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Full Length Research Paper

Replacement of Fish Meal with Poultry By-product Meal (PBM) and its effects on the Survival, Growth, Feed Utilization, and Microbial Load of European Seabass, *Dicentrarchus Labrax* fry

Srour, T. M.[1], Essa, M. A.[2*], Abdel-Rahim,M. M.[2]., Mansour, M. A[1].

[1] Faculty of Agriculture - Saba Basha, Alexandria University, Egypt.
[2] National Institute of Oceanography and Fisheries (NIOF), Alexandria, Egypt.

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Replacement of fishmeal with poultry by-product meal (PBM) on an ideal *isonitrogenous*/*isocaloric* diets for seabass was evaluated. Four diets were formulated to contain 48% protein, 14% lipid and 4.8 kcal/g. In these diets, fish meal was replaced with PBM at levels 0, 20, 40, and 60%. Fish fry (0.73 gm/pcelW) were stocked in 12 fiberglass tanks at density of 25 fry/m^3. Fish were fed at 10% BW/day through three meals and gradually decreased to 6% during the last week of experiment that continued for 70 days. Results of survival, growth, feed utilization, carcass composition and microbial load were evaluated. Growth performance results showed a non-significant differences between control and both 20% PBM and 40%PBM. However, the best growth rate was obtained at 60% PBM giving the best SGR (3.52%/day). Contrary, survival decreased significantly ($P < 0.05$) at 60% PBM (94%), while other treatments showed no mortalities. FCR, PER, PPV, EU values revealed that replacing fish meal with PBM did not negatively affect the efficiency of feed utilization and the best results were obtained at 60% PBM. Protein and lipid content in fish carcass increased in PBM diets. Aeromonas, salmonella, and E. coli in fish carcass and rearing water were less than control. Results clearly demonstrated that, replacement of fish meal with PBM up to 60% of Seabass diets is acceptable. However, replacing 40% PBM instead of fish meal is the optimum level in terms of keeping both fish survival and fish welfare in the best levels.

Keywords: European Seabass, Nutrition, Poultry By-Product (PBM), survival, growth, feed utilization, microbial load.

INTRODUCTION

European seabass (*Dicentrarchus Labrax*) is common valuable cultivated fish exists all over the Mediterranean, the Black Sea and the North Eastern Atlantic from Norway to Senegal. It inhabits coastal marine waters, as well as brackish waters in estuarine areas and occasionally it can

*Corresponding Author's Email: messa51@yahoo.com;
Tel: +201006635376.

be found in rivers. European Seabass is one of the most important marine fishes for the Egyptian marine aquaculture. In Egypt, despite the rapid growth of aquaculture, still marine aquaculture contributes with less than 5% of the total Egyptian fish production (GAFRD, 2014). Total production of fish from aquaculture and fisheries was 1,481882, aquaculture sector shared with 1,137,091 tons (76.73%), while the total production of Seabass was 16447 tons contributing 1.11 % of the total Egyptian fish production (aquaculture and Fisheries), and 15167 tons from fish farms (1.33 % from aquaculture sector) (GAFRD, 2015). One of the most important reasons for this situation of marine aquaculture in Egypt is because of the high price of the artificial feeds (Essa, 2013).

Feed manufacturing of marine aquaculture is highly dependent on protein ingredient supplies, especially on fishmeal, that is being considered the indispensable source of protein (Badillo *et al*., 2014) and fish oil. According to Tacon and Metian (2008), 75% and 80% (FAO, 2014) of the world's fish stocks are currently considered as fully exploited or overexploited, including many small pelagic fish species used to produce fishmeal for feed formulation worldwide. Since fishmeal production is forecast to be unable to support the growth of the world aquaculture sector, the search for alternative ingredients as protein sources and the optimization of dietary protein content is an important goal (NRC, 2011). Marine fish meals and oils are superior sources of essential amino and fatty acids and may enhance diet palatability for some carnivorous species (Hertrampf and Piedad-Pascual, 2000). However, adding high contents of those ingredients in the diets of marine fishes is the main reason for its high cost. Therefore, the replacement of fish meal with alternative less cost protein sources in the diets of marine valuable fishes, such as Seabass and sea bream still welcomes and required topic in the area of fish nutrition to reduce the production cost (Uran *et al*., 2008).

