

## PARTIAL REPLACEMENT OF FISHMEAL WITH TUNA LIVER MEAL IN DIETS FOR COMMON CARP FRY, *CYPRINUS CARPIO* L. 1758

E. GÜMÜŞ, Y. KAYA, B. A. BALCI AND B. B. ACAR

Faculty of Fisheries, Akdeniz University, 07058, Antalya, Turkey

### ABSTRACT

Tuna liver meal (TLM) was tested to replace fish meal (FM) in diets for carp fry, *Cyprinus carpio* Linneus, 1758. Triplicate groups of fish with average weight of  $0.32 \pm 0.65$ g were fed each of six isonitrogenous (42%), isolipidic (16%) and isoenergetic (18 KJ DE g<sup>-1</sup>) diets prepared to include 0, 10, 20, 30, 40 and 50% (diets 1- 6, respectively) of FM protein being substituted by TLM. The control diet contained fish meal (17.14%) and soybean meal (46.9%) as the main sources of dietary protein. After 13 weeks of feeding, fish fed diet 2 and diet 3 exhibited the highest ( $P < 0.05$ ) values for live weight gain, specific growth rate, condition factor, better feed conversion ratio and protein efficiency ratio compared to the others. Survival range was from 70 to 87.5%. However, fish fed diet 6 exhibited lower growth and survival than those fed other diets. Apparent protein and lipid digestibility values for fish fed diet 2 or diet 3 were higher than the others. There were no significant ( $P > 0.05$ ) differences in the crude protein and ash content in the whole-body. However, moisture and lipid contents were significantly ( $P < 0.05$ ) affected by dietary treatments. These results indicate that up to 20% of FM protein in fish diet can be replaced by TLM without adverse effects on fish growth, feed utilization and body composition.

**Key words:** *Cyprinus carpio*, fish meal, tuna liver meal, growth, digestibility.

### INTRODUCTION

Diets containing high levels of proteins are necessary for the economical growth of fish in intensive rearing conditions. Ratios of protein levels in fish feeds generally vary from 25 to 60% and are supplied from a variety of protein sources of both vegetable and animal origin (Gümüş and Ikiz, 2009). Supplementary feeding plays an important role in the growth of fish (Afzal *et al.*, 2008).

Fishmeal (FM) is considered the major animal protein ingredient in aquaculture diets for most cultivable fish species because of its nutritional quality (Hardy and Tacon, 2002). However, FM is one of the most expensive macro-ingredients in an aquaculture diet because it is used in high proportions. Moreover, its availability is limited by various factors such as climatic phenomena, the increasing demand for FM use in animal feed, especially in aquaculture diets, and the overexploitation and decline of fish stocks that are used to produce FM. Restricted FM supplies can no longer meet the needs of the expanding fish feed industry as a result of aquaculture development (Dong *et al.*, 1993). It is crucial to reduce the FM used in aquaculture feed by replacing it with other protein sources. The efficiency of less expensive animal and/or plant protein sources as partial or complete substitute for FM has been evaluated in fish diets, e.g. poultry by-products (Yang *et al.*, 2006), sunflower meal (El-Saidy and Gaber, 2002a), soybean (Webster *et al.*, 2000),

gambusia meal (Abdelghany, 2003), broad bean meal (Gaber, 2006), turkey meal (Muzinic *et al.*, 2006) and krill (Olsen *et al.*, 2006).

