J. Ocean Univ. China (Oceanic and Coastal Sea Research)
DOI 10.1007/s11802-016-2934-8
ISSN 1672-5182, 2016 15 (4): 660-666
http://www.ouc.edu.cn/xbywb/
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Effects of Dietary Carbohydrate-to-Lipid Ratio on the Growth Performance and Feed Utilization of Juvenile Turbot (*Scophthalmus maximus*)

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(Received April 20, 2015; revised June 25, 2015; accepted April 21, 2016)

Abstract A 9-week feeding trial was conducted to investigate the effects of dietary carbohydrate to lipid ratio (CHO:LIP) on the growth performance and feed utilization of juvenile turbot *Scophthalmus maximus* (initial body weight 8.75 g ± 0.04 g). Four isonitrogenous and isoenergetic low protein level (39%) diets were formulated with increasing ratios of dietary carbohydrate to lipid (2:18, 6:18, 18:12 and 28:6). A high protein level (50%) diet with the 2:12 ratio of carbohydrate to lipid was used as the control. Results showed that the survival rate, contents of moisture, crude protein and ash in muscle were not significantly affected by dietary treatments. With the dietary CHO:LIP ratio increased from 2:18 to 18:12, weight gain rate significantly increased ($P<0.05$). Higher dietary CHO:LIP ratio (28:6) resulted in the significantly decreased weight gain rate ($P<0.05$). Meanwhile, this treatment also resulted in the highest daily feed intake and liver glycogen content, as well as the lowest feed efficiency ($P<0.05$). Muscle glycogen content in fish fed the diet with 2:12 or 2:18 CHO:LIP ratio was significantly lower than those fed with the other three diets ($P<0.05$). The present results confirmed that the juvenile turbot can utilize carbohydrate. Furthermore, the appropriate ratio of dietary carbohydrate to lipid was important to the growth and feed utilization of turbot. The proper CHO:LIP ratio based on the growth performance in the present study was determined to be 18:12 when the dietary protein level was 39%.

Key words turbot; *Scophthalmus maximus*; carbohydrate; lipid; nutrition

1 Introduction

Turbot (*Scophthalmus maximus*), with high economic value, delicious meat, and rapid growth, is widely farmed in Europe and Asia. Its estimated production in 2012 was 77000 tons (FAO, 2014). It is also one of the main farmed species in northern China. The dietary protein requirement for its optimal growth was 50% (Lee *et al*., 2003a). Higher dietary protein level is typically more expensive, and is also associated with negative environmental impacts (Enes *et al*., 2006). Therefore, it is necessary to take measures to reduce the protein level in turbot diet.

If available, dietary carbohydrates and lipids will spare protein use as an energy source (Hemre *et al*., 2002). Although lipids are well utilized by most fishes, excessive levels may reduce fish growth or produce fatty fish. The carbohydrate in the diet can be used effectively in reducing feed costs (Wilson, 1994). However, excessive dietary carbohydrate has been demonstrated to cause negative effect on fish health through metabolic disturbances (Polakof *et al*., 2012). Moreover, fish physiological alterations were also observed, such as prolonged hyperglycemia (Hatlen *et al*., 2005), triggering hepatic anti-oxidative response (Azaza *et al*., 2013), high fat deposition in whole body and liver (Hemre *et al*., 2002), low red blood cells and hemoglobin (Abdel-Tawwab *et al*., 2010), increased liver histopathology (Russell *et al*., 2001), and impaired bone development (Tan *et al*., 2009). Therefore, an appropriate dietary ratio of non-protein energy sources is important for maximizing growth and feed efficiency, considering dietary protein requirement and utilization is closely related to energy availability (Erfanullah and Jafri, 1998).

Nijhof *et al*. (1994) and Garcia-riera *et al*. (1996) found that turbot was capable of metabolizing dietary carbohydrates to a certain extent. Miao *et al*. (2013) found that turbot could utilize dextrin better than glucose and sucrose. The present study was conducted to investigate the effects of dietary carbohydrate-to-lipid (CHO:LIP) ratio on the growth performance, feed utilization and metabo-

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lism response of juvenile turbot.