Poultry by-product meal (PBM) is also a viable propitious protein source for carnivorous fishes, because of it is high content in protein and a favorable profile of indispensable essential amino acids (IAA) for monogastric animals and fishes (Tacon, 1993; Gaylord and Rawles, 2005). In addition, high availability and lower price render PBM as an ideal candidate for replacing fish meal in aquafeeds. The composition of PBM depends on processing conditions and the source of raw materials (Johnson and Parsons, 1997). Some PBM have a very high protein content (75-90%) with relatively low contents of ash (less than 10%) and fat (less than 15%). Other PBM are of lesser quality, with a protein content (55-75%), higher levels of ash (up to 15%) and fat (more than 15% and up to 30%) (Heuzé et al., 2015). However, some Poultry by-products are deficient in one or more essential amino acids (Davies *et al*., 1991). The most important and valuable use for poultry by-product meal is as feed ingredients for livestock, poultry and aquaculture **(**Meeker, 2006**;** Laban, *et al.,* 2014)**.**

Poultry by-product meal has been tested in a wide range of fish species, including Common carp (*Cyprinuscarpio* L.) (Paixao *et al*., 1989); Grass Carp (Tabinda and Butt, 2012); Chinook salmon *Oncorhynchus tshawytscha* (Fowler, 1991), palmetto bass Morone saxatilis × Morone chrysops (Gallagher and La Douceur, 1995), Sunshine bass Morone chrysops × Morone saxatilis (Webster *et al*., 2000), largemouth bass (Tidwell *et al*., 2005), Black Sea Turbot (Turker *et al*., 2005), Gilt-head Sea bream (*Sparus aurata*) (Lupatsch *et a*l., 1997), African catfish *Clarias gariepinus* (Abdel-Warith *et al*., 2001), catfish (*Pangasianodon hypophthalmus*) (HAO and YU, 2003), rainbow trout (Pfeffer *et al*., 1995; Badillo et al., 2014), Nile tilapia (*Oreochromis niloticus)* (Metwalli, 2008; El-Husseiny *et al*., 2006; Yones, and Metwalli, 2015), Tilapia Zilli (Yildrim *et al.,* 2009), *Totoaba macdonaldi* (Zapata *et al*., 2014), Malabar grouper (Wang *et al*., 2008); Hybrid Striped Bass, (Rawles *et al*., 2009) and Golden Pompano, (Ma *et al*., 2014). Generally, partial replacement of fish meal with PBM can be successfully applied in aquafeeds, but amino acid supplementation may be required. Reduced digestibility of protein and essential amino acids from PBM may limit its use in diets for marine fish to a certain limit (Portz and Cyrino, 2004). Recommended inclusion levels of poultry by-product meal for fish are in the range of 5-25% (Tacon *et al*., 2009).

In Egypt, more than five hundred thousand tons of PBM is available yearly (MALR, 2015). This huge quantity of PBM is causing some serious environmental problems such as vectors for insects, vermin, bacteria and viruses, which may result in water contamination (leaching of nutrients and pathogenic microorganisms) and air pollution (noxious gases and nuisance odorants) (FAO, 2011). Therefore, we intended from this study to treat this problem, add value to this by-product and reduce the cost of marine aquafeeds. In this study, we investigated the replacement of fish meal with different levels of Poultry By-product Meal (PBM) and its effects on fish survival, growth, feed utilization, Carcass composition and microbial load of European Seabass (*Dicentrarchus labrax*) fry.