Liver as a feedstuff is a by-product of the slaughtered animals. Since liver is used as food for human consumption, mainly discarded livers not fit for human consumption are used as a feedstuff. Fresh or frozen liver is used as the first exogenous food for fish larvae. Before use as feed for larvae, juvenile or older fish, the processed liver meal is minced together with other feed ingredients. Liver meal may be produced by warm-blooded or large aquatic animals such as whales and bluefin Tuna fish. Because of its nutritional quality, Tuna fish liver meal (TLM) may be an important animal protein ingredient for use in aquaculture diets to replace FM. During the past five-year in Turkey, as throughout the world, bluefin tuna fish rearing industry has rapidly expended. Tuna fish production by culture for the Mediterranean basin has almost totalled 29500 tonnes in 2007, of which Turkey has totalled 918 tonnes (ICCAT, 2007). TLM is a by-product of the Tuna fish rearing industry. However, it is still an ingredient to be considered for use in aquaculture larvae diets. Generally, TLM is cheaper than FM because of its reputation as a by-product. Presently, no available data exist on the use of TLM in aquaculture larvae diets. The aim of this investigation was to assess growth, body composition and apparent digestibility of carp fry grown in aquaria, when fed diets with TLM partially replacing fishmeal.

## MATERIALS AND METHODS

### Feed ingredients

Tuna fish liver meal (TLM) was prepared at the laboratory of Fisheries Faculty of Akdeniz University, Turkey. Fresh Tuna fish (*Thynnus tunnus*) livers were obtained after harvesting from the Tuna Cage Farm, Ak-Tuna, Gazipaşa, Antalya, Turkey. They were oven-dried for 24 h at 70°C, packed in plastic bags, sealed and stored at -20°C until used in the diet production. The proximate composition of each of the principal dietary protein sources is given in Table 1.

### Diet preparation and feeding regime

Six diets were prepared to be isonitrogenous, isolipidic and isoenergetic in terms of crude protein (42%, dry wt.), lipid (16%, dry wt.) and digestible energy (18 KJ DE g<sup>-1</sup>), following NRC (1993). The digestible energy (DE) content of the diet was estimated using the DE values 20.9 KJ g<sup>-1</sup> protein, 37.7 KJ g<sup>-1</sup> lipid and 14.6 kJ g<sup>-1</sup> carbohydrate (Chiou and Ogino, 1975); fibre was not included in the calculation. Formulation and ingredient composition of each of the experimental diets is presented in Table 2.

**Table 1: Main sources of protein used in the experimental diets<sup>a</sup> (% dry wt.)**

Ingredient	Crude protein	Ether extract	Crude fibre	Dry matter	Ash	NFE <sup>b</sup>
Peruvian fish meal (FM)	73.79	9.76	0.76	92.47	14.83	0.86
Tuna liver meal (TLM)	30.10	65.23	0.79	90.94	2.88	1.00
Soybean meal	53.58	3.99	4.31	90.70	7.71	30.41

<sup>a</sup>Values are means of three analyses; <sup>b</sup>Nitrogen-free extract.

**Table 2: Formulation and proximate composition of the experimental diets**

Ingredients (%)	% Replacement of Tuna liver meal for protein supplied by fish meal and corresponding diets numbers					
	0	10	20	30	40	50
	1 (control)	2	3	4	5	6
Fish meal	17.14	15.42	13.71	11.99	10.28	8.57
Tuna liver meal	0	4.04	8.09	12.13	16.18	20.23
Corn meal	5.74	5.74	5.74	5.74	5.74	5.74
Corn gluten	7.23	7.23	7.23	7.23	7.23	7.23
Soybean meal	46.9	46.9	46.9	46.9	46.9	46.9
Starch	10.27	10.12	10.06	9.93	9.90	9.83
Fish oil	11.12	8.95	6.7	4.52	2.24	0
Vitamin premix <sup>1</sup>	0.2	0.2	0.2	0.2	0.2	0.2
Mineral premix <sup>2</sup>	0.3	0.3	0.3	0.3	0.3	0.3
L-methionine	0.5	0.48	0.46	0.44	0.42	0.4
Iodized salt	0.1	0.1	0.1	0.1	0.1	0.1
Chromic oxide	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100

