conducted studies have shown some promising results on the utilization of olive by-products for feeding ruminants (Ben-Salem *et al*., 2003; Molina-Alcaide & Yanez-Ruiz, 2008). However, only limited information is available on the use of olive waste and by-products in fish feeding. If olive waste is to be used as a feed ingredient in the preparation of tilapia diets, it must not only have the desired nutritional qualities but should also be available at a cost that may help to develop cost effective Aquafeeds. The present study was conducted to evaluate the possibility of using olive waste as a substitute feed ingredient in practical diets for Nile tilapia (*Oreochromis niloticus*).

MATERIALS AND METHODS

Preparation of experimental diets: Olive waste was obtained from the Olive Oil Squeezer at El-Gobah Valley in Shaka city, while all the other ingredients were obtained from ARASCO FEED MILL, Riyadh, Saudi Arabia. Five experimental diets, in which the wheat bran was replaced with olive waste (OW) at 0, 25, 50, 75 and 100%, respectively were prepared through extrusion process (Table I). The pellets were dried in a hot air oven at 45^{o}C for 24 h and then stored under refrigeration during the trial period. All the ingredients (fish meal, wheat bran, soybean meal & olive waste) as well as the experimental diets were analyzed for their proximate chemical composition (Tables II & III) according to the methods as described by AOAC (1995). The dietary gross energy content was calculated on the basis of the crude protein, total fat (ether extract) and carbohydrate (nitrogen free extract, NFE) contents as described by Hepher *et al*. (1983). The dietary metabolizable energy was calculated according to NRC (1993).

Experimental design: *Oreochromis niloticus* fry (mean body weight 4.04 ± 0.31 g & average length 6.05 ± 0.36 cm) were obtained from the Fish Seed Hatchery of King Abdulaziz City for Sciences and Technology (KACST), Deerab, Riyadh, Saudi Arabia. The acclimatization period lasted for two weeks, during, which the fish were fed the same larvae diet as used previously at the hatchery. Thirty randomly captured fish (divided into three replicate groups of 10 fish in each) were killed rapidly by immersion in iced water at the start of the experiment. After recording the body weight and length of these fish, they were stored at –30ºC for determining their initial body composition at a later stage. One hundred and fifty fish were then randomly divided into 15 replicate groups with 10 fish in each. The five experimental diets were then randomly assigned to three replicate groups on each diet. The diet 0% OW acted as a control. The fish were kept in glass aquaria (100 × 50 × 45 cm) containing de-chlorinated and well-aerated tap water under standard fish rearing and management practices. The experiment was conducted under artificial light with a light and dark cycle of 12:12 h. The water quality parameters were monitored biweekly and were kept within the optimal ranges as described earlier by Ali *et al*. (2008). The fish were fed twice daily in the morning and afternoon. To quantify the actual amount of feed intake, any feed refusal was recorded. The fish were weighed on biweekly basis and the daily feed intake was adjusted to 3% of their total biomass. The experiment lasted for 10 weeks (70 days).

Proximate chemical analysis: At the end of the experimental period all fish were killed and their body weight and length was recorded. The proximate chemical composition of fish muscles was determined according to the methods of AOAC (1995). The specific growth rate (SGR), condition factor (k), feed conversion ratio (FCR) and protein efficiency ratio (PER) were calculated as previously described by Ali *et al*. (2008).

Statistical analysis: The statistical analysis of the data was done using one way analysis of variance (ANOVA) technique. The means were separated by Fisher's LSD test and compared using Duncan's Multiple Range Test (DMRT) as described by Snedecor and Cochran (1989). Significant differences were defined at $P < 0.05$.

RESULTS AND DISCUSSION

Growth performance and nutrient utilization: Table IV shows the average values for initial body weight, final body weight, total weight gain, specific growth rate (SGR), condition factor (k), feed intake, protein consumption, feed conversion ratio (FCR) and protein efficiency ratio (PER). The results indicated significant ($P < 0.05$) differences in the average weight gain and growth performance of fish fed various experimental diets (Table IV). There was a progressive decrease in the weight gain and growth performance of fish with the increasing level of olive waste in their diets. However, the fish fed on 25% OW did not show any significant ($P > 0.05$) difference in the growth performance as compared to control group i.e., fish fed 0% OW. The fish fed 100% OW exhibited the lowest specific growth rate and total weight gain. Significant ($P < 0.05$) differences were also observed in the feed intake of fish fed

Table I: Ingredient Composition of the Experimental Diets (%)