MATERIALS AND METHODS

Experimental Research Station

The present study was carried out at El-Max Research Station, National Institute of Oceanography and Fisheries (NIOF), Alexandria, with co-operation with Department of Animal and Fish Production, Faculty of Agriculture (Saba-Basha), Alexandria University, Egypt.

Experimental Fish and Rearing Facilities

European sea bass (*Dicentrarchus Labrax*) fry with average initial size of 0.73 ± 0.01 gm/fish were purchased from the Marine Finfish Hatchery, K21, GAFRD and stocked in fiberglass tanks at initial density of 25 fry/m^3. The. Twelve fiberglass tanks each of 1 m^3water volume were used to evaluate four experimental diets. The tested treatments were: control, 20, 40, and 60 % replacement of fish meal with poultry by product meal (PBM). The rearing condition were as follows: water temperature averaged 20±1°C, continuous aeration using air blower and the daily water exchange rate was 50% with saline underground water with salinity 32±1 ppt. This experiment lasted for 70 days.

Experimental diets

All the feed ingredients used in this study were purchased from local market. According to information provided by the manufacturer, PBM used consists of chicken slaughter wastes including viscera, heads, legs and feather, and was produced by exposing to 150-200 °C under a 2.5 bar (atmospheric pressure) for ten hours. Both fish meal and PBM were analyzed for proximate composition prior to the formulation of diets. The chemical analyses of PBM showed the following values: 57.05% Crude Protein, 48.02% Digestible protein, 23.20% fat, 10.13% carbohydrate, 11.85% fiber, 2.24% calcium, and 1.12% phosphorus. The experimental diets were prepared in El-Max Research Station. All feed ingredients were finely ground, mixed well, manufactured to the tested diets with suitable mesh sizes, and then dried and kept in refrigerator until were offered to the experimental fish. The Ingredients and chemical composition (%) of the tested diets are shown in Table (1).

Feeding regime

Experimental Fish were hand-fed three meals per day at 9:00 a.m., 12.00 and 15:00 p.m., seven days a week at 10% daily feeding rate. After that Feeding rate was re-adjusted depending on the live fish body weights every two weeks according to (Moretti *et al.,* 1999).

Isolation and identification of pathogenic Bacteria

Samples from internal organs of the examined fish were cultured in brain heart infusion broth (Oxoid) and Rappaport Vassilidis soya broth and incubated at 37ºC for 18-24 hrs. After incubation, loopful of cultured broth were streaked onto Aeromonas agar medium, XLD and EMB (Lab M) plate, then incubated at 37ºC for 24-48 hr. The growing colonies were picked up in pure form and the identification of all isolates was done by cultural, morphological and biochemical characters according to Quinn *et al*. (2002) and Austin and Austin (2007).

Measured parameters

Survival and Growth performance

Final body weight (FW), weight gain (WG), average daily gain (ADG), specific growth rate (SGR), survival rate, length, length gain and condition factor were measured according to the following equations:

Weight gain (g/fish): $WG = W_t - W_0$

Where: W_0: initial mean weight of fish in grams, W_t: final mean weight of fish in grams.

Average daily gain (g/fish/day): $ADG = W_t - W_0/n$

Where: n: experimental period.

Specific growth rate (%/day): $SGR = 100 \times (\ln W_t - \ln W_0)/$ days

Where: ln: natural logarithm.

Survival rate (%) = 100 × (final number of fish/initial number of fish)

Length gain (cm)= $L_t - L_0$

Where:L_0: initial mean length of fish in cm,L_t: final mean length of fish in cm.

Condition factor = 100 × (BW (g)/ L^3 (cm)).

Feed Utilization

Feed Intake (g/fish): This is the amount of feed given or supplied during the experimental period for each fish per gram.

Feed conversion ratio (**FCR)** and protein efficiency ratio (**PER**) were estimated according to the following equations:

Feed conversion ratio (FCR) = dry matter feed intake/ gain.

Protein efficiency ratio (PER) = gain/protein intake.

