

Inclusion of a commercial poultry by-product meal as a protein replacement of fish meal in practical diets for African catfish *Clarias gariepinus* (Burchell 1822)

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Abstract

This study examined the full and partial replacement of the fish meal component of practical diets with commercial poultry by-product meal (PBM) for juvenile African catfish *Clarias gariepinus* (Burchell 1822). Six test diet formulations were based on a low temperature Norwegian fish meal (LT94) as the reference protein source. Dietary inclusion levels of PBM at 20%, 40%, 60%, 80% and 100% substitution of total protein content were compared with the fish meal-based control diet (100% fish meal). All diets were iso-nitrogenous and isocaloric in gross terms. The results showed that there were significant differences ($P < 0.05$) between the final average weights of fish at the end of a 10-week feeding trial. Growth performance, feed intake, feed conversion ratio, protein efficiency ratio and specific growth rate were all depressed for catfish fed the highest levels of PBM diets compared with the fish meal group. It was inferred that amino acid profile and digestibility of protein were mainly responsible for these effects, together with reduced palatability. PBM may successfully replace up to 40% of the protein component in practical feeds for this species. Histological examination of liver tissue showed alterations in hepatic structure of those fish fed diets with higher levels of PBM.

Introduction

Much of the costs of aquafeed production are due to the extensive use of fish meal in the feed (Tacon & Jackson 1985; Tacon 1994; Higgs, Dosanjh, Prendergast, Beames, Hardy, Riley & Deacon 1995). Therefore numerous attempts have been made to develop economical alternative protein and energy sources for inclusion in fish diets. Considerable efforts are also being made to improve fish meal quality so that maximum nutritive value can be obtained from this expensive dietary component. Historically, fish meal has been the most desirable protein concentrate in formulated feeds; however, this is an expensive resource for use in warm water fish diets. The demand for fish meal is expected to raise the price of compounded feeds even higher than present levels due to the rapid expansion of aquaculture (Tacon 1996). This will become an increasing problem in countries that rely on the impact of this resource for fish feed manufacture. Alternative feed ingredients will become even more cost effective for incorporation in diets, especially those destined for warm water fish species such as carp, tilapia and catfish (Rumsey 1993).

Gouveia (1991) critically reviewed the use of animal by-products in diets for salmonids, and described the use of various protein concentrates that originated from the rendering industries.

Table 1 Composition and proximate analysis of the control and test diets (g 100 g⁻¹ dry weight)

Ingredients	International	Control	20% PBM	40% PBM	60% PBM	80% PBM	100% PBM
	Feed No.						
Fish meal ¹	5-02-000	40.00	32.00	24.00	18.00	10.00	3/4
PBM	5-03-798	3/4	9.00	17.00	24.00	33.00	45.50
Wheat meal ²	4-05268	38.00	38.00	40.00	41.00	41.00	38.00
Blood meal	5-00-381	2.00	2.00	2.00	2.00	2.00	2.00
Corn oil ³		4.41	4.12	3.91	3.66	3.37	2.94
Cod liver oil ⁴		4.41	4.12	3.91	3.66	3.37	2.94
Vitamin premix ⁵		2.00	2.00	2.00	2.00	2.00	2.00
Mineral premix		1.00	1.00	1.00	1.00	1.00	1.00
Binder ⁶		2.00	2.00	2.00	2.00	2.00	2.00
Cellulose ⁷		6.18	5.76	4.18	2.68	2.26	3.62
Proximate composition (dry matter basis)							
Moisture		4.81	4.44	4.73	3.25	4.38	4.54
Protein		34.98	36.63	35.60	36.25	35.92	35.94
Lipid		17.49	17.6	17.44	16.50	15.11	15.28
Ash		7.35	8.03	7.96	8.69	8.93	9.72

¹Fish meal LT94, Nutreco Salmon and trout fry diets. Trouw Aquaculture (Ltd).

²Wheat meal, Kalpro STM Orsan, Paris, France.

³Mazola – pure corn oil.

⁴Fish oil – Seven Seas (pure cod liver oil).

⁵Vitamin premix, Colborne Dawes Nutrition Ltd, providing the following per kg of dry feed; vitamin A, 1600 IU; vitamin D, 2400 IU; vitamin E, 160.0 mg; vitamin K, 16.0 mg; thiamin, 36.0 mg; riboflavin, 48.0 mg; pyridoxine, 24.0 mg; niacin, 288.0 mg; pantothenic acid, 96.0 mg; folic acid, 8.0 mg; biotin, 1.3 mg; cyanocobalamin, 48.0 µg; ascorbic acid (ascrbyl-phosphate), 720.0 mg; choline chloride, 320.0 mg; calcium, 5.2 g; cobalt, 3.2 mg; iodine, 4.8 mg; copper, 8.0 mg; iron, 32.0 mg; manganese, 76.0 mg; zinc, 160.0 mg; endox (antioxidant), 200.0 mg

⁶Carboxymethyl Cellulose.

