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Partial and total replacement of fishmeal with poultry by-product meal in diets for gibel carp, *Carassius auratus gibelio* Bloch

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Abstract

Triplicate groups of gibel carp *Carassius auratus gibelio* Bloch (initial body weight: 4.89 g) were fed for 8 weeks at 24.8–30.8 °C with nine isonitrogenous and isoenergetic diets. The control diet (F1) used white fishmeal (FM) as the sole protein source. In the other eight diets (F2–F9), 40.5–100% of FM protein was substituted by poultry by-product meal (PBM) at 8.5% increments. The specific growth rate (SGR), feed efficiency ratio, protein efficiency ratio, protein retention efficiency and energy retention rate for fish fed PBM diets (F2–F9) were all higher, but not always significantly, than those for fish fed F1. All apparent digestibility coefficients for fish fed PBM diets were lower than those for fish fed F1. Fish fed F1 had a significantly higher hepatosomatic index value than fish fed PBM diets ($P < 0.05$). No significant ($P > 0.05$) effect of diet was found in whole-body moisture and fat content. Whole-body protein and energy content for fish fed PBM diets were slightly higher than that for fish fed F1. The optimal replacement level of FM by PBM was estimated by second-order polynomial regression to be 66.5% in protein.

Keywords: fishmeal, poultry by-product meal, replacement, gibel carp *Carassius auratus gibelio*

Introduction

Aquaculture is the fastest expanding food production system in the world. This rapid development largely depends upon the increased production of aquafeeds, which traditionally rely on fishmeal (FM) as the main protein source (Hardy & Tacon 2002). Global FM production varies around 6–7 million tonnes year^{-1} because of the fisheries conservation, but continuously the increasing demand for FM use in animal feed, especially in aquafeed, has resulted in FM becoming difficult to obtain and more expensive. As the largest importer of FM, China consumed 1 237 866 tonnes of FM in 2001, which accounted for about 20% of the world' total production of FM (FAO 2001). The statistical data showed that quantity of the imported FM in June 2005 was 721 040 tonnes (http://www.feedtrade.com.cn/market/fishmeal/fishmeali.stat/200507/79546.html).

It is urgent to reduce the FM used in aquafeed by replacing it with other protein sources. Among alternative animal protein sources, poultry by-product meal (PBM) has been shown to be of high nutritional quality, and is considered as a valuable source of protein for many species, especially for carnivorous fish species (Nengas, Alexis & Davies 1999). Initially, PBM could not replace more than 50% of FM in fish diets (Fowler 1981, 1982, 1991; Gallagher & Degani 1988; Steffens 1994). However, in recent years, PBM has replaced 75% or even 100% of FM, without a significant reduction in fish performance (Alexis 1997; Nengas *et al.* 1999; Takagi, Hosokawa, Shimeno & Ukawa 2000). This progress may be partly attributed to improvements in manufacturing technique applied to the meal, which in turn improved its digestibility and nutritive quality (Bureau, Harris & Cho 1999). On the other hand, as vast quantities of by-product waste generated in commercial poultry abattoirs are discarded indiscriminately into the environment in some Asian countries (Fagbenro & Fasakin 1996),

utilizing PBM as a feed protein source may make a contribution to the protection of the natural environment (Steffens 1994). Thus, the potential of these products should be evaluated further.

However, most of these studies have focused on salmonid species. There have been few studies on the replacement effects of PBM in diets for crucian carp, *Carassius auratus*, one of the most important aquaculture species in China. Gibel carp, *C. auratus gibelio,* is an improved strain of crucian carp and, in recent years, has almost replaced the latter in aquaculture in China because of its higher growth rate. The total production of gibel carp in China was more than 1.7 million tonnes in 2003 and keeps increasing. The protein requirement for the juveniles is around 38% (Qian 2001), and dietary protein covers about 60% of feed cost.

The objectives of the present study were to evaluate the effect on growth performance of the replacement of FM by different levels of PBM in the diet for gibel carp.