Ingredients	Diets (wheat brawn replaced with olive waste; OW)				
	0%	**25%**	**50%**	**75%**	**100%**
Fish meal (FM)	15.00	15.00	15.00	15.00	15.00
Soybean meal (SBM)	47.26	47.26	47.26	47.26	47.26
Wheat bran (WB)	37.24	27.80	18.62	9.44	0.00
Olive waste (OW)	0.00	9.44	18.62	27.80	37.24
Mineral and Vitamin mixture[1]	0.50	0.50	0.50	0.50	0.50
Total	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**

1. Vitamins and Minerals mixture (ARASCO) contains: Vitamin A 2 million IU/kg; D 0.3 million IU/kg; E 40,000 IU/kg; C 60 g/kg; K 1.2 g/kg; B_1 0.4 g/kg; B_2 4 g/kg; B_6 2.4 g/kg; B_{12} 10 mg/kg; Niacin 2.4 g/kg; Pantothenic acid 5 g/kg; Folic acid 0.8 g/kg; Biotin 0.2 g/kg; antioxidants 20 g/kg; Copper 1 g/kg; Iron 4 g/kg; Magnesium 15 g/kg; Zinc 1.5 g/kg; Cobalt 0.2 g/kg and Selenium 20 mg/kg

Table II: Chemical composition (%) of feed ingredients used for the preparation of Tilapia diets

Items	Moisture	Crude protein	Lipids	Ash	Crude fiber	NFE[1]	GE[2] (kcal/100 g)
Olive waste (OW)	2.77	5.80	10.41	1.86	39.62	39.54	297.21
Wheat bran	7.00	14.48	5.25	5.16	7.95	60.16	384.09
Soybean meal (SBM)	6.55	46.20	2.91	5.41	2.63	36.35	440.96
Fish meal (FM)	3.61	65.08	13.33	13.54	1.15	3.39	509.80

1. Nitrogen free extract (NFE) = 100 - (moisture + CP + EE + CF + Ash)
2. Gross energy (GE) was calculated according to Hepher *et al.* (1983) using the equivalent factors of 5.65, 9.45 and 4.2 kcal/g for CP, EE, and NFE, respectively

Table III: Proximate chemical composition of the experimental diets (% on as such basis)

Parameters	Diets (wheat brawn replaced with olive waste; OW)				
	0%	**25%**	**50%**	**75%**	**100%**
Moisture (%)	6.66	6.01	6.05	6.50	6.30
Crude protein	37.20	36.45	35.72	34.32	33.41
Crude fibre	4.51	7.79	9.28	13.35	16.01
Total fat	5.00	5.41	5.66	6.16	6.75
Ash	7.64	7.02	6.74	6.17	5.79
Nitrogen free extract (NFE)[1]	38.99	37.32	36.55	33.57	31.74
Gross energy GE, (kcal/100g)[2]	421.25	413.71	408.83	393.14	385.89
Metabolizable energy, ME[3] (kcal/100g)	247.51	245.29	243.14	236.78	235.13
P/E ratio[4]	150.3	148.6	146.9	144.9	142.1

1. Nitrogen free extract (NFE) = 100 - (moisture + CP + EE + CF + Ash)
2. Gross energy (GE) was calculated according to Hepher, *et al.* (1983) using the equivalent factors of 5.65, 9.45 and 4.2 kcal/g for CP, EE, and NFE, respectively
3. Metabolizable energy (ME) was calculated according to NRC (1993) using the equivalent factors of 3.9, 8.0 and 1.6 kcal/g for CP, EE, and NFE, respectively
4. Protein to energy (P/E) ratio = mg protein/kcal ME

various experimental diets. There was a direct correlation between the feed intake of fish and the level of olive waste in diets. The lowest total feed intake (40.88 g/fish) was observed in fish fed 100% OW. However, no significant ($P < 0.05$) differences were observed in the feed intake of fish fed 0 and 25% OW. The overall decrease in feed intake with increasing level of olive waste in tilapia diets also decreased their protein consumption. Similarly both the feed conversion ratio and protein efficiency ratio were affected with the increasing level of olive waste in fish diets. However, no significant differences were observed in the FCR and PER values in fish fed 0 and 25% OW.

These results indicated that substitution of wheat bran with olive waste at a level above 25% in tilapia diets significantly reduced their growth performance and efficiency of feed utilization. The increase of olive waste in tilapia diets increased the overall crude fibre contents of the diets (0% OW, 4.51%; 25% OW, 7.79%; 50% OW, 9.28%; 75% OW, 13.35% & 100% OW, 16.01%), which could be attributed to the higher crude fiber content of olive waste (39.62%; Table II). The increase in dietary fibre content might have affected the overall digestibility of the fish diets that could be held responsible for reduction in growth performance and efficiency of feed utilization. Tilapia is considered to efficiently utilize various types of feed ingredients and by-products to achieve a rapid growth and weight gain. However, tilapia cannot digest the cellulose and lignin because it has no caecum or vermiform appendix in their digestive tract. The high fiber content in tilapia diets may have an inhibitory effect on the digestion of other dietary components. The nutritive value of mixed diets depends on the nutrient composition of individual feed ingredients and the ability of the fish to digest and absorb the nutrients from these feeds (Riche *et al*., 2001; Koprucu & Ozdemir, 2005). Al-Asgah and Ali (1999) reported that the increasing levels of dried poultry excreta in tilapia diets

