Mediterranean Aquaculture Journal 2014 (6); ***14-20.***

Original Article

Use of Black Pepper Seeds as a Growth Enhancer for Juvenile of Florida Hybrid Red Tilapia, *Oreochromis niloticus* (Linnaeus) X *Oreochromis mosambicus* (Peters)

Shalaby, S.M.M.[1]; Sakr, E. A. [2]; Abdel Moniem, A.I. [3] and El-Dakar, A.Y. [1]

1- Aquaculture Department, Faculty of Fisheries, Suez University, Suez, Egypt
2- Aquaculture Division, National Institute of Oceanography and Fisheries, Alexandria, Egypt
3- Animal and Fish Production Department, Faculty of Agriculture, Alexandria University, Alexandria, Egypt.

Abstract

Nutritionists are encouraging use of natural growth promoters in livestock feeds due to their ability to prevent side effects that causes by chemical agents. Black pepper seeds are spices that may use as feed additives to enhance survival, and growth of fish. In the present study, the effects of graded levels of black pepper seeds meal (BPSM) on growth performance, feed and nutrient utilization, body composition and cost-benefit analysis of Florida hybrid red tilapia juveniles were studied. Eight glass aquaria with the dimensions of 70 x 30 x 40 cm were used to stock 15 fish averaging 8.6 g/fish. Four experimental diets were formulated to contain 0, 0.5, 1.0,and 2 % of BPSM as feed additive in red tilapia diets. All the diets were isonitrogenous (29-30% crude protein) and isocaloric (457-463 kcal GE/100 g diet). The feed amount was given three times daily, six days a week for 60 days. Fish were weighed biweekly and feed amounts were adjusted on the basis of the new fish weight. Body weight, weight gain and specific growth rate (SGR) of fish fed the control diet was relatively lower than all diets had BPSM except the diet containing 2% BPSM. Fish fed 0.5% BPSM was significantly higher ($P<0.05$) SGR, feed conversion ratio (FCR), protein efficiency ratio (PER), productive protein value (PPV %) and energy retention (ER %) than other tested black paper levels. No significant differences ($P>0.05$%) were found in moisture, protein, fat, ash and energy contents among all the fish groups fed BPSM levels and the control diet. In addition, diet containing 0.5% BSM was economically superior to other tested levels of black paper in diets. It seemed to save about 28% of feed cost consumed to produce one kilogram fish gain (incidence cost). It was recommended to use black pepper seeds meal in fish feeds on the commercial scale at level of 0.5%.

Key words: *Black paper seeds, red tilapia, growth, feed conversion ratio and incidence cos*t

Received: August 2014
Accepted: December 2014

Corresponding author: Asmaa I. Abdel Moniem
Animal and Fish Production Department, Faculty of Agriculture, Alexandria University, Alexandria, Egypt

Introduction

Natural feed additives including some spices or medicinal and aromatic plants in aquafeeds used to minimize the adverse effects of chemical agents (El-Dakar et al., 2005). Recently, marjoram, basil, licorice roots, black seeds, peppermint leaves, fenugreek seeds and caraway seeds were successfully use in fish feeds (Abd Elmonem et al., 2002; Shalaby et al., 2003; El-Dakar et al., 2004a, b, c; Shalaby, 2004; El-Dakar, 2004; El-Dakar et al., 2005 and El-Dakar et al., 2012)). The spices may be used as environmental-friendly feeds in shrimp diets in order to reduce the amount of pollutants in the effluent water (Lawerance et al., 2001). In this context, incorporation of some natural plants may be caused a reduction of uneaten feed percent by aquatic cultured animals (Sakr, 2003; El-Dakar 2004; El-Dakar et al., 2004a,b,c), about third of feed introduced to cultured fish is wasted into the water (Axler et al., 1996) causing adversely effects on water quality. Previous studies indicated that use of marjoram, basil and peppermint in tilapia diets decreased feed waste to 3-16% in fish culture units (El-Dakar et al., 2004a,b). The reduction of feed waste may be due to attract of fish to diets through the flavor of spices as feed additives. Flavor is a most important characteristic property, which governs the acceptability of any food product. Black pepper is one of the most spices has strong flavor which is also known as the 'king of spices' or even as 'black gold'. Different products from black pepper available are ground pepper, pepper oil and oleoresin (Ravindran and Johny, 2001). Black pepper is an excellent source of manganese, iron and vitamin K (Purseglove et al., 1981). Black pepper has demonstrated impressive antioxidant and antibacterial effects**; i**t is remarkably beneficial for digestion. Pepper is associated with a number of functional properties such as antioxidant effects and antimicrobial properties (Kapoor et al., 1993). Studies on the use of black pepper as a feed additive in fish nutrition are very scarce. Therefore, the aim of this study is to evaluate the effect of black pepper as a feed additive in fish diets and study their effects on growth performance, feed conversion, nutrient retention efficiencies, body composition and cost-benefit analysis for red tilapia hybrid Red Tilapia, *Oreochromis niloticus* (Linnaeus) x *Oreochromis mosambicus* (Peters).

