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Use of black pepper seed as growth enhancer in *Labeo rohita*

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Abstract

Nutritionists are continuously in search and use of natural growth promoters for their respective animal feeds because of their use results in no side effects which are imminent by the use of various chemicals. The field of fish nutrition also needs some breakthrough if some natural growth promoter is discovered. Black pepper seed is natural flavor that could be explored as a feed additive to enhance growth, disease resistance and survival in fish. In the present study, we studied the effect of graded levels of black pepper seeds (BPS) on growth performance, feed and nutrient utilization, body composition in *Labeo rohita*. The trial was conducted in four cemented rectangular tanks. Each tank was further sub-divided into three replicates having dimensions 2.896 x 0.762 x 0.914 (length x width x depth). There were 12 fishes *(Labeo rohita)* stocked (with pre-recorded their morphometric) in every replicate with average weight of 9.9 grams. The fish feed was formulated, prepared, pelleted and dried having 30% CP level. It was further categorized into experimental feeds on the basis of BPS i.e. 0.0, 0.5, 1.0, and 2.0 % of BPS as feed additive. The feed was dispensed twice a day, six days a week, and for 60 days. The physic-chemical parameters were maintained and recorded on daily basis. Each fish was caught every fortnightly and its weight and length was recorded. Fish fed with 0.5% (BPS) exhibited significantly higher ($P<0.05$) SGR, feed conversion ratio (FCR), % weight gain. Fish fed 0.5% BPS was had significantly higher ($P<0.05$) protein, fat, ash, fiber contents. Anyhow, no significant difference ($P>0.05$%) was found in Phosphorus contents. Therefore, it is recommended to use black pepper seeds in feeds as growth enhancer in fish at commercial scale @ 0.5% BPS.