### Proximate composition (% dry weight)<sup>3</sup>

Moisture	5.62	4.89	5.16	5.03	5.68	4.97
Crude protein	42.41	42.06	42.55	42.07	42.44	42.00
Ether extract	16.18	16.20	16.03	15.95	16.07	16.10
Ash	7.99	7.68	7.06	6.92	6.71	6.64
Crude fibre	2.40	2.39	2.42	2.43	2.47	2.47
NFE	30.99	31.64	31.91	32.61	32.30	32.77
DE kJ (g diet) <sup>-1</sup>	18.08	18.09	18.07	18.08	18.06	18.05

<sup>1</sup>Vitamin premix (mg or IUkg<sup>-1</sup> of diet): Vit-A 7000 IU; Vit-D3, 1000; Vit-E, 100; Vit-K3, 4.8; Vit-B1, 12; Vit-B2, 20; Vit-B6, 12; Vit-B12, 0.04; Vit-C, 200; Niacin, 120; Folic acid, 3.2; Calcium D- Pantothenate, 30; Biotin, 0.4; Inositol, 200; Endox D Dry, 100.

<sup>2</sup>Mineral premix (mg kg<sup>-1</sup> of diet): Iron, 18; Copper, 3.6; Manganese, 36; Cobalt, 3; Zinc, 45; Iodine, 1.2; Selenium, 0.24; Magnesium, 90.

<sup>3</sup>Values are mean of triplicate analysis.

Diets were made based upon the results of the chemical composition of the ingredients. The control diet contained fish meal (17.14%) and soybean meal (46.9%) as the principal sources of dietary protein. In six diets, the fish meal was gradually replaced by Tuna liver meal substituting 0, 10, 20, 30, 40 and 50% of the fish meal protein. Diets for the digestibility assay included 0.5% chromic oxide as inert marker. Prior to preparing the experimental diets, all ingredients were ground in a hammer mill and passed through a 0.5 mm mesh sieve. All the dry ingredients of each diet were thoroughly mixed in a food mixer until they were homogenous. The fish oil and water were then added to the mixed ingredients and thoroughly mixed. The resulting mixture was made into pellets using a meat grinder and a 2 mm die. The pellets were dried in an oven at 70°C for 24 h. The pellets were then crumbled (0.8-1 mm diameter) in suitable size for the fry, sealed in plastic bags and kept at -20°C until feeding.

### Rearing conditions of fish

The experiment was carried out at the Laboratory of Fisheries Faculty of Akdeniz University, Antalya, Turkey, from 1 February to 31 April, 2007. Carp fry were obtained from the Institute of Mediterranean Aquacultural Research, Antalya, Turkey. Prior to the start of the experiment, all fry were acclimated to laboratory conditions for 2 weeks in a 250-L glass aquarium. During this period, fry were fed twice daily a commercial feed (Abaloğlu Yem-Soya A.Ş., Denizli, Turkey) containing 44% protein. At the beginning of the experiment, 18 glass aquaria (65 L) were each stocked with 20 fry with an average weight of  $0.32 \pm 0.65$  g fish<sup>-1</sup>. The aquaria were filled with dechlorinated tap water throughout the study and in order to avoid accumulation of metabolites, two-thirds of the aquarium water was changed daily. Each aquarium was aerated with air produced by a central compressor. Water temperature was also maintained constant with a 2000-W automatic bayonet titanium heater set at 24-26°C. Water temperature and dissolved oxygen were recorded daily, using a Model WTW Oxi 330i multi-oxygen meter (WTW Wissenschaftlich-Weilheim, Germany). A photoperiod of 12:12 h light/dark cycle using fluorescent lighting was maintained throughout the experiment. The water temperature ranged from 24 to 26°C, dissolved oxygen from 4.5 to 5.2 mg L<sup>-1</sup> and the pH ranged from 7.8 to 8.4. Water quality parameters remained within the acceptable ranges for carp fry growth during the experimental period.

Each dietary treatment consisted of three replicates (aquarium) and was randomly assigned to groups of fish. The feeding rate consisted of 8% of the biomass and the ratio was adjusted each time the fry were weighed. Feeding was carried out twice a day throughout the experiment.