Protein productive value (PPV %) =100 * (Pt –P0/protein intake (gm)). Where: P0: Protein content in fish carcass at the start.

Pt: Protein content in fish carcass at the end.

Energy utilization (EU, %) = 100 [Energy gain (Kcal/100g)/ Energy intake (Kcal/100g)].

Statistical Analyses

Mean values and standard Error (mean ± S.E) for every studied parameter were first calculated. Results were subjected to one-way analysis of variance (ANOVA), using IBM SPSS statistics 22 software package (SPSS® IBM, Chicago, IL, USA) to evaluate the effect of the tested diets on survival, growth performance, and feed utilization. Differences between variables were further examined using LSD test if significant differences were detected in ANOVA according to SPSS (version 22). The significant level was set at $P<0.05$ 9 (Steel and Torrie, 1980).

Table 1: Ingredients and Chemical Composition (%) of the Experimental Diets.

Ingredients, gm	*PBM Replacement, %*			
	0	*20*	*40*	*60*
Fish Meal	*47.35*	*37.88*	*28.41*	*18.94*
poultry by product meal	*0*	*9.47*	*18.94*	*28.41*
Soybean Meal, 48%	*10.98*	*12.22*	*13.35*	*14.58*
Corn gelatin, 60%	*10.98*	*12.22*	*13.35*	*14.58*
Wheat flour	*13.92*	*14.78*	*12.78*	*12.03*
Vitamins and Minerals primex1	*6.15*	*6.15*	*6.15*	*6.15*
Fish oil.	*10.61*	*8.81*	*7.01*	*5.30*
Total	*100*	*100*	*100*	*100*
Chemical composition (%) on dry matter basis				
Dry matter (DM)	*92.44*	*92.24*	*92.04*	*91.85*
Crude protein (CP)	*48.37*	*48.41*	*48.35*	*48.36*
Ether extract (EE)	*14.29*	*14.28*	*14.27*	*14.35*
Crude fiber (CF)	*1.81*	*2.98*	*4.15*	*5.32*
NFE	*18.67*	*17.82*	*17.07*	*16.17*
Ash	*9.30*	*8.75*	*8.20*	*7.65*
Gross energy (kcal /kg DM)2	*4844.4*	*4810.7*	*4775.6*	*4746.7*
*Gross energy (MJ /kg DM)*3	*20.28*	*20.14*	*19.99*	*19.87*

[1]Composition of vitamin mineral mixture of 1 kg: Vitamin A - 50,00,000 IU; Vitamin D_3 - 10,00,000 IU; Vitamin B_2 - 2.0 g; Vitamin E - 750 units; Vitamin K - 1.0 g; Calcium pantothenate 2.5 g; Nicotinamide - 10.0 g; Vitamin B_{12} - 6.0 g; Choline Chloride - 150.0 g; Calcium - 750.0 g; Manganese - 27.5 g; Iodine - 1.0 g; Ion - 7.5 g; Zinc - 15.0 g; Copper - 2.0 g; Cobalt - 0.45 g.

[2] GE: Gross energy calculated on the basis of 5.64, 9.44 and 4.11 kcal gross energy/g protein, ether extract and NFE, respectively (NRC, 1993).

3 GE: Gross energy calculated on the basis of 23.6, 39.4 and 17.2 k joule grossenergy/ gprotein, ether extract and NFE respectively.

Table 2: Effect of Replacement of Fish Meal with Poultry By-product Meal (PBM) on the Growth Performance of Sea Bass, *Dicentrarachus labrax* fry.