⁷Sigma Chemical Co., Poole, Dorset. UK

One of the more promising sources is poultry by-product meal (PBM), the rendered product of poultry processing waste, made from inedible portions of poultry, excluding feathers. PBM has been previously evaluated in diets for chinook salmon *Oncorhynchus tshawytscha* (Brannon, Roley & Roley 1976; Roley, Roley, Hardy & Brannon 1977; Westgate 1979; Fowler 1981a,b, 1990, 1991), coho salmon *Oncorhynchus kisutch* (Markert, Higgs, Dye & MacQuarrie 1977; Higgs, Markert, MacQuarrie, McBride, Dosanjh, Nichols & Hoskins 1979) and Atlantic salmon *Salmo salar* (Bergström 1973). PBM has been tested as a partial fish meal replacement in the diets of channel catfish *Ictalurus punctatus* (Brown, Strange & Robbins 1985) and rainbow trout *Oncorhynchus mykiss* (Alexis, Papaparaskeva & Theochari 1985).

However, compared with fish meal, animal by-products may be deficient in one or more essential amino acids (Davies, Williamson, Robinson &

Bateson 1991). More recently, Nengas, Alexis & Davies (1999) reported that diets containing poultry meat meal (PMM), poultry feather meal (PFM) mixture and PBM at 40% replacement of fish meal in gilthead seabream *Sparus aurata* indicated that the first limiting essential amino acid (EAA) was methionine. Investigations concerning the use of animal by-product meals have been achieved with other freshwater fish species, mainly carp and tilapia, with good results (Rodríguez-Serna, Olvera-Novoa & Carmona-Osalde 1996). There is little information available for the use of such products in practical diet formulations for the African catfish *Clarias gariepinus*. A PBM available in the UK was evaluated as the alternative protein source for African catfish.

The objective was to determine the optimum level consistent with good growth performance without compromising the feed utilization efficiency and health of these fish.

Table 2 Essential amino acid composition of protein component (g 16N⁻¹) of experimental diets

	D1 (Control)	D2 (20% PBM)	D3 (40% PBM)	D4 (60% PBM)	D5 (80% PBM)	D6 (100 PBM)
Arginine	6.51	5.13	4.83	4.57	4.64	4.59
Histidine	3.38	2.81	2.58	2.36	2.23	2.13
Isoleucine	4.20	3.40	2.66	2.80	2.41	2.59
Leucine	8.43	6.70	5.80	5.69	5.24	5.58
Lysine	6.53	4.94	4.38	3.94	3.74	2.85
Methionine + Cystine	2.83	2.71	2.78	1.96	1.98	1.88
Phenylalanine + Tyrosine	7.97	6.49	6.40	5.72	5.65	5.48
Threonine	5.06	3.73	3.38	3.02	2.93	3.14
Valine	5.33	4.03	3.50	2.90	2.90	3.16
Tryptophan	ND ¹	ND	ND	ND	ND	ND

¹ND, not detected.

Materials and methods

Experimental diets

The experimental diets were formulated by using a least-cost linear programme (AGM System Ltd). Six experimental diets were formulated to contain a variable proportion of PBM to partially and totally replace the fish meal component of the diet. PBM was obtained from a commercial source (Sun Valley Poultry Ltd). It is defined as the milled dry rendered material originating from the processing of ground, rendered clean parts of the carcass of slaughtered chicken, turkey and duck carcass. This is inclusive of heads, feet, undeveloped ova and intestines (exclusive of feathers) as offal. The material is cooked at a temperature of 125 °C at 1 Atm for approximately 3 h.

The product typically consists of 16% ash and 50%–60% crude protein with a fat content of about 13%. All diets were designed to be isocaloric and isonitrogenous in gross nutrient terms and were adjusted at appropriate levels to contain 37% crude protein and 15% lipid.

Table 1 shows the proximate feed formulation and composition of the experimental diets. A control diet based on fish meal (LT94) served as the reference source of dietary protein used to substitute with PBM.

Table 2 shows the EAA composition of experimental diets as determined. A ground wheat meal was also included as the main carbohydrate source and bulk filler component. Five diets were formulated with an incremental substitution of fish meal with PBM up to 100% replacement.

Feeding regime and experimental protocol

Fish were fed for 1 week for acclimation to each test diet and the experimental system and to free their gastrointestinal tract from the pre-experimental diet (Trouw 00, salmon starter – 54% CP and 15% lipid) until the feeding response was uniform.

The trials were conducted in an automatically controlled recirculation system. The fiberglass tanks were approximately 75 L, and were suspended over a 1000-L bio-filter compartment. Water entered each tank via a spray bar after filtration and an aerator was placed in the centre of the tank to supply continuous oxygen. Partial water changes amounted to approximately 20% of the system's volume per week. Filters of the systems were cleaned daily to avoid the accumulation of nitrate (NO₃⁻) levels.

A 12-h light cycle was provided by fluorescent lighting and 10% of the water was replaced daily. The water temperature was maintained at 28 ± 1 °C by a thermostatically controlled immersion heater and pH, ammonia (NH₃), nitrite (NO₂⁻), nitrate (NO₃⁻) and dissolved oxygen were monitored and remained at acceptable levels throughout the experimental period.

At the end of the acclimation period, the fish were weighed and then started on the respective experimental diets. Each tank was randomly assigned to the dietary treatments.

Twenty fish with an average weight of 16.5 g were graded and transferred to each tank. All dietary treatments were tested in duplicate groups of graded fish of uniform size.