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Introduction

Nutritionists are encouraging use of natural growth promoters in live stock 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. Studied the effects of graded levels of black pepper seeds meal (BSM) on growth performance, feed and nutrient utilization, body composition and cost-benefit analysis of Florida hybrid red tilapia juveniles were studied. (Shalaby S. M.M, *et al* 2013). This successful use of piperine to increase bioavailability of certain drugs has created interest in the area of nutrient and food absorption, since nutritional deficiencies due to poor gastrointestinal absorption are an increasing problem in developing countries as well as in Western nations. In developing countries, overall gross malnutrition may be the culprit. However, in Western nations, poor gastrointestinal absorption is increasing due to a larger percentage of elderly people in the population, as well as an increasing incidence of "junk food diets", allergies, gastric ulcers, and chronic yeast infections (Majeed *et al*, 2000).Black pepper (*Piper nigrum*) belongs to the family Piperaceae. It is a perennial woody climbing liana. It is native to India, Indonesia, Malaysia, South America and West Indies but is also widely cultivated in the tropical regions. Black pepper is a universal table condiment used to flavor all types of cuisines worldwide. It is christened as the 'King of Spices' (Srinivasan, 2007; Mathew *et al*., 2001). The spicy taste is mainly due to the presence of a compound Piperine. Piperine is a pungent alkaloid (Tripathi *et al*,1996) that enhances the bioavailability of various structurally and therapeutically diverse drugs. (Khajuria *et al.,* 2002). It also contains small amounts of safrol, pinene, sabinene, limonene, caryophyllene and linaonol compound. Black pepper is also an important traditional medicine and used to treat asthma, chronic indigestion, colon toxins, obesity, sinus, congestion, fever (Ravindran,2000), intermittent fever, cold extremities, colic, gastric ailments and diarrhoea (Ao *et al*., 1998). It has been shown to have antimicrobial activity (Dorman & Deans, 2000). Both aqueous and ethanolic extracts of black pepper have been screened for antibacterial activity against *Bacillus cereus* and *Bacillus subtilus* (Singh *et al*., 2005; Ghori & Ahmad, 2009). Aqueous decoction of black pepper exhibited maximum effect against *Staphylococcus aureus* and found to be most active antibacterial agent against all bacterial isolates except *Salmonella typhi*, *Streptococcus intermedius*, *Streptococcus mitis*, *Streptococcus salivarius*, *Streptococcus sanguis* and *Streptococcus uberis*. (Nazia Masood *et al*., 2006). Fish is highly nutritive and rich source of animal proteins. For the improvement of fisheries and to achieve maximum yields from resources of fresh water, it is necessary to provide artificial feed, by which fish grows rapidly and attains maximum weight in shortest possible time. Among commonly used feed ingredients, fish meal is considered to be the best ingredients, due to its compatibility with the protein requirement of fish (Alam *et al*. 1996). Replacement of fish meal with cheaper ingredients of plant origin in fish feed is necessary because of rising cost and uncertain availability of fish meal (Higgs *et al*. 1995). Inclusion of feedstuffs with relatively high levels of carbohydrate in formulated fish feed is preferred in view of its protein-sparing action that may make the diet more cost effective (Hidalgo *et al*. 1993). According to Rumsey (1993), increased use of plant protein supplements in fish feed can reduce the cost of fish meal. The research has focused on utilizing less expensive and readily available resources to replace fish meal, without reducing the nutritional quality of feed (EI-Sayed 1999).The apparent digestibility of protein, energy and individual amino acids are of prime consideration as the basis for feed formulation in fish, with information gained for different raw materials, such as plant by-products commonly utilized in the feed manufacturing industry. Numerous investigations have been applied to variety of fish species for several decades with digestibility data obtained for most nutrients (Tacon 1994). Attempts to use the natural materials such as medicinal plants could be widely accepted as feed additives to enhance efficiency of feed utilization and animal productive performance (Mohamed *et al*., 2003). Using medicinal herbs in human feeding is a well-known culture thousands of years in ancient Egypt, India and China. In this concern, Harada (1990) stated that caraway has strong attractant effect depending on its concentration used. The most recent studies showed successful use of spices and natural herbs in fish nutrition including marjoram, licorice roots, black seeds, peppermint, caraway seed, fennel seed, Marjoram, fenugreek seeds, ginger, Cresson, Alpinia and Ipeedeuanha as reported by Abd Elmonem *et al*. (2002); Sakr (2003); Shalaby *et al*., (2003); El-Dakar *et al*., (2004 a and b); El-Dakar (2004); Shalaby (2004); El_Dakar *et al*., (2007), El Dakar *et al*., (2008), Khalil *et al*. (2009), Al- Absawy (2010) and Abdelhamid (2010). A comparison between these plants is needed to determine the nutritional potential of each and its possibility to be used in the commercial fish feed industry. Conducted 84-day feeding and digestibility trial to evaluate the use of a pea seed derived meal in experimental diets for European Sea bass fingerlings with initial weight 10 g. (Gouveia *et al,* 1998). The effects of adding Chinese herbal medicine, enzymes preparations, cysteamine on growth performance and non-specific immune function of Eel *Anguilla anguilla*. *Anguillaanguilla* was fed with basal diet without or with adding Chinese herbal medicine **(**Jian-bin *et al,* 2010).evaluate the use of caraway seed meal (*Carumcarvi L*; CSM) as a feed additive on growth performance, feed utilization, and whole body composition of Nile tilapia (*Oreochromisniloticus*). (Ahmad *et al*, 2011) interpreted immune modulator effects of de-caffeinated green tea extract on rainbow trout (Sheikhzadeh *et al*, 2011**)** experiment to evaluate the use of *Epilobiumhirsutum* extract in diet of common carp, *Cyprinus carpio*.(Pakravan *et al*, 2011). Herbal additives enhance the growth of fishes and protect them from diseases. Inclusion of herbal additives in diets often provides cooperative action to various physiological functions. Beneficial role of vitamins C and E have been reported in fish nutrition, reproduction, growth and related indices (Govind *et al*, 2012). The efficacy of green tea (*Camellia sinensis*) on the growth performance, immune and antioxidant systems and cytokine gene expression in rainbow trout tissues (Nootash *et al*, 2013).The present study was conducted to evaluate the effect of Black pepper seed, at three levels (0.5; 1.00 and 2.00 % BPS) on growth performance, feed conversion, feed utilization, whole body composition fingerlings reared in tanks.

Materials and methods

The present study was conducted at Research and Training Facilities of Department of Fisheries and Aquaculture, University of Veterinary & Animal Sciences, Ravi Campus, Pattoki. The experiment was conducted in the fish hatchery premises of the same department.

The fish was procured from fish production ponds at UVAS Ravi campus, Pattoki. Feed ingredients were collected from Choburgi, Lahore. Trials was conducted in cemented tanks located inside the hatchery building having dimensions 2.896x0.762x0.914 (length x width x depth).