Table IV: Growth performance and feed utilization of Nile tilapia (*Oreochromis niloticus*) fed experimental diets

Characteristics	Diets (wheat brawn replaced with olive waste; OW)				
	0%	25%	50%	75%	100%
Mean initial body weight (g)	4.09 ± 0.08 [NS]	4.07 ± 0.02[NS]	4.06±0.08 [NS]	4.00±0.08 [NS]	4.01 ± 0.04 [NS]
Final body weight(g)	43.80 ± 4.05[a]	41.98 ± 2.80[a]	36.43±0.23[b]	30.57±1.17[c]	28.13 ± 0.77[c]
Total weight gain(g)	39.55 ± 4.15[a]	37.90 ± 2.78[a]	32.36±0.15[b]	26.57±1.11[c]	24.11 ± 0.80[c]
Feed intake(g/fish)	55.53 ± 5.05[a]	52.17 ± 3.45[ab]	48.90±0.85[b]	43.01±1.15[c]	40.88 ± 0.74[c]
Protein consumption (g/fish)	20.66 ± 1.08[a]	19.02 ± 0.96[a]	17.47 ± 1.21[a]	14.76 ± 1.01[a]	13.66 ± 0.99[a]
SGR[1]	3.37±0.14[a]	3.32±0.09[a]	3.13±0.01[b]	2.89±0.04[c]	2.77±0.05[c]
K[2]	1.82±0.05[a]	1.81±0.04[a]	1.76±0.03[ab]	1.67±0.09[b]	1.67±0.03[b]
FCR[3]	1.39±0.10[c]	1.37±0.04[c]	1.50±0.02[b]	1.61±0.02[a]	1.69±0.05[a]
PER[4]	1.92±0.13[a]	1.98±0.07[a]	1.84±0.02[b]	1.79±0.03[bc]	1.76±0.06[c]

NS = non-significant

a, b, c, d, e, f, g = Different superscripts in the same row mean significant differences between the values at 5% (n =3).

[1] Specific growth rate (SGR) = 100 x [ln final BW (g) - ln initial BW (g)]/experimental period (days)

[2]Condition factor (k) = body weight (g)/body length (cm^3) × 100

[3]Feed conversion ratio (FCR) = Feed intake (g)/body weight gain (g).

[4]Protein efficiency ratio (PER) = Body weight gain (g)/protein intake (g).

Table V: Proximate chemical composition of fish muscles of Nile tilapia fed experimental diets (% fresh weight basis)[1]

Diets / Parameters	Diets (wheat brawn replaced with olive waste; OW)				
	0%	25%	50%	75%	100%
Moisture	77.96±0.11[NS]	78.07±0.17[NS]	78.12±0.40[NS]	78.28±0.07[NS]	78.23±0.23[NS]
Protein	19.52±0.13[NS]	19.39±0.20[NS]	19.43±0.42[NS]	19.22±0.21[NS]	19.19±0.13[NS]
Lipid	0.79±0.03[b]	0.73±0.00[b]	0.73±0.05[b]	0.74±0.04[b]	1.13±0.23[a]
Ash	1.24±0.07[b]	1.17±0.06[b]	1.30±0.04[a]	1.26±0.02[ab]	1.22±0.02[b]

1 = Initial composition of fish slaughtered at the beginning of the experiment (moisture, 78.17%; crude protein, 19.31%; fat, 1.11%; ash 1.26%)

NS = non-significant

a, b, c, d = Different superscripts in the same row mean significant differences between the values at 5% (n = 3)