Initially 30 fish were killed using a lethal concentration of benzocaine and stored at -20 °C

in order to determine initial carcass composition. Six fish ($n = 6$) were randomly collected from each treatment at the end of the feeding trial to analyse the carcass and muscle composition.

Fish were fed twice daily by hand at a rate of 4% body weight per day for the first 4 weeks and this was subsequently reduced to 3% for the remaining of 4-week period and finally to 2.5% in the last 2 weeks.

The fish were weighed bi-weekly and were not fed on the day prior to weighing. The experiment was undertaken for a total period of 10 weeks. On termination of the study, the final weight of the fish was measured following a 24-h starvation period.

Proximate composition

Proximate compositions of diets and fish tissue were performed according to AOAC (1990) for moisture content and ash. Crude protein (%N 6.25) was determined by the micro Kjeldahl method. Total lipid was determined by a modification of the Folch method (1957).

Determination of amino acids

The amino acid contents of the diets were determined in acid hydrolysates. Approximately 20–25 mg of ground sample were weighed into 5 mL vials with 4 mL (6.6 M) of HCl and 1 mL (0.1 M) phenol, to protect tyrosine each vial was sealed and placed in an oven for 22 h at 110 °C.

Amino acids were assayed at Loughborough University, Department of Chemical Engineering, using a Kontron Chromakon 500 automatic amino acid analyser [column 250 × 4.6 mm, cation ion-exchange resin material (AS70)].

The mobile phase was a gradient of sodium citrate-based buffers according to the following composition: (1) 0.22 M sodium citrate, adjusted to pH 3.2 with concentrated HCl, +8% v/v methanol; (2) 0.067 M sodium citrate + 0.5 M sodium chloride adjusted to pH 3.79 with concentrated HCl; (3) 0.067 M sodium citrate + 1.4 M sodium chloride adjusted to pH 4.3 with concentrated HCl.

Detection was by a post column reaction with ninhydrin (in 4 M lithium acetate buffer pH 5.2 flow rate 12 mL h⁻¹) at 115 °C in a reaction oven followed by visible absorption measurement at 570 nm and 440 nm to produce a mean signal for quantitative integration. Dilution was made by loading buffer (2.2 pH sodium citrate buffer) and

suitable 100 µL aliquots injected into the rheodyne automatic injection valve.

The amino acid compositions (expressed as g 16 gN⁻¹) are presented in (Table 2).

Histological studies

Histological preparation and staining techniques

At the termination of the feeding trial, five fish from each group were sacrificed and their livers removed for histological examination. Hepatic tissue was sampled and processed according to the following procedures. All livers were dissected to give an equal size piece of tissue with 5 mm long square faces. This piece of tissue was immediately fixed in buffered formol saline.

Subsequently all the pieces were processed together, separately labelled and encased in a Shandon Elliot Hypercentre II tissue processor (Shandon Southern Products Ltd, Runcorn, UK). This allowed dehydration in a graded series of alcohols, clearing in xylene and infiltration in Fibrowax. Then the pieces were embedded in Fibrowax Mpt 56°C. Sectioning was at 5 µm using disposable blades and a rotary microtome (Reichert–Jung). Short ribbons of sections were placed into a heated water bath to flatten the sections. Then they were mounted onto glass slides and dried before staining. Samples were then stained by Mallory's trichrome according to Peacock (1973) in order to elucidate the general histology.

Image analysis

Stained slides were examined with a Zeiss photomicroscope II and the images captured using an Hitachi 3CCD colour camera. The analogue signal was then imported into a Quantimet Q570 image analyser (Cambridge Instruments, Cambridge, UK). Once the binary image was created it was measured for lipid area using a macro to ensure reproducibility of results.

Photomicrographs

Photomicrographs were taken using the Zeiss photomicroscope II and a Nikon 950 coolpix digital camera at an objective magnification of ×40 and photo-eyepiece ×2.5. A green filter was used for all photomicrographs and the condenser iris position kept constant.

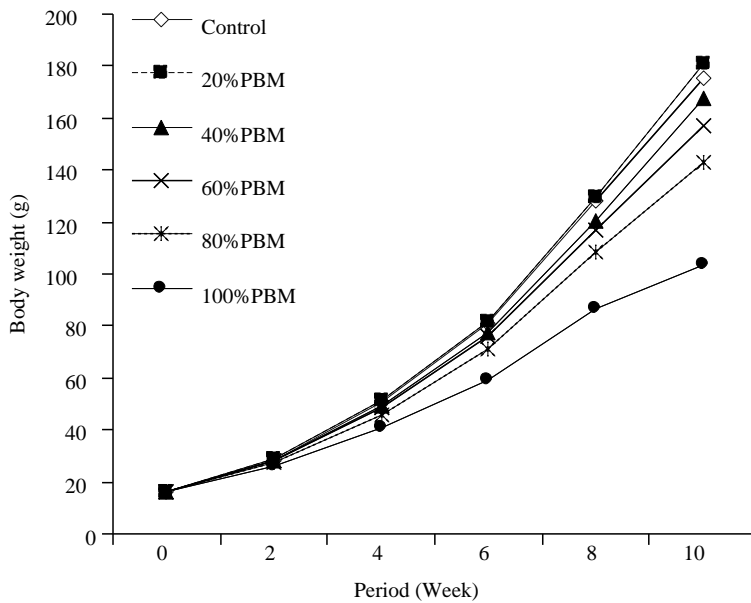


Figure 1 The relationship between average live weight (g) and time (weeks) of *C. gariepinus* fed the six test diets containing graded levels of PBM.