2 Materials and Methods

2.1 Experimental Diets

In the present study, four isonitrogenous and isoenergetic low protein level (39%) diets were formulated with increasing ratios of dietary carbohydrate to lipid (2:18, 6:18, 18:12 and 28:6). These four diets were named as D1, D2, D3 and D4, respectively. A higher protein level (50%) diet with the 2:12 ratio of carbohydrate to lipid was used as the control (Table 1). Dextrin was used as the dietary carbohydrate source. Fish oil was used as the main dietary lipid source. White fish meal, casein and gelatin were used as the dietary protein sources.

Table 1 Ingredients and compositions of the experimental diets (% of dry-matter)

Ingredient	Control	D1	D2	D3	D4
	2:12	2:18	6:18	18:12	28:6
White fish meal	36.00	36.00	36.00	36.00	36.00
Casein	21.20	12.80	12.80	12.80	12.80
Gelatin	5.30	3.20	3.20	3.20	3.20
Dextrin	0.00	0.00	5.00	15.00	28.00
Soybean lecithin	2.00	2.00	2.00	2.00	2.00
Fish oil	11.00	17.00	15.00	11.00	5.70
Carboxymethyl cellulose	1.10	1.10	1.10	1.10	1.10
Microcrystalline cellulose	20.50	25.00	22.00	16.00	8.30
Vitamin premix	0.50	0.50	0.50	0.50	0.50
Mineral premix	1.00	1.00	1.00	1.00	1.00
Choline chlorine (95%)	0.25	0.25	0.25	0.25	0.25
Calcium dihydrogen phosphate	0.50	0.50	0.50	0.50	0.50
Ethoxyquin	0.05	0.05	0.05	0.05	0.05
Calcium propionate	0.10	0.10	0.10	0.10	0.10
Attractant	0.50	0.50	0.50	0.50	0.50
Proximate analysis					
Crude protein	50.22	39.02	39.88	40.44	40.58
Crude lipid	12.36	18.29	16.93	12.39	6.70
Soluble carbohydrate	1.91	2.05	6.51	15.86	28.72
Ash	11.23	11.50	11.81	11.72	11.72
Gross energy (kJ g^{-1})†	17.09	16.81	17.24	17.18	17.17

Notes: Vitamin premix (mg (kg diet)$^{-1}$): Vitamin A, 32 mg; Vitamin D, 5 mg; Vitamin E, 240 mg; Vitamin K, 10 mg; Vitamin B_1, 25 mg; Vitamin B_2, 45 mg; Nicotinic acid, 200 mg; Vitamin B_6, 20 mg; Biotin, 60 mg; Inositol, 800 mg; Calcium pantothenate, 60 mg; Folic acid, 20 mg; Vitamin B_{12}, 10 mg; Vitamin C, 2000 mg; Microcrystlline cellulose, 1473 mg. Mineral premix (mg (kg diet)$^{-1}$): $CuSO_4 \cdot 5H_2O$, 10 mg; Na_2SeO_3, 20 mg; $MnSO_4 \cdot H_2O$, 45 mg; $CoC_{l2} \cdot 6H_2O$ (1%), 50 mg; $ZnSO_4 \cdot H_2O$, 50 mg; $Ca(IO_3)_2$, 60 mg; $FeSO_4 \cdot H_2O$, 80 mg; $MgSO_4 \cdot 7H_2O$, 1200 mg; Zeolite powder, 8485 mg. Attractants: Taurine/Glycine/Betaine = 1/3/3. † Based on the analyzed proximate composition of the completed feeds.

All ingredients were ground into fine powder through a 246-μm mesh. The diets were prepared by thoroughly mixing the dry ingredients with fish oil, and then gradually adding water to produce a stiff dough. The dough was then pelleted with an experimental feed mill (F-26(II), South China University of Technology, China), then dried for about 12 h in a ventilated oven at 50℃. The dietary pellets (1.5×3.0 mm) were sealed in a sample bag and stored at −20℃ until used.