### Growth studies

The trial was carried out during 13 weeks. At the beginning and every 15 days, all fry in each aquarium were collected, anaesthetized with quinaldine (1/20.000), weighed and returned to their corresponding aquarium. At the beginning of the experiment, 40 fry of similar average body weight to the experimental fish were sampled for analysis and kept frozen. At the end of the 13 weeks growth study, 15 fish were randomly removed from each aquarium, killed for carcass composition analysis and frozen at -20°C. For each experimental group, the livers were removed from 10 fish randomly selected and weighed for hepatosomatic index (HSI) calculation.

### Digestibility studies

Fry within each aquarium were fed twice daily with the experimental diets containing 0.5% chromic oxide as a marker. Fish were allowed to feed on the chromic oxide diets for 1 week before any faecal samples were taken. Faeces were collected, using the siphoning method, until the end of the experimental study. After each feeding, in the morning and in the evening, uneaten feed was removed from the aquarium by siphon after 1 h feeding, and then faeces from each aquarium were siphoned through a fine mesh netting (80 µm) 5 h after each feeding, and separately collected in individual jars. Pooled faeces from each treatment group were homogenized in individual jars and then stored at -20°C until analysis. Faecal samples were dried in an oven at 70°C for 24 h, ground and prepared for chemical analysis. Apparent digestibility coefficients (ADC<sub>s</sub>) of diet composition were calculated according to the following formula (Maynard and Loosli, 1969):

ADC of the dry matter (%) =

$$100 - [(\%Cr_2O_3 \text{ in feed} / \%Cr_2O_3 \text{ in faeces}) \times 100];$$

ADC of nutrients (%) =

$$100 - 100 [(\%Cr_2O_3 \text{ in feed} / \%Cr_2O_3 \text{ in faeces}) \times (\% \text{nutrient in faeces} / \% \text{nutrient in feed})].$$

### Chemical analysis

Proximate diet composition and chemical composition of the faeces and fish were analysed for dry matter by drying in an oven at 105°C for 24 h; crude protein was established (as g N x 6.25) by the Kjeldahl method after acid digestion; fat by the soxhlet method after ethylether extraction; ash by combustion at 550°C in a muffle furnace for 24 h; crude cellulose after an alkali and acid digestion and nitrogen-free extract (NFE) by the difference [NFE = 100 - (moisture + protein + lipid + ash + fibre)] according to the methods of AOAC (1995). Chromic oxide (Cr<sub>2</sub>O<sub>3</sub>) in the diets and faeces samples was measured using a spectrophotometer procedure involving perchloric acid digestion (Furukawa and Tsukahara, 1966).

### Statistical analysis

Data analysis was performed by one-way analysis of variance (ANOVA) using SPSS 15.0 (SPSS INC. Chicago, IL, USA). Differences among the means were compared using Duncan's post hoc test at 5% probability level (Steel *et al.*, 1996).

### RESULTS

Changes in growth performance and feed utilization for carp fry fed diets containing TLM are shown in Table 3. There were significant differences in final body weight (FBW), weight gain (WG) and specific growth rate (SGR) among the treatments ( $P < 0.05$ ). Initial body weight did not differ among treatments. The mean FBW, WG and SGR of fish fed the diets containing 10 or 20% TLM were significantly higher than those of fish fed TLM levels of  $\geq 30\%$ . However, the fish fed diet 6 (50% TLM) showed lower FBW, WG and SGR compared with those fed the other diets.