Materials and Methods

This work was carried out in Fish Nutrition Laboratory, Aquaculture Division, National Institute of Oceanography and Fisheries (NIOF), Alexandria Branch Egypt. Eight glass aquaria with dimensions of 70 x 30 x 40 cm, total net volume for each aquarium was 70 L, were used in this study. All aquaria were filled with dechlorinated tap water stored 24 hours before used. Fingerlings of red tilapia were obtained from Marine Fish Hatchery (MFH), (21 Km west of Alexandria) and transferred to Fish Nutrition Lab NIOF, Anfoushy, Alexandria, Egypt. After arrival, all fish were kept for one week to alleviate stresses and to be adapted to the new conditions. Ten fish in the same initial weight (8.6 g/fish) were selected randomly and allotted into the experimental aquarium. Fish were fed the control diet for two weeks, during this period healthy fish of the same weight replaced dead one. Four experimental diets were formulated to contain 0, 0.5, 1 and 2% of black pepper. Each diet was fed to two randomly assigned duplicate aquaria. Ingredient composition of the experimental diets is presented in Table (1). Fishmeal was home made by collecting small fish and non-saled fish named locally "Wazafa" dried at 60 ºC. Grounded and sieved prior to keep at –1 ºC. Soybean meal 43% was used as a plant protein source. Yellow corn, wheat bran, wheat milling by-product and sunflower oil were served as energy sources. The experimental diets were prepared by mixing dry ingredients with water and then pelleted using a meat mincer with a 1-mm diameter. The pellets were air dried and stored at -1 ºC until use. The pelleted feed were slightly broken.

Table 1. Ingredients composition of the experimental diets

Ingredient	Diet No.			
	1	2	3	4
Fish meal	10	10	10	10
Soybean meal	30	30	30	30
Corn gluten	10	10	10	10
Wheat milling by-product	23	22.5	22	21
Wheat bran	10	10	10	10
Yellow corn	10	10	10	10
Sunflower oil	5	5	5	5
Vitamin & Mineral mix.[2]	2	2	2	2
Black pepper	0	0.5	1	2

into particles during the first two weeks. Fish were fed 4 % of the body weight daily. The feed amount was given three times a day (900, 1100 and 1500) in equal proportions. Feeding was performed for six consecutive days, for 60 days. Fish were weighed biweekly and feed amounts were adjusted on the basis of the new weight. Fish were reared in 29±1 °C and for temperature and 8.5±0.2 pH. Third water volume was exchanged daily except the weighing day that changed about two thirds water volume.

Diets and fish samples were analyzed (Table 2) according to (AOAC, 1990) for dry matter, crude protein, ether extract, crude fiber, nitrogen free extract (NFE) and ash. The gross energy contents of the experimental diets and fish samples were calculated by using factors of 5.65, 9.45 and 4.2 kcal/g of protein, lipid and carbohydrate, respectively (NRC, 1993). Digestible energy content was calculated from standard physiological fuel values as 4 and 9 kcal/g of protein and carbohydrate and lipid, respectively (Garling and Wilson, 1976).