reduced their growth performance and efficiency of feed utilization. They concluded that only 5% of dried poultry excreta could be included in the diets of *O. niloticus* fry without any adverse effects on their growth. Rojas and Verreth (2003) reported that increasing levels of coffee pulp in the diets of blue tilapia (*O. aureus*) affected their growth, which was directly correlated with their dietary fibre contents. The poor growth performance of tilapia with increasing levels of OW in their diets observed in this study might not only be due to high dietary fibre contents but could also be due to some dietary limiting amino acids in particular of methionine. Garcia *et al*. (2003) reported that methionine could be the limiting amino acid in olive by-products (14.1 & 18.2 g/kg of total nitrogen for two stage dried olive cake & the olive leaves, respectively). Ghazalah *et al*. (2002) found that using date stone meal in Nile tilapia diets over 25% as an energy source instead of yellow corn decreased their growth performance. Hassanen *et al*. (2005a) reported that the incorporation some field wastes, such as haulms of tomatoes, cantaloupe, cucumber and squash, leaves of egg plant and pepper or olive pulp, as non-conventional energy sources instead of wheat flour in hybrid tilapia diets, decreased their daily weight gain, final weight and specific growth rate. Hassanen *et al*. (2005b) also studied the effects of replacing 30 to 35% of a conventional energy source (wheat flour) in hybrid tilapia diets with some non-conventional sources (such as wild plants in North Sinai, Aaear, ghasool, Agram, Rotreyte, Quallam, Metnan, Ikreish & Acacia leaves) on their growth performance. They reported that the fish fed with diet containing Ikreish showed significantly heavier body weights and higher gains as compared to other groups. Also El-Gendy (2006) reported that replacing 25% of soybean meal protein with leaves of broad bean, cucumber leaves and squash in diets of Nile tilapia did not produce any adverse effects on their growth and feed utilization parameters. Al-Asgah (1988) recommended that date palm seeds can replace up to 75% the commonly used wheat bran-barley mixture in carp diets without any significant effect on their final body weight. The condition factor (k) of fish also decreased with the increasing levels of dietary olive wastes. Tilapia is an isometric fish, which with increasing body weight often faces an increase in body length (Younis, 2004).

The decrease in the FCR and PER values in fish fed 50, 75 and 100% OW may be attributed to their relatively lower protein and energy intakes. Soltan *et al*. (2002) and Abdel-Warith (2008) demonstrated that the feed consumption in Nile tilapia increased with the increase in dietary protein level as well as with increasing dietary P/E ratio. Higher dietary fibre levels are considered to interfere with the digestion and absorption of other nutrients and have been regarded as the one of the major factor for the poor performance of fish (Al-Asgah & Ali, 1996; Ali *et al*., 2008). The increasing level of olive waste in the experimental diets not only increased the dietary fibre contents but also reduced their dietary protein levels that overall affected their dietary protein to energy (P/E) ratio. Any inadequate dietary P/E ratio may result in lower growth as well as low protein and energy utilization (Ai *et al*., 2004; Ali *et al*., 2008). The poor FCR and PER values obtained in this study on diets C, D and E can be explained in this context and might also be related to their lower dietary P/E ratio.

Chemical composition of fish muscles: The results on the proximate chemical composition of fish muscles are presented in Table V. No significant ($P < 0.05$) differences were observed in the moisture and crude protein contents of fish muscles fed different experimental diets. The lipid and ash contents of fish muscles fed different experimental diets differed significantly ($P < 0.05$). However, the changes in the muscle ash content were not consistent. The fish fed on diet E showed significantly ($P < 0.05$) higher lipid contents in their muscles as compared to fish fed all other diets (0, 25, 50 & 75% OW). This might be related to higher fat content in 100% OW, in which wheat bran was completely replaced with olive waste that contained higher fat levels. The body as well as the muscle composition of fish is mainly influenced by both the endogenous and exogenous factors, which operate simultaneously (Haard, 1992; Shearer, 1994). The dietary lipid content is regarded as one of the most important factor influencing the muscle and carcass lipid levels in fish (Hanley, 1991; Ali & Al-Asgah, 2001). The higher lipid contents observed in the muscles of fish fed 100% OW can be related to higher oil content of olive waste. The changes in muscle lipid and ash contents observed in this study appear to be associated with the level of dietary olive waste.

CONCLUSION

Olive wastes represent one of the important agricultural by-product and waste in KSA. Overall the results of this study indicated that locally produced olive waste in Saudi Arabia can be used as a substitute to wheat bran only up to 25% level in tilapia diets without compromising their growth and the efficiency of feed utilization. However, to obtain more conclusive results on the utility of olive wastes in tilapia diets further studies are required to determine the amino acid profile and digestibility of olive waste, which may provide a better insight in assessing its nutritive value in tilapia diets.

Acknowledgement: We thank to King Abdulaziz City for Science and Technology (KACST), Riyadh, Saudi Arabia for providing the fish for this study. We acknowledge the cooperation of Olive Oil Squeezers at El-Gobah Valley in Shaka city, Saudi Arabia for the supply of olive wastes. Thanks are also due to ARASCO Feed Mills, Riyadh, for the supply of other feed ingredients. This work was supported by the Centre of Excellence for Biodiversity Research, College of Science, King Saud University, Riyadh, Saudi Arabia"

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(Received 24 May 2010; Accepted 07 July 2010)