Materials and methods

Experimental diets

High-quality PBM, with 70.9% crude protein and 14.8% crude fat, was supplied by the Asian regional office of the National Renderers Association (Causeway Bay, Hong Kong). White FM from the American Seafood Company (Seattle, WA, USA) was purchased in a local market. FM and PBM were analysed for proximate composition (Table 1) before the formulation of diets. Nine approximately isonitrogenous (39% crude protein) and isoenergetic (18 MJ kg^{-1}) diets were prepared, with PBM replacing FM at different levels. The control diet (F1) was prepared with white FM as the only protein source, the remaining eight diets (F2–F9) were prepared with 40.5–100% of FM protein substituted by PBM at an 8.5% increment in PBM between diets (Table 2). This regime was based on a finding that PBM could replace 50% FM protein in diets for gibel carp without any adverse effects (Yang, Xie, Cui, Zhu, Yang & Yu 2004). All diets were formulated to contain 38% protein and 9.5% lipid. Chromic oxide (1%) was added to determine the apparent digestibility coefficient (ADC). The diets were made into sinking pellets using a laboratory pellet machine (SLH-200, Zhanwang Machine Ltd., Shanghai, China), oven-dried at 60 °C and stored at 4 °C until use. The chemical composition of the experimental diets is shown in Tables 2 and 3.

Table 1 Comparison of the proximate composition of fishmeal (FM) and poultry by-product meal used in the experimental diets (% dry matter)

Ingredient	Crude protein	Crude lipid	Ash	Moisture
White FM (American Seafood)	70.96	8.04	19.57	5.75
Poultry by-product meal (National Renderers Association)	70.91	14.28	15.33	2.23

Experimental conditions, fish and feeding

The experiment was conducted in an indoor semi-recirculation system consisting of 27 circular fibreglass tanks with flat bottoms (diameter 70 cm, water volume 90 L). Water was treated in a biological filter, aerated intermittently in a reservoir for 30 min per hour and then distributed to each tank at a rate of 3 L min^{-1}. During the experiment, the water quality was checked periodically. The water temperature ranged from 24.2 to 31.4 °C, and pH from 7.0 to 7.2; photoperiod was 12D:12L, with the light period from 08:00 to 20:00 hours. Dissolved oxygen was > 6.9 mg L^{-1}, and NH_4^+-N was < 0.1 mg L^{-1}.

Young-of-the-year gibel carp were obtained from the Guanqiao hatchery of the Institute of Hydrobiology (Wuhan, China). They were held in the experimental system, 50 fish per tank, for acclimation. Fish were fed a practical pellet (Tianan 906 crucian carp feed, Tianan, Haida, Wuhan, China) containing 39% protein for 1 month and then an equal mixture of experimental diets 1 week before the experiment. At the beginning of the experiment, fish were starved for 1 day and pooled. Twenty fish were randomly selected, batch weighed and stocked into each tank. Then, three groups of seven fish were sampled randomly from the remaining stock of fish and stored frozen at -20 °C for analyses of the initial whole-body composition.

Three tanks were randomly assigned to each diet. The mean initial weight of fish in all tanks was 4.89 g, and did not differ significantly between treatments ($P > 0.05$). Fish were fed to satiation twice a day (at 9:00 and 15:00 hours), 7 days $week^{-1}$. At each feeding, an excess amount of feed was weighed and fed to each tank. Uneaten feed was collected 1 h after feeding, dried at 70 °C and reweighed. Leaching rate was estimated by placing weighed feeds in tanks without fish for 1 h and then recollecting, drying and re-weighing the feeds. The average leaching rate was used to calibrate the amount of uneaten feed.