Statistical treatment of data

Statistical evaluation of the data was conducted using the computer software application Statgraphics plus (Statistical Graphics Corporation, USA). ANOVA was used to identify any statistical differences ($P < 0.05$) in weight, resulting from feeding each test diet formulation. Duncan's New Multiple Range Test was subsequently used to identify the significance differences between treatment mean values for selected parameters.

Results

Growth performance

The growth performance and feed utilization data for African catfish fed the six respective diets are shown in Fig 1 and Table 3. There was a significant difference between the final average body weights amongst the fish fed each of the experimental diets. Fish fed the fish meal (LT94)-based control diet demonstrated the highest mean final body weight (175.5 g) resulting in a 10-fold increase in weight from the start of the study.

However, the lowest value (103.8 g) was observed for catfish fed the 100% inclusion level of PBM in the diet replacing the entire fish meal component.

The control diet supported the highest weight gain of 158.9 g, while fish fed the PBM 100% diet

exhibited the lowest weight gain of 87.3 g. It was apparent that above 40% inclusion, PBM resulted in a significant reduction in growth performance with a final mean weight of less than 160 g.

The specific growth rate (SGR) values further supported this trend, with SGR reduced from 3.47 for the fish fed the control diet to 2.83 for the fish fed the 100% PBM diet. Fish fed the 40% and 60% level of inclusion of animal protein source (PBM) performed better than those on the 80% and 100% level of PBM, whereas the 20% level inclusion group was very similar to the control group. No mortality was observed during the experimental period and the overall health of the fish appeared normal.

Feed consumption and feed utilization

All diets were well accepted by the catfish, except Diet 6, containing the maximum amount of PBM. Mean daily feed intake ranged between 3.93 and 3.02 g 100 g⁻¹ fish. There was a noticeable effect of the higher inclusion of alternative protein sources on feed intake (Table 3). Feed intake for catfish fed the diet containing the highest amount of fish meal (LT94) was significantly better than those observed for fish fed diets including PBM.

Feed conversion ratio (FCR) values also differed significantly amongst the groups of fish. The poorest FCR was obtained for catfish fed the test diet containing 100% PBM with a value of 2.25.

Table 3 Weight increase, feed consumption, nutritive utilization of feed and protein, protein retention and HSI

	CON	20% PBP	40% PBP	60% PBP	80% PBP	100% PBP
Mean initial weight (g)	16.5 ± 0.12 ^a	16.5 ± 0.10 ^a	16.5 ± 0.14 ^a	16.4 ± 0.10 ^a	16.5 ± 0.11 ^a	16.5 ± 0.10 ^a
Final weight (g)	175.5 ± 2.7 ^{de}	180.76 ± 2.5 ^e	167.46 ± 2.6 ^{cd}	157.37 ± 2.0 ^c	143.1 ± 1.5 ^b	103.82 ± 1.9 ^a
SGR ³	3.57 ^{de}	3.68 ^e	3.56 ^{cd}	3.48 ^c	3.32 ^b	2.83 ^a
Weight gain (g)	158.9	164.3	151.0	140.9	126.6	87.3
Mean daily feed intake (g fish ⁻¹ day ⁻¹)	3.93	3.99	3.78	3.78	3.52	3.02
FCR ¹	1.61	1.58	1.63	1.74	1.86	2.25
PER ²	1.78	1.73	1.72	1.58	1.50	1.24
HSI ⁴	1.58	1.50	1.63	1.31	1.40	1.74

¹FCR: feed intake (g)/body weight gain (g).

²PER: body weight gain (g)/protein intake (g).

³SGR: [Ln final bw (g) – Ln initial bw (g)]/feeding days × 100.

⁴HSI: liver weight (g)/fish weight (g) × 100. Values with the same superscript are not significantly different ($P > 0.05$).

Superior FCR (< 2) were obtained for the remaining diets (control diet and 20% PBM showed the best FCRs, at 1.61 and 1.58, respectively).

Protein efficiency ratio (PER) was noticeably different between treatments. The fish fed the control diet displayed superior PER (1.78), whereas fish receiving the highest level of PBM had a PER of 1.24.

It should be noted that the essential amino acid profile of the experimental diets clearly showed a declining level of each amino acid at each PBM increment. This was especially apparent for the total sulphur amino acids (Meth. + Cys.) and also lysine above a 60% inclusion of PBM.

The hepatosomatic index (HSI) did not reflect any trend in catfish sampled at the end of the study, although fish fed the highest level of PBM showed the highest value for HSI (1.74). Fish fed the other diets revealed no significant relationship after 10 weeks.

