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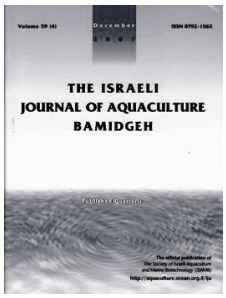

The Dietary L-Methionine Requirement of the Juvenile Yellow Catfish *Pelteobagrus fulvidraco*

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Abstract

An 8 week feeding trial was conducted to investigate the effects of dietary methionine levels on growth, feed utilization, body composition, and morphometric parameters of juvenile yellow catfish (*Pelteobagrus fulvidraco*), and to determine their dietary methionine requirement. Six isonitrogenous and isoenergetic diets (405.0 g/kg crude protein and 18.0 kJ/g gross energy) were formulated, and crystalline L-methionine was added to obtain dietary methionine levels of 5.5, 6.7, 7.5, 8.7, 9.5 and 11.3 g/kg dry diet. Fish with an initial weight of 1.4 g were randomly distributed into six groups and fed the respective formulated diets. Increasing amounts of dietary methionine up to 8.7 g/kg dry diet resulted in increased final body weight, rate of weight gain, specific growth rate, feed conversion efficiency and protein efficiency ratio however, a further increase in methionine level resulted in a decrease in these parameters. No significant difference in survival percentages was detected among the dietary treatments. A one-slope quadratic broken-line analysis model, based on specific growth rate and dietary methionine levels, indicated that the dietary L-methionine requirement of juvenile *P. fulvidraco* is 10.5 g/kg dry diet (accounting for 26.0 g/kg of dietary protein).

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Introduction

Protein is the most important nutrient in fish feed. The gross dietary protein requirement is directly affected by the composition of amino acids in the diet. Determining the essential amino acid requirements of cultured fish is important because of the effects of these nutrients on growth, feed costs and nitrogen pollution (Small and Soares, 1998). Methionine is an essential amino acid required by various fish species and terrestrial vertebrates for normal growth and metabolic function; it is the most limited amino acid, other than lysine, in plant protein sources. With the rising cost of fish meal, cutting back on its use in fish diets has become a priority. Alternative plant protein sources, such as soybean meal, peanut meal and copra meal, are generally used to replace all or part of the fish meal because of their higher levels of protein, steady supply, low cost and lower nitrogen excretion. The supplementation of methionine (and/or other essential amino acids if necessary) in plant protein diets can improve the growth response in many fish species (Mukhopadhyay et al., 2001;Takagi et al., 2001).

The yellow catfish *Pelteobagrus fulvidraco* is a popular food fish cultured in China and a potentially important aquaculture species because of its strong adaptability and fast growth. They are suitable for export and are therefore an economically important species (Pan et al., 2008). However, few studies have been conducted on the nutrient requirements of this species. In a previous study, we found that dietary lysine at 33.1 g/kg dry diet (83.2 g/kg dietary protein) could improve the growth performance of this fish (Cao et al., 2012). However for this species, the quantitative requirement for the other nine essential amino acids was not determined. Hence, the objective of this study was to investigate the effect of dietary methionine levels and dietary methionine requirement (with a constant level of cysteine) on growth, feed utilization, body composition, hematological parameters, and morphometric parameters of juvenile yellow catfish.

Materials and Methods

Diets. A diet containing 22.5% fish meal, 10.8% soybean protein, and 18.0% CAA mixture without crystalline L-methionine was used as the basal diet for comparison with the experimental diets which were supplemented with graded levels of crystalline L-methionine (0–0.75% dry matter with an increment of 0.15%) and a constant level of cysteine. Using an automatic amino acid analyzer, the methionine levels in the six experimental diets were 0.55, 0.67, 0.75, 0.87, 0.95 and 1.13% of the dry diet (indicated as D0.55–D1.13), respectively. A mixture of glycine and aspartic acid was added to all diets to make them isonitrogenous and isoenergetic. The ingredient composition and analyzed chemical composition are shown below in Table 1.