Treatments (PBM, %)*	Initial Weight(g)	Final Weight(g)	Weight Gain (g)	Average Daily gain (g/fish/day)	Specific growth Rate (%/day)
0	0.73 ± 0.00	6.94 ± 0.04^{b}	6.21 ± 0.04^{b}	0.09 ± 0.00^{ab}	3.22 ± 0.01^{b}
20	0.75 ± 0.02	6.71 ± 0.38^{b}	5.96 ± 0.36^{b}	0.08 ± 0.01^{b}	3.13 ± 0.06^{b}
40	0.73 ± 0.06	7.34 ± 0.03^{ab}	6.61 ± 0.08^{ab}	0.10 ± 0.01^{ab}	3.31 ± 0.11^{ab}
60	0.71 ± 0.01	8.28 ± 0.10^{a}	7.57 ± 0.11^{a}	0.11 ± 0.00^{a}	3.52 ± 0.03^{a}

*Means with different superscript letters within a column are significantly different (P<0.05)

RESULTS

Growth Performance

Final results of final weight, weight gain, average daily gain, and specific growth rate revealed that replacement of fish meal with PBM increased significantly ($P < 0.05$) the growth performance of Seabass fingerlings and the best value was obtained at 60% PBM replacement without significant ($P > 0.05$) differences with treatment of 40% PBM (Table 2).

Survival Rate and Condition Factor

Incorporating seabass feed with 60% PBM instead of fish meal gave the lowest survival rate (94%)with significant($P < 0.05$) differences with the other treatments, where there were no mortalities (Table 3). Results of

Table 3: Effect of Replacement of Fish Meal with Poultry By-product Meal (PBM) on the survival rate and condition factor of Sea Bass, *Dicentrarachus labrax* fry.

Treatments (PBM, %)*	Survival (%)	Final length (cm)	Length Gain (cm)	Condition Factor (CF)
0	100.0 ± 0.00^a	8.20 ± 0.10^b	3.99 ± 0.10^b	1.26 ± 0.04
20	100.0 ± 0.00^a	8.60 ± 0.06^a	4.39 ± 0.06^a	1.06 ± 0.08
40	100.0 ± 0.00^a	8.60 ± 0.10^a	4.39 ± 0.10^a	1.16 ± 0.04
60	94.0 ± 2.00^b	8.80 ± 0.10^a	4.59 ± 0.10^a	1.22 ± 0.03

*Means with different superscript letters within a column are significantly different (P<0.05)

Table 4: Effect of Replacement of Fish Meal with Poultry By-product Meal (PBM) on the feed utilization of European Sea Bass, *Dicentrarachus labrax* fry.

Treatments (PBM, %)*	FCR	PER	PPV	ER	EU
0	2.58 ± 0.18	0.82 ± 0.06	13.84 ± 1.22	43.54 ± 2.95	7.06 ± 0.02
20	2.87 ± 0.25	0.74 ± 0.06	12.00 ± 0.87	32.59 ± 5.16	4.98 ± 0.67
40	2.69 ± 0.28	0.79 ± 0.08	12.47 ± 0.71	40.94 ± 0.15	6.12 ± 0.69
60	2.24 ± 0.18	0.94 ± 0.08	15.13 ± 1.17	42.00 ± 1.01	6.51 ± 0.28

*Means with different superscript letters within a column are significantly different (P<0.05)

Table 5: Effect of Replacement of Fish Meal with Poultry By-product Meal (PBM) on the Carcass analysis of European Sea Bass.

Treatments (PBM, %)*	Dry Matter (%)	Crude Protein (%)	Crude Lipids (%)	Crude Ash (%)
0	28.04 ± 0.0^b	54.21 ± 0.00	12.36 ± 0.00	18.89 ± 0.00^a
20	28.85 ± 0.4^{ab}	56.65 ± 1.23	16.06 ± 1.92	17.54 ± 0.69^{ab}
40	29.25 ± 0.0^a	54.21 ± 1.06	17.28±0.77	17.12 ± 0.76^{ab}
60	29.25 ± 0.0^a	53.10 ± 0.58	16.95 ± 0.76	15.67 ± 0.48^b

*Means with different superscript letters within a column are significantly different (P<0.05)

Condition factors (CF) revealed that treatments supplemented with PBM, improved condition factor but still lower than the control one with the value of CF (1.22) at 60% PBM treatment and 1.26.for control without any significant (P > 0.05) differences between treatments.