Fish body composition

Initial and final carcass composition of the fish fed the experimental diets is presented in Table 4. The final carcass composition showed a treatment effect on proximate composition with respect to protein and ash content only. Fish fed the fish meal-based control diet and 20%, 40%, 60% PBM diets did not yield any variations in the protein content. Whilst fish fed the 80% and 100% PBM diets had a slightly lower protein level in their final carcass composition.

Fish fed the elevated level of PBM resulted in little differences in lipid content compared with lower PBM diets or the fish meal fed control group, respectively (Table 4). Similarly, there was no obvious trend in the carcass moisture content ($P > 0.05$).

However, the ash content in the carcasses of fish fed the control diets was appreciably low (2.63%) compared with catfish fed on higher levels of PBM. This was found to be significant ($P < 0.05$) above the 40% inclusion level with values ranging from 3.10 to 3.65.

Histological studies

Sections of liver tissue showed alterations in the liver structure of those fish fed diets with higher levels of PBM, which is shown in Figs 2 and 3(a,b) for typical gross architecture of hepatic tissue from representative fish. Hepatic cells of fish fed the control diet and 20% PBM were well defined in shape and well organized, and there was no sign of shrinkage or necrosis. Catfish fed diets with high inclusion levels of PBM (80%, 100%) showed appreciable alterations in the liver tissue.

The relative size of hepatocytes increased as the proportion of the PBM in the diets increased, and this was associated with a much greater hepatic lipid deposition. Polarization and isolated necrosis in hepatocytes were also observed when the diets included higher levels of PBM (80% and 100%). These changes were all quantified by the use of

Table 4 Fish carcass composition (g 100 g⁻¹ wet weight) of whole fish fed experimental diets

Proximate composition (%)	Initial fish	CON	20% PBM	40% PBM	60% PBM	80% PBM	100% PBM
Moisture	77.62	69.83 ± 0.11 ^a	70.67 ± 0.15 ^b	70.89 ± 0.16 ^b	69.56 ± 0.12 ^a	69.37 ± 0.13 ^a	69.55 ± 0.10 ^a
Protein	11.95	15.86 ± 0.03 ^c	15.81 ± 0.03 ^c	15.39 ± 0.05 ^b	15.89 ± 0.02 ^c	15.18 ± 0.03 ^b	14.7 ± 0.06 ^a
Lipid	6.77	10.31 ± 0.08 ^a	10.3 ± 0.05 ^a	10.10 ± 0.06 ^a	10.63 ± 0.04 ^a	11.01 ± 0.04 ^a	10.53 ± 0.04 ^a
Ash	2.07	2.63 ± 0.02 ^a	2.97 ± 0.04 ^b	3.10 ± 0.04 ^b	3.42 ± 0.05 ^c	3.51 ± 0.04 ^c	3.65 ± 0.04 ^c

Values with the same superscript are not significantly different ($P > 0.05$).

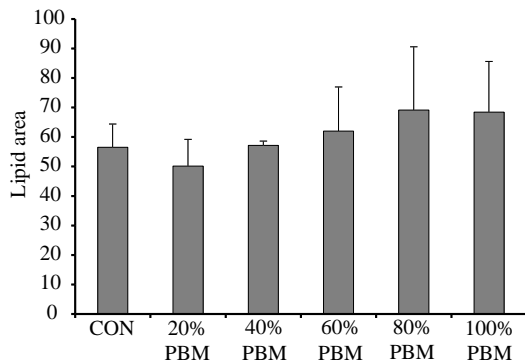


Figure 2 Mean area (μm^2 , \pm SD, $n = 5$) of intracellular lipid deposition in the liver from African catfish fed on experimental diets.

the image analyser and the mean relative area (μm^2) of lipid deposition is shown in Figs 2 and 3(a,b).

Discussion

The inclusion of alternative protein sources for the partial or direct replacement of fish meal has been studied in previous investigations for numerous fish species. These have concluded that increasing animal-derived protein to replace fish meal has a detrimental effect on growth rate and feed utilization above certain constraints, although partial substitution is quite feasible. The feasibility of poultry by-products in fish diets was found to depend on fish species and size as well as composition and processing techniques. Lu & Kevern (1975) found that a diet containing 30% PBM and 70% salmon feed lowered the growth rate of channel catfish, *Ictalurus punctatus*. On the other hand, up to 75% of fish meal could be replaced by defatted PBM in coho salmon, *Oncorhynchus kisutch*, diets without

adverse effects on growth (Higgs *et al.* 1979). At a 100% substitution level, growth was significantly compromised. However, a mixture of PBM and feather meal supplemented with essential amino acids effectively replaced fish meal in rainbow trout diets (Gropp, Koops, Tiews & Beck 1976; Tiews, Gropp & Koops 1976).

However, Higgs *et al.* (1979) reported at least 28% of PBM may be included in the diet of coho salmon, without amino acid supplementation.

In the present study, the results for African catfish *Clarias gariepinus* demonstrate that PBM can replace up to 40% of a high-quality fish meal protein without amino acid supplementation, whilst not compromising growth performance and feed utilization. Sadiku & Jauncey (1995) investigated the nutritional value of a feather meal and PBM blend for *C. gariepinus*. These authors reported similar conclusions to our investigations. The highest mean final weight, SGR, PER and FCR values were recorded for fish fed the control diet and 20% PBM diets. The improved performance of these diets was probably due to a more favourable EAA balance than fish meal alone.