Feed Formulation

The following feed formula was used during the course of this experiment.

Fish was analyzed for its growth efficiency, proximate composition after completion of the experimental trials.

Experimental Protocol

There were 03 treatments groups (T1, T2, and T3) and one control (T0). Control group was fed on same diet (mentioned in Table 1) but it was devoid of Black pepper seed whereas the treatment groups were fed with above given feed recipe with 0.5 %,(T1) 01 % (T2) and 02 %(T3) of Black pepper seed in treatments one, two and three respectively. Twelve *Labeorohita* having size ranges from 10-12 g of each were stocked in each treatment tank. Each treatment group had two replicates. At the time of stocking, the morphometric characteristics of fish viz. wet fish body weight and total length were measured and recorded.

Feeding

Fish was regularly fed at the rate of 4% of its body weight twice a day.

Physico-chemical parameters

Physico-chemical parameters such as dissolved oxygen, pH, electrical conductivity and water temperature, total dissolved solids and salinity were monitored and recorded on daily basis by using DO meter (YSI 55 Incorporated, Yellow Springs, Ohio, 4387, USA), pH meter (LT-Lutron pH-207 Taiwan) and electrical conductivity meter (Condi 330i WTW 82362 Weilheim Germany), respectively was measured in the morning and evening on daily basis.

Growth Parameters

At the end of each week, all the fish were caught for morpho-metric records and were released back to their respective tanks. Fishes were measured and weighed and mortality was also recorded if found. At the end of feeding trial, water was totally drained off and all the fish were collected for final morpho-metric measurements. Other growth parameters like Feed Conversion Ratio (FCR) and Specific Growth Rate (SGR) were calculated according to following formulas:

FCR= feed given / weight gain

SGR= $(\ln W_f - \ln W_i \text{ x}100) / T$

Where W1, W2, and T are the initial weight, final weight, and number of days in the feeding trial, respectively.

Proximate analysis

At the end of trial five fish from each tank were caught and used for the estimation of following proximate records.

- Crude protein (CP)
- Fat Estimation
- Ash Contents
- Fiber Consents
- Phosphorus

All these records were noted using NIR (Buchi, made in Switzerland) at Buffalo Research Institute, Pattoki.

Statistical analysis

The data obtained was analyzed using SAS 9.1 version statistical software.

The data on different variables was statistically analyzed by using Analysis of Variance (ANOVA) technique under Duncan's
Multiple Range Test.

Results

The present study resulted into three types of results including the physic-chemical properties, growth estimation as a result of different concentrations of Black Pepper Seed and the last step was the estimation of proximate analysis of fish meat raised during the trail. The results can be discussed under the following headings.

Table 1. Fish Feed Formula used with different doses of Black Pepper Seed powder.

Sr. No.	Ingredient Used	Amount Used
1	Fish Meal	300 g
2	Soybean Meal	500 g
3	Rice Polish	440 g
4	Cotton Seed Meal	380 g
5	Corn Glutton	340
6	Vitamin Premix	2 g
7	Chromic Oxide	2 g
8	Black Pepper Seed	0.0 %, 0.5%, 01 %, 02 %

Physico-Chemical Properties

During this study, following physico-chemical factors were recorded and are discussed as under In the present study, the physico-chemical parameters were considered important during the course of this experiment. In this study, all the parameters were found highly significant, such as the pH was noted as highly significant in T 1 as compared to To, T2 and T3 but there were no significant differences between the all treatments shown is (Table 2). Same as in this study the Table 2. Summarized results of Physico-Chemical parameters Temperature were study in the all treatment they were highly significant but there were no significant variation. Same as they DO in which no significant difference were found shown in the (Table 1).about TDS were study which were more significant in the T1 as compare to the To,T2 and T3 shown in the (Table 2).but in this no significant variation in the TDS. Salinity were also studied the highly significant in the T1.but there were no significant variation in the salinity as shown in the (Table 2).

Table 2. Summarized results of Physico-Chemical parameters.