Feed Utilization and Carcass composition

The results of feed utilization efficiency were illustrated in Table (4). Data showed that, there were no significant (P>0.05) differences in FCR, PPV, PER, and ER between treatments. Generally, the lowest value of FCR and the highest values of protein utilization efficiency (PER and PPV) were achieved at 60% PBM treatment, and the highest values of energy utilization efficiency (ER and EU) was in favor of the control treatment.

Chemical composition of the Seabass at the end of this experiment illustrated higher content of moisture, protein and lipids and lower content of ash for PBM treatments compared with the control (Table 5). The differences between treatments were significant (P < 0.05) in moisture, and ash, but not-significant (P> 0.05) for protein and lipid contents. Generally, data of crude protein revealed that adding 20% PBM gave the best protein content (56.65%) compared with 54.21% for the control treatment. However, adding 40% PBM gave the best Crude Lipids(17.28 %).

Table 6: Aeromonas, Salmonella, and E. Coli content in water samples and carcass of Seabass fish fed with diets tested with different concentrations of PBM.

Treatments	Fish samples			Water samples		
Bacteria	Aeromonas	Salmonella	E. Coli	Aeromonas	Salmonella	E. Coli
Control	+ +	+	-	-	-	-
PBM 20	-	-	-	-	-	-
PBM 40	+	-	-	-	-	-
PBM 60	-	-	-	-	-	-

Microbial Load

Aeromonas, Salmonella, and E. Coli were examined in fish and water samples under the tested treatments as shown in Table 6. Escherichia coli (*E. coli*)was not detected in both fish and water samples for all treatments, while Aeromonas was detected only under control and 40% PBM treatments. On the other hand, Salmonela was detected only in the control treatment. Treatments of 20 and 60% PBM were free from any pathogenic bacteria in both fish and water samples.

DISCUSSION

Replacement of fish meal with PBM to a certain level could increase the growth performance of Seabass fish during the sensitive nursing stage under intensive culture system. In the present study, the obtained results of growth performance agree with the findings of many authors, such as Paixao *et al.* (1989), Turker, *et al.* (2005), Rawles *et al.* (2009) and Badillo *et al.* (2014) for different species of fish. Badillo *et al.* (2014) found that Rainbow Trout (*Oncorhynchus mykis*) fed the 67% PBM diet had a significantly higher final weight than the rest of the treatments, whereas the 100% PBM had a significantly lower final weight than the other treatments (0, 33, and 67%). Also, Rawles *et al.* (2009) found no significant differences in the growth rate of Hybrid Striped Bass, (*Morone chrysops * Morone saxatilis*) fed fish meal supplemented diets replaced with poultry by-product meal (PBM) at levels of 0, 35, 70, and 100%. With the same trend, fish meal could be replaced up to 75% PBM in commercial feed for common carp and the highest daily and final weight gain were obtained at this level of inclusion (Paixao *et al.*, 1989).Generally, fish fed the 35% replacement diet (35PBM) performed as well as fish fed the generic diet, whereas fish fed the 70PBM diet not. Tidwell *et al.* (2005) used 33.4% PBM to replace fish meal in diets for largemouth bass without impairing growth. Abdel-Warith *et al.* (2001) reported 40% replacement of fish meal with poultry by product meal without effectively altering growth in the diet of African catfish. However, Hao and Yu (2003) evaluated 80% replacement of fish meal with poultry by product meal and meat and bone meal in the diet of juvenile catfish without any harmful effect.