Table 2 Formulation and proximate composition of the experimental diets

	F1	F2	F3	F4	F5	F6	F7	F8	F9
Ingredient Replacement of FM CP%	0.00	40.50	49.00	57.50	66.00	74.50	83.00	91.50	100.00
FM	52.97	31.52	27.10	22.51	18.01	13.51	9.00	4.51	0.00
PBM	0.00	21.47	25.97	30.48	34.98	39.49	43.99	48.50	53.00
Wheat flour	18.20	18.20	18.20	18.20	18.20	18.20	18.20	18.20	18.20
Fish oil	4.21	2.74	2.46	2.18	1.90	1.61	1.33	1.05	0.77
Cellulose	17.12	18.58	18.86	19.13	19.41	19.69	19.97	20.24	20.53
Vitamins premix*	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Mineral premix†	6.00	6.00	6.00	6.00	6.00	6.00	6.00	6.00	6.00
Cr_2O_3	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Proximate composition									
Moisture (%)	6.40	7.14	6.23	6.85	6.71	7.54	7.40	7.25	6.52
Protein (%)	38.13	37.36	36.48	36.76	36.57	39.70	39.27	39.75	37.91
Fat (%)	9.63	9.27	10.56	9.50	9.12	10.44	10.15	9.58	9.39
Ash (%)	14.66	15.13	14.21	13.25	12.93	13.03	12.89	13.31	12.33
Energy ($J\,mg^{-1}$)	18.05	18.24	17.89	18.43	17.69	17.68	17.56	18.48	18.58

*Vitamin premix contained the following vitamins per kilogram feed: vitamin A (as vitamin A acetate and vitamin A palmitate, 1:1), 5500 IU; vitamin D_3, 1000 IU; vitamin E (as DL-α-tocopheryl acetate), 50 IU; vitamin K_3(as menadione sodium bisulphite), 10 IU; choline (as choline chloride), 550 mg; niacin, 100 mg; riboflavin, 20 mg; pyridoxine, 20 mg; thiamin, 20 mg; D-calacium pantothenate, 50 mg; biotin, 0.1 mg; foliacin, 5 mg; vitamin B_{12}, 20 mg; ascorbic acid, 100 mg; inositol, 100 mg.

†Mineral premix contained the following minerals as milligram per kilo gram feed: NaCl, 257; $MgSO_4 \cdot 7H_2O$, 3855; $Na_2H_2PO_4 \cdot 2H_2O$, 6425; KH_2PO_4, · 8224; $Ca(H_2PO_4)_2 \cdot H_2O$, 5140; $C_6H_{10}CaO_6 \cdot 5H_2O$, 899.5; $FeC_6H_5O_7 \cdot 5H_2O$, 642.5; $ZnSO_4 \cdot 7H_2O$, 90.7; $MnSO_4 \cdot 4H_2O$, 41.6; $CuSO_4 \cdot 5H_2O$, 7.97; $CoCl_2 \cdot 6H_2O$, 0.26; KIO_3, 0.77.

FM, fishmeal; PBM, poultry by-product meal.

Table 3 Amino acid compositions of experimental diets (% in dry matter)

Diet	F1	F2	F3	F4	F5	F6	F7	F8	F9
ASP	3.60	3.66	3.75	3.62	3.48	3.59	4.20	3.92	3.28
SER	2.12	1.83	1.48	1.62	1.59	1.71	1.68	1.98	1.89
GLU	7.08	5.42	5.98	5.72	5.79	5.89	7.08	7.48	6.58
GLY	2.78	2.90	2.42	3.03	3.05	3.39	3.48	3.90	4.16
HIS	0.75	0.78	0.80	0.72	0.66	0.75	0.88	0.95	0.95
ARG	3.91	3.57	3.39	3.97	3.24	4.16	4.00	4.37	2.84
THR	0.96	0.87	0.73	1.07	0.81	0.81	1.08	0.97	1.63
ALA	3.34	2.70	2.99	2.45	2.69	2.96	3.20	3.80	3.33
PRO	2.12	2.17	2.38	2.14	2.38	2.66	2.69	3.21	3.13
CYS	0.03	0.04	0.04	0.06	0.08	0.04	0.08	0.10	0.08
TYR	1.45	1.42	1.24	1.24	1.10	1.43	1.25	1.41	1.24
VAL	2.12	1.78	1.83	1.79	1.92	2.08	2.07	2.33	2.22
MET	1.28	0.89	1.09	1.16	1.13	1.32	1.20	1.33	1.18
LYS	2.75	2.06	2.09	1.74	1.96	2.09	2.14	2.34	2.00
ILE	1.70	1.64	1.46	1.32	1.47	1.46	1.52	1.75	1.70
LEU	3.03	2.77	2.48	2.46	2.55	2.74	2.77	3.06	2.84
PHE	1.85	1.68	1.51	1.58	1.62	1.80	1.66	1.92	1.80
TRP	Not determined								
Total	40.88	36.20	35.65	35.71	35.51	38.89	40.98	44.80	40.86

Faeces were collected 1 h after the collection of uneaten feed 5 days after the start of the experiment and throughout the experimental period. To minimize nutrient leaching in faeces, only fresh and intact faeces were collected and dried to a constant weight at 70 °C. During the trial, dead fish were removed and weighed.