There were significant effects of diets on the feed conversion ratio (FCR) and the protein efficiency ratio ( $P < 0.05$ ). The FCR values of fish fed TLM levels of  $\geq 30\%$  were poorer than those fed the other diets. The better FCR value was 1.53 in the diet 2, and the poorer was 3.52 in the diet 6. FCR values of the fish fed diet 1 (control) and diet 3 did not differ significantly from those fed the diet 2. The protein efficiency ratios (PER) were similar in diets 1-3 and significantly higher than those of fish fed diets 4-6. At the end of the experiment, the hepatosomatic index (HSI) and carcass weight (CW) did not differ among treatments ( $P > 0.05$ ). The highest HSI value was obtained in fish fed diet 2. Survival of fish ranged from 70 to 87.5% and was affected ( $P < 0.05$ ) by different experimental diets (Table 3). Percentage survival of fish fed TLM as replacements was significantly lower than that of the control group ( $P < 0.05$ ).

The apparent digestibility coefficients (ADC) of the experimental diets are given in Table 4. Replacement of fishmeal by the TLM affected digestibility of protein, lipid and dry matter ( $P < 0.05$ ). The highest protein digestibility coefficients were observed in fish fed the diet 2, which were significantly higher than those observed for diets 5 and 6. Diet 5 presented the highest ADC of lipid. There was a significant effect of diet on the ADC of dry matter ( $P < 0.05$ ), with the values for diet 4 or diet 5 being significantly higher than the values for other diets.

Initial and final body composition values for fish fed with experimental diets are given in Table 5. No significant differences were found in the whole body protein (16.04-17.17%) and ash (1.08-1.32%) contents of fish fed different experimental diets ( $P > 0.05$ ). Whole body lipid (5.79-9.02%) and moisture (72.43-76.07%) contents were affected by diet treatments ( $P < 0.05$ ). A

tendency was noted for body lipid level to decrease, whereas for body moisture contents to increase, as TLM contents increased in the diets.

### DISCUSSION

The results of the present study clearly indicate that the growth performance or feed utilization of common carp fry was affected by different experimental diets ( $P < 0.05$ ). Final weight, weight gain and SGR values of fish fed diets 2 or 3 in which up to 20% TLM replaced 10 or 20% of FM protein improved when compared with those of fish fed the diets containing  $> 20\%$  replacement. Highest growth was obtained with diet 2. Growth was reduced when the TLM content of the diet was higher than 20%, suggesting that high TLM levels led to poor gains. Liver meal has been found to yield good growth and survival for larvae of cyprinids (Charlon and Bergot, 1984) and post-larvae freshwater prawn, *Macrobrachium rosenbergii* (Garces and Heinen, 1993; Molina-Vozzo *et al.*, 1995). Several other ingredients of animal origin have been tested with various fish or crustacean species as substitute ingredients for fish meal with variable success (Webster *et al.*, 2000; Gaber, 2006; Muzinic *et al.*, 2006; Yang *et al.*, 2006; Afzal *et al.*, 2008). Growth of fish or crustacean species tends to be reduced when a high proportion or all the fish meal was substituted by various feed ingredients in the diets. Similar results have been obtained in the present study in which common carp fry were fed diets used at high inclusion levels of TLM.

Feed utilization efficiencies such as SGR, FCR and PER were influenced by dietary treatments. In general SGR, FCR and PER were negatively influenced with increasing TLM level in feed. The higher SGR and better FCR were obtained by diet 2. SGR, FCR and PER of common carp fry were improved slightly when they were fed the diet containing up to 20% TLM level, while with the increase in TLM replacement level from 20 to 50%, SGR, FCR and PER decreased significantly ( $P < 0.05$ ). PER and FCR are generally related to digestibility of nutrients. In our study, apparent protein and lipid digestibilities were decreased by inclusion of dietary TLM. The enrichment of the digestible energy content of diets by addition of lipid up to 20% did not result in improvement in growth performance nor protein utilization. Moreover, the dietary essential fatty acid (EFA) requirement of freshwater fish for n-3 polyunsaturated fatty acids (PUFA) may be lower than that of marine fish. The growth suppressing effects of n-3 PUFA were also noted when the common carp was fed with diets containing more than 0.5% of the diet (NRC, 1993; Kaushik, 1995). N-3 PUFA was not measured in the TLM used in this study; however, TLM (65.23%, dry weight, ether extract) in general contain more n-3 than n-6 PUFA. The reason for the decline in feed

