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Effect of dietary conjugated linoleic acid (CLA) on growth performance, body composition and hepatic intermediary metabolism in juvenile yellow catfish *Pelteobagrus fulvidraco*

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ABSTRACT

The present study was conducted to determine the effect of dietary conjugated linoleic acid (CLA) on growth performance, body composition and hepatic intermediary metabolism in juvenile yellow catfish *Pelteobagrus fulvidraco* (initial body weight: 1.87 ± 0.04 g, mean ± SD). The five isonitrogenous (35% crude protein) experimental diets were formulated to contain graded CLA levels of 0 (control), 0.5, 1, 1.5 and 2%, respectively. Three replicated groups of fish were fed to satiation, twice a day, over a period of 8 weeks with CLA oil, containing mainly the bioactive cis-9, trans-11 and trans-10, and cis-12 isomers. The increase of dietary CLA levels reduced growth performance, feed conversion rate (FCR), condition factor, hepatosomatic index and viscerosomatic index of yellow catfish. Increasing dietary CLA levels significantly reduced lipid contents in the whole body and liver. The dietary inclusion of CLA modified total percentages of the main groups of fatty acids. Increasing saturated fatty acid (SFA) content and reduced mono-unsaturated fatty acid (MUFA) contents in the whole body were observed with increasing dietary CLA inclusion. A gradual reduction of 16:1 and 18:1 fatty acids was depicted in the whole body with increasing CLA levels. Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) profiles showed no significant differences among the treatments. Total polyunsaturated fatty acids (PUFA) tended to increase with increasing dietary CLA levels. Dietary CLA supplementation resulted in a significant increase of the trans-10, cis-12 and cis-9, and trans-11 CLA isomers in the whole body, and also significantly influenced several hepatic enzymatic activities, such as succinate dehydrogenase (SDH), lactate dehydrogenase (LDH), malic dehydrogenase (MDH), lipoprotein lipase (LPL) and hepatic lipase (HL) activities. These indicate that despite promoting the CLA and PUFA deposition in the whole body, dietary CLA supplementation should be carefully evaluated in intensive yellow catfish farming as it reduces growth performance and feed utilization.

1. Introduction

Conjugated linoleic acid (CLA) is the generic designation for a group of positional or geometric derivatives of linoleic acid (18:2n−6) containing two conjugated cis/trans double bonds that are found in a wide variety of food products, including meat, poultry, seafood, cheese, butter, milk, and vegetable oils, with the two main naturally occurring biologically active cis-9, trans-11 and trans-10, and cis-12 isomers (Pariza et al., 2001). CLA has been shown to have many biological effects for terrestrial animals, including remarkable inhibition of atherosclerosis (Lee et al., 1994), anti-cancer (Ha et al., 1990). In addition, there is increasing evidence that dietary CLA decreases body fat and increases lean body mass (Thiel-Cooper et al., 2001; Tischendorf et al., 2002), and thus attenuates obesity in several animal models (Delany and West, 2000; Wang and Jones, 2004). This has been suggested to be a positive effect in a variety of farmed species and animal disease models and by extension, humans (Thiel-Cooper et al., 2001; Wang and Jones, 2004). Dietary CLA could be beneficial to fish culture if these effects on body composition and lipid metabolism could be reproduced in farmed fish without negative effects on growth performance. In fish, studies have shown that dietary CLA may alter growth responses, feed efficiency and lipid concentration (Twibell et al., 2000, 2001; Yasmin and Takeuchi, 2002; Twibell and Wilson, 2003), and result in high CLA deposition levels (Twibell et al., 2000, 2001; Berge et al., 2004; Kennedy et al., 2005; Bandarra et al., 2006; Valente et al., 2007a). Fish being an important source of protein and n−3 PUFA, and a further increase in its CLA content could be of great interest to enhance

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the nutritional value of cultured fish for human consumption. Accordingly, CLA is of particular interest for fish nutrition research.

