

## Effect of partially replacing fish meal by poultry by-product meal on growth performance and feed utilization of European eels, *Anguilla anguilla* (Linné 1758)

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### Abstract

A 60-days feeding study was conducted to evaluate the effect of partial substitution (0, 20, 40 and 60%) of dietary fish meal (FM) with poultry by-product meal (PBM) on growth performance and feed utilization of European elver eels, *Anguilla anguilla*. A total of 80 *A. anguilla* with an average initial weight of  $1.64 \pm 0.13$  g were equally divided into the eight glass aquaria (100 L each, duplicate per treatment). All aquaria were filled with well water. Four isonitrogenous (45% CP) and isocaloric 20.0 MJ GE kg<sup>-1</sup> test diets were formulated. A diet containing FM (D<sub>1</sub>) as the main source of protein was considered as the control diet. Three tested diets were partially (20 (D<sub>2</sub>), 40 (D<sub>3</sub>) and 60% (D<sub>4</sub>)) substituted for dietary FM protein from control diet with PBM, respectively. In a substitution experiment, the best feed conversion ratio (FCR) and survival rate were obtained with the D<sub>1</sub> (100% FM), while protein efficiency ratio (PER), protein productive value (PPV) and energy retention (ER) were obtained with the D<sub>1</sub> and D<sub>2</sub>. The present results indicate that without any significant effect on the PER, PPV and ER, PBM could be used as dietary alternative protein replacement of FM up to the level of 20% for European elver eels, *A. anguilla*.

**Key words:** alternative protein source, *Anguilla anguilla* eel, fish meal replacement

### Introduction

Eels are essentially carnivorous with dietary protein requirement about 45%. Fish meal is the dominant ingredient in commercially prepared diets for eel fish. As a consequence of rapid growth in fish and shellfish farming, fish meal prices have increased significantly in the past few years and are likely to increase further with continued growth in demand (Hardy & Tacon 2002). As with general aquaculture nutrition, a priority area of major research is the reduction and possible elimination of fish meal and fish oil (Hardy & Gatlin 2002). The uncertain future of fish meal availability and its potential high cost has forced to investigate alternative protein sources of good nutritional quality, which are ideally readily available and more cost effective than fish meal. This will reduce production costs and create a good quality product suitable for any small or large-scale fish production system.

Poultry by-product meal (PBM) as rendered animal protein have considerable potential in fish and shrimp feeds (Bureau, *et al.* 2003; Goda *et al.* 2007; Burr *et al.* 2013). This animal by-product have long been used in compound feeds for terrestrial monogastric animals such as poultry and swine (Parsons & Fisher-Vanden, 1997). Many studies in recent years have also shown that rendered animal protein ingredients are useful for fish feed formulation and comparatively less expensive than fish meal (Steffens, 1994; Rodriguez-Sena *et al.*, 1996; Bureau *et al.*, 1999; Abdel-Warith *et al.*, 2001;

Samocha *et al.* 2004; Cruz-Suarez *et al.* 2007; Muralisankar and Bhavan 2013).

Relatively little research has been focused on dietary requirements of European eel, *A. anguilla* due to the critical problem in early age culture related to acceptance of artificial feed (weaning) compared other fish. Therefore, this study was designed to investigate the effect of partial substitution of dietary FM with three poultry by-product meal (PBM) inclusion levels (20, 40 and 60%) on growth performance and feed utilization of European elver eel, *A. anguilla*.

### Material and methods

#### Fish Husbandry and Experimental Design

The experiment was conducted at the Experimental Fish Farm at El-Kanater El-Khayria, National Institute of Oceanography and Fisheries (NIOF), Delta Barrage, Kalubiya Governorate, Egypt.

European elver eel, *A. anguilla* with an initial body weight of  $1.64 \pm 0.13$ g were obtained from the wild catch of Edku Lake, Alexandria Governorate, Egypt. The fish were stocked at a rate of 10 fish aquarium<sup>-1</sup> into eight indoor glass aquaria (80×40×60cm, 100 L each) representing the four experimental treatments, the control diet (100% FM) and other three inclusion levels by PBM (20,40,60%). Aquaria were supplemented with well water which was exchanged once daily after the second feeding time.