The dietary ingredients, ground to a fine powder through 250-μm mesh, were mixed beginning with the smallest sized ingredients and gradually adding the larger ingredients. The pH was adjusted to 7.0-8.0 by adding 6 N NaOH. Fish oil, soybean oil, and distilled water (0.3 l/kg) were slowly added to the premixed dry ingredients and thorough mixed until the mixture was homogenous. The 2.5-mm pellets were wet-extruded and air-dried until the moisture content was less than 10.0%. The dry pellets were packed into plastic bags and stored at -20.0°C until use.

Fish and feeding conditions. The experiment was conducted in an indoor recirculating aquaculture system at the Guangdong Academy of Agricultural Sciences (Guangzhou, China). Experimental fish were obtained from a commercial farm in Guangzhou, China and acclimated to the laboratory conditions prior to the experiment. They were fed the basal diet without supplemented methionine for 2 weeks. After acclimation, fish of similar sizes (with an initial weight of 1.4 g) were selected and randomly distributed into 24 tanks (0.3 m^3), at 40 fish/tank with flow-through freshwater of 28.0 ± 0.1°C. Each diet was assigned to four tanks in a completely randomized design. The fish were fed to apparent satiety twice a day at 0900 and 1600 h. The amount consumed in each tank was recorded daily and adjusted to the amount consumed the previous day. During the experimental period of 8 weeks, the water temperature was 28.0 ± 0.1°C, the pH level was maintained at 7.4-7.8, the ammonia-nitrogen level was lower than 0.03 mg/l, and the dissolved oxygen content was at least 7.0 mg/l. Excess feed and feces were removed from the tanks daily, and one-third of the tank water was replaced.

Table 1. Formulation and proximate chemical composition of the experimental diets.

Ingredients (% diet)	*Diets*					
	D0.55	*D0.67*	*D0.75*	*D0.87*	*D0.95*	*D1.13*
Fish meal[1]	22.50	22.50	22.50	22.50	22.50	22.50
Soybean protein concentrate[2]	10.80	10.80	10.80	10.80	10.80	10.80
Cornstarch[1]	22.00	22.00	22.00	22.00	22.00	22.00
Crystalline amino acids (CAA)[3]	18.00	18.00	18.00	18.00	18.00	18.00
L-Methionine[3]	0.0	0.15	0.30	0.45	0.60	0.75
L-Aspartic acid:L-glycine (1:1)[3]	0.75	0.60	0.45	0.30	0.15	0.0
Fish oil[1]	2.50	2.50	2.50	2.50	2.50	2.50
Soybean oil[1]	2.50	2.50	2.50	2.50	2.50	2.50
Carboxy methyl cellulose[1]	4.00	4.00	4.00	4.00	4.00	4.00
Carrageenan[1]	2.50	2.50	2.50	2.50	2.50	2.50
Cellulose[1]	11.31	11.31	11.31	11.31	11.31	11.31
$Ca(H_2PO_4)_2$[1]	2.00	2.00	2.00	2.00	2.00	2.00
Vitamin premix[4]	0.10	0.10	0.10	0.10	0.10	0.10
Mineral premix[5]	0.50	0.50	0.50	0.50	0.50	0.50
Vitamin C ester[1]	0.04	0.04	0.04	0.04	0.04	0.04
Choline[1]	0.30	0.30	0.30	0.30	0.30	0.30
Sodium chloride[1]	0.20	0.20	0.20	0.20	0.20	0.20
Proximate comp.(% dry matter diet, n=2)						
Crude protein	37.71	38.37	37.95	38.06	38.08	37.93
Crude lipid	7.08	7.68	7.34	7.56	7.32	7.57
Ash	9.09	9.07	8.84	9.22	9.14	8.86
Moisture	6.12	6.08	6.22	6.40	6.35	6.44
Energy (kJ/g)	18.2	17.6	18.1	18.0	18.0	18.0
Methionine	0.55	0.67	0.75	0.87	0.95	1.13
Cystine	0.36	0.37	0.40	0.39	0.38	0.39