The growth trial lasted for 8 weeks. At the end of the trial, fish were starved for 1 day and batch weighed. Five fish were selected randomly from each tank and dissected, and the liver was removed and weighed. Another batch of five fish from each tank were selected at random and stored frozen at − 20 °C for subsequent proximate composition analysis.

Chemical analysis

The ingredients, diet and pooled fish samples from each tank were analysed for proximate composition and energy content. Diet and faeces were also analysed for chromic oxide.

Dry matter was determined by drying to a constant weight at 105 °C. Nitrogen was determined using the semi-Kjeldahl method and the protein content was calculated from the nitrogen content multiplied by 6.25. Lipid was determined by ether extraction using a Soxtec system (Soxtec System HT6, Tecator, Hgans, Sweden), ash using combustion at 550 °C (AOAC 1984), energy by bomb calorimetry (Phillipson microbomb calorimeter, Gentry Instruments, Aiken, SC, USA) and Cr_2O_3 content using the method described by Bolin, King and Klosterman (1952). At least duplicate measurements were made for each sample.

One-way analysis of variance was used to test the effects of the diets. Duncan's procedure was used for multiple comparisons. Differences were regarded as significant when $P < 0.05$. Second-order polynomial regression analysis of the relationship between the specific growth rate (SGR) and the replacement levels of protein of PBM was used to estimate the optimal replacement level of protein of FM by PBM in the diets for gibel carp.

Results

Digestibility

Data on apparent digestibility coefficients (ADC) are presented in Table 4. All ADC values obtained in fish fed the PBM diets were lower than those observed for the control diet, F1. There was a significant effect of diet on the ADC of dry matter ($P < 0.05$), with the values for F2, F4, F5 and F6 diets being significantly lower than the value for the control F1. Apparent digestibility coefficient values of protein (ADC_P) in all PBM groups were significantly lower than that of F1 except for F8 ($P < 0.05$). Apparent digestibility coefficient values of energy in all PBM groups were significantly lower than that of F1 except for F3 ($P < 0.05$).

Growth performance and diet utilization

All diets were well accepted by gibel carp. Feed intake was not significantly ($P > 0.05$) affected by the treatments (Table 5). The final body weight and SGR in diets containing PBM were significantly higher than those in the control group ($P < 0.05$).

Table 4 Apparent digestibility coefficients (ADC, %) of nine experimental diets

ADC (%)	F1	F2	F3	F4	F5	F6	F7	F8	F9
ADC_D*	66.25 ± 2.09^{a}	57.80 ± 0.25^{bc}	62.92 ± 0.56^{ab}	56.86 ± 0.94^{cd}	58.85 ± 1.38^{bc}	54.22 ± 2.09^{c,d}	51.88 ± 2.5^{d}	64.09 ± 3.12^{ab}	64.47 ± 1.11^{ab}
ADC_P†	89.92 ± 0.71^{a}	81.89 ± 0.53^{b}	85.33 ± 0.97^{bc}	82.16 ± 0.42^{c}	83.00 ± 1.18^{c}	82.62 ± 1.88^{c}	82.29 ± 0.65^{c}	87.39 ± 1.20^{ab}	84.95 ± 0.75^{bc}
ADC_E‡	76.00 ± 1.81^{a}	68.39 ± 0.65^{b}	72.39 ± 1.13^{ab}	67.56 ± 1.61^{cd}	69.68 ± 1.43^{bc}	63.79 ± 2.04^{d}	59.9 ± 0.68^{d}	70.87 ± 2.06^{bc}	68.78 ± 0.77^{bc}

^{a,b,c,d}Values in the same row with different superscripts are significantly different ($P < 0.05$).

