Effect of replacement of dietary fish meal by meat and bone meal and poultry by-product meal on growth and feed utilization of gibel carp, *Carassius auratus gibelio*

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Abstract

Triplicate groups of gibel carp *Carassius auratus gibelio* (initial body weight: 5.25 ± 0.02 g) were fed for 8 weeks at 20–25 °C on five isonitrogenous (crude protein: 400 g kg^{-1}) and isoenergetic diets (gross energy: 17 kJ g^{-1}). Meat and bone meal (MBM) or poultry by-product meal (PBM) were used to replace fish meal at different levels of protein. The control diet contained fish meal as the sole protein source. In the other four diets, 150 or 500 g kg^{-1} of fish meal protein was substituted by MBM (MBM$_{15}$, MBM$_{50}$) or PBM (PBM$_{15}$, PBM$_{50}$). The results showed that feeding rate for the MBM$_{50}$ group was significantly higher than for other groups except the PBM$_{50}$ group ($P < 0.05$). Growth rate in the MBM$_{15}$ group was significantly higher than that in the control ($P < 0.05$), while there was no significant difference in growth between the control and other groups ($P > 0.05$). Feed efficiency and protein efficiency ratio in MBM$_{50}$ was significantly lower while that in MBM$_{15}$ was significantly higher ($P < 0.05$). Replacement of fish meal by MBM at 500 g kg^{-1} protein significantly decreased apparent dry matter digestibility (ADC$_D$) and gross energy (ADC$_E$) while apparent protein digestibility (ADC$_P$) was significantly decreased by the replacement of MBM or PBM ($P < 0.05$). The results suggest that MBM and PBM could replace up to 500 g kg^{-1} of fish meal protein in diets for gibel carp without negative effects on growth while 150 g kg^{-1} replacement by MBM protein improved feed utilization.

KEY WORDS: *Carassius auratus gibelio*, feed utilization, growth, meat and bone meal, poultry by-product meal

Introduction

Rapid development of global aquaculture has resulted in increased production of aquafeed which traditionally relies on fish meal as the main protein source (Tacon & Jackson 1985). However, fish meal is becoming limited due to increasing demand and decreasing marine fishery resources. Therefore, alternative proteins, including plant and animal protein, have been studied by many fish nutritionists and the feed industry (Tacon & Jackson 1985). However, plant protein inclusion has normally been limited due to deficiencies in essential amino acid, anti-nutrients factors and/or poor palatability (Gomes *et al.* 1995). Meat and bone meal (MBM) and poultry by-product meal (PBM) are two potential animal protein sources because of their high protein contents and low price compared to fish meal (Tacon & Jackson 1985). Generally the substitution of MBM for fish meal is less than 300 g kg^{-1} in fish feeds (Pongmaneerat & Watanabe 1991; Robaina *et al.* 1997; Bureau *et al.* 2000; Kureshy *et al.* 2000), but even higher replacement with positive results has been reported in gilthead seabream *Sparus aurata* Linnaeus (Alexis 1997), Mozambique tilapia *Oreochromis mossambicus* Peters (Davies *et al.* 1989) and rainbow trout *Oncorhynchus mykiss* Walbaum (Watanabe & Pongmaneerat 1991). Previous studies have shown that PBM cannot replace more than 500 g kg^{-1} of fish meal in fish diets (Gallagher & Degani 1988; Fowler 1991; Steffens 1994). Other studies have shown that as PBM quality has been significantly improved in recent years it could replace 750 g kg^{-1} or even 1000 g kg^{-1} of fish meal without significant decrease in fish growth performance (Alexis 1997; Nengas *et al.* 1999; Takagi *et al.* 2000). Differences could be partly caused by the inconsistent quality of MBM/PBM which depends on the processing method and/or the materials. Better manufacturing practices may improve the quality of animal by-products (Bureau *et al.* 1999, 2000).

The potential of these products should therefore be re-evaluated.

Most studies on evaluation of MBM and PMB have focused on salmonids and related species while there have been fewer on cyprinidae, the most important aquaculture species in China and other countries (Welcomme 1988). Gibel carp, *Carassius auratus gibelio* is an improved strain of crucian carp and is becoming increasingly popular as an aquaculture species in China due to its excellent taste and high growth rates compared with other crucian carps (Xue & Cui 2001). The present study was designed to investigate the effect of replacement of fish meal by MBM or PBM on growth and feed utilization of gibel carp.