[1] Obtained from Guangzhou Fishtech Fisheries Science & Technology Co., Ltd., Institute of Animal Science, Guangdong Academy of Agricultural Sciences (Guangzhou, China).
[2] Purchased from Guangzhou Green Bank Trade Co., Ltd.
[3] Crystalline amino acids were purchased from Yuan Ju Bio-Technology Co., Ltd. (Shanghai, China).
[4] Provided by Institute of Animal Science, Guangdong Academy of Agricultural Sciences (Guangzhou China). Vitamin premix (g/kg premix): A, 3,200,000 IU; D, 1,600,000 IU; E, 16 g; K, 4.0 g; B_1, 4.0 g; B_2, 8.0 g; B_6, 4.8 g; B_{12}, 0.016 g; niacin nicotinic acid, 28.0 g; calcium pantothenate, 16.0 g; folacin, 1.3 g; biotin, 0.064 g; inositol, 40.0 g; moisture ≤ 10.0%.
[5] Provided by Institute of Animal Science, Guangdong Academy of Agricultural Sciences (Guangzhou China). Mineral premix (g/kg premix): Ca, 230.0; K, 36.0; Mg, 9.0; Fe, 10.0; Zn, 8.0; Mn, 1.9; Cu, 1.5; Co, 0.25; I, 0.032; Se, 0.05; moisture ≤ 10.0%.

Sample collection and chemical analyses. At the end of the 8 week feeding trial, all fish were counted and weighed to calculate weight gain (WG), specific growth rate (SGR), protein efficiency ratio (PER), feed conversion efficiency (FCE) and survival rate (SR). Ten fish from each group were euthanized by cold shock (Nickum et al., 2004) and stored at -20.0°C for whole-body composition analysis. Five fish from each tank were individually weighed, and their body length, and weights of the viscera, hepatopancreas and mesenteric fat were determined for calculation of the condition factor (CF), viscerosomatic index (VSI), hepatosomatic index (HSI), and intraperitoneal ratio (IPR). Samples of white muscle from the backs of five fish from each tank were sealed in plastic bags and then frozen at -20.0°C for analysis of muscle nutrient composition.

The crude protein, lipid, moisture, and ash contents in the diets, hepatopancreas, muscle, and whole body were measured in triplicate. Moisture content in the diets was determined by oven drying at 105°C for 24 h, and moisture content in the muscle, whole body and hepatopancreas were determined after these tissues were lyophilized, pulverized, and stored at -80 °C. Crude protein (N × 6.25) content was determined by the Kjeldahl method using a semi-automatic Kjeldahl System (1030 Autoanalyzer, Tecator, Hoganos, Sweden) after acid digestion. Crude lipid content was determined by ether-extraction method using a Soxhlet System HT (Soxtec System HT6, Tecator, Sweden), and ash content was determined by burning the samples at 550°C in a muffle furnace for 24 h (AOAC, 1995).

To determine the dietary contents of methionine and cysteine, the samples were oxidized with performic acid [30% hydrogen peroxide:88% formic acid; 1:9 (v/v)] for 18 h at 0°C, resulting in the stable formation of cysteic acid and methionine sulfone. The samples were then hydrolyzed with 7.2 N HCl for 24 h at 110 °C and analyzed with an automatic amino acid analyzer (Hitachi 835-50, Japan). The gross energy content was determined on an IKA ballistic bomb calorimeter (C2000, Germany).

Statistical analysis. Data are presented as means ± SD. All data were analyzed by one-way analysis of variance (ANOVA) and tested with Duncan's multiple-range test using SPSS software (version 13.0) to determine the effect of the experimental diets. The significance level was $P<0.05$. A one-slope quadratic broken-line analysis model in SAS software (version 8.1) was used to estimate the dietary L-methionine requirement (Portz et al., 2000; Parr et al., 2003).

Results

Growth performance. No pathological signs or anomalies were observed during or at the end of the 8 week trial. Table 2 shows the WG, SGR, FE, PER, MFI (mean feed intake), MI (methionine intake) and SR of juvenile yellow catfish fed different graded levels of dietary methionine.