**Table 3: The growth performance of common carp fry fed experimental diets for three months**

Parameters	Diets (%TLM replacing FM in diets)					
	1 (control)	2 (10%)	3 (20%)	4 (30%)	5 (40%)	6 (50%)
Initial body wt. (g)	0.326 ± 0.006	0.328 ± 0.001	0.328 ± 0.001	0.328 ± 0.001	0.332 ± 0.001	0.324 ± 0.003
Final body wt. (g)	2.44 ± 0.14 <sup>ab</sup>	3.05 ± 0.22 <sup>a</sup>	2.76 ± 0.14 <sup>a</sup>	1.82 ± 0.42 <sup>bc</sup>	1.80 ± 0.30 <sup>bc</sup>	1.46 ± 0.51 <sup>c</sup>
Weight gain <sup>1</sup> (g)	2.12 ± 0.15 <sup>ab</sup>	2.72 ± 0.22 <sup>a</sup>	2.43 ± 0.14 <sup>a</sup>	1.49 ± 0.42 <sup>bc</sup>	1.47 ± 0.31 <sup>bc</sup>	1.14 ± 0.51 <sup>c</sup>
SGR (%day <sup>-1</sup> ) <sup>2</sup>	2.03 ± 0.07 <sup>ab</sup>	2.24 ± 0.07 <sup>a</sup>	2.14 ± 0.04 <sup>ab</sup>	1.71 ± 0.23 <sup>bc</sup>	1.70 ± 0.17 <sup>bc</sup>	1.49 ± 0.35 <sup>c</sup>
FCR <sup>3</sup>	1.88 ± 0.10 <sup>bc</sup>	1.53 ± 0.14 <sup>c</sup>	1.80 ± 0.15 <sup>bc</sup>	2.92 ± 1.39 <sup>abc</sup>	3.18 ± 0.81 <sup>ab</sup>	3.52 ± 2.18 <sup>a</sup>
PER <sup>4</sup>	1.32 ± 0.07 <sup>a</sup>	1.63 ± 0.15 <sup>a</sup>	1.39 ± 0.11 <sup>a</sup>	0.87 ± 0.12 <sup>b</sup>	0.81 ± 0.20 <sup>b</sup>	0.72 ± 0.16 <sup>b</sup>
HSI (%) <sup>5</sup>	2.25 ± 0.51 <sup>a</sup>	2.74 ± 1.22 <sup>a</sup>	2.62 ± 0.19 <sup>a</sup>	2.41 ± 0.03 <sup>a</sup>	2.65 ± 0.38 <sup>a</sup>	2.55 ± 0.26 <sup>a</sup>
CF <sup>6</sup>	1.50 ± 0.09 <sup>ab</sup>	1.74 ± 0.19 <sup>a</sup>	1.46 ± 0.02 <sup>b</sup>	1.40 ± 0.04 <sup>b</sup>	1.39 ± 0.04 <sup>b</sup>	1.41 ± 0.11 <sup>b</sup>
CW(% <sup>7</sup> )	89.43 ± 0.39 <sup>a</sup>	89.38 ± 0.87 <sup>a</sup>	88.25 ± 0.39 <sup>a</sup>	89.75 ± 0.52 <sup>a</sup>	89.59 ± 0.84 <sup>a</sup>	89.82 ± 0.02 <sup>a</sup>
Survival (%) <sup>8</sup>	87.5 ± 3.53 <sup>a</sup>	82.5 ± 3.53 <sup>ab</sup>	85.0 ± 0.00 <sup>ab</sup>	80.00 ± 0.00 <sup>ab</sup>	77.5 ± 3.53 <sup>bc</sup>	70.0 ± 7.07 <sup>bc</sup>

Each value is the mean (± SD) of three replicates. <sup>a,b,c</sup>Values in the same row with different superscripts are significantly different from each other (P<0.05).