Analysis of variance (ANOVA) was carried out according to Snedecor and Cochran (1982) using a completely randomized design (CRD). Differences were subjected to Duncan′s Multiple Range-Test (Duncan, 1955) at a significance level of 0.05. All statistical tests were performed using the MSTAT-C (1988) software.

Results and Discussion

The natural plants have incorporated in feeds as feeding attractants for some species (Shalaby et al., 2003; Shalaby, 2004; Abd-Elmonem, et al., 2002; El-Dakar et al., 2004). The results for growth and feed efficiency of experimental fish are presented in Table (3). Fish fed the diets containing 0.5-1% BPSM grew significantly ($P<0.05$) faster than those fed the control diet. However, fish fed diet had 2% BPSM were not significantly ($P>0.05$) than those fed the control diet in body weight and weight gain. While specific growth rate of fish received 2% BPSM being relatively lower value than the control diet. Similar phenomena was obtained with different spices as peppermint (Sakr, 2003), anise (Hanafy, 1995; Sakr, 2003)**,** licorice roots (Shalaby et al., 2003), marjoram (El-Dakar et al., 2004), chamomile (Abd-El-Maksaud et al., 2002) and Carawya (El-Dakar, 2004). The improvement of growth performance of fish fed diet containing BPSM upto 1% level may be attributed to black pepper constituents include essential oils, phytoenzyme, minerals and vitamins. Tainter et al. (2001) reported that pepper seeds having piperine, eugenol, as essential oils, lipase, calcium, potassium, selenium and vitamins A and K. Similar results are found by Musenga, et al. (2007). Srinivasan (2007) reported that black pepper has vitamin-A (Beta Carotene), which is very beneficial for ocular health as well as is a very good antioxidant.

Table 2. Proximate analysis of the experimental diets

Item	Diet No.			
	1	2	3	4
Dry matter %	94.3	94.22	93.98	94.01
% on the DM basis:-				
Crude protein	29.8	29.71	29.63	29.47
Ether extract	5.63	5.61	5.59	5.56
Crude fiber	3.83	2.82	3.8	3.78
NFE	44.57	46.25	44.86	45.12
Ash	11.17	0.5.61	11.12	11.07
Gross energy	457	463	457	457
Digestible energy	393	399	393	393
P/E ratio mg CP/kcal DE	75.83	74.46	75.39	74.99

Table 3. Growth performance and feed utilization efficiency of red tilapia fingerlings fed diets containing different levels of BPSM.

Item	Diet No.			
	1	2	3	4
Growth performance				
Initial weight g/fish	8.7	8.6	8.68	8.46
Final weight g/fish	19.44 ^{c}	25.25 ^{a}	22.47 ^{b}	17.7 ^{c}
Gain[2] g/fish	10.74 ^{b}	16.65 ^{a}	13.79 ^{a}	9.25 ^{b}
SGR[3] % /day	1.34 ^{bc}	1.8 ^{a}	1.59 ^{ab}	1.23 ^{c}
Feed and nutrient intake				
Feed intake g/fish	23.48 ^{b}	26.34 ^{a}	26.27 ^{a}	22.71 ^{b}
Protein intake g/fish	6.60 ^{b}	7.37 ^{a}	7.31 ^{a}	6.29 ^{b}
Energy intake kcal	101 ^{ab}	115 ^{a}	112.5 ^{ab}	97.5 ^{b}
Feed and nutrient efficiency				
FCR[4]	2.09 ^{ab}	1.5 ^{b}	1.8 ^{ab}	2.32^{a}
PER[5]	1.64 ^{b}	2.26 ^{a}	1.89 ^{ab}	1.48 ^{b}
PPV[6] %	26.2 ^{b}	36.45 ^{a}	29.48 ^{b}	26.59^{b}
ER[7] %	19.75 ^{ab}	23.86 ^{a}	21.2 ^{ab}	18.22 ^{b}

*Values in the row having a common superscript letter are not significantly different (P>0.05).