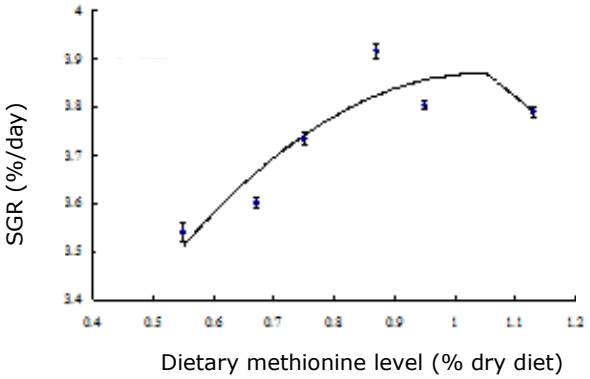
Table 2. Effect of dietary methionine levels on growth and feed utilization in juvenile yellow catfish (mean±SD, n = 4).

	Groups						
Indices	*D0.55*	*D0.67*	*D0.75*	*D0.87*	*D0.95*	*D1.13*	*Pooled SEM*
IBW (g)	1.38±0.01	1.39±0.01	1.38± 0.01	1.38±0.01	1.39±0.01	1.39±0.00	0.011
FBW (g)	10.06±0.12^{a}	10.41±0.06^{ab}	11.19±0.38^{bc}	12.42±0.47^{d}	11.68±0.34^{cd}	11.59±0.29^{cd}	0.43
WG (%)	626.43±7.92^{a}	651.6±2.31^{ab}	710.6±26.54^{bc}	798.5±37.28^{d}	742.8±22.78^{cd}	736.3±20.4^{cd}	27.83
SGR(%/dd)	3.54±0.02^{a}	3.60±0.01^{ab}	3.73±0.06^{bc}	3.92±0.08^{d}	3.80±0.05^{cd}	3.79±0.04^{cd}	0.061
PER	1.58±0.05^{ab}	1.61±0.11^{ab}	1.74±0.09^{a}	1.79±0.09^{a}	1.62±0.13^{ab}	1.42±0.11^{b}	0.12
FCE (%)	59.4±4.01^{ab}	61.3±2.1^{ab}	66.07±1.92^{a}	68.3±0.6^{a}	61.71±7.0^{ab}	53.9±4.83^{b}	4.92
MFI(g/fish/d)	0.25±0.0	0.26±0.0	0.27±0.0	0.27±0.0	0.28±0.0	0.28±0.0	0.0
MI (g/fish/d)	0.0014±0.0	0.0017±0.0	0.0020±0.0	0.0023±0.0	0.0028±0.0	0.0032±0.0	0.0
SR (%)	92.5±2.7	87.5±5.1	96.25±2.17	85.0±4.89	86.88±3.87	89.38± 3.87	4.79

Different superscript letters in a column indicate significant differences ($P < 0.05$).
[1] initial body weight (IBW); final body weight (FBW); weight gain (WG); specific growth rate (SGR); protein efficiency ratio (PER); feed conversion efficiency (FCE); mean feed intake (MFI); methionine intake (MI); survival rate (SR).
The variables were calculated as follows:
WG = 100 × (final body weight-initial body weight)/initial body weight.
SGR = 100 × [ln(final average body weight)-ln(initial average body weight)]/days of the experiment.
FCE = 100 × weight gain (g)/total feed intake (dry matter, g).
PER = body weight gain (g)/protein intake (g).
SR = 100 × (final number of fish)/(initial number of fish).
MI = mean feed intake × dietary methionine content (%).

$$\text{Mean feed intake (MFI)} = \frac{1}{56} \times \sum_{1}^{56} \frac{\text{Consumption in the tank on day } i}{\text{Number of fish in the tank on day } i}$$

The lowest WG and SGR were observed in yellow catfish fed the basal diet (D0.55: 0.55% dry diet methionine from intact protein). The highest WG, SGR, PER and FCE values were observed in yellow catfish fed the D0.87 diet containing 0.87% dry diet methionine. The SR was high for all dietary treatments and was not significantly affected by methionine levels. The growth curve showed WG increased with increasing dietary methionine levels up to the peak, after which WG decreased as dietary methionine content further increased (Fig. 1).



X<1.05159, Y=3.8708-1.4246 x (1.05159-X)^2
X>1.05159, Y=3.8708-1.0193 x(X-1.5159)
R^2*= 0.9356*

Fig.1. Relationship between the dietary methionine level and specific growth ratio (SGR) of juvenile yellow catfish based on one-slope, quadratic broken-line analysis model.