Treatments	pH	Temperature C°	DO mg/l	TDS mg/l	Salinity
To (0%)	8.21±0.19^{aa}	24.33±0.44^{aa}	4.6±0.45^{aa}	2345.45±896.77^{aa}	1.44±0.33^{aa}
T1 (0.5%)	8.01±0.13^{a}	24.54±0.41^{aa}	3.64±0.63^{aa}	1231.85±945.64^{a}	1.11±0.23^{a}
T2 (1%)	8.34±0.23^{aa}	25.45±0.66^{aa}	4.33±0.55^{aa}	2453.56±963.66^{aa}	1.22±0.43^{aa}
T3 (2%)	8.23±0.29^{aa}	25.11±0.47^{aa}	4.12±0.60^{aa}	1891.36±951.83^{aa}	1.33±0.32^{aa}

Dissolved Oxygen (DO)

Dissolved oxygen recorded during the trail is represented via as chart as shown in the figure 1. It describes that the highest value for DO was noted in the treatment T 3 whereas the lowest was recorded in the control tank. It also shows that overall values for DO from 3.59 to 3.70 mg/l.

Table 3. Proximate composition of fish samples.

Treatment	Fats	Fiber	Ash	Phosphorus	Protein
T o (Pre-Treatment)	2.89± 0.03^{d}	2.51± 0.14^{d}	10.61 ±0.08^{b}	0.40±0.14^{aa}	44.83±0.12^{c}
T 1 (1%)	28.77± 0.15^{aa}	9.29± 0.07^{c}	2.14±0.13^{d}	0.33±0.15^{aa}	46.37±0.36^{b}
T 2 (2%)	23.42±0.31^{b}	9.95±0.03^{b}	9.36±0.40^{c}	0.45±0.02^{aa}	44.14±0.12^{d}
T 3 (0.5%)	28.39±0.06^{a}	16.39±0.07^{aa}	0.26±0.04^{e}	0.42±0.02^{a}	47.37±0.24^{a}
T 4 (0%)	8.30±0.43^{c}	16.19±0.04^{a}	16.19±0.04^{a}	0.29±0.06^{cs}	44.27±0.09^{dd}

Table 4. Weight, Length, FCR, and SGR Relationship for all treatments.

Parameters	T_0 0%	$T_1$1% B.PP	$T_2$2% B.PP	$T_3$0.5% B.PP
Initial wt	9.91± 0.04^{b}	10.39±0.07^{a}	10.49±0.07^{aa}	9.40±0.07^{c}
Final wt	19.61±0.01^{d}	27.08±0.05^{b}	25.19±0.07^{c}	28.23±0.14^{a}
Initial length	8.13±0.05^{b}	8.49±0.09^{a}	8.56±0.07^{aa}	7.92±0.04^{c}
Final length	13.44±0.04^{bb}	14.11±0.02^{a}	13.17±0.07^{c}	13.39±0.08^{b}
FCR	3.71±0.03^{a}	2.73± 0.08^{ccb}	2.91±0.06^{b}	2.67±0.09c
SGR	0.52± 0.00^{c}	0.70±0.06^{bb}	0.65± 0.02^{ccb}	0.85±0.07^{a}
% WT GAIN	97.17± 0.07^{c}	166.19±0.20^{b}	138.74± 0.25^{d}	183.18±0.05^{a}

Temperature

Temperature plays a very crucial role in the fish culture either in the laboratory conditions or in pond culture system. It greatly affects the feed intake in the fish. The same phenomenon is observed during this research trail. The highest value of temperature was recorded as 24.83 on average in the control tank but on the whole, there was a very slight change in the temperature in all the four treatments as shown in the figure 2.

pH

Figure 3 shows the pH records of the research experiment. Average pH recorded in all the four treatment tanks ranges from 7.90 to 8.09. It also shows very slight difference on the whole. But control tank showed the highest value of pH as compared to other research tanks.

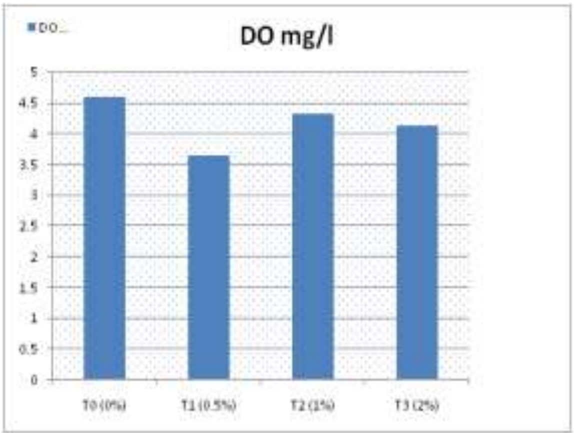

Fig. 1. Showing Dissolved Oxygen (mg/l) recorded during experiments.