2.2 Fish, Experimental Condition and Sample Collection

Juvenile turbot were obtained from a commercial hatchery (Jiaonan, Shandong, China). Firstly, fish were acclimated to the system for 2 weeks by feeding with a commercial diet (Qingdao Great Seven Bio-Tech, Qingdao, China). At the beginning of the feeding trial, fish were fasted for 24 h, then 420 turbot juveniles (initial body weight 8.75±0.04 g) were randomly distributed into 15 cylindrical fiberglass tanks (500 L) with a flow-through water system, 28 fish individuals each tank and 3 tanks each diet. Fish were fed by hand to apparent satiation twice daily at 07:00 and 18:00, respectively. During the 9-week feeding trial, feed consumption, mortality and feeding behavior were recorded every day. The water temperature was 19±1℃, pH was 7.7±0.1, salinity was 29.2±1, and dissolved oxygen concentration was not less than 7.0 mg L^{-1}.

At the end of the feeding trial, fish were starved for 24 h. Then all fish in each tank were counted and anaesthetized with MS-222. The body length and weight of all fish were recorded to calculate weight gain rate (WGR), daily feed intake (DFI), feed efficiency (FE), and condition factor (CF). Then all fish were dissected. Liver and visceral mass were separated and weighed to calculate hepatosomatic index (HSI) and viscerosomatic index (VSI). Samples of muscle were collected from 5 fish per tank to determine the proximate composition. Blood samples were collected from the caudal vein of 5 fish per tank by using heparinized syringe 24 h after the feeding. Blood was centrifuged at 3500×*g* for 5 min, and plasma was separated and stored at −80℃. Before frozen, plasma was divided into aliquots to determine glucose concentration, and the activities of alanine minotransferase (ALT) and aspertate aminotransferase (AST).

The calculation formulae for above parameters were as follows:

$$WGR\ (\%) = 100 \times [(\text{Final body weight} - \text{Initial body weight})/\text{Initial body weight}],$$

$$DFI\ (\%) = 100 \times \text{Feed intake in dry matter}/[(\text{Initial body weight} + \text{Final body weight} + \text{Dead fish weight})/2]/\text{Days},$$

$$FE = \text{Wet weight gain}/\text{Feed intake in dry matter},$$

$$VSI\ (\%) = 100 \times (\text{Visceral weight}/\text{Fish weight}),$$

$$CF = 100 \times \text{Final body weight}/\text{Final body length}^3,$$

$$HSI\ (\%) = 100 \times (\text{Liver weight}/\text{Fish weight}),$$

where *WGR* is weight gain rate, *DFI* is Daily feed intake, *FE* is feed efficiency, *CF* is condition factor, *HIS* is Hepatosomatic index, *VSI* is Viscerosomatic index.

2.3 Chemical Analysis

Chemical composition of the dietary ingredients, experimental diets and fish muscle were determined with the methods of the Association of Official Analytical Chemist (AOAC, 1995). Crude protein was determined by using the Kjeldahl method and estimated by multiplying nitrogen by 6.25. Crude lipid was measured after diethyl ether extraction using Soxhlet method. Ash was examined by combustion in a muffle furnace at 550℃ for 24 h. Gross energy was analyzed using the Parr 6100 Automatic Bomb Calorimeter (Parr, Moline, IL, USA).

Carbohydrate content in dietary ingredients and diets was determined with the methods of anthrone colorimetry (Hedge and Hofreiter, 1962). Content of glycogen in muscle and liver was measured as described by Plummer (1987). Plasma glucose concentration was determined by the glucose oxidase method using the kit manufactured by Sigma (Product No.510; Sigma Chemicals, St. Louis, USA). Activities of the ALT and AST were measured by the automatic biochemistry analyzer (Hitachi 7020, Tokyo, Japan). All analyses were done in triplicate.

2.4 Statistical Analysis

All statistical analyses were performed using the SPSS 13.0 for Windows. Results were presented as mean ± S.E.M. The one-way ANOVA was used to compare the difference among treatments. When significant difference ($P<0.05$) was found, Tukey's test was used to compare the mean values between treatments.