According to the analysis of the one-slope quadratic broken-line model, the optimal requirement of dietary methionine was estimated to be 1.05% of the diet (accounting for 2.60% of dietary protein). Taking the dietary cysteine content (0.39%) into consideration, the requirement for sulfur-containing amino acids (methionine + cysteine) in this fish species was estimated to be 1.44% of the diet, corresponding to 3.56% of the dietary protein.

Whole-body, white-muscle and hepatopancreatic composition and morphometry index. The whole body, white-muscle and hepatopancreatic composition of the juvenile yellow catfish fed graded levels of dietary methionine are shown in Table 3.

Table 3. Effect of dietary methionine levels on chemical composition of whole body, white muscle and hepatopancreas in juvenile yellow catfish (%, mean ± SD, n = 4).

Groups	*Moisture*	*Crude protein*	*Crude lipid*	*Ash*
Whole body				
D0.55	73.83 ± 0.63	13.77 ± 0.21^{a}	7.20 ± 0.34	3.73 ± 0.15
D0.67	73.65 ± 0.46	13.99 ± 0.26^{ab}	7.27 ± 0.22	3.65 ± 0.09
D0.75	73.53 ± 0.12	13.81 ± 0.08^{ab}	7.15 ± 0.11	3.64 ± 0.05
D0.87	73.39 ± 0.38	14.44 ± 0.26^{b}	6.88 ± 0.31	3.54 ± 0.07
D0.95	73.81 ± 0.06	14.01 ± 0.16^{ab}	7.26 ± 0.04	3.57 ± 0.1
D1.13	73.75 ± 0.48	13.84 ± 0.13^{ab}	7.55 ± 0.42	4.03 ± 0.33
Pooled SEM	0.50	0.26	0.34	0.21
White muscle				
D0.55	80.06 ± 0.25^{bc}	16.51 ± 0.2^{a}	1.55 ± 0.07	1.10 ± 0.02^{a}
D0.67	79.65 ± 0.05^{b}	16.80 ± 0.2^{a}	1.52 ± 0.05	1.15 ± 0.02^{ab}
D0.75	79.61 ± 0.27^{c}	16.63 ± 0.3^{a}	1.62 ± 0.04	1.08 ± 0.03^{a}
D0.87	78.23 ± 0.27^{a}	18.18 ± 0.3^{b}	1.73 ± 0.1	1.16 ± 0.03^{ab}
D0.95	78.40 ± 0.21^{a}	18.02 ± 0.2^{b}	1.73 ± 0.11	1.20 ± 0.02^{b}
D1.13	78.89 ± 0.09^{b}	16.73 ± 0.1^{a}	1.55 ± 0.14	1.12 ± 0.03^{ab}
Pooled SEM	0.26	0.28	0.11	0.031
Hepatopancreas				
D0.55	76.99 ± 1.31	11.07 ± 0.59	4.74 ± 0.36	
D0.67	75.80 ± 1.13	11.30 ± 0.52	4.89 ± 0.54	
D0.75	76.92 ± 0.51	11.31 ± 0.48	4.52 ± 0.06	
D0.87	72.94 ± 3.88	12.73 ± 1.99	5.66 ± 0.78	
D0.95	74.22 ± 2.94	12.33 ± 1.7 2	5.11 ± 0.53	
D1.13	76.53 ± 0.58	11.30 ± 0.31	46.2 ± 0.43	
Pooled SEM	2.61	1.402	0.61	

Different superscript letters added in a column indicate significant differences ($P < 0.05$).

In the whole body of fish fed the D0.87 diet, highest protein content ($P<0.05$), and lowest lipid and ash contents were observed compared to the other dietary treatments. For the other dietary treatments, the protein content increased with increasing dietary methionine level but decreased when the dietary methionine level surpassed the optimal value. The moisture, lipid and ash contents showed the opposite trend.