The fatty acid composition of fish body is generally determined both by the type of dietary lipid ingested and the ability of the individual fish species to modify that dietary input via both pathways of catabolism and conversion including desaturation and elongation (Henderson and Tocher, 1987; Sargent et al., 2002). In fish, liver is attributed preferentially to a sensitive organ reflecting dietary lipid change and plays a central role in lipid metabolism including fatty acid synthesis and degradation through enzyme regulation (Likimani and Wilson, 1982; Kiessling and Kiessling, 1993). Despite the fact that the major biochemical and metabolic pathways have been identified in fish (Cowey and Walton, 1989), effects of dietary CLA on hepatic intermediary metabolism are still poorly understood.

Yellow catfish *Pelteobagrus fulvidraco* is an omnivorous, freshwater species of fish with increasing interest in Chinese inland aquaculture. Recently, we have conducted several studies on nutrient requirements of this fish, such as optimal dietary protein to carbohydrate ratio (Ye et al., 2009), optimal dietary linolenic acid to linoleic acid ratio (Tan et al., 2009), dietary copper (Tan et al., 2010) and phosphorus requirements (Luo et al., 2010). Still, the effects of dietary CLA on the performance of yellow catfish have not yet been clarified. Thus, the main objective of this study was to evaluate the effects of various dietary levels of CLA (0, 0.5, 1.0, 1.5 and 2%) on yellow catfish responses, such as growth performance, body composition, whole body fatty acid profiles, and hepatic intermediary metabolism. Here, we investigated the effect of dietary CLA on hepatic intermediary metabolism through examining liver activities of succinate dehydrogenase (SDH, EC 1.3.99.1), lactate dehydrogenase (LDH, EC 1.1.1.27), malic dehydrogenase (MDH, EC 1.1.1.40), lipoprotein lipase (LPL, EC 3.1.1.34) and hepatic lipase (HL, EC 3.1.1.3) (as representative enzymes involved in glycolysis and lipid catabolism). To our knowledge, this is the first to demonstrate, at the biochemical levels, the effect of dietary CLA on these hepatic enzymatic activities, which is important for further exploration in nutrient metabolism for yellow catfish.

2. Materials and methods

2.1. Ingredients and experimental diets

The five isonitrogenous (35% CP) experimental diets were formulated to contain increasing CLA levels of 0 (control), 0.5, 1, 1.5 and 2%, respectively. The ingredients and proximate analysis of the five diets are shown in Table 1. CLA oil provided by Dalian Youyou Commercial Developmental Co. Ltd. (Dalian, China), was added to the diets at the cost of fish oil to maintain diets isolipidic (7.5% lipid), and 0.01% butylated hydroxytoluene (BHT) was used as an antioxidant. The composition of CLA, together with fatty acid profiles of experimental diets, is shown in Table 2. Analysis of the CLA revealed that the total CLA isomers were 89.0% of total lipids [44.7% 18:2 (cis-9, trans-11), 44.3% 18:2 (trans-10, cis-12)].

The experimental diets were produced according to the methods described in our recent study (Tan et al., 2009). Briefly, all dry ingredients were ground to pass a 120-μm sieve, weighed, and mixed to homogeneity. CLA oil was added to fish oil and mixed until homogenous. The oil mix was then added to homogenous dry ingredients and mixed thoroughly. Then the pre-weighed distilled water (10%, v/w) was added to form a dough. The diets were extruded at 130 °C and at 30 atm using a laboratory model Muyang Extruder Machine (Jiangsu, China) for about 40 s to make the diet water stable, and then ground through a 1.0-mm sieve. The resulting strands were dried with forced air circulation at room temperature, crumbled to approximately 2 mm length and kept in the freezer at −20 °C until used.

Table 1
Ingredients and proximate analysis of experimental diets.

Ingredients (%)	1	2	3	4	5
White fish meal	45	45	45	45	45
Soybean meal	10	10	10	10	10
Fish oil	4	3.5	3	2.5	2
CLA oil	0	0.5	1	1.5	2
Wheat flour	18	18	18	18	18
$NaH_2PO_4 \cdot 2H_2O$	1	1	1	1	1
Vitamin premix	0.5	0.5	0.5	0.5	0.5
Mineral premix	0.5	0.5	0.5	0.5	0.5
Ascorbyl-2-polyphosphate	1	1	1	1	1
Binder	2	2	2	2	2
Betaine	1	1	1	1	1
BHT	0.01	0.01	0.01	0.01	0.01
Cellulose	16.99	16.99	16.99	16.99	16.99
Proximate analysis (percentage on a dry matter basis)					
Moisture	10.18	10.49	10.32	10.50	10.18
Crude protein	35.48	35.15	35.96	35.70	35.08
Lipid	7.76	7.46	7.41	7.32	7.81
Ash	8.72	9.40	9.11	9.21	8.90