Materials and methods

Experimental diets

MBM and PBM were provided by the Asian Regional Office of the National Renderers Association (NRA), USA, and white fish meal from Russia was used as the reference protein in experimental diets. All ingredients were analysed for chemical composition (Table 1) before diet formulation. Five isonitrogenous [400 g kg^{-1} crude protein content] and iso-energetic (17 kJ g^{-1}) diets were formulated. The control diet contained white fish meal as the only protein source. In the other four diets, 150 or 500 g kg^{-1} of fish meal protein were substituted by MBM (MBM$_{15}$, MBM$_{50}$) or PBM (PBM$_{15}$, PBM$_{50}$) (Table 2). All diets were formulated to contain 400 g kg^{-1} protein and 75 g kg^{-1} lipid. Chromic oxide (10 g kg^{-1}) was added as a marker to determine apparent digestibility coefficient (ADC). Diets were made into sinking pellets (2 mm in diameter) by a pelleter, oven-dried at 60 °C and stored at 4 °C.

Experimental conditions, fish and feeding

The experiment was conducted in an indoor recirculation system. The system contained 15 fibreglass tanks with cyl-

Table 1 Proximate composition of the proteins used in the experimental diets (g kg^{-1} dry matter)

Ingredients	White fish meal (FM)	Meat and bone meal (MBM)	Poultry by-product meal (PBM)
Crude protein	700.4	498.6	529.3
Crude lipid	68.2	106.3	183.5
Ash	214.7	296.1	201.4
Moisture	69.7	78.5	17.6

Table 2 Formulation and chemical composition of the experimental diets (g kg^{-1} dry matter)

Ingredient	Control	MBM$_{15}$	MBM$_{50}$	PBM$_{15}$	PBM$_{50}$
White fish meal	543.1	462.2	272.3	462.6	272.4
MBM	–	140.5	466.3	–	–
PBM	–	–	–	108.2	359.0
Wheat flour	171.2	171.2	171.2	171.2	171.2
Fish oil	46.2	37.1	16.3	33.7	–
Cellulose	165.2	115.4	–	151.7	123.9
Vitamin premix[1]	5.0	5.0	5.0	5.0	5.0
Mineral premix[2]	60.0	60.0	60.0	60.0	60.0
Cr$_2$O$_3$	10.0	10.0	10.0	10.0	10.0
Chemical composition (g kg^{-1} dry matter)					
Dry matter	984.2	983.3	977.1	971.8	984.9
Crude protein	388.2	391.4	416.4	393.7	402.1
Lipid	72.3	78.0	76.3	74.4	73.5
Ash	163.2	192.6	263.7	172.4	185.3
Gross energy(kJ g^{-1})	17.02	17.31	16.52	16.94	17.23

MBM, meat and bone meal; PBM, poultry by-product meal.
[1] Vitamin premix contained the following vitamins (kg^{-1} 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 bisulfite), 10 IU; choline (as choline chloride), 550 mg; niacin, 100 mg; riboflavin, 20 mg; pyridoxine, 20 mg; thiamin, 20 mg; D-calcium pantothenate, 50 mg; biotin, 0.1 mg; foliacin, 5 mg; vitamin B$_{12}$, 20 mg; ascorbic acid, 100 mg; inositol, 100 mg.
[2] Mineral premix contained the following minerals (mg kg^{-1} feed): NaCl, 257; MgSO$_4$·7H$_2$O, 3855; N$_a$H$_2$PO$_4$·2H$_2$O, 6425; KH$_2$PO$_4$, 8224; Ca(H$_2$PO$_4$)$_2$·H$_2$O, 5140; C$_6$H$_{10}$CaO$_6$·5H$_2$O, 899.5; Fe-C$_6$H$_5$O$_7$·5H$_2$O, 642.5; ZnSO$_4$·7H$_2$O, 90.7; MnSO$_4$·4H$_2$O, 41.6; CuSO$_4$·5H$_2$O, 7.97; CoCl$_2$·6H$_2$O, 0.26; KIO$_3$, 0.77.

indroconical bottoms (diameter: 80 cm, water volume: 300 L). Each tank was connected to a filter tank filled with zeolite and active carbon. Water was recirculated through the filter at a rate of 4 L min^{-1}. Additional aeration was provided 30 min h^{-1} by an inflator controlled by a timer. During the experiment water temperature ranged from 19.7 to 25.7 °C (mean 23.6 °C), photoperiod was 12D : 12L with the light period from 08:00 to 20:00. Dissolved oxygen was above 6 mg L^{-1}, and ammonia-N (NH$_4^+$–N plus NH$_3$–N) was less than 0.5 mg L^{-1}.