3 Results

3.1 Survival, Growth Performance and Feed Utilization

Survival rate of turbot was not significantly affected by the dietary CHO:LIP ratio (Table 2). However, dietary CHO:LIP ratio significantly affected the WGR ($P<0.05$). With the dietary CHO:LIP ratio increasing from 2:18 to 18:12, WGR significantly increased from 255.45% to 267.51%. However, when dietary CHO:LIP ratio was up to 28:6, the WGR significantly decreased to 244.79%. Moreover, there was no significant difference in WGR between the control and D3 group ($P>0.05$).

Table 2 Survival, growth performance and feed utilization of juvenile *Scophthalmus maximus* fed the experimental diets

Diet	Initial weight (g)	Final weight (g)	Weight gain rate (WGR, %)	Feed intake (DFI, d^{-1})	Feed efficiency (FE)	Survival rate (%)
Control	8.82±0.22	32.12±0.44^{b}	272.47±3.65^{d}	1.39±0.06^{a}	1.30±0.02^{c}	98.81±1.19
D1	8.74±0.27	33.24±0.60^{b}	255.45±6.38^{ab}	1.54±0.05^{ab}	1.15±0.01^{b}	96.43±2.06
D2	8.88±0.29	31.75±0.21^{b}	257.47±3.99^{bc}	1.51±0.08^{ab}	1.15±0.05^{b}	96.43±2.06
D3	8.46±0.30	31.11±0.08^{ab}	267.51±2.53^{cd}	1.60±0.11^{b}	1.11±0.09^{b}	96.43±0.00
D4	8.92±0.12	30.78±0.07^{ab}	244.79±3.58^{a}	1.90±0.05^{c}	0.95±0.06^{a}	95.24±1.19

Note: Means in the same column with different superscripts are significantly different ($P<0.05$) as determined by Tukey's test.

The feed efficiency (FE) of fish fed with the control diet was significantly higher than those fed with the other four diets ($P<0.05$). The daily feed intake (DFI) of fish fed the control diet was significantly lower than that fed with D3 and D4 ($P<0.05$). There was no significant difference in DFI and FE among fish fed with D1, D2 and D3. Fish fed with D4 had significantly higher DFI and lower FE than those fed the other diets ($P<0.05$).

3.2 Muscle Compositions

The proximate compositions of muscle are shown in Table 3. The contents of moisture, crude protein and ash in muscle were not significantly affected by dietary treatments. Crude lipid content in muscle of fish fed with D1 and D2 was significantly higher than that fed with the control diet, D3 or D4 ($P<0.05$).

Table 3 Proximate compositions of muscle of juvenile turbot *Scophthalmus maximus* fed the experimental diets (wet basis)

Composition	Diet				
	Control	D1	D2	D3	D4
Moisture (%)	76.74±1.54	76.01±1.15	76.42±1.07	76.42±1.51	75.70±0.40
Crude protein (%)	20.80±1.27	20.72±0.94	20.03±1.06	21.35±1.48	21.87±0.30
Crude lipid (%)	1.66±0.26^{a}	2.52±0.16^{b}	2.67±0.09^{b}	1.41±0.07^{a}	1.51±0.04^{a}
Ash (%)	0.15±0.01	0.15±0.01	0.15±0.01	0.15±0.01	0.15±0.01

Note: Means in the same line with different superscript letters are significantly different ($P<0.05$) as determined by Tukey's test.

Muscle glycogen content in fish fed with the control and D1 diet was significantly lower than that fed with diet D2, D3 or D4 ($P<0.05$). There was no significant difference in muscle glycogen content among the fish fed with D2, D3 and D4 (Fig.1). The highest glycogen content in liver was found in fish fed with diet D4 (Fig.2). The second highest liver glycogen content was found in the D3 group. There was no significant difference in liver glycogen content among the control, D1 and D2 group ($P>0.05$).