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## Experimental Diets

All experimental diets were fed as wet paste Table 1 and 2. The daily ration was divided into two equal amounts and offered two times a day (8:00 and 14.00 h). The fish were fed, one of four experimental diets, for 60 days. All fish (100%) from each replicate were weighed every 10 days and the amount of daily allowance was adjusted accordingly.

**Table 1.** Chemical composition of diet ingredients.

	Fish meal	Poultry by-product meal
<b>Chemical analysis (determined on dry matter basis)</b>		
Dry Matter	90	89
Ether Extract	13	13.1
Crude protein	66	68
Total Carbohydrates	1	2
Ash	10	5.9
Gross Energy (Kcal/Kg)	4966	5130

**Table 2.** Composition and proximate analysis of the experimental diets

Ingredients	Experimental diets			
	Diet <sub>1</sub>	Diet <sub>2</sub>	Diet <sub>3</sub>	Diet <sub>4</sub>
Fish meal	61	49	37	24
Poultry by-product meal	--	12	24	36
Corn gluten meal	7	6	5	8
Wheat bran	7	7	8	6
Corn starch	14	15	15	15
CMC <sup>1</sup>	3	3	3	3
Fish oil	2	2	2	2
Soybean oil	2	2	2	2
Vit. & Min. premix <sup>2</sup>	4	4	4	4
Vitamin C (g/Kg diet)	1	1	1	1
Chemical analysis (determined on dry matter basis)				
Dry matter (DM)	94.0	92.5	93.0	92.4
Crude protein (CP)	45.5	44.3	44.5	45.4
Digestible protein (DP) <sup>3</sup>	34.24	33.00	29.62	28.33
Ether extract (EE)	13.0	12.6	13.6	12.7
Total carbohydrate <sup>4</sup>	9.0	8.5	7.0	6.5
Ash	26.5	27.1	27.9	27.4
GE (Mj kg <sup>-1</sup> diet <sup>-1</sup> ) <sup>5</sup>	20.38	20.11	20.70	20.55
DE (Mj kg <sup>-1</sup> diet <sup>-1</sup> ) <sup>6</sup>	15.02	14.98	14.25	13.65
P/E ratio(Kcal kg <sup>-1</sup> diet <sup>-1</sup> )	93.39	92.12	89.92	92.59
Feed cost (LE.) Kg <sup>-1</sup> gain	34.40	35.27	65.8	105.1
Amino acid profile				
Arginine	6.56	6.47	6.20	5.80
Histidine	2.42	2.29	2.10	1.90
Isoleucine	4.62	4.49	4.23	4.01
Leucine	8.21	7.95	7.45	7.31
Lysine	7.40	6.91	6.21	5.34
Methionine	2.98	2.76	2.45	2.15
phenylalanine	4.15	3.96	3.66	3.45
Threonine	4.18	4.00	3.70	3.38
Tryptophan	1.10	1.04	0.96	0.84
Valine	6.21	5.94	5.49	5.02

<sup>1</sup>Carboxy methyl cellulose

<sup>2</sup>Vitamins and minerals mixture each 2Kg of mixture content: Per kg mix: 4 000 000 IU vitamin A, 480 000 IU vitamin D3, 40 000 mg vitamin E, 2400 mg vitamin K3, 4 000 mg vitamin B1, 6 000 mg vitamin B2, 40 000 mg niacin, 10 000 mg Ca-D-pantothenate, 4 000 mg vitamin B6, 10 mg vitamin B12, 100 mg D-biotin, 1 200 mg folic acid, 40 000 mg vitamin C ve 60 000 mg inositol, 23 750 mg Mn, 75 000 mg Zn, 5 000 mg Zn, 2 000 mg Co, 2 750 mg I, 100 mg Se, 200 000 mg.

<sup>3</sup>, 6 Digestible protein (DP) and Digestible energy (DE, Mj kg<sup>-1</sup> diet<sup>-1</sup>) calculated according to results of digestibility trails (Table 5).

<sup>4</sup>Total carbohydrate =100-(CP + EE+ Ash)

<sup>5</sup>Calculated using gross calorific values of 23.63, 39.52 and 17.15 kj g<sup>-1</sup> for protein, fat and carbohydrate, respectively according to Brett (1971).

### Statistical Analysis

Data were statistically analyzed by ANOVA using SAS ANOVA procedure (SAS Institute Inc., 2004).