1. *Standard error of the means derived from the analysis of variance*
2. Gain = *(final weight - initial weight).*
3. Specific growth rate = *(Ln final weight - Ln initial weight)/ day).*
4. Feed conversion ratio = *DM intake/weight gain.*
5. Protein efficiency ratio = *weight gain/protein intake.*
6. Productive protein value = *100x(protein gain / protein intake).*
7. Energy retention % = *100 x (gross energy gain / gross energy intake).*

Furthermore, increase performance of tilapia fed BPSM may be due to the spice's effect in improvement of the digestibility's of protein and energy (Sakr, 2003; El-Dakar, 2004 and El-Dakar et al., 2004a and b); enhancement growth and feed conversion (Abd El-Maksoud et al., 2002; Abd Elmonem et al., 2002; Shalaby et al., 2003; El-Dakar et

al., 2004a,b,c and Shalaby, 2004). The present study confirmed the previous studies that revealed a good response of spices to tilapia sp e.g., garlic, onion, marjoram, caraway, basil, anise, fennel, licorice, black seeds and fenugreek). Fish fed 0.5-1% BPSM were consumed more feed, protein and energy than those fed other tested groups. These results are in agreement with those obtained by El-Dakar et al. (2004a) who noticed that use marjoram leaves in tilapia feeds led to increase palatability and feed intake. Schanenberg and Paris (1977) explored that spices have gastric, aromatic and appetizing effect; they caused improving in palatability index and redaction of uneaten feed amounts (Mir et al., 1998; El-Dakar et al., 2004a, b). Feed waste decreased from 33% with control diet to 3% with diet containing 2% dried marjoram leaves (El-Dakar et al., 2004a). Similar results were obtained by El-Dakar (2004) and El-Dakar et al. (2004 a,b,c). However, uneaten feed percent has been decreased to only 3% with dried marjoram leaves (El-Dakar et al., 2004a); 8% with of dried basil leaves (El-Dakar et al., 2004b). While the feed waste percent was not influenced by caraway seeds (El-Dakar, 2004).The present study confirmed the above findings where feed intake increased when fish fed on diets containing 0.5-1% BPSM than the control diet. These results may be due to its strong feeding attraction activity of black pepper flavor. The best FCR found in fish fed diet containing 0.5% BPSM because black pepper may have health benefits, particularly in enhancing digestive tract function. This agree with studies of Capasso et al. (2002) and Vazquez-Olivencia et al. (1992) who reported that black pepper has piperine that can stimulate digestive enzymes, modify stomach secretions, alter gastrointestinal feed transit time, and inhibit diarrhea in rat and mice. Inclusion of 2% BPSM in diet resulted in a decline in the FCR that may be due to stimulate stomach secretions and its effects on the rapidity of movement of food through the digestive tract (Vazquez-Olivencia, et al., 1992). Protein efficiency ratio, protein productive value and energy retention of fish fed 0.5% BPM was significantly ($P<0.05$) higher than other test levels. Protein and energy utilization value were decreased with the increasing of BPSM levels that may be due to the presence of piperine in pepper which increase the body's expenditure of energy, apparently by affecting Black pepper has demonstrated impressive antioxidant and antibacterial effects--yet another way in which this wonderful seasoning promotes the health of the digestive tract. the production of hormone-like chemicals that regulate energy balance. (Westerterp-Plantenga, et al., 2006).

Chemical composition of whole body fish fed different levels of BPSM in diets of Nile tilapia is showed in Table (4). There was a significance ($P<0.05$) higher in ash content of fish fed 2% BPSM than those fed the other tested or control diets. However, there were no significant ($P>0.05$) differences in protein and ether extract contents. Different spice plants gave similar results (Shalaby, 2004).

Cost-benefit analysis (Table 5) showed that the using of 0.5% BP in red tilapia diet resulted in significance decrease in the incidence cost and increase in profit index than other diets.

Table 4. Body composition of red tilapia fed different levels of black pepper.