In the white muscle of fish fed the D0.87 diet, protein, lipid and ash contents were higher than those in fish fed the other diets, whereas moisture content showed the opposite trend; the protein and ash contents increased and moisture content decreased with increasing levels of dietary methionine ($P<0.05$). The protein and ash contents of the white muscle decreased and the moisture content increased once the dietary

methionine level reached the optimal value. The lipid content of the white muscle was not affected by dietary methionine levels.

No significant effect of dietary methionine level was observed on the moisture, crude protein or crude lipid contents of the hepatopancreas.

VSI, HSI, CF and IPR. There was a significant effect of dietary methionine levels on HSI, CF and IPR (P<0.05), but there was no significant difference in the VSI among dietary treatments. The HSI decreased with an increase in methionine level to 0.87% (P<0.05), and then increased when the methionine level was further increased. The same trend was observed for IPR. The CF increased significantly to a dietary methionine level of 0.87%, and decreased significantly thereafter (Table 4).

Table 4. Effect of dietary methionine levels on morphological parameters in juvenile yellow catfish (mean ± SD, n = 4).

Groups[1]	*VSI*	*HSI*	*CF*	*IPR*
D0.55	8.58±0.25	2.11±0.02^{c}	0.91±0.02^{a}	1.91±0.15^{b}
D0.67	8.56±0.31	1.96±0.05^{bc}	0.95±0.04^{ab}	1.67±0.05^{ab}
D0.75	8.64±0.47	1.90±0.01^{bc}	0.94±0.01^{a}	2.02±0.07^{b}
D0.87	8.03±0.15	1.59±0.09^{a}	1.11±0.01^{c}	1.10±0.09^{a}
D0.95	8.11±0.21	1.85±0.02^{b}	1.04±0.04^{bc}	1.99±0.23^{b}
D1.13	9.13±0.51	2.36±0.1^{d}	0.96±0.02^{ab}	2.23±0.3^{b}
Pooled SEM	0.42	0.073	0.032	0.21

Different superscript letters in a column indicate significant differences (P < 0.05).
[1] VSI, viscerosomatic index; HSI, hepatosomatic index; CF, condition factor; IPR, intraperitoneal ratio. The variables were calculated as follows:
VSI=100×viscera weight (g)/ body weight(g).
HSI=100×[hepatopancreas weight (g)]/[body weight(g)]
CF=100×body weight (g)/[body length (cm)]^{3}.
IPR=100×[intraperitoneal fat weight (g)]/[body weight (g)].

Discussion

In the present study, the juvenile yellow catfish fed diets with lower methionine levels grew more slowly and exhibited lower FCE than those fed diets containing higher methionine levels. Significantly higher values of WG, SGR and FCE were observed in fish fed up to an optimal dietary methionine level, but these values declined when the dietary methionine exceeded that level. These results indicate that methionine is an essential amino acid for the growth of juvenile yellow catfish, and that they can make good use of methionine in its crystalline form. These results are similar to those found in the channel catfish *Ictalurus punctatus* (Murai et al., 1982), the yellowtail *Seriola quinqueradiata* (Watanabe et al., 2001), and the yellow croaker *Pseudosciaena crocea* R. (Mai et al., 2006). A balanced amino acid content in the diet is necessary for optimal fish growth (Wilson and Halver, 1986). The growth response of yellow catfish fed increasing levels of dietary methionine was consistent with previous studies reporting that fish growth increases with an increase in methionine up to a certain level, then decreases when that level is exceeded. This trend may be attributed to toxicity of ketones and other toxic metabolites, imbalance in amino acids, poor palatability resulting from excess total sulfur-containing amino acids (TSAA) (Griffin et al., 1994), and differential utilization and absorption rates of CAA and intact dietary proteins with adverse effects on growth.