### Result and Discussion

Dietary protein supply is one of the major factors that influence the growth of fish. Like other fish species, the reported protein requirements of European eel are relatively high compared to terrestrial animals (NRC, 1993). The protein requirement of European eel is ranging from 30-48% (Spannhof and Kuhne, 1977; Degani *et al.* 1985). Poultry products are becoming plentiful in countries with extensive chicken and turkey production industries such as Egypt. If this abundant and inexpensive source of protein could be substituted for fish meal, much less expensive fish feeds could be produced.

Chemical analysis of the experimental diets indicated that the formulated protein levels was achieved. Analyzed crude protein levels (% as DM) in the diets were 45.5 (control diet); 44.5 (diet<sub>2</sub>); 44.5 (diet<sub>3</sub>) and 45.4 (diet<sub>4</sub>).

Differences in growth of the fish fed the experimental diets became apparent after 60 days of feeding where the mean weight of fish either control or 20% PBD diets were significantly higher ( $P \geq 0.05$ ) than those fed other experimental diets. This difference persisted for the duration of the experiment Table. 3. Fish fed control diet showed the highest significant ( $P \geq 0.05$ ) values for average daily gain and survival (%) followed by eel fed 20% PBM diet. Dietary PBM level had a significant effect on eel weight gain, specific growth rate, survival (%) and feed conversion ratio. Replace dietary FM with PBM more than 20% for eel resulted in the lowest final body weight, specific growth rate, average daily gain and survival (%). The present results recorded for a daily weight gain values within the range of previous values reported of the same life stage. Gallagher and Degani, (1988) recorded an average daily gain for eel ranged between (0.01-0.024 g/ day) and (0.02-0.05 g/ day) where the initial weight were 1.42 and 2.4 g, respectively. For eels with initial weight of 2.2 g, Engin and Carter (2005) recorded an average daily gain of 0.01 g/day. Average daily weight gain ranging from 0.002 to 0.004 g/day was recorded for glass eel of initial weight 0.1 g (Okorie *et al.* 2007).

**Table 3.** Growth performances and survival (%) of eel fed different experimental diets

	Experimental diets				SE ±*
	Diet <sub>1</sub>	Diet <sub>2</sub>	Diet <sub>3</sub>	Diet <sub>4</sub>	
Initial body weight (g)	1.66	1.63	1.65	1.61	0.13
Final body weight (g)	4.11 <sup>a</sup>	3.63 <sup>a</sup>	2.39 <sup>b</sup>	1.98 <sup>b</sup>	0.18
Average weight gain (g/fish/day)	0.040 <sup>a</sup>	0.033 <sup>b</sup>	0.012 <sup>c</sup>	0.006 <sup>d</sup>	0.03
Specific growth rate (%/day)	1.51 <sup>a</sup>	1.33 <sup>a</sup>	0.61 <sup>b</sup>	0.34 <sup>b</sup>	0.09
Survival rate (%)	95 <sup>a</sup>	90 <sup>b</sup>	85 <sup>c</sup>	80 <sup>d</sup>	0.5

Diet<sub>1</sub>: Control diet; Diet<sub>2</sub>: 20% PBM diet; Diet<sub>3</sub>: 40% PBM diet; Diet<sub>4</sub>: 60% PBM diet.

A, b, ... etc. means that in the same row with different superscription are significantly different ( $P \leq 0.05$ ).

\*SE± standard error. Calculated from residual mean square in the analysis of variance.

The best feed conversion ratio (FCR) was recorded for eel fed the control diet. This FCR was significantly better ( $P \geq 0.05$ ) than when eels were fed the higher dietary PBM level diets more than 20% Table 4. Fraga, *et al.* (2003); Luzzana, *et al.* (2003); Nielsen and Prouzet (2008) recorded FCR ranged between 4.5-10 for European eel, *A. anguilla*. while, was 3.64 for American eel, *A. rostrata*. The same trend was observed in the present results.

Protein productive value and energy retention values did not differed significantly between fish fed either control or 20% PBM diets, while the lowest values was recorded with increasing inclusion levels of PBM in elver diets (40 and 60 % PBM) Table 4. The same trend was observed for protein efficiency ratio and feed intake.