*ADC_D: ADC of dry matter = (1 − % Cr_2O_3 in diet/% Cr_2O_3 in faeces) × 100.

†ADC_P: ADC of protein = (1 − % protein in faeces × % Cr_2O_3 in diets/(% protein in diets × % Cr_2O_3 in faeces)) × 100.

‡ADC_E: ADC of energy = (1 − % energy content in faeces × % Cr_2O_3 in diets/(% energy content in diets × % Cr_2O_3 in faeces)) × 100.

Values are mean ± standard errors ($n = 3$).

Table 5 Growth performance of gibel carp fed five experimental diets for 56 days

	F1	F2	F3	F4	F5	F6	F7	F8	F9
IBW*	4.89 ± 0.00	4.90 ± 0.01	4.87 ± 0.01	4.87 ± 0.00	4.88 ± 0.02	4.89 ± 0.01	4.91 ± 0.01	4.89 ± 0.01	4.88 ± 0.01
FBW†	20.81 ± 0.60^{a}	28.50 ± 0.49^{b}	30.65 ± 1.60^{bc}	30.00 ± 0.50^{bc}	31.15 ± 0.32^{bc}	32.61 ± 1.04^{c}	29.72 ± 0.58^{bc}	28.96 ± 0.31^{b}	28.32 ± 1.47^{b}
SGR‡	2.58 ± 0.05^{a}	3.15 ± 0.03^{b}	3.28 ± 0.09^{bc}	3.24 ± 0.03^{bc}	3.31 ± 0.02^{bc}	3.39 ± 0.06^{c}	3.22 ± 0.04^{bc}	3.17 ± 0.02^{b}	3.14 ± 0.09^{b}
FR§	3.73 ± 0.09	3.76 ± 0.04	4.18 ± 0.50	3.77 ± 0.14	3.84 ± 0.04	3.93 ± 0.14	4.22 ± 0.10	4.15 ± 0.07	4.02 ± 0.14
FER¶	59.37 ± 1.46^{a}	67.08 ± 0.33^{bc}	69.91 ± 1.49^{c}	68.76 ± 1.84^{c}	67.91 ± 0.81^{bc}	67.77 ± 3.15^{bc}	61.09 ± 0.75^{a}	61.27 ± 0.91^{a}	63.52 ± 1.52^{ab}
PER ‖	1.66 ± 0.04^{d}	1.93 ± 0.01^{ab}	2.04 ± 0.04^{a}	1.99 ± 0.07^{a}	1.99 ± 0.02^{a}	1.83 ± 0.08^{bc}	1.67 ± 0.03^{d}	1.66 ± 0.02^{d}	1.76 ± 0.03^{cd}
PRE**	22.29 ± 1.81^{b}	26.79 ± 1.76^{ab}	30.41 ± 0.91^{ab}	28.34 ± 1.46^{ab}	30.63 ± 1.86^{a}	26.42 ± 0.81^{ab}	23.87 ± 0.69^{b}	24.73 ± 1.00^{ab}	27.53 ± 1.27^{ab}
ERR††	27.30 ± 0.04	30.07 ± 0.31	32.22 ± 0.57	30.2 ± 1.65	32.46 ± 1.01	31.73 ± 0.80	29.67 ± 1.02	29.02 ± 0.28	29.69 ± 0.73
HIS‡‡	7.30 ± 0.29^{a}	5.05 ± 0.12^{bc}	5.11 ± 0.21^{bc}	4.82 ± 0.15^{c}	5.15 ± 0.33^{bc}	4.86 ± 0.05^{c}	5.16 ± 0.14^{bc}	5.67 ± 0.10^{b}	5.13 ± 0.17^{bc}

^{a,b,c,d}Values in the same row with different superscripts are significantly different ($P < 0.05$).

*IBW (g): initial body weight.

†FBW (g): final body weight.

‡SGR (% day^{-1}): specific growth rate = 100 × ((ln FBW − ln IBW)/56 days).

§FR (% bw day^{-1}): feeding rate = 100 × dry feed intake/(56 days × (FBW+IBW)/2).

¶FER (%): feed efficiency ratio = 100 × wet weight gain/dry feed intake.