Poultry by-product meal (PBM) is an ideal protein source for the partial substitution of fishmeal. According to Tacon and Metian (2008), PBM substitutes up to 30% of fishmeal in diets for salmon, sea trout and shrimp. A 30% replacement in rainbow trout diets yielded a similar growth performance to those fed a diet with fishmeal as the only protein source (Cruz-Sua´rez *et al.*, 2007; EL-Haroun *et al.*, 2009). Partial replacement of fishmeal with PBM has been shown to yield a similar growth performance to that obtained with 100% fishmeal in aquafeeds (NRC, 2011; Rossi and Davis, 2012). For omnivorous fishes, Fish meal could be fully replaced by poultry offal meal without adverse effect on carp growth (Hasan and Das, 1993) but the best growth occurred when poultry offal meal provided 75% of the supplemental protein when combined with fish meal (Hasan, 1991). Turker *et al.* (2005) found that there was no significant reduction in growth performance of the turbot fed the 25% replacement diet compared to the control diet (only fishmeal). On the other hand, Rawles *et al.* (2006) found that diet composition significantly ($P < 0.05$) influenced final weight, weight gain, yield, hepatosomatic index (HSI) and intraperitoneal fat (IPF) ratio, but did not significantly alter feed conversion and muscle ratio of for hybrid striped bass (*Morone chrysops* × *M. saxatilis*).

In the current study, results of survival rate harmonized with the findings of many researchers, such as Rawles *et al.* (2009), Tabinda and Butt(2012), Badillo et al. (2014), and Ma *et al.* (2014). Rawles *et al.* (2009) found no significant differences in survival rate between Hybrid Striped Bass, *(Morone chrysops * Morone saxatilis*) fed 100% fish meal diets replaced with diets supplemented with poultry by-product meal (PBM) at 45, 70, and 100%, and the overall survival rate was more than 95% without significant differences between treatments. Similar results were obtained by Ma *et al.*, (2014) for Golden Pompano, *(Trachinotus ovatus)* with survival percent more than 99% and Tabinda and Butt (2012) with 100% survival for Grass carp, and Turker *et al*. (2005) with 100% survival for Turbot. The most negative point related with 60% PBM treatment is the lowest survival rate (94%). This may be attributed to the highest content of fiber (*5.32%)* in the tested diets compared with the other treatments, specially the tested fish size was very small.

Results of Condition factors (CF) revealed that treatments supplemented with PBM kept the condition factor at high value, but still lower than the control without any significant differences. According to Adeyemi *et al*., (2009), negative allometric growth pattern in fish implied that the weight increases at a lesser rate than the cube of the body length. Idodo-Umeh (2005) and Abowei and Hart (2009) reported that the length – weight relationship of fish also known as growth index, is an important management tool used in estimating the average weight at a given length growth. The mean condition factor in the intensive farms often be less than 1, while the values of condition factors in the natural water resources are better than in the intensive rearing tanks (Okan and Gamsiza, 2010) with mean condition factor of 2.058. Therefore, the obtained values (e.g. CF= 1.22 at 60% PBM) in this study are in agreement with other output researches.

The results of feed utilization efficiency of the present study are in agreement with the findings obtained by many authors, like Tidwell *et al*. (2005), Rawles *et al*. (2009) and Badillo *et al*., (2014). Rawles *et al*. (2009) found that Hybrid Striped Bass, *(Morone chrysops * Morone saxatilis)* fed fish meal diets replaced with PBM at 0, 35, 70, and 100%, showed no significant differences in the final values of FCR and PER with the control treatment. Also, Badillo *et al*., (2014) found that the PER was also similar among treatments (0, 33, 67, and 100%) with values around 2.8 g body weight g protein intake. With the same trend, poultry by-product meal could replace up to 75% fish meal in the artificial feed of common carp diets and the highest apparent feed and protein efficiency were obtained at this level of inclusion (Paixao *et al*., 1989). In contrast, Pfeffer *et al*. (1995) found that increasing the dietary proportion of poultry slaughter by-products decreased protein and lipid digestibility's in rainbow trout. Reduced digestibility of protein and essential amino acids from PBM may limit its use in diets for Largemouth Bass (Portz and Cyrino, 2004). Turker, *et al*. (2005) found that there was a severe decrease in feed intake, growth performance, feed utilization, protein efficiency ratio, and apparent net protein utilization at the replacement levels of 50, 75, and 100% PBM compared with 25% PBM. However, High-fat PBM (16% fat) is prone to rancidity, which may result in various symptoms including diarrhoea, low feed intake (anorexia), poor animal performance, myopathy, hepatomegaly, steatitis, haemolytic anemia and deficiencies in vitamins A and E in non-ruminants (El Boushy and Van der Poel, 2000).