Item	initial fish	Diet No.			
		1	2	3	4
Moisture	78.12	73.96	74.24	74.58	73.31
Crude protein	12.51	14.48	14.93	14.44	15.46
Ether extract	3.46	7.11	6.33	6.8 3	6.67
Ash	3.93	3.49 [b]	3.93 [ab]	3.73 [ab]	4.21 [a]

*Values in the row having a common superscript letter are not significantly different ($P>0.05$).

Table 5. Cost-benefit analysis of red tilapia fed different levels of black pepper.

Diet	Diet No.				
	1	2	3	4	
Cost per kg feed	2.68	2.69	2.77	2.91	
Incidence cost[1]	5.6	4.04	4.99	6.49	
Change %	100	72.14	89.11	110	
Profit index[2]	1.25	1.73	1.4	1.08	
Change %	100	138.4	112	86.4	

1- *Incidence cost = feed cost to produce 1 kg fish*

2- *Profit index = value of fish /cost of feed consumed, 1 kg fresh fish equals 7 LE*

References

Abd El-Maksoud, A.M.S.; Aboul-Fotoh, G.E.; Allam, S.M. and Abou Zied, R.M. 2002. The response of Nile tilapia to animal protein free diets supplemented with some free amino acids and some medicinal plants. Proc. 1th Conf. Aquacult., 13-15 December, El-Arish, Egypt. Aquacult. Soc.: 233-260.

Abd –Elmonem, A.; Shalaby, S.; and El-Dakar, A.2002. Response of red tilapia to different levels of some medicinal plants by-products: black seed and roquette seed meals.The1 st Scientific Conference of Egyptian Aquaculture Society, 13-15 December pp. 247-260.

AOAC, 1990. Official Methods of Analysis of Association of Official Analytical Chemists. 15th Ed. Published by the Association of Analytical Chemists. Virginia, 2220, USA.

Axler, R. C.;Larcen,C.; Tekkanen,C.; McDonald,M. and Yokom,S. 1996. Water quality issues associated with aquaculture. A case study in mine pit takes. Water Environment Research 68: 995-997.

Capasso, R.; Isso, A.; Borrelli, F. and et, al. 2002. Effect of piperine, the active ingredient of black pepper, on intestinal secretion in mice. Life Sci. 71:2311Y2317.

Duncan, D.B. 1955. Multiple range and multiple F-test. Biometrics, 11:1-42

El-Dakar, A.Y.;Hassanen, G.D.;Shalaby S. M.; Ghonieam, S.I. and Abed-Kader, T.S.2012. Assessing the use of propiotic (*Bacillus subtilus*) and spices oil extracts.

El-Dakar, A.Y. 2004. Growth response of hybrid tilapia, *Oreochromis niloticus* x *Oreochromis auraus*, to diets supplemented to different levels of caraway seeds. Agric. Sci. Mansoura Univ. 29 (11): 6083-6094.

El-Dakar, A.Y.; Hassanien, G. D.I.; Gad, S.S. and Sakr, S.E. 2004a. Use of medical and aromatic plants in fish diets: I. Effect of dried marjoram leaves on performance of hybrid tilapia *Oreochromis niloticus x Oreochromis auraus*, Fingerlings. J. Egypt. Acad. Soc. Environ. Develop. (B. Aquaculture), 5 (1): 67-83.

El-Dakar, A.Y.; Hassanien G. D.I.; Gad, S.S. and Sakr, S.E. 2004b. Use of medical and aromatic plants in fish diets: 2. Effect of dried basil leaves on performance of hybrid tilapia *Oreochromis niloticus x Oreochromis auraus*, Fingerlings. 3 rd Inte. Conf. on Anim. Production and Health in Semi-Arid Areas, Suez Canal Univ.265-277.

El-Dakar,A.Y.;Shalaby,S.M.M.; Abd Elmonem, A.I. and Wahbi, O.M. 2004c. Enhancement of performance using fennel seeds meal as feed additive for Nile tilapia *Oreochromis niloticus.* J. Egypt. Acad. Environ. Develop. (B-Aquaculture), 5 (2): 43-67.

El-Dakar, A. Y.; Shymaa, S. M. Abead,S.M. and Khalafala M. M., 2005. Use of spices as feeding stimulants in diets of marine shrimp, *Penaeus japonicus.* Egyptian J. Nutrition and Feeds, 8 (1):1065-1076.