The methionine and lysine requirements for the African catfish have been recently determined by Fagbenro, Balogun & Fasakin (1998) and Fagbenro, Balogun, Fasakin & Bello-Olusoji (1998), and are 3.2% and 5.7% of the total protein, respectively, based on semipurified diets. In the present study, these specific amino acids were appreciably lower for all PBM inclusions above 40% of the protein component of the diet. It should also be noted that several other amino acids became progressively reduced at high PBM inclusions, and may have contributed to the inferior protein utilization (PER) and growth performance of catfish.

Fowler (1991) was successful in rearing chinook salmon, *Oncorhynchus tshawytscha*, with a diet

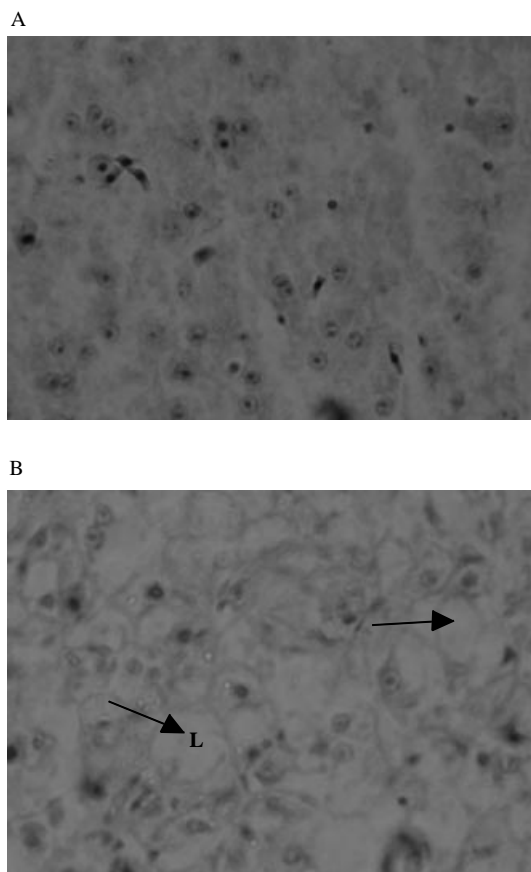


Figure 3 (a) Section of liver from *C. gariepinus* fed the fish meal-based control diet showed no visible intracellular lipid deposition. (b) Fish fed Diet 6 (100% PBM) showed severe intracellular lipid deposition (L). Nikon camera CoolPix 950 with Adapter MD lens- MEIJI techno. Company; magnification $\times 40$.

containing 20% PBM meal without additional amino acid supplementation.

Alexis, Papaparaskeva & Theochari (1985), working with rainbow trout, *Salmo gairdneri*, obtained very good results with a feed containing 20% PBM meal, with methionine supplementation.

The inferior performance of the fish receiving the higher PBM diets compared with groups receiving fish meal in most studies is possibly a result of the lower availability of nutrients and amino acid imbalance. Alexis *et al.* (1985) listed the digestibility of some of the main animal by-products tested in their study. These workers gave values of about 85% digestible protein for fishmeal but only 60% for PBM in the rainbow trout. Gaylord & Gatlin (1996) also reported the protein digestibility coeffi-

cients of selected animal protein sources, including meat and bone meal, and found them to be close to values previously reported for various fish species.

The protein digestibility of PBM in their study appeared low at 49% for red drum, *Sciaenops ocellatus*, compared with values for rainbow trout and chinook salmon of 68% and 74%, respectively (NRC 1993). There have been few studies to determine the digestibility coefficients for protein and energy in ingredients for tropical fish such as tilapia and catfish. Hanley (1987) reported such values for tilapia *O. niloticus* and found that the digestibility of protein and energy was appreciably lower (74% and 59%, respectively) compared with fish meal (86% and 80%, respectively). In the present study this is probably one explanation for the reduced growth performance associated with increased levels of PBM for African catfish.

Investigations are currently in progress in our laboratory to define the digestibility coefficient profiles for protein, individual amino acids and energy for a range of raw materials for catfish and tilapia feeds (unpublished data).

The palatability of low fish meal diets for fish is also a problem and should be addressed for even omnivorous fish such as the African catfish. At high inclusion levels of PBM in the current study there was a reduction in feed intake.

The carcass composition of catfish was not affected by substitution of fish meal with PBM with respect to either moisture content or lipid. Also the protein component remained consistent for each of the groups receiving the experimental diets. This was consistent with the similar protein and energy levels employed in the feed formulations.

It should be noted that Belal, Al-Owairer & Al-Dosari (1995) also found that the replacement of fish meal with chicken offal silage for tilapia *O. niloticus* did not compromise growth performance or carcass composition. There was, however, a progressive significant increase in the ash content of catfish fed PBM. This elevated mineral retention reflected the inorganic component of the diets. Future studies should also address the mineral composition of whole carcass and selected tissues.

The accumulation of lipid observed in the liver histology of African catfish fed the higher level PBM diets could be related to a dietary imbalance between saturated and unsaturated fatty acids, as a consequence of the high ratio of saturated fatty acids present in this ingredient.