3.3 Body Index

Dietary CHO:LIP ratio significantly affected the body index (Table 4). The HSI of fish fed with D1 and D3 diet was significantly higher than that with the control or D4 diet ($P<0.05$). The VSI of fish fed with D1 was signify-

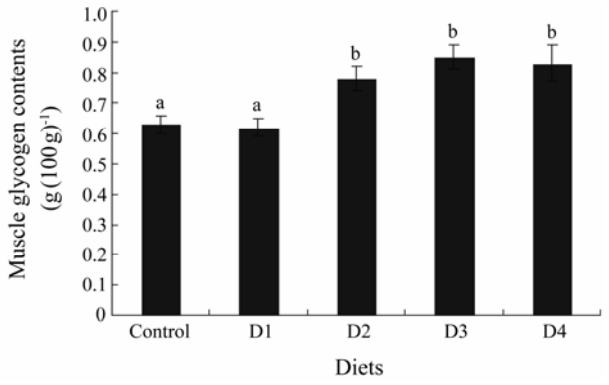

Fig.1 The content of muscle glycogen in juvenile turbot *Scophthalmus maximus* fed the experimental diets. Values are means ± SE (n =3). Bars with different letters were significantly different by Tukey's test (P<0.05).

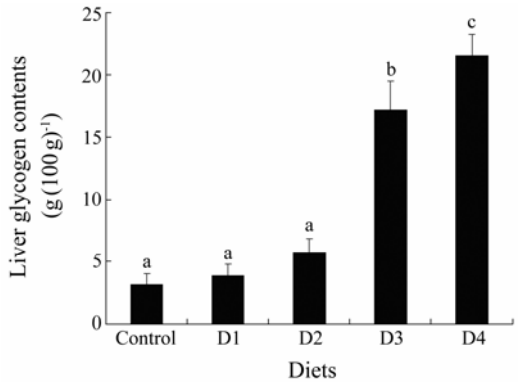

Fig.2 The content of liver glycogen in juvenile turbot *Scophthalmus maximus* fed the experimental diets. Values are means ± SE (n =3). Bars with different letters were significantly different by Tukey's test (P<0.05).

cantly higher than that with D2 (P<0.05). There was no significant difference in VSI among other groups. The CF of fish fed with D4 was significantly lower than that with D1, D2 or D3 diet (P<0.05). However, there was no significant difference in CF of fish between control and other four groups (P>0.05).

Table 4 Somatic parameters of juvenile turbot *Scophthalmus maximus* fed the experimental diets

Diet	*HSI* (%)	*VSI* (%)	*CF*
Control	1.07 ± 0.03^{a}	5.98 ± 0.44^{ab}	3.56 ± 0.14^{ab}
D1	1.48 ± 0.19^{b}	6.56 ± 0.25^{b}	3.69 ± 0.11^{b}
D2	1.23 ± 0.05^{ab}	5.73 ± 0.12^{a}	3.63 ± 0.07^{b}
D3	1.50 ± 0.16^{b}	6.28 ± 0.29^{ab}	3.89 ± 0.27^{b}
D4	1.17 ± 0.21^{a}	6.23 ± 0.09^{ab}	3.17 ± 0.07^{a}

Notes: Means in the same column with different superscript letters are significantly different (P<0.05) as determined by Tukey's test. *HSI*: Hepatosomatic index, *VSI*: viscerosomatic index, *CF*: condition factor.

3.4 Plasma Parameters

The plasma parameters of turbot are shown in Table 5. There was no significant difference in plasma glucose concentration among all the groups (P>0.05). Alanine aminotransferase (ALT) showed significantly lower values in fish fed with the control diet (P<0.05), while no difference was found among the other four groups (P>0.05). The lowest aspertate aminotransferase (AST) activity was found in fish fed the control diet, and the highest one was found in D4 group (P<0.05).