Garling, D.L. and Wilson,R.P. 1976. Optimum dietary protein to energy ratio for channel catfish fingerlings, *Ictalurus punctatus*. J. Nutr. *106*: 1368- 1375.

Hanafy, M.M. 1995. The use of anise, fennel and ginger as a feed additive in the nutrition of broilers, M.Sc. Thesis, Fac. of Agric., Alex. Univ.

Kapoor,V.K.; Chawla, A.S.; Kumar, M. and Kumar,P.1993. Search for anti-inflammatory agents. Indian Drugs 30, 481–493.

Lawerance, A.L; Castille, F.; Velasco, M. and Samocha, T. 2001. Environmental friendly or least pollutingfeedmanagementprograms for shrimp farming. Book of Abstracts of Aquaculture 2001, January 21-25, 2001. Orlando, Florida, USA. pp 352.

Mir,Z.P.S.;Acharya, S.N.; Zaman, M.S.; Taylor, W.G.; Mears, T.A.; Mcallister, T.A.and Gonewardene, L.A. 1998. Composition of *alfalfa* and *fenugreek silabes* supplemented with barley grain on performance of growing steers. Canadian J. Anim. Sci. 78:343-348.

MSTAT-C 1988. MSTAT director, Crop and Soil Science Department, Michigan State University, USA.

Musenga,A.; Mandrioli. R.; Ferranti, A.; D'Orazio, G.; Fanali, S. and Raggi, M. 2007. Analysis of aromatic and terpenic constituents of pepper extracts by capillary lectrochromatography. J. Sep Sci.;30:612Y619.

NRC, 1993. Nutrition Requirements of Fish. National academy press, Washington, DC, USA.

Purseglove, J.W.; Brown, E.J.; Green, C.L. and Robbins, S.R.J. 1981. Spices, vol. I. Longman Inc., New York.

Ravindran, P.N., Johny, A.K., 2001. Black pepper. In: Peter, K.V. (Ed.), Handbook of Herbs and Spices. CRC Press/Woodhead Publishing Ltd., Cambridge, New York, pp. 62–64

Sakr, S.E. 2003. Studies on the feeding attractants for fish. M.Sc. Thesis. Fac. of Environ. Agric. Sci., El-Arish, Suez Canal Univ., Egypt.

Snedecor, G.W. and Cochran, W.G.1982. Statistical methods 6 th ed. Iowa State University. Press, Ames. Iowa

Schanenberg, D.S. and Paris, R. R.1977. Precis de matiere medical, Tomes Masson, Paris. Pp, 256

Shalaby,S.M. 2004. Response of Nile tilapia, *Oreochromis niloticus*, fingerlings to diets supplemented with different levels of fenugreek seeds (Hulba). J. Agric. Mansoura Univ. 29: 2231-2242.

Shalaby,S.M.; Abd-Elmonem,A.I. and El-Dakar, A.Y. 2003. Enhancement of growth performance, feed and nutrient utilization, of Nile tilapia, *Oreochromis niloticus*, using of licorice roots (Erksous) as a feed attractive. J. Egypt. Acad. Soc. Environ. Develop. (B-Aquaculture), 4 (2): 119-142.

Srinivasan, K. 2007. Black pepper and its pungent principle-piperine: a review of diverse physiological effects. Crit Rev Food Sci Nutr.;47:735Y748.

Tainter,D. and Grenis,A. 2001.Spices and Seasonings,2001. A Food Technology Handbook. 2nd ed. Hoboken, NJ: Wiley-IEEE; 2001.

Vazquez-Olivencia,W.;Shah,P.andPitchumoni, C.1992. The effect of red and black pepper on orocecal transit time. J. Am Coll Nutr. 11:228Y231.

Westerterp-Plantenga,M.;Diepvens,K.; Joosen, A.; Berube-Parent, S. and Tremblay, A. 2006. Metabolic effects of spices,teas, and caffeine. Physiol Behav.;89:85Y91