White fish meal: Peru.
Soybean meal: Shenyang Hefeng Feed Co., Ltd. Shenyang China.
Fish oil: Wuhan Coland Feed Co., Ltd., Wuhan, China.
CLA oil: Dalian Youyou Commercial Developmental Co. Ltd., Dalian, China.
Wheat flour: locally available in the market, Panjin, China.
$NaH_2PO_4 \cdot 2H_2O$: Sinopharm Chemical Reagent Co. Ltd., Shanghai, China.
Ascorbyl-2-polyphosphate: Wuhan Yuancheng Technology Development Co., Ltd., Wuhan, China.
Betaine: Sigma Aldrich, USA.
Binder: Qingdao Crystal Rock Biology Development Co., LTD, Qingdao, China.
BHT: Nanjing Ningkang Chemical Co., Ltd., Nanjing, China.
Cellulose: Anhui Shanhe Pharmaceutical Co., Ltd., Hefei, China.
Vitamin premix (mg or IU per kg diet): retinylacetate 10,000 IU; cholecalciferol 1000 IU; all-rac-a-tocopheryl acetate 30 IU; menadione nicotinamide bisulfite 7; thiamine hydrochloride 6; riboflavin 3; pyridoxine hydrochloride 12; D-calcium pantothenate 30; niacin 50; biotin 1; folic acid 6; cyanocobalamine 0.03.
Mineral mixture (mg per kg diet): $Ca(H_2PO_3)_2 \cdot H_2O$, 1000; $FeSO_4 \cdot 7H_2O$ 40; $ZnSO_4 \cdot H_2O$ 100; $MnSO_4 \cdot H_2O$ 40; $CuSO_4 \cdot 5H_2O$ 2; $CaIO_3 \cdot 6H_2O$ 3; Na_2SeO_3 0.05; $CoSO_4$ 0.05.

2.2. Experimental procedures

The experiment was conducted in an indoor static aquarium system of Panjin Guanghe Fishery Co., Ltd, Panjin, China, the same as in our recent study (Tan et al., 2009). Eight hundred juvenile yellow catfish *P. fulvidraco* were collected from a local fish pond (Panjin, China) and kept in ten, 300-L circular fiberglass tanks for 14-day acclimatization. During the acclimatization period, the fish were fed

Table 2
Main fatty acid profiles (% of total fatty acids) and CLA contents (% of total lipids) of CLA oil and experimental diets with different CLA incorporation levels (0 (control), 0.5, 1.0, 1.5 and 2%).

	CLA oil	Diet 1	Diet 2	Diet 3	Diet 4	Diet5
16:0	0.4	20.08	19.62	18.25	17.77	16.54
18:0	0.07	4.02	3.96	3.96	3.93	3.88
SFA		31.38	30.22	28.53	27.71	25.94
16:1		7.52	7.04	6.85	6.23	5.52
18:1	3.7	11.32	11.57	11.62	12.04	12.38
MUFA		24.52	24.01	23.50	22.97	22.16
18:2	1.8	5.36	5.28	5.20	5.22	5.14
Cis-9, trans-11 CLA	44.7	0.00	1.96	4.08	6.42	8.92
Trans-10, cis-12 CLA	44.3	0.00	1.92	4.05	6.22	8.46
20:4		0.16	0.14	0.12	0.11	0.08
n−6		5.52	9.30	13.45	17.97	22.60
18:3		1.72	1.64	1.48	1.40	1.32
20:5		14.49	13.86	13.22	12.04	11.45
22:6		16.84	15.62	14.91	13.04	12.02
n−3		33.05	31.12	29.61	26.48	24.79
Total CLA (% total lipids)	93.9	0.42	5.56	10.43	17.04	21.75

SFA: saturated fatty acid; MUFA: monounsaturated fatty acids; n−6 fatty acids: included total CLA; total CLA: sum of isomers trans-10, cis-12 and cis-9 and trans-11.