‖ PER: protein efficiency ratio = 100 × weight gain/crude protein intake.

**PRE: protein retention efficiency = 100 × (final body protein − initial body protein)/protein intake.

††ERE: energy retention efficiency = 100 × (final body energy − initial body energy)/energy intake.

‡‡HSI: hepato-somatic index = hepatopancreas weight/somatic weight.

Values are mean ± standard errors ($n = 3$).

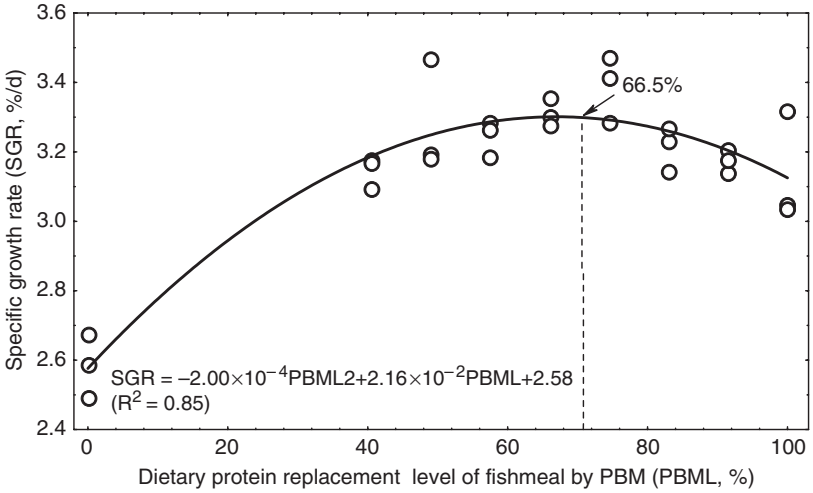

Figure 1 Effect of different dietary protein replacement levels of fishmeal by poultry by-product meal (PBML) on the specific growth rate (SGR) of gibel carp. A growth peak was reached when the replacement level was 66.69% (SGR $= -2.00 \times 10^{-4}$PBML^2+2.16 $\times$ 10^{-2}PBML+2.58, $R^2 = 0.85$). At this level 35.5% PBM was included in the diet.

There was a significant effects of diet on the feed efficiency ratio (FER) and the protein efficiency rate (PER) ($P < 0.05$). Groups F2–F6 all had significantly higher FER and PER than the control, F1. The protein retention efficiency (PRE) in diets F2–F9 was higher than that of the control diet. The energy retention efficiency (ERE) of fish fed diets F2–F9 was not significantly different from the controls ($P > 0.05$). Fish fed the control diet had a significantly higher mean hepatosomatic index than those fed PBM diets ($P < 0.05$).

A second-order polynomial regression analysis expressed the relationship between SGR and the dietary protein replacement level of FM by PBM (PBML), with a growth maxim reached when the PBM inclusion level was 35.5% of the diet (SGR $= -2.00 \times 10^{-4}$PBML^2+2.16 $\times$ 10^{-2}PBML+2.58, $R^2 = 0.85$) (Fig. 1). At this level, 66.5% FM protein was replaced by PBM.

Whole-body composition

Table 6 gives the whole-body composition of fish at the beginning and at the end of the trial. No significant difference ($P > 0.05$) was found in the final whole-body moisture content for fish fed the different diets. The final whole-body protein content of fish fed diets (F2–F9) was higher than that of fish fed the control diet (F1). Whole-body fat content in the fish fed nine experimental diets was similar and significantly higher ($P < 0.05$) than in initial fish samples. The highest value for whole-body ash content was seen in fish fed diet F8. The gross energy content of fish fed experimental diets was significantly higher ($P < 0.05$) than that in initial fish samples, and increased with increasing level of PBM in diets.

