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Optimum Dietary Protein Requirement of a Thai Strain of Climbing Perch, *Anabas testudineus* (Bloch, 1792) Fry

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Abstract

The optimum dietary protein requirement of a Thai strain of climbing perch (*Anabas testudineus*, Bloch, 1792) fry was investigated by feeding six semi-purified diets containing varying levels of dietary protein within a range of 25-50% over an 8 week feeding period. Fish meal was used as protein source. The experiment was conducted in a recirculatory system consisting 18 glass aquaria of each 65 litre capacity. Each treatment had three replicates. Each of the aquaria was stocked with 12 fingerlings of *A. testudineus* with a mean initial weight of 1.04±0.02 g. Fish were fed to apparent satiation three times daily. Statistical analysis of growth data showed that percent weight gain, feed conversion ratio (FCR), protein efficiency ratio (PER) and body composition were significantly ($P<0.05$) affected by dietary protein levels. The weight gain and specific growth rate (SGR) of fish in all dietary groups increased proportionally with the increase in dietary protein concentration to level of 40% and thereafter decreased with further increase in dietary protein levels. Fish fed diet 4 containing 40% protein level showed significantly ($P<0.05$) the highest weight gain. The FCR values ranged between 1.95 and 5.73 and the best FCR value was obtained with diet 4. The PER decreased with increasing dietary protein levels. The PER values ranged between 0.64 and 1.27 and the apparent net protein utilization (ANPU%) values ranged between 9.76 and 19.80%. In general, there was a progressive decrease in carcass protein and an increase in carcass lipid. On the basis of observed weight gain, SGR and FCR, a diet containing 40% protein with a protein to energy (PE) ratio of 92.63 (mg protein/Kcal gross energy) is recommended for maximum growth of *A. testudineus* under experimental condition used in this study.

Keywords: Climbing perch, semi-purified diets, protein requirement, growth.

Introduction

Among the live fishes, climbing perch commonly known as koi (*Anabas testudineus*) is one of the most popular fishes in Bangladesh because of its taste and high market demand. This species is considered a valuable item of diet for sick and convalescents. It contains high amount of available iron and copper essentially needed for haemoglobin synthesis. In addition, it also contains all the essential amino acids (Saha, 1971).

A. testudineus encompasses a wide range of geographical distribution. This freshwater fish is found in the beels, haors, baors, flooded water bodies, ponds, streams and rivers of Bangladesh (Siddique *et al*., 2000). This species naturally occurs in India, Pakistan, Burma, Sri-Lanka, Thailand, China, Hong Kong, Philippines, Polynesia and Malaysia (Jayaram, 1981).

Once upon a time *A. testudineus* was abundantly available in almost all freshwater systems of Bangladesh, but its recent trends showing a continuous dilapidated tendency. One decade ago the contribution of koi was 2.83% in the total pond catch of Bangladesh but latter it turned down to 0.85% (DoF, 1999). The reasons for such drastic decline are ecological degradation, indiscriminate fishing, use of pesticides and fertilizers, habitats alteration, obstruction of breeding migration etc. As the natural population of koi is declining fast. Fish Biologists are thinking for its cultivation through intensive farming (DoF, 2002).

Since the augmentation of fish production from canals, beels, haors, rivers and estuaries is generally considered to be complicated, intensification of pond aquaculture is necessary. The induced breeding, larval and fry rearing technique for commercial aquaculture of *A. testudineus* are successful at farmers' level. Recently attempts have been made to boost up the aquaculture production through incorporation of an exotic strain of *A. testudineus* known as Thai koi, which was introduced in Bangladesh by private sector in 2001 from Thailand (Mr. Nurul Haque, personal communication). The higher growth and bigger size of Thai koi and tolerance of wide range of environmental condition soon caught the attention of farmers for its potential aquaculture.

Climbing perch encompasses a divergence feeding behaviour. It is a fierce predator, and will also eat other fish if they can master them. Some are omnivorous, feeding on invertebrates, fish and plants. Climbing perch appear to be visual feeders, feeding primarily during the day (Patra, 1993). To intensify the culture of this species provision of nutritionally balanced diet is necessary. So, it is essential to know the minimum protein requirement for optimum growth in formulating a balanced ration since protein is the most important major nutrient providing for growth and other metabolic activities, as well as having a higher cost than carbohydrate and fat (Wee and Ngamsnae, 1987). There is not much information available in on the nutritional requirement of climbing perch and the only report available in the literature regarding the protein requirement of *A. testidineus* is by Chareontesprasit and Jiwyam (1996). Thus, the present study was undertaken with a view to determine the optimum dietary protein requirement of *A. testudineus.*

Materials and Methods

Experimental System

The experiment was conducted in a recirculatory system in the Wet Laboratory of the Department of Aquaculture, Bangladesh Agricultural University, Mymensingh for a period of 8 weeks during the month of April to June, 2006. The recirculatory system consists of 20 rectangular glass aquaria (each of size 46 × 41 × 41 cm^3) containing about 65 liter of water. The flow rate in each aquarium was maintained at about 1.5 Litre per minute. Altogether 18 aquaria were used for the study. All the aquaria were kept on a rack made of iron bar to facilitate better observation and accessibility. Water from a deep tube well was used in the aquaria during experimental period. An adequate level of oxygen in each aquarium was maintained through artificial aeration using an air pump (Johnson pump, MDR-Series). Natural photoperiod of 12 hr light and 12 hr dark was maintained throughout the experimental period.