Chemical composition of Seabass fish in the present study, revealed that the retention of protein was best at 20% PBM and lipid retention was higher for treatments supplemented with PBM compared with control. These results were in concordant with those obtained by other researchers, such as Lupatsch *et al*., (1997) for Gilthead Seabream, Wang, et al., (2008) for Malabar grouper, Rawles *et al*. (2009) for Hybrid Striped Bass, Badillo *et al*. (2014) for Rainbow Trout, Ma *et al*. (2014) for Golden Pompano, and Yones, and Metwalli (2015) for Tilapia. Lupatsch *et al*., (1997) observed a high protein digestibility (80%) for poultry by-product meal in Gilt-head sea bream (*Sparus aurata*). According to the results of Badillo *et a*l. (2014), the 33% and 67% PBM diets showed a slightly higher level of retention of nitrogen from the poultry by product meal than what was included in each formulation. The 33% and 67% PBM dietary treatments showed a 35.9 and 78.2% retention.

Pathogenic Bacterial load (Aeromonas, Salmonella, and E. Coli) in both water and fish samples in the present study, revealed that PBM used in this experiment was safe to be incorporated in seabass feed. That is because of the presence of potential pathogenic bacteria. For example, E. coli and Salmonela were not detected in both fish and water samples in any treatment, while Aeromonas was detected only under control and 40% PBM treatment. This means that, the PBM ingredient could be eliminated of any potential pathogens if dried well under appropriate temperature for sufficient exposure time. So, there is no fear of adding it for marine fish feeds. Unfortunately, the research papers dealt with this point are still few. However, presence of Aeromonas in the fish sample under treatment 40% PBM replacement, may be related to post-drying re-contamination from the environment (Kinley *et al*., 2010; Laban *et al.,* 2014). NRA (2003) stated that ground raw parts of slaughtered poultry carcasses as heads, feet, undeveloped eggs and intestine are highly contaminated with microorganisms including bacteria viruses, virus-like particles, fungi, yeast and associated microbial toxins that constitute a potential risk to animal and human illness (Crump *et al*., 2002;ARA, 2012) specially Salmonella serotypes. That is why the efficiency of cooking is very crucial to eradicate the content of pathogenic microbes in PBM. If PBM raw materials are cooked at a predetermined, continuously monitored temperature and atmospheric pressure in batch steam cookers (115°C to 145°C for 40 to 90 minutes), that inactivate many bacteria, viruses and molds (NRA. 2006). Also, in the present study, cooking PBM at 150-200 °C under a 2.5 bar (atmospheric pressure) for ten hours could get rid of many potential pathogenic bacteria efficiently. Laban, *et al.* (2014) found that raw PBM exposed to treatment of 140°C and pressure of 2 bars for 40-90 minutes resulted a reduction of 99.96% in

Total Bacterial Counts (TBC), 99.99% in total coliforms count (TCC), 100% in *Campylobacter spp.* Count. However, *Salmonella* spp. was reduced from 70% to 10% and, fungal count (TFC) reduced only by 60%.

CONCLUSION

From the above mentioned results, it could be concluded that replacing fish meal with Poultry By-product Meal (PBM) in the artificial feeds of European Sea Bass, *Dicentrarachus labrax* fry up to 60%, is acceptable, but replacing 40% of PBM instead of fish meal is the optimum in terms of keeping both fish survival rate and fish welfare in the best levels. Therefore, switching from 100% fish meal diets to PBM supplemented diets is highly recommended for marine fishes to reduce the cost of production.

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