Discussion

Poultry by-product meal is a rendered product obtained from the waste of poultry production and processing plants. It is usually made from inedible portions of poultry, excluding feathers. It has been studied as a FM replacement in the diets for chinook salmon *Oncorhynchus tshawytscha* (Fowler 1981, 1982, 1991), rainbow trout *Oncorhynchus mykiss* (Alexis, Papaparaskeva-papoutsoglou & Theochari 1985; Steffens 1994), coho salmon *Oncorhynchus kisutch* (Higgs, Markert, Macquarrie, Mcbride, Dosanjh, Nichols & Hoskins 1979), European eels *Anguilla anguilla* (Gallagher & Degani 1988), gilthead seabream *Sparus aurata* (Alexis 1997; Nengas *et al.* 1999), red sea bream *Chrysophrys major* (Takagi *et al.* 2000), sunshine bass *Morone chrysops* × *M. saxatilis* (Webster, Tiu & Morgan 1999), red drum *Sciaenops ocellatus* (Kureshy, Davis & Arnold 2000) and Pacific white shrimp *Litopenaeus vannamei* (Davis & Arnold 2000).

Early studies had shown that PBM, if used alone, could generally replace not more than 50% of FM protein, or growth was compromised (Fowler 1981, 1982; Steffens 1994). If supplemented with amino

Table 6 Body composition (expressed on a wet weight basis) of gibel carp fed the experimental diets

	Initial	F1	F2	F3	F4	F5	F6	F7	F8	F9
Moisture (%)	72.87 ± 0.27	68.70 ± 0.30	69.29 ± 0.08	68.64 ± 0.85	68.92 ± 0.60	68.62 ± 0.23	69.33 ± 0.51	68.20 ± 0.29	67.44 ± 0.69	67.72 ± 0.25
Protein (%)	13.77 ± 0.20^{ab}	12.81 ± 0.78^{a}	13.35 ± 0.81^{ab}	14.21 ± 0.36^{ab}	13.68 ± 0.34^{ab}	14.68 ± 0.84^{ab}	13.93 ± 0.55^{ab}	13.76 ± 0.17^{ab}	14.23 ± 0.68^{ab}	14.81 ± 0.50^{b}
Fat (%)	6.72 ± 0.06^{a}	10.53 ± 0.23^{b}	9.94 ± 0.25^{b}	10.34 ± 0.74^{b}	10.14 ± 0.21^{b}	10.80 ± 0.26^{b}	10.20 ± 0.71^{b}	11.30 ± 0.24^{b}	11.01 ± 0.49^{b}	10.63 ± 0.40^{b}
Ash (%)	4.60 ± 0.18^{ab}	4.40 ± 0.12^{b}	4.58 ± 0.04^{ab}	4.53 ± 0.15^{b}	4.50 ± 0.07^{b}	4.44 ± 0.10^{b}	4.41 ± 0.05^{b}	4.51 ± 0.11^{b}	4.87 ± 0.06^{a}	4.66 ± 0.07^{ab}
Energy (J mg^{-1})	5.83 ± 0.06^{a}	7.31 ± 0.14^{b}	7.29 ± 0.05^{b}	7.42 ± 0.24^{bc}	7.30 ± 0.13^{b}	7.56 ± 0.14^{bc}	7.45 ± 0.12^{bc}	7.60 ± 0.09^{bc}	7.73 ± 0.09^{bc}	7.85 ± 0.26^{c}

^{a,b,c}Values in the same row with different superscripts are significantly different ($P < 0.05$).
Values are mean ± standard errors ($n = 3$).

acids (lysine, methionine or tryptophan) or combined with other protein, PBM could show a more pronounced nutritional potential (Webster *et al.* 1999). These tests were carried out with a PBM that normally contained not more than 60% crude protein and 16–22% ash. However, an improvement in the nutritional quality of PBM has been achieved in recent years. High-quality PBM now contains about 70% crude protein and a relatively low ash content (Nengas *et al.* 1999; Davis & Arnold 2000; Kureshy *et al.* 2000), and can even be used without supplementation, replacing 75% or even 100% of the FM without a significant depression in fish performance (Alexis 1997; Nengas *et al.* 1999; Takagi *et al.* 2000). This improvement can be mainly ascribed to better refining practices used in the production of this ingredient to meet the stringent requirements of the pet food industry, the main market for this ingredient (Bureau *et al.* 1999).

In comparison with white FM, the PBM product used in this study had an equally high protein content (71%), a lower ash content, but a higher lipid content. All the experimental diets were well accepted by gibel carp.