Young-of-the-year gibel carp (initial body weight 5.26 g) were obtained from the hatchery farm of the Institute of Hydrobiology, the Chinese Academy of Sciences. They were held in the experimental system at 50 fish per tank for acclimatization. During acclimatization, fish were fed a practical pellet containing 400 g kg^{-1} protein and about 450 g kg^{-1} fish meal for 2 months and then an equal mixture of experimental diets for 1 week prior to the experiment. At the beginning of the experiment, fish were starved for 1 day and pooled. Twenty fish were randomly selected, batch weighed and allocated into each tank. Three tanks were randomly assigned to each diet. Initial body weight of the fish was 5.25 ± 0.02 g (mean ± SE.).

During the experiment, fish were fed to satiation twice daily (9:00 and 15:00 hours), 7 days per week. At each feeding, an excess amount of feed was fed to fish and uneaten feed were collected 1 h after feeding, dried at 70 °C and reweighed. Leaching rate of uneaten feed in tanks was estimated by placing weighed feeds into a tank without fish for 1 h and then recovering, drying and reweighing. The average leaching rate was used to calibrate the amount of uneaten feed. Faeces were collected after uneaten feed collection 5 days after the start of the experiment and through all the experiment period. To minimize nutrient leaching in faeces, only fresh and intact faeces were collected.

The growth trial lasted for 8 weeks. At the end of the trial, the fish were starved for 1 day and batch weighed.

Chemical analysis

Dry matter was determined by drying to constant weight at 105 °C [Association of Official Analytical Chemists (AOAC) 1984], nitrogen was determined using an elemental analyzer (Perkin Elmer 2400; Perkin-Elmer Corporation, Norwalk, CT, USA) and protein content was calculated from nitrogen (N) content by multiplying by 6.25. Lipid was determined by ether extraction using a Soxtec system (Soxtec System HT6; Tecator, Höganäs, Sweden), ash by combustion at 550 °C, gross energy by bomb calorimetry (Phillipson Microbomb Calorimeter; Gentry Instruments Inc., Aiken, SC, USA), and Cr$_2$O$_3$ content by concentrated nitric and perchloric acid digestion (Bolin et al. 1952). At least duplicate measurements were made for each sample.

One-way analysis of variance was used to test the effect of diets. Tukey's procedure was used for multiple comparisons. Differences were regarded as significant when $P < 0.05$.

Results

There were only two mortalities recorded during the experiment which were not related to any specific problem.

Table 3 showed that feeding rate (FR) of fish fed diets containing 500 g kg^{-1} MBM (MBM$_{50}$) was significantly higher while FR for fish fed the control diet, MBM$_{15}$ and PBM$_{15}$ was lower ($P < 0.05$). There was no significant difference between FR in PBM$_{50}$ and other groups ($P > 0.05$).

Table 3 Growth and feed utilization of gibel carp fed different diets (mean ± SE)[1]

	Control	MBM$_{15}$	MBM$_{50}$	PBM$_{15}$	PBM$_{50}$
IBW (g)[2]	5.28 ± 0.01	5.25 ± 0.02	5.26 ± 0.02	5.25 ± 0.01	5.25 ± 0.03
FBW (g)[3]	22.93 ± 0.52^b	25.91 ± 0.97^a	24.91 ± 0.80^{ab}	22.90 ± 0.31^b	22.04 ± 0.21^b
FR (% bw day^{-1})[4]	3.14 ± 0.06^b	3.19 ± 0.09^b	3.99 ± 0.15^a	3.20 ± 0.11^b	3.57 ± 0.24^{ab}
SGR (% day^{-1})[5]	2.62 ± 0.04^{bc}	2.85 ± 0.06^a	2.78 ± 0.05^{ab}	2.63 ± 0.02^{bc}	2.56 ± 0.01^c
FE (%)[6]	71.37 ± 1.05^{ab}	74.32 ± 1.17^a	58.82 ± 1.41^c	70.09 ± 2.61^{ab}	62.07 ± 3.97^{bc}
PER[7]	1.76 ± 0.02^{ab}	1.90 ± 0.03^a	1.40 ± 0.05^c	1.79 ± 0.07^{ab}	1.54 ± 0.10^{bc}