Source of Fry and Acclimation

The induced bred fry of Thai strain of climbing perch, *A. testudineus* (1.04±0.02 g) were collected from Freshwater Station, Bangladesh Fisheries Research Institute (BFRI), Mymensingh during the month of April, 2006. The collected fish were given a prophylactic treatment with salt (3% NaCl) solution for 10 minutes. During treatment sufficient oxygen supply was maintained through artificial aeration. Before starting the experiment, the fish were acclimatized to the experimental condition for one week and fed commercial shrimp nursery diet (42% protein).

Diet Formulation and Preparation

Six semi-purified diets containing 25, 30, 35, 40, 45 and 50% level of protein were formulated for determining the dietary protein requirement of *A. testudineus.* Fish meal was used as the protein source and was of Malaysian origin. Prior to formulation of diets, the fish meal was analyzed for proximate composition and it contained 66% protein, 11.06% lipid, 20.34% ash and 0.5% crude fibre. The experimental diets were formulated to be as iso-energetic (gross-energy) as possible by adding soybean oil and α -cellulose as bulking or filler material (Table 1).

In order to prepare diets, all the dietary ingredients were finely ground and sieved to pass through 0.5 mm mesh. All the dietary ingredients including vitamin-mineral premix were weighed as per formulae (Table 1) and mixed well with an electrical mixture machine. During mixing oil was gradually poured into the mixture and mixed thoroughly to ensure homogeneity. Then required amount of water was added to moisten the mixture and the mixture was extruded through 1 mm diameter die of a pellet machine (Hobart Mixture Machine, Model A200). The pellets were dried at 45°C in an oven overnight. The pellets were broken and sieved to bite size pellet for the fry. All the diets were packed in air-tight polyethylene bag and stored in a deep freeze

Table 1. Formulation (%) of the experimental diets

Ingredients	Diets					
	1	2	3	4	5	6
Fish meal	37.82	45.38	52.95	60.51	68.07	75.64
Soybean oil	5.82	5.00	4.15	3.30	2.47	1.63
Starch	43.00	37.00	30.00	25.50	17.00	14.00
Vitamin & mineral premix*	2.00	2.00	2.00	2.00	2.00	2.00
Binder**	3.00	3.00	3.00	3.00	3.00	3.00
α-cellulose	8.36	7.62	7.90	7.69	7.46	6.73
Total	100.00	100.00	100.00	100.00	100.00	100.00

* Obtained from SK+F, Eskayef Bangladesh Limited
** Carboxymethyl cellulose.

for further use. The diets were subjected to proximate analysis and the results are shown in Table 2.

Experimental Design, Feeding and Sampling of Fish

Eighteen aquaria were divided into 6 treatments viz. T_1, T_2, T_3, T_4, T_5 and T_6 each having three replicates. The fishes were then randomly released into different treatment groups. Twelve fishes were stocked in each aquarium. The fishes were hand fed to apparent satiation three times, delivered at 9.00, 13.00 and 17.00 hr daily. A record of supplied feed was kept for determining the food conversion ratio (FCR), protein efficiency ratio (PER) and apparent net protein utilization (ANPU %). Sampling was done weekly interval. Prior to weighing, fish were caught with a fine mesh scoop net and excess water was then removed from fish body by gently blotting on a soft tissue paper. Weight of fish in each sampling was measured by bulk weighing them using a digital electronic balance (KERN, Model No EMB-2000-0). During sampling fish were handled very carefully. Any mortality of fish during the study period was recorded. The aquaria were washed and cleaned during sampling time. At the beginning of the experiment twenty fish from the stock was randomly sacrificed and used for initial proximate composition analysis. At the end of the experiment five fish from each replicate were sacrificed and used for final carcass composition analysis.

Water Quality Parameters

The water quality parameters such as water temperature, dissolved oxygen (DO), pH and total ammonia were monitored weekly throughout the experimental period. The ranges were: temperature 26.0–29.5°C, dissolved oxygen 6.5–8.4 mg/L, pH 6.9–8.4, and total ammonia 0.10-0.25 mg/L.

Analytical Methods and Statistical Analysis

The proximate composition of fish samples, feed ingredients, and experimental diets was analyzed in triplicate according to AOAC (1990). Specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER) and apparent net protein utilization (ANPU%) were calculated according to Castell and Tiews (1980). Data on growth performance and feed utilization were compared by one-way analysis of variance (ANOVA) followed by Duncan's multiple range tests (Duncan, 1955). A significance level of $P<0.05$ was used.