Our study indicated that the diets including PBM were better than the control diet (F1), containing only FM. Takagi *et al.* (2000) had found that yearling red sea bream fed diets with up to 100% FM replaced by PBM showed a growth performance and feed utilization similar to or better than fish fed FM-based control diets. Davis and Arnold (2000) reported that replacement of 80% FM protein in practical diets for *L. vannamei* resulted in a significant increase in weight gain and FER. These varying results may relate to the species tested, but are more likely to be a consequence of the different quality of PBM production, which varies among producers (Dong, Hardy, Haard, Barrows, Rasco, Fairgrieve & Forster 1993; Burearu *et al.* 1999). Dong *et al.* (1993) found that there were significant differences in proximate composition and protein digestibility in PBM samples obtained from six different manufacturers.

All variables related to feed utilization, including PER, FER, PRE and ERE, estimated for gibel carp fed diets containing PBM were higher than those in fish fed the control diet F1. Protein efficiency ratio and FER were generally related to protein ADC. Published ADC values for fish fed diets containing PBM are scare. In our study, ADC_P values for gibel carp fed diets with different levels of PBM ranged from 81.89 to 87.39. These values are comparable with values obtained for gilthead seabream fed diet containing

PBMa, a high-fat PBM available on the Greek market (Nengas *et al.* 1999), but higher or lower than those of gilthead seabream fed seven other diets containing different PBM products (Nengas *et al.* 1999). Apparent digestibility coefficient values of protein values for juvenile red sea bream (88.3–93.6%, Takagi *et al.* 2000) fed five diets with PBM replacing 0–100% FM were all higher than our values. These differences may have been largely caused by variation in the quality of raw materials of PBM (Dong *et al.* 1993) and differences in the methods of faeces collection (Watanabe & Pongmaneerat 1991; Bureau *et al.* 1999; Takagi *et al.* 2000). Although it is well documented that high ash contents in meat and bone meal could reduce protein digestibility (Alexis 1997; Robaina, Moyano, Izquierdo, Socorro, Vergara & Montero 1997; Kureshy *et al.* 2000), the similar conclusion could not be made in the current study and other similar experiments (Nengas *et al.* 1999; Yang *et al.* 2004).

In the present study, a variation in ADC_P similar to that of FE and PER was observed among fish fed diets with PBM. However, the lower FE and PER observed for fish fed the FM-based control diet (F1) could not be explained by a reduction in ADC_P, because the highest ADC_P occurred in F1. This may imply that ADC values are not a valuable index for the evaluation of the nutritional quality of protein sources, because their different effects might be masked by a high feed intake or good palatability (Hardy & Masumoto 1990).

There was no difference in the whole-body moisture and fat content of fish fed the experimental diets. Whole-body protein and energy content for fish fed PBM diets were slightly higher than those for fish fed the control diet. In rainbow trout (Alexis *et al.* 1985; Steffens 1994) chinook salmon (Fowler 1991) and European eel (Gallagher & Degani 1988), whole-body fat content of fish fed diets containing PBM increased, because PBM generally has a high fat content. PBM used in the present study also contained a much higher fat content than that of FM, but no effect of dietary PBM on whole-body fat content of gibel carp was observed. Takagi *et al.* (2000) did not find any effect of different rates of dietary inclusion of low-fat PBM (with 6.7% fat) on whole-body composition in yearling red sea bream. Different whole-body compositions, including fat content, were observed for juvenile red sea bream fed diets with the same PBM content (Takagi *et al.* 2000). This finding suggests that differences in utilization and transformation capacity for PBM are shown by different species, or even in the different growth stages of fish.

Polynomial regression analysis has been suggested as a method for describing and approximating the relationship between growth and essential nutrient intake (Zeitoun, Ullrey & Magee 1976). In the present study, this method suggested that the optimal replacement level of FM by PBM was about 66.5%, with PBM forming about 35.5% of the feed.

In conclusion, this study has suggested that high-quality PBM could replace 100% FM in diets for gibel carp without adversely affecting growth performance and feed utilization, but an optimal replacement level of 66.5% is suggested.

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