Salinity

Salinity is measured in mg/l and is very important phenomenon which affects the fish growth to a little extent. In the present study, highest salinity level noted was in the control tank and the lowest was noted in the treatment 3 on the average.

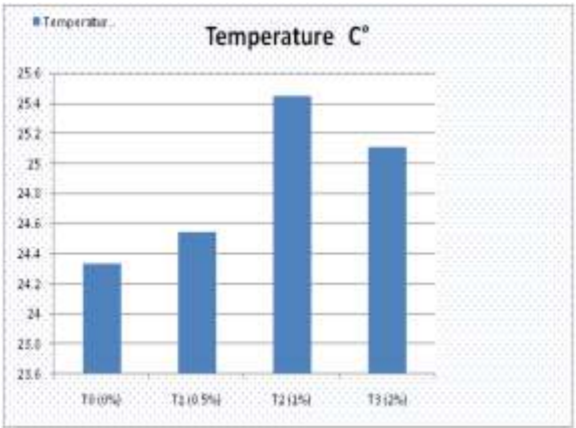

Fig. 2. Temperature records during experiment.

Total Dissolved Solids (TDS)

TDS showed significantly varied vales during this study. The highest value on the average was recorded in the control and treatment tank number one whereas the lowest was recorded on the treatment 2.

Proximate records

The significant ($p<0.05$)difference was found in the whole body fat content of fish fed with different experimental diets containing Black pepper seed (BPS) as micro-additive. Whole body fat content was observed significantly higher ($p<0.05$)in T1 & T3 compared to T0, T2 & T4, respectively. Whereas in pretreated fish fat contents are significantly lower than all the treated groups (Table 3).

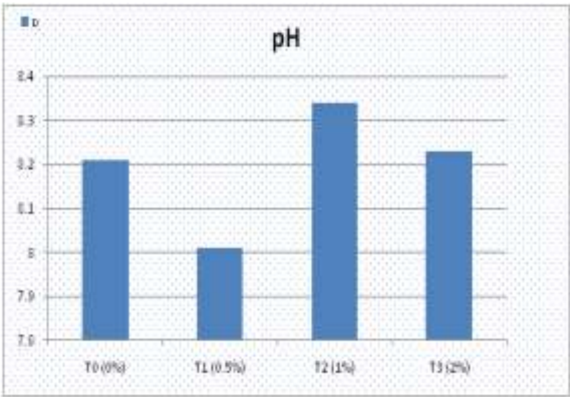

Fig. 3. pH records during experiment.

Ash contents also revealed significant variations among various treatment groups such as using the different fed diets such as the higher significant ($p<0.05$) T4, T0 and T2 than T1 and T3 which contain 0% ash (Table 3).

In this proximate analysis the protein was also estimated. There were large variations observed in the protein value using different diets. The significantly higher ($p<0.05$) values of the protein were in the T3 and T1 followed by T0 lower in T2 & T4, respectively (Table 2).

Fig. 4. Salinity records during experiment.

Whole body fiber was significantly different in the different diets such as T3 and T4 were significantly higher followed by T1, T2 and T0 (Table 3).

On the other hand, whole body Phosphorus content of fish numerically varied between treatments but statistically non-significant except T4 which exhibited significantly lower value (Table 3).

In this study, fish growth was enhanced significantly (P<0.05) with Black pepper seed (BPS) supplemented as feed additive as compared to the control diet (Table 4). Moreover, the highest final weight and weight gain percentage were obtained at a diet containing 0.5% BPS diet ($28.23\pm\ 0.14^{a}$g) and ($27.08\pm\ 0.05^{b}$g), respectively. In addition, the difference in fish performance at 0% and 2% BPS were insignificant. No significant differences were observed in survival among the treatments since its range was 25.19 ± 0.07^{c} (P<0.05); Table 4). The lowest and higher growth performances (final weight and weight gain %) were observed for fish fed the diets containing 0.0 (control) and 2% BPS diet, respectively.