<sup>1</sup>Weight gain = Final body wt. (g) - initial body wt. (g)

<sup>2</sup>Specific growth rate (SGR) = [final body wt. (g) - initial body wt. (g)] / days x100.

<sup>3</sup>Feed conversion ratio (FCR) = Feed consumed (g) / wet weight gain (g).

<sup>4</sup>Protein efficiency ratio (PER) = Weight gain (g) / protein consumed (g).

<sup>5</sup>Hepatosomatic index (HSI) = Wet liver wt. (g) / wet body wt. (g) x100.

<sup>6</sup>Condition factor (CF) = Final body weight (g) / (total body length, cm)<sup>3</sup> x 100.

<sup>7</sup>Carcass weight (CW) = [Final body wt. (g) - visceral weight (g)] / final body wt. (g) x 100

<sup>8</sup>Survival = Final fish number/initial fish number x 100.

**Table 4: Effects of experimental diets on apparent digestibility coefficients (ADC, %) of nutrients**

Parameters	Diets (%TLM replacing FM in diets)					
	1 (control)	2 (10%)	3 (20%)	4 (30%)	5 (40%)	6 (50%)
Protein	89.84 ± 0.03 <sup>a</sup>	90.03 ± 0.87 <sup>a</sup>	88.89 ± 0.64 <sup>ab</sup>	88.70 ± 0.62 <sup>ab</sup>	88.27 ± 0.21 <sup>bc</sup>	87.18 ± 0.07 <sup>c</sup>
Lipid	72.57 ± 2.44 <sup>c</sup>	73.70 ± 0.23 <sup>bc</sup>	74.14 ± 3.62 <sup>b</sup>	78.49 ± 1.05 <sup>ab</sup>	81.66 ± 2.49 <sup>a</sup>	81.01 ± 2.06 <sup>a</sup>
Dry matter	63.78 ± 0.98 <sup>b</sup>	65.76 ± 0.63 <sup>ab</sup>	65.57 ± 1.60 <sup>ab</sup>	66.65 ± 0.54 <sup>a</sup>	66.96 ± 0.19 <sup>a</sup>	66.44 ± 1.50 <sup>ab</sup>

Each value is the mean (± SD) of three replicates. <sup>a,b,c</sup>Values in the same row with different superscripts are significantly different from each other (P<0.05).

**Table 5: Initial and final body composition (% wet basis) of carp fry fed with experimental diets**

Parameters	Initial	Diets (%TLM replacing FM in diets)					
		1 (control)	2 (10%)	3 (20%)	4 (30%)	5 (40%)	6 (50%)
Protein	13.40 ± 0.18	16.04 ± 0.63 <sup>a</sup>	16.25 ± 0.22 <sup>a</sup>	17.17 ± 0.37 <sup>a</sup>	16.94 ± 1.31 <sup>a</sup>	16.91 ± 0.30 <sup>a</sup>	16.95 ± 0.25 <sup>a</sup>
Lipid	2.99 ± 0.10	8.21 ± 1.79 <sup>a</sup>	8.16 ± 0.91 <sup>a</sup>	9.02 ± 1.38 <sup>a</sup>	8.87 ± 0.11 <sup>a</sup>	6.98 ± 0.24 <sup>ab</sup>	5.79 ± 0.52 <sup>b</sup>
Ash	2.74 ± 0.01	1.14 ± 0.12 <sup>a</sup>	1.18 ± 0.01 <sup>a</sup>	1.32 ± 0.18 <sup>a</sup>	1.26 ± 0.11 <sup>a</sup>	1.15 ± 0.13 <sup>a</sup>	1.08 ± 0.18 <sup>a</sup>
Moisture	81.36 ± 0.16	73.61 ± 1.11 <sup>a</sup>	73.99 ± 0.61 <sup>ab</sup>	72.43 ± 1.86 <sup>a</sup>	72.62 ± 0.96 <sup>a</sup>	74.41 ± 0.47 <sup>ab</sup>	76.07 ± 0.04 <sup>b</sup>

Each value is the mean (± SD) of triplicate analysis; <sup>a,b,c</sup>Values in the same row with different superscripts are significantly different from each other (P<0.05).

utilization could be a decrease in digestibility of nutrients by the inclusion of TLM in the diets, possibly as an effect of increased n-3 PUFA. Similar results were also observed in various fish or crustacean species which were fed several other ingredients of animal or plant origin (Kim *et al.*, 1995; Webster *et al.*, 2000).