Table 5 Plasma parameters of juvenile turbot *Scophthalmus maximus* fed the experimental diets

Diet	Glucose (mmol L^{-1})	ALT (IU L^{-1})	AST (IU L^{-1})
Control	0.29 ± 0.11	14.23 ± 0.97^{a}	28.20 ± 1.00^{a}
D1	0.23 ± 0.07	21.73 ± 2.41^{b}	42.23 ± 1.24^{b}
D2	0.31 ± 0.15	21.30 ± 1.91^{b}	45.37 ± 1.45^{b}
D3	0.23 ± 0.05	19.23 ± 1.07^{b}	40.40 ± 2.04^{b}
D4	0.28 ± 0.06	20.98 ± 1.01^{b}	52.17 ± 1.21^{c}

Note: Means in the same column with different superscript letters are significantly different (P<0.05) as determined by Tukey's test.

4 Discussion

Compared with the control diet (CHO:LIP=2:12), diet D3 with the CHO:LIP of 18:12 resulted in the statistically same growth rate. According to the experimental feed formulation, the differences in feed composition between these two groups were the protein content and carbohydrate content. Thus it would be a good approach to look for the protein sparing effect of dietary carbohydrate based on the results of these two groups. This result was similar to those previous findings in other species, such as tilapia *Oreochromis niloticus* ×*O. aureus* (Shiau and Peng, 1993), ruhu *Labeo rohita* (Erfanullah and Jafri, 1995), cobia *Rachycentron canadum* (Webb *et al*., 2010). Whereas, the growth and feed utilization of turbot *S. maximus* was significantly decreased when fed diet D4. It was suggested that turbot had a limitation of dextrin utilization.

Moreover, different dietary CHO:LIP ratio significantly affected turbot growth and feed utilization. Group of D3 had higher growth rate than D1 and D2. This result could be related to two aspects. On one hand, the dietary lipid requirement for turbot was estimated to be 9% (Peng, 2013). The negative effect of larger amounts of dietary lipids on growth has been proved in turbot. Regost *et al*. (2001) discovered that there was a decrease in the growth of turbot with the increasing dietary lipid levels (11.3%–27.7%). On the other hand, the protein-sparing effect obtained by increasing lipid and/or carbohydrate levels in the diets has been reported in several fish species, such as flounder *Paralichthys olivaceus* (Lee *et al*., 2003b), Senegalese sole *Solea senegalensis* (Rubio *et al*., 2009), and catfish jundiá, *Rhamdia quelen* (Giovanni *et al*., 2010). However, the utilization of carbohydrates and lipids by fish is species-specific. For Nile tilapia *Oreochromis niloticus*, both carbohydrates and lipids were equally acceptable within the levels used on an energy equivalent basis (Ali and AL-Asgan, 2001). However, the growth performance of turbot in the present study suggested that it can possibly utilize dietary dextrin as an energy source more efficiently than lipids, which was in line with that of flounder *Paralichthys olivaceus* (Lee *et al*., 2003b). Moreover, fish fed with diet D4 got lower weight

gain rate than those fed with other diets. This result was somewhat in agreement with that of previous studies on other fish species, which also showed a worse utilization of the excessive dextrin levels (Erfanullah and Jafri, 1998; Hutchins *et al*., 1998). Walking catfish *Clarias batrachus* fed with diets containing more than 36% dextrin tended to produce lower growth (Erfanullah and Jafri, 1998). Juvenile sunshine bass *Morone chrysops* ♀ × *M. asxatilis* ♂ fed with diet containing 40% dextrin had significantly lower weight gain rate than that fed with diet containing 20% dextrin (Hutchins *et al*., 1998). However, the down-regulated growth of turbot fed with diet D4 could be due to the lipid deficiency. In order to meet the requirement of the essential fatty acids (EFA) for normal growth, appropriate dietary lipid level was necessary for aquaculture fish. Peng (2013) pointed out that the lipid requirement for optimum growth of turbot was 9%, whereas only 6.7% lipid in diet D4 in the present study. Dietary lipid deficient also resulted in the decreased growth of Atlantic salmon *Salmo salar* in the previous study (Young *et al*., 2006). The overall total feed utilization by turbot was significantly affected by the dietary CHO:LIP ratios. Fish fed with D4 had significantly higher DFI and lower FE than those in the other four treatments. In order to meet the energy requirement, turbot adjusted to have a larger feed intake, which could be also the main cause of FE decreased with the increasing dietary dextrin levels.