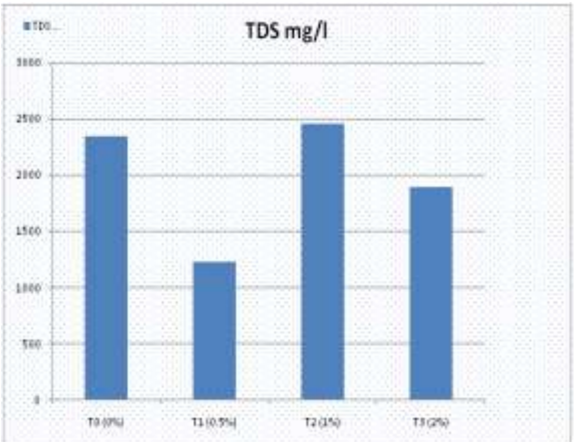

Fig. 5. TDS records during experiment.

In table (Table 4) showed that the most suitable BPS level for maximum growth was determined to be 0.5%. Feed intake increased significantly, while FCR decreased significantly (P<0.05) when fish fed BPS- as additive diets compared to that fed on a control diet (Table 4). Feed intake and apparent protein utilization showed similar trend with final weight and weight gain % and their relationships with dietary BPS levels. The significant difference was observed for final body weight (FBW) and for specific growth rate (SGR) among fish fed diets 0.0%, 0.5%, 1% and 2% which had the best growth rates (Table4). The significant FBW and SGR were in the 0.5%, 1% BPS diets but the FBW of the 0% was the lowest but they was highly significant. The 2% BPS fed diet showed a slightly lower FBW and significant SGR and the values were different from the other groups (Table 4).

Discussion

Black pepper has longer been used as an important spice used in luxurious cooking style all over the world. In the present study, fish feed containing Black pepper seed (BPS) had significantly better growth and feed utilization as compared to fish fed the control diet with 0% BPS. This proved that dietary supplementation of BPS enhanced the fish growth. This is in agreement with significant increased weight gain, feed efficiency, protein efficiency ratio (PER) and specific growth rate (SGR) in the *Labeo rohita* when fed diet containing BPS powder. Furthermore, feeding diet with 0.5% BPS resulted /kg diet in the highest growth performance in the *Labeo rohita*. It was observed a positive improvement in biomass and specific growth rate with BPS supplementation. Although growth is enhanced with BPS supplementation, high dose of BPS in fish may reduce feed intake as a result of its unpleasant odor. It can be an arguable statement that the effect of BPS as growth promoter for fish, the present results suggest that dietary BPS for fingerling of *Labeo rohita* could positively affect growth and survival. Black pepper seed is believed to cure illness such as constipation, diarrhea, earache, gangrene, heart disease, hernia, hoarseness, indigestion, insect bites, insomnia, joint pain, liver problems, lung disease, oral abscesses, sunburn, tooth decay, and toothaches, skin care agent, impotency, muscular strains, etc pyorrhea. Black pepper is a rich source of manganese, iron, potassium, vitamin-C, Vitamin-K and dietary fiber. Various sources from the 5th century onward also recommend pepper to treat eye problems, often by applying salves or poultices made with pepper directly to the eye. The U.S. Food and Drug Administration (FDA) has approved black pepper, black pepper oil, black pepper oleoresin, piper dine, and Piperine as Generally Recognized as Safe (GRAS) for use in foods in the United States. The improve survival rate may be due to the enhanced immune response resulting from improved defense mechanism. In the present study it is concluded that BPS affects the protein level as the protein were noted the highest in 0.5% BPS additive.

References

Ahmad HM, Abdel-Tawwab M. 2011. The use of caraway seed meal as a feed additive in fish diets: Growth performance, Feed utilization, and whole-body composition of Nile tilapia, *Oreochromisniloticus* (L.) fingerlings. Aquaculture **314,** 110–114.

Arias Irigoyen J, Talavera Fabuel A, Maranon Lizana F. 2003. Occupational rhinoconjunctivitis from white pepper. J InvestigAllergolClinImmunol **13(3),** 213-215.

Boonyaratpalin M, Suraneiranat P, Tunpibal T. 1998. Replacement of fish meal with various types of soybean products in diets for the Asian sea bass, *Latescalcarifer*. Aquaculture **161,** 67–78.