Some studies have reported that the mean condition factor (CF), carcass weight (CW) or HSI values were significantly affected by the dietary nutritional factors, especially main nutrients, such as carbohydrates, lipids and proteins (Jover *et al.*, 1999; Kim and Lall, 2001). In the present study, CW and HSI of fish were not significantly affected by the replacement level of fish meal protein replaced by TLM.

The apparent protein digestibility was significantly different for TLM replacement of the FM protein. The high protein digestibility was observed in diets 1-2 and these values were significantly higher than diets 5 and 6. The low protein digestibility was recorded with diets 5 and 6. This suggests that the apparent protein digestibility of TLM is slightly lower than that of FM for common carp fry. Apparent digestibility coefficients for dietary proteins were in the range of 87.18-90.03% in the present study. These findings are slightly higher than the findings of Wee and Shu (1989), who reported digestibility protein values of 74.3-86.6 for Nile tilapia, but similar to Takagi *et al.* (2000), who reported digestibility protein values of 88.3-93.6% for juvenile red sea bream. The differences observed among studies

may have been largely caused by variations in the quality or kind of raw materials (Dong *et al.*, 1993) and differences in the methods of faeces collection (Takagi *et al.*, 2000). Apparent dry matter and lipid digestibility values obtained in the present study indicate that inclusion of TLM in the experimental diets reduced feed utilization when dietary TLM was included in excess of 20%.

Partial replacement of FM with TLM affected whole-body composition in terms of protein, fat, ash or moisture compared with the initial fish. Fish fed all experimental diets had remarkably higher percentages of protein and lipid contents, whereas ash and moisture contents were lower than initial fish. These suggest that common carp fry efficiently ingested, digested and assimilated TLM proteins.

The total crude protein and ash contents of whole-body common carp fry were not influenced by dietary treatments. Similarly, Pongmaneerat *et al.* (1993) in carp, Abdelghany (2003) in red tilapia, Gaber (2006) in Nile tilapia and Muzinic *et al.* (2006) in sunshine bass did not find any effects of the dietary mixtures of replacements on the whole-body protein content. Whole-body moisture increased and whole-body lipid contents decreased in fish fed with diets containing the high TLM. The fat was slightly lower in fish fed diet 5 or 6, whereas moisture was higher in fish fed diet 5 or 6. This agrees with the results of Siddhuraju and Becker (2001), who reported a similar reduction in whole-body lipid content of common carp. Findings of the present study are in agreement with the suggestions of Pongmaneerat *et al.* (1993), Abdelghany (2003), Gaber (2006) and Yang *et al.* (2006), who investigated the effect of protein replacements on whole-body composition (protein, lipid, ash and moisture) of various species.

The results of the present study indicate that up to 20% TLM can be included in common carp diet without affecting the growth and nutrient utilization. This observation is supported by the ADC of protein for diets containing mixtures of TLM. In addition, these TLM sources are locally available unlike imported FM, because TLM is a by-product of Tuna fish. The potential for considering this animal protein source for carp fry culture is therefore promising. TLM at 10 or 20% replacement level is suggested for use in diets for common carp fry in order to obtain good growth performance. Further research will be required to determine the feasibility of using TLM or other by-products of Tuna fish composed of different combinations of ingredients and to examine the effects of TLM or by-products of Tuna fish in diets on different species of fish under field conditions.

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