By the end of the experiment, survival (%) of eels fed control diet was higher than the other experimental diets. All mortalities observed were small eels (mean weight, 1.74 g) that displayed bite marks on their bodies. Based on this, it is felt that the higher growth rate in the control experimental fish led to higher variation between large and small eels and, ultimately, more aggressive behaviour within these aquaria. Degani and Levanon (1983 and 1984) reported that the study of the initial feeding of the glass eel is difficult due to the different rates of growth due to distinguishable groups among glass eels, *A. anguilla* during the initial period. Glass eels that did not adapt to eat, lost weight and died (20%). Glass eels adapted to culture conditions could be divided into three main groups: fast-growing elvers (3% of the total sample), moderately fast-growing elvers (25%) and the other 52% were slow-growing

elvers (Degani 1986). Few studies dallied with PBM as protein source in eel fish diets. Degani *et al.* (1984) found that poultry by-products could be used to support growth of glass eels, *Anguilla anguilla*, but a more precise study is needed to find out the optimum dietary levels should be used in eel diets in the initial period.

Some other fish species with high protein requirement noticed for accepting PBM as fish meal substitution. Fowler, (1991) reported that PBM replaced 20% of dietary fish meal in juvenile fall Chinook salmon, *Oncorhynchus tshawytscha* successfully, while negative effect on weight gain and feed efficiencies values was associated with increasing substitution up to 30%. The ability to replace fish meal up to 25% by poultry by-product meal, without causing reduction in growth performance, nutrient utilization and nitrogen retention was recorded for juvenile black sea turbot, *Psetta maeotica* (Yigit *et al* 2006). More dietary inclusion level of PBM for low protein requirement

of fish species was suggested by many findings (up to 40% for African catfish, *Clarias gariepinus* by Abdel-warith *et al.* 2001; up to 66% for major carp and rainbow trout *Oncorhynchus mykiss* by Cheng *et al.* (2002).

Amino acid profile of poultry by product meal could be one of the main limitations for more inclusion in eel diets. As with more inclusion of PBM in elver diets more unbalanced dietary amino acid profile occur, which lead to suppression in elver growth performance. It was reported that, amino acid addition to PBM can improve fish growth performance (Rawles *et al.*, 2009). Lysine and methionine supplementation, helped to achieve complete replacement of fish meal by PBM in diet for hybrid striped bass (Rawles *et al.* 2009). The amino acid availability was higher for most of the essential amino acids from FM compared to PBM with the exception of threonine, histidine and methionine for hybrid striped bass (Gaylord and Rawles, 2005).

**Table 4.** Feed utilization of eel fed different experimental diets

	Experimental diets				SE ±*
	Diet <sub>1</sub>	Diet <sub>2</sub>	Diet <sub>3</sub>	Diet <sub>4</sub>	
Feed consumption (g/day/fish)	0.17	0.15	0.12	0.10	0.05
Feed conversion ratio (g feed/g gain)	4.08 <sup>c</sup>	4.58 <sup>b</sup>	9.51 <sup>a</sup>	16.98 <sup>a</sup>	0.51
Protein efficiency ratio	0.53 <sup>a</sup>	0.49 <sup>a</sup>	0.23 <sup>b</sup>	0.12 <sup>b</sup>	0.05
Protein productive value	8.12 <sup>a</sup>	7.34 <sup>a</sup>	3.55 <sup>b</sup>	1.86 <sup>b</sup>	0.40
Energy retention%	11.91 <sup>a</sup>	11.16 <sup>a</sup>	6.28 <sup>b</sup>	5.39 <sup>b</sup>	0.64

Diet<sub>1</sub>: Control diet; Diet<sub>2</sub>: 20% PBM diet; Diet<sub>3</sub>: 40% PBM diet; Diet<sub>4</sub>: 60% PBM diet.

A, b, ...etc. means that in the same row with different superscription are significantly different ( $P \leq 0.05$ ).

\*SE± standard error. Calculated from residual mean square in the analysis of variance.

## Conclusion

In a substitution experiment, the best feed conversion ratio (FCR) and survival rate were obtained with the D<sub>1</sub> (100% FM), while protein efficiency ratio (PER), protein productive value (PPV) and energy retention (ER) were obtained with the D<sub>1</sub> and D<sub>2</sub>. The present results indicate that without any significant effect on the PER, PPV and ER, PBM could be used as dietary alternative protein replacement of FM up to the level of 20% for European elver eels, *A. anguilla*.

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