Burel C, Boujard T, Tulli F, Kaushik SJ. 2000. Digestibility of extruded peas, extruded lupin, and rapeseed meal in rainbow trout (*Oncorhynchusmykiss*) and turbot (*Psetta maxima).* Aquaculture **188,** 285–298.

Carter CG, Hauler RC. 1999. Fish meal replacement by plant meals in extruded feeds for Atlantic salmon, (*Salmosalar*L). Aquaculture **185,** 299–311.

Chaudhry NM, Tariq P. 2006. Bactericidal activity of black pepper, bay leaf, aniseed and coriander against oral isolates. Pak J Pharm Sci **19(3),** 214-218.

Ebihara T, Ebihara S, Maruyama M. 2006. A randomized trial of olfactory stimulation using black pepper oil in older people with swallowing dysfunction. J Am GeriatrSoc; **54(9),** 1401-1406.

D'Souza P, Amit A, Saxena VS. 2004. Antioxidant properties of Aller-7, a novel polyherbal formulation for allergic rhinitis. Drugs ExpClin Res; **30(3),** 99-109.

Fontaínhas-Fernandes A, Gomes E, Reis-Henriques MA, Coimbra J. 1999. Replacement of Fish Meal by Plant Proteins in the Diet of Nile Tilapia: Digestibility and Growth Performance. Aqua. Inter **7,** 57-67.

Gomes EF, Rema P, Kaushik SJ. 1995. Replacement of fish meal by plant proteins in the diet of rainbow trout (*Oncorhynchusmykiss*): digestibility and growth performance.Aquaculture **130,** 177–186.

Gouveia A, Davies SJ. 1998. Preliminary nutritional evaluation of pea seed meal (*Pisumsatium*) for juvenile European sea bass (*Dicentrarchuslabrax).* Aquaculture **166,** 311–320.

Govind P, Madhuri S, Sahni YP. 2012. Beneficial effects of certain herbal supplements on the health and disease resistance of fish. Novel Science International Journal of Pharmaceutical Science **1(7),** 497 -500.

Hu Z, Yang X, Ho PC. 2005. Herb-drug interactions: a literature review. Drugs; **65(9),** 1239-1282.

Ingkaninan K, Temkitthawon P, Chuenchom K. 2003. Screening for acetylcholinesterase inhibitory activity in plants used in Thai traditional rejuvenating and neurotonic remedies. J Ethnopharmacol **89(2-3),** 261-264.

Jian-bin L, Qing-guo Z, Ping L, Du-huang C, Zhi-qing Q, Chun-xiang A. 2010. Effects of different feed additives and combinations on growth and non-specific immunity of European Eel (*Anguilla anguilla*). J Shanghai Ocean University 06.

Kaushik SJ, Cove SD, Dutto G, Blanc D. (2004).Almost total replacement of fish meal by plant protein sources in the diet of a marine teleost, the European seabass, *Dicentrarchuslabrax*. Aquaculture

230, 391–404.

Lambert JD, Hong J, Kim DH. 2004. Piperine enhances the bioavailability of the tea polyphenol (-)-epigallocatechin-3-gallate in mice. J Nutr; **134(8),** 1948-1952.

Mbahinzireki GB, Dabrowski K, Lee KJ, El-Saidy D, Wisner ER. 2001. Growth, feed utilization and body composition of tilapia (Oreochromis sp.) fed with cottonseed meal-based diets in a recirculating system. Aqua Nut. **7,** 189-200.

McNamara FN, Randall A, Gunthorpe MJ. 2005. Effects of piperine, the pungent component of black pepper, at the human vanilloid receptor (TRPV1). Br J Pharmacol; **144(6),** 781-790.

Nootash S, Sheikhzadeh N, Baradaran B, Oushani AK, Moghadam MRM, Nofouzi K, Monfaredan A, Aghebati L, Zare F, Shabanzadeh F. 2013. Green tea (*Camellia sinensis*) administration induces expression of immune relevant genes and biochemical parameters in rainbow trout (*Oncorhynchus mykiss*). Fish & Shellfish Immunology **35,** 1916-1923.

Oliva-Teles A, Gouveia AJ, Gomesb E, Remac P. 1994. The effect of different processing treatments on soybean meal utilization by rainbow trout (*Oncorhynchus mykiss)*. Aquaculture **124,** 343-349.