Crude lipid content in the muscle of turbot fed with diet D1 and D2 was significantly higher than those in the other groups. However, the muscle glycogen content was lower. These results indicated that the body lipid was directly affected by dietary lipid intake, and turbot may have limited metabolic ability to synthesize lipid from glucose. The similar result was observed in catfish *R. quelen* (Giovanni *et al*., 2010). However, the inverse relationship between dietary carbohydrate and the whole-body lipid content was also found in other species, such as Atlantic halibut *H. hippoglossus* (Hamre *et al*., 2003), Piracanjuba *Brycon orbignyanus* (Borba *et al*., 2006), silver barb *Puntius gonionotus* (Mohanta *et al*., 2007) and European sea bass *Dicentrarchus labrax* (Moreira *et al*., 2008). Moreira *et al*. (2008) found that the whole-body lipid content of European sea bass *D. labrax* fed with diets containing 46% crude protein and 12% crude lipid increased with dietary carbohydrate levels (10%–30%). In the present study, it was indicated that there was a positive relationship between dietary dextrin content and liver glycogen content. Previous studies showed that the high-level-starch diet elevated liver glycogen content in rainbow trout, eel *Angilla anguilla* (Suárez *et al*., 2002) and European sea bass (Enes *et al*., 2006).

However, a different trend was found in HSI of turbot. Fish fed with D4 got the lowest HSI. HSI is often used as an indicator of condition and nutritional status of fish. Moreira *et al*. (2008) reported that the increase of HSI is related to the increased glycogen and lipid deposition in the liver of fish. Rueda-Jasso *et al*. (2004) reported that hepatocyte vacuolization was due to the storage of lipid and glycogen. In gadoid fish, an increase in liver lipid content is associated with an increase in HSI (Nanton *et al*., 2001). Lipid content, just like glycogen, could also affect HSI of turbot.

Carnivorous fish fed with diets containing high level of starch seem to have a poor ability in response to the excess glucose (Hemre *et al*., 1993, 1995; Moon, 2001). It was assumed that these fish were under constant metabolic stress (Pieper and Pfeffer, 1980). Thereafter, it may result in suppressed immune functions (Ellis, 1981; Maule *et al*., 1989; Wiik *et al*., 1989). The enzymes of ALT and AST are quantitatively the most important aminotransferases in the fish liver (Cowey and Walton, 1989). When the liver cells are damaged or their permeability increased, AST and ALT will be released into the blood, resulting in elevated blood transaminase activity (Ming *et al*., 2012). Hence, the increased activities of aminotransferases in fish means that some damages have happened to liver (Bell *et al*., 2006; Montero *et al*., 2010). Therefore, the activities of AST and ALT in the blood can be used to monitor the health status of fish. No difference in ALT activities was observed in fish fed different graded CHO:LIP ratio, whereas significantly increased AST was observed in fish fed diet D4 in the present study. It indicated that high dietary CHO:LIP ratio affected turbot health. This phenomenon is similar to the results reported in other fish species. For example, activities of plasma AST and ALT were significantly increased in juvenile obscure puffer *Takifugu obscurus* fed the high-amylose diet (Liu *et al*., 2014). However, the HSI in D4 group, in the present study, did not significantly differ from that in the control group. Further study is needed to make it clear how the relative high dietary carbohydrate level influences the fish health.

In a conclusion, the present results confirmed that the juvenile turbot can utilize carbohydrate. Furthermore, the appropriate ratio of dietary carbohydrate to lipid was important to the growth and feed utilization of turbot. The proper CHO:LIP ratio based on the growth performance in the present study was determined to be 18:12 when the dietary protein level was 39%.

Acknowledgements

This research was financially supported by the National Basic Research Program (973 program, No. 2014CB 138600) and the National Natural Science Foundation of China (No. 31072219).

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(Edited by Qiu Yantao)