These results agree with those reported by Shimeno, Masumoto, Hujita & Ueno (1993), who found a higher liver lipid content in yellowtail seabream *Seriola quinqueradiata* fed meat and bone meal compared with fish fed corn gluten meal diets at the same dietary inclusion level.

Interestingly, in the present experiment, isolated necrosis in hepatocytes were found in livers of fish fed higher levels of PBM in the diet, indicating possible irreversible effects on fish health due to nutritional imbalances (Mosconi-Bac 1987, 1990). Also Robaina, Izquierdo, Moyano, Socorro, Vergara & Montero (1998) have reported that the increase in the *n-3/n-6* fatty acid ratio with about 30% soybean meal improved the utilization of liver lipids, thus reducing liver histological alterations in gilt-head sea bream.

It should be noted that the high fat content of PBM (18%–20%) contributes significantly to the total lipid content in diet formulations in which the PBM inclusion rate is above 25%. Since PBM is predominately a source of *n-9* (oleic acid), this could have an important consequence for the longer term feeding of such ingredients. However, the supplementary fish/vegetable oils remained relatively high in our diets and would have provided the *n-3*, *n-6* fatty acid requirements for this species.

In conclusion, the results from this study would indicate that PBM is an acceptable ingredient for the partial replacement of fish meal protein in practical diets for juvenile African catfish. PBM can be used in balanced diet formulations for this species with up to 40% replacement of fish meal protein before limitations in growth performance and health criteria are observed.

Further work is required to obtain reliable digestibility data for protein, amino acids, lipid and energy components for this ingredient to realize its full potential in practical diets. This would necessitate investigations with various size classes of fish representing the complete production cycle.

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References

- Alexis M.N., Papaparaskeva. E. & Theochari V. (1985) Formulation of practical diets for rainbow trout (*Salmo gairdneri*) made by partial or complete substitution of fish meal by poultry by-products and certain plant by-products. *Aquaculture* **50**, 61–73.
- AOAC (Association of Official Analysis Chemists) (1990) *Official Methods of Analysis*, 15th edn. AOAC, Arlington, Virginia, 1298pp.
- Belal E.H., Al-Owafair A. & Al-Dosari M. (1995) Replacing fish meal with chicken offal silage in commercial *Oreochromis niloticus* (L.) feed. *Aquaculture Research* **26**, 855–858.
- Bergstrom E. (1973) The role of nutrition in growth and survival of young hatchery reared Atlantic salmon. *The International Atlantic Salmon Symposium 1972*, St Andrews, NB, Canada. Spec. Publishers Ser., **4** (1), 265–282.
- Brannon E.L., Roley D.D. & Roley S.E. (1976) Alternate protein sources to supplement the University of Washington standard hatchery diet for chinook salmon *Oncorhynchus Tshawytscha*, *Annual Report of College Fisheries University Washington, Seattle, WA*, 64–65.
- Brown P.B., Strange R.J. & Robbins K.R. (1985) Protein digestibility coefficients for yearling channel catfish fed high protein feedstuffs. *Progress in Fish Culture* **47**, 94–97.
- Davies S.J., Williamson J., Robinson M. & Bateson R. (1991) Practical inclusion levels of common animal by-products in complete diets for tilapia (*Oreochromis Mossambicus*, Peters). In: *Fish Nutrition in Practice* (ed. by S.J. Kaushik & P. Luquet), 325–332. INRA, Paris.
- Fagbenro O.A., Balogun A.M. & Fasakin E.A. (1998) Dietary methionine requirement of the African catfish, *Clarias Gariepinus*. *Journal of Applied Aquaculture* **8** (4) 47–54.
- Fagbenro O.A., Balogun A.M., Fasakin E.A. & Bello-Olusoji O.A. (1998) Dietary lysine requirement of the African catfish, *Clarias Gariepinus*. *Journal of Applied Aquaculture* **8** (2) 71–77.
- Folch J., Lee M. & Sloane-Stanley G.H. (1957) A simple method for the isolation and purification of total lipids from animal tissues. *Journal of Biological Chemistry* **226**, 497–509.
- Fowler L.G. (1981a) Tests of vegetable protein replacements and alternate ingredients in Abernathy diet. *Abernathy Salmon Culture*. Development Centre, Techn. Transfer Ser, no. **81**–1, 14pp.
- Fowler L.G. (1981b) Effect of lipid and amino acid supplementation in chinook salmon diets containing poultry by-product meal. *Abernathy Salmon Culture*.