MBM, meat and bone meal; PBM, poultry by-product meal.
[1] Values are mean ± SE ($n = 3$).
[2] Initial body weight.
[3] Final body weight.
[4] Feeding rate = 100 × dry feed intake/[56 days × (FBW + IBW)/2].
[5] Specific growth rates = 100 × [(ln FBW–ln IBW)/56 days].
[6] Feed efficiency = 100 × wet weight gain/dry feed intake.
[7] Protein efficiency ratio = weight gain/crude protein intake.
[a,b,c] Values in the same row with same superscripts are not significantly different ($P > 0.05$).

Specific growth rate (SGR) for MBM_{15} was significantly higher and that for PBM_{50} lower ($P < 0.05$). Feed efficiency (FE) and protein efficiency ratio (PER) was significantly higher for MBM_{15} and lower for MBM_{50} ($P < 0.05$).

Apparent dry matter (ADC_D) and gross energy (ADC_E) digestibility coefficients were significantly lower for MBM_{50} ($P < 0.05$). Apparent crude protein digestibility coefficients (ADC_P) were significantly lower for groups with inclusion of MBM or PBM than in the control ($P < 0.05$) (Table 4).

Discussion

Meat and bone meals are generally considered inferior animal proteins and rarely included in commercial diets as main protein sources. Watanabe & Pongmaneerat (1991) evaluated the nutritional quality of MBM used as the sole protein source at different protein levels in diets for rainbow trout. They indicated that MBM was not an ideal protein source compared with fish meal due to its relatively poor amino acid profile. Replacement of fish meal with MBM generally compromised the growth performance of the fish. Kikuchi *et al.* (1997) reported that replacement of fish meal with MBM at levels higher than 200 g kg^{-1} resulted in reduced per cent weight gain in Japanese flounder *Paralichthys olivaceus*. Yellowtail *Seriola quinqueradiata* showed reduced growth performance when fed with practical diets with 192 g kg^{-1} fish meal substituted by MBM (Shimeno *et al.* 1993). Red drum *Sciaenops ocellatus* Linnaeus could only tolerate less than 50 g kg^{-1} MBM (replacement 167 g kg^{-1} fish meal) (Kureshy *et al.* 2000).

In the present study, however, growth performance of gibel carp fed diets with MBM was similar (in MBM_{50} group) or superior (in MBM_{15} group) to that fed the control diet in which fish meal was the only protein source. This result is similar to Robaina *et al.* (1997), who reported that 280 g kg^{-1} MBM (replacement 400 g kg^{-1} of fish meal protein) could be included in diets for gilthead seabream without

adverse effects on growth performance of fish. This positive effect could largely be due to good palatability of MBM which could increase feed intake (Mohson & Lovell 1990; Watanabe *et al.* 1993; Wu *et al.* 1999). Although several indispensable amino acids (e.g. lysine and methionine) were reported to be deficient in MBM and MBM was considered to be of low nutritional value (Watanabe & Pongmaneerat 1991), high inclusion levels up to 150 g kg^{-1} of fish meal protein by MBM showed similar growth and feed utilization compared with the control. It was suggested that limited inclusion might not result in poor essential amino acid profile.

Although the highest feed intake occurred in the MBM50 group, the lowest FE and PER were also observed in this group. Yamamoto *et al.* (2002) reported in juvenile rainbow trout that high dietary ash resulted in higher feed intake and lower feed efficiency. The high ash content of MBM may produce a faster gut transit rate, thus providing an increased feed intake. This would result in increased growth but poor feed efficiency. Similar results were also reported in rainbow trout (Watanabe & Pongmaneerat 1991; Bureau *et al.* 2000) and silver perch *Bidyanus bidyanus* (Stone *et al.* 2000) that feed efficiency decreased at high inclusion levels of MBM while fish showed good growth. Feed efficiency was generally related to protein digestibility. In the present study, apparent protein digestibility (ADCp) for diets with MBM was significantly lower than that of the control. It is well documented that high ash contents in MBM could reduce protein digestibility (Pongmaneerat & Watanabe 1991; Alexis 1997; Robaina *et al.* 1997; Kureshy *et al.* 2000).