Dose-response experiments with increasing contents of amino acids constitute a well-accepted method for determining amino acid requirements. The equation, according to the one-slope quadratic broken-line analysis model of the relationship between SGR and dietary methionine level, was: Y = 3.8708-1.4246 × z1 × z1-1.0193 × z2 (if X<1.05159, Y = 3.8708-1.4246 × z1 × z1, Y = 3.8708-0193 × z2); where z1 = 1.05159-X; z2 = X-1.05159; and R^2 = 0.9356. The methionine requirement was calculated to be 1.05% of the diet (accounting for 2.60% of dietary protein). This result was similar to results reported for other fish species, such as the red drum *Sciaenops ocellatus* (1.06%; Moon and Gatlin, 1991) and yellowtail (1.11%; Ruchimat et al., 1997). Fish have a TSAA requirement, rather than a specific methionine or cysteine requirement (Ravi and Devaraj, 1991). In the present study, if the cysteine level (0.39%) was considered, the calculated TSAA requirement of juvenile yellow catfish was 1.44% of the diet, corresponding to 3.56% of the dietary protein. The TSAA requirement for juvenile yellow catfish was similar to that of *Catla catla* (3.6% of dietary protein; Ravi and Devaraj, 1991), lower than that of the juvenile milkfish *Chanos chanos* Forsskal (4.37%; Borlongan and Coloso, 1993) and yellow croaker (4.02%; Mai et al., 2006), and higher than that of juvenile groupers *Epinephelus coioides*, (2.73%; Luo et al., 2005) and channel catfish (2.34%; Harding et al., 1977). These differences may occur because juvenile yellow catfish are carnivorous, with a high dietary protein requirement that

results in a relatively higher methionine requirement than fish with lower dietary protein requirements (Zhou et al., 2006). We considered that the analytical model has an important role in determining the dietary methionine requirement for this fish.

In the present study and in our previous study evaluating dietary lysine requirements of juvenile yellow catfish (Cao et al., 2012), we selected the optimal analytical model by comparing the correlation coefficients of different analytical models — a second-order polynomial regression analysis model, one-slope straight broken-line analysis model and one-slope quadratic broken-line analysis model. The latter model was found to be best. More research is needed to determine the required dietary amino acids for other fish using the above analytical models, to confirm the optimality of the model used herein. Some studies have reported that diets containing high amounts of CAA frequently result in much lower growth rates in fish compared to diets in which all amino acids are derived from intact protein (Robinson et al., 1981; Walton and Wilson, 1986). Using a mixture of fish meal, soybean meal, yeast, and wheat meal, 84.0% dietary protein was supplied in experimental diets; 16.0% of the dietary protein was provided by CAA (Mai et al. 2006). The higher intact protein levels observed by Mai et al. (2006) may have resulted in a relatively higher growth rate and feed-conversion efficiency(FCE) when compared with other studies (Moon and Gatlin, 1991; Alam et al., 2001). In the present experiment, the intact protein was derived from fish meal and soybean meal and constituted 63.7% of the dietary protein; the supplemented CAA accounted for approximately 37.3% of the dietary protein. These diet compositions may to some extent have affected the dietary methionine requirement of juvenile yellow catfish. The conversion of methionine to cysteine in the diets may influence the calculation of the dietary methionine requirement for fish (Moon and Gatlin, 1991). This issue was not examined in the current study but warrants future investigation.

In this study, the dietary methionine level significantly affected the whole-body protein content but had no significant effect on the moisture, ash or crude lipid contents. These results are similar to those found in other species (Kim et al., 1992; Ruchimat et al., 1997). Moreover, dietary methionine levels were found to have a significant effect on HSI and CF, but had no effect on the VSI. In juvenile groupers, the HSI was reported to increase with increasing dietary methionine levels up to the optimal required level, and to remain unchanged beyond that point (Luo et al., 2005). In rainbow trout fed a low-methionine diet, higher HSI values were observed, suggesting that dietary methionine and cystine levels affect HSI (Walton et al., 1982). However, in juvenile Asian seabass, there appears to be no correlation between dietary methionine content and HSI (Coloso et al., 1999). In the present study, the IPRs varied among the different dietary treatments and were not related to dietary methionine levels.

According to the one-slope quadratic broken-line analysis model of SGR and dietary methionine levels, we conclude that the dietary L-methionine requirement of juvenile yellow catfish is 1.05% of the diet (accounting for 2.60% of the dietary protein). Accounting for cysteine content in the dry diet (0.39%), the sulfur-containing amino acid (methionine + cysteine) requirement for juvenile yellow catfish is estimated to be 1.44% of the diet, corresponding to 3.56% of the dietary protein.

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