- Development Center, Techn. Transfer Ser. no. **81**–4, 12pp.
- Fowler L.G. (1990) Feather meal as a dietary protein source during parr-smolt transformation in fall chinook salmon. *Aquaculture* **89**, 301–314.
- Fowler L.G. (1991) Poultry by-product meal as a dietary protein source in fall chinook salmon diets. *Aquaculture* **99**, 309–321.
- Gaylord T.G. & Gatlin D.M. (1996) Determination of digestibility coefficients of various feedstuffs for red drum (*Sciaenops ocellatus*). *Aquaculture* **139**, 303–314.
- Gouveia A.J.R. (1991) The use of animal by-products in diets for salmonids. *Série 'Monografias' No 5. Instituto De Zoologia 'Dr. Augusto Nobre'*. Universidade De Porto, Porto, Portugal.
- Gropp J., Koops H., Tiews K. & Beck H. (1976) Replacement of fish meal in trout feeds by other feedstuffs. FAO Technical Conference on Aquaculture. *FAO-FIR Aq\Conference\76 E* **24**, 596–601.
- Hanley F. (1987) The digestibility of foodstuffs and the effects of feeding selectively on digestibility determinations in tilapia *Oreochromis niloticus* L. *Aquaculture* **66**, 163–179.
- Higgs D.J., Dosanjh B.S., Prendergast A.F., Beames R.M., Hardy R.W., Riley W. & Deacon G. (1995) Use of rapeseed/canola protein products in finfish diets. In: *Nutrition and Utilization Technology in Aquaculture* (ed. by C.E. Lim & D.J. Sessa), 130–156. AOCS Press, Champaign, IL.
- Higgs D.A., Markert J.R., MacQuarrie D.W., McBride J.R., Dosanjh B.S., Nichols C. & Hoskins G. (1979) Development of practical dry diets for coho salmon, *Oncorhynchus kisutch*, using poultry by-product meal, feather meal, soybean meal and rapeseed meal as major protein sources. In: *Proceedings of the World Symposium of Finfish Nutrition and Fish Feed Technology, Hamburg 20–23 June, 1978, Vol. II*, 191–218
- Lu J.D. & Kevern N.R. (1975) The feasibility of using waste materials as supplemental fish feed. *Progress in Fish-Culture* **37**, 241–244.
- Markert J.R., Higgs D.A., Dye H.M. & MacQuarrie D.W. (1977) Influence of bovine growth hormone on growth rate, appetite, and food conversion of yearling coho salmon *Oncorhynchus kisutch*, fed two diets of different composition. *Canadian Journal of Zoology* **55**, 74–83.
- Mosconi-Bac N. (1987) Hepatic disturbances induced by an artificial feed in the sea bass (*Dicentrarchus labrax*) during the first year of life. *Aquaculture* **67**, 93–99.
- Mosconi-Bac N. (1990) Reversibility of artificial feed-induced hepatocyte disturbances in cultured juvenile sea bass (*Dicentrarchus labrax*): An ultrastructural study. *Aquaculture* **88**, 363–370.
- Nengas I., Alexis M. & Davies S.J. (1999) High inclusion levels of poultry meals and related By-products in diets for gilthead seabream (*Sparus aurata* L.). *Aquaculture* **179**, 13–23.
- NRC (National Research Council). (1993) *Nutrient Requirements of Fish*. National Academy Press, Washington, DC.
- Peacock H.A. (1973) *Elementary Microtechnique*, 4th edn, 278pp.
- Robaina L., Izquierdo M.S., Moyano F.J., Socorro J., Vergara J.M. & Montero D. (1998) Increase of the dietary n-3/n-6 fatty acid ratio and addition of phosphorus improve liver histological alterations induced by feeding diets containing soybean meal to gilthead seabream, (*Sparus aurata*). *Aquaculture* **161**, 281–293.
- Rodríguez-Serna M., Olvera M.A. & Carmona-Osalde C. (1996) Nutritional value of animal by-product meal in practical diets for Nile tilapia *Oreochromis niloticus* (L.) fry. *Aquaculture Research* **27**, 67–73.
- Roley D.D., Roley S.E., Hardy R.W. & Brannon E.L. (1977) Feather meal and candida yeast as substitutes for fish meal in the diets of chinook salmon fry (*Oncorhynchus Kisutch*). *Annual Report of College Fisheries University Washington, Seattle, WA*.
- Rumsey G.L. (1993) Fish meal and alternate sources of protein in fish feeds update 1993. *Fisheries* **18**, 14–19.
- Sadiku S.O.E. & Jauncey K. (1995) Digestibility, apparent amino acid availability and waste generation potential of soybean flour: poultry meat meal blend based diets for tilapia, *Oreochromis niloticus* (L.), fingerlings. *Aquaculture Research* **26**, 651–657.
- Shimeno S., Masumoto T., Hujita T. & Ueno S. (1993) Alternative protein sources for fish meal in diet of young yellowtail. *Nippon Suissan Gakkaishi* **59**(1), 137–143.
- Tacon A.G.J. (1994) Feed ingredients for carnivorous fish species. Alternatives to fish meal and other fishery resources. *Fish Circular*, no. 881. FAO, Rome.
- Tacon A.G.J. (1996) Commercial aquafeed production and use of fishery resources. *Feed Milling International* September, 17–32.
- Tacon A.G.J. & Jackson A. (1985) Utilization of conventional and non-conventional protein sources in practical feeds. In: *Nutrition and Feeding in Fish* (ed. by C.B. Cowey, A.M. Mackie & J.G. Bell), 119–145. Academic Press, London.
- Tiews K., Gropp J. & Koops H. (1976) On the development of optimal rainbow trout pellet feeds. *Archiv Fischereiwiss, Beih* **27**, 1–29.
- Westgate J.W. (1979) Hatchery biology – Columbia River Fishery Development Programme. *Annual Progress Report*. Oregon Department Fish Wildlife, Portland, OR.