In the present study, replacement level of fish meal by MBM in diets for gibel carp at 150 g kg^{-1} protein level resulted in similar growth and feed utilization to the fish meal diet. Davies *et al.* (1989); Watanabe & Pongmaneerat (1991) and Millamena (2002) successfully replaced 750, 800 and 900 g kg^{-1} fish meal protein with MBM combined with other

Table 4 Apparent digestibility coefficient (ADC) of experimental diets in gibel carp[1]

	Control	MBM_{15}	MBM_{50}	PBM_{15}	PBM_{50}
ADC_D[2] (%)	71.8 ± 1.3^a	67.7 ± 2.2^a	60.2 ± 1.3^b	67.7 ± 2.2^a	68.8 ± 1.3^b
ADC_P[3] (%)	90.8 ± 0.4^a	86.6 ± 1.3^b	79.8 ± 1.3^c	86.4 ± 1.2^b	84.9 ± 0.7^b
ADC_E[4] (%)	82.9 ± 1.0^a	82.6 ± 0.8^a	78.3 ± 1.7^b	80.6 ± 1.7^{ab}	80.6 ± 0.5^{ab}

[1] Values are mean ± SE ($n = 3$).
[2] ADC of dry matter = $(1 - \% \, Cr_2O_3 \text{ in diet}/\% \, Cr_2O_3 \text{ in faeces}) \times 100$.
[3] ADC of protein = $[1 - \% \text{ protein in faeces} \times \% \, Cr_2O_3 \text{ in diets}/(\% \text{ protein in diets} \times \% \, Cr_2O_3 \text{ in faeces})] \times 100$.
[4] ADC of energy = $[1 - \% \text{ energy content in faeces} \times \% \, Cr_2O_3 \text{ in diets}/(\% \text{ energy content in diets} \times \% \, Cr_2O_3 \text{ in faeces})] \times 100$.
^{a,b,c} Values in the same row with same superscripts are not significantly different ($P > 0.05$).

protein sources in diets for Mozambique tilapia, grouper *Epinephelus coioide* and rainbow trout, respectively. The different acceptable levels in fish might be due to differences in quality of MBM and animal protein blends tested.

Poultry by-product meal has been extensively studied as an alternative protein source in fish diets. Most results show that PBM could replace more than 500 g kg^{-1} of fish meal protein (Fowler 1991; Steffens 1994; Webster *et al.* 1999).

Growth performance and feed utilization of gibel carp in the present study were not significantly affected by replacement of fish meal with PBM up to 500 g kg^{-1} protein. This is similar to the report by Fowler (1991) that 200 g kg^{-1} PBM inclusion could efficiently substitute 500 g kg^{-1} fish meal in a practical diet for chinook salmon *O. tschawytscha* Walbaum. A similar result was also observed in gilthead seabream fed diets including a locally produced PBM (Nengas *et al.* 1999). In recent years, a number of studies have indicated that many species could tolerate up to 1000 g kg^{-1} replacement of fish meal (Steffens 1994; Alexis 1997; Nengas *et al.* 1999; Kureshy *et al.* 2000; Takagi *et al.* 2000). These different findings reflect the fact that the utilization of PBM differs considerably depending on the quality of the products (Dong *et al.* 1993; Bureau *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. On the other hand, the progressive tendency could be largely attributed to better manufacturing practices improving the quality of this animal by-product (Bureau *et al.* 1999, 2000).

Conclusion

The present study indicated that MBM in this experiment could be used as a main protein to replace fish meal up to 150 g kg^{-1}, while PBM up to 500 g kg^{-1} of dietary protein without negative effect on the growth and feed utilization. MBM, combined with other proteins or reduced ash by improving processing method, may give even more positive effects. Optimal replacement of fish meal with PBM in diet for gibel carp requires further investigation.

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