Results

Analyzed Composition of the Experimental Diets

The analyzed proximate composition of the experimental diets is shown in Table 3. The protein content in different diets varied between 25.58% and 50.36%. The variations of protein levels in all experimental diets from the expected level were very small. The variations in lipid content among different experimental diets were also minimal. However, there was slight variation in ash content. Diets 5 and 6 contained 15.80 and 16.10% ash respectively whereas diet 1 and 2 contained 10.46 and 11.71% ash respectively. The dietary crude fibre contents ranged between 6.18 and 7.68%. The gross energy content of experimental diets varied between 4.29 and 4.51 Kcal/g. The protein to energy (PE) ratio of different experimental diets varied between 59.62 and 111.66. The calculated essential amino acid (EAA) compositions of the experimental diets are also shown in Table 4. The EAA requirement value for channel catfish was used as reference values since no EAA requirement values for *A. testudineus* is not available.

Acceptability of Diets

The acceptability of different experimental diets was judged by a subjective behavioral assessment of the feeding responses. All fish were acclimated to the experimental diets from the start of feeding. The acceptability of all the diets was more or less similar. Fish were observed to feed actively throughout the trial.

Table 2. Analyzed proximate composition of the experimental diets (% dry matter basis)

Components	Diets					
	1	2	3	4	5	6
Dry matter	91.51	92.65	92.88	92.91	93.31	93.54
Protein	25.58	30.50	36.53	40.48	45.41	50.36
Lipid	10.15	10.19	10.41	10.20	10.43	10.53
Ash	10.46	11.71	13.32	15.26	15.80	16.10
Crude fibre	7.68	6.59	6.72	6.75	6.46	6.18
NFE*	46.13	41.01	33.02	27.41	21.55	16.83
Gross energy (Kcal/g)**	4.29	4.35	4.38	4.37	4.41	4.51
PE ratio***	59.62	70.11	83.40	92.63	102.97	111.66

*Nitrogen free extract calculated as 100 – %(moisture + protein + lipid + ash + crude. fibre)
**According to Jauncey and Ross (1982)
***Protein to energy ratio (mg protein/ Kcal energy).

Table 3. Growth performance and feed utilization of *A. testudineus* fed experimental diets for 8 weeks

Parameters	Diets					
	1	2	3	4	5	6
Mean initial weight (g)	1.07^{a}±0.01	1.02^{a}±0.04	1.06^{a}±0.03	1.06^{a}±0.03	1.03^{a}±0.03	1.03^{a}±0.03
Mean final weight (g)	2.63^{d}±0.17	3.73^{c}±0.23	4.84^{b}±0.04	7.79^{a}±0.17	7.61^{a}±0.17	7.13^{a}±0.06
Weight gain (g)	1.56^{d}±0.18	2.71^{c}±0.19	3.79^{b}±0.06	6.74^{a}±0.15	6.58^{a}±0.16	6.11^{a}±0.08
%Weight gain	145.59^{d}±17.79	265.16^{c}±10.76	358.18^{b}±12.92	637.98^{a}±14.58	638.72^{a}±23.43	595.15^{a}±21.87
SGR (% day)	1.65^{d}±0.06	2.31^{b}±0.06	2.76^{b}±0.07	3.57^{a}±0.04	3.57^{a}±0.06	3.46^{a}±0.06
FCR	5.73^{f}±0.32	3.86^{e}±0.09	2.36^{d}±0.12	1.95^{a}±0.05	2.09^{b}±0.04	2.24^{c}±0.09
PER	0.64^{d}±0.07	0.83^{c}±0.01	0.91^{b}±0.04	1.27^{a}±0.03	1.05^{ab}±0.03	0.89^{b}±0.04
ANPU (%)	9.76^{f}±0.73	12.78^{e}±0.24	14.04^{c}±0.56	19.80^{a}±7.10	16.48^{b}±0.39	13.63^{d}±0.70
Mortality (%)	25.0	16.6	-	-	-	-

*Figures in the row having same superscripts are not significantly different (P>0.05)
* Mean values ±standard deviation

Table 4. Calculated essential amino acid (EAA) contents of experimental diets*

Essential amino acids (EAA)	Diets						Requirement for channel catfish**
	1	2	3	4	5	6	
Arginine	1.90	2.27	2.66	3.03	3.41	3.80	1.03
Histidine	0.68	0.81	0.95	1.08	1.22	1.36	0.37
Leucine	2.13	2.55	2.98	3.41	3.83	4.26	0.84
Isoleucine	1.28	1.54	1.80	2.06	2.32	2.57	0.62
Lysine	2.20	2.65	3.08	3.52	3.95	4.40	1.50
Methionine	0.86	1.03	1.20	1.38	1.55	1.71	0.56
Cystine	0.30	0.36	0.42	0.50	0.56	0.61	-
Phenylanine	1.11	1.33	1.56	1.78	2.00	2.22	1.20
Tyrosine	0.90	1.08	1.27	1.45	1.62	1.80	-
Threonine	1.20	1.43	1.68	1.91	2.16	2.40	0.53
Tryptophan	0.31	0.38	0.43	0.50	0.56	0.62	0.12
Valine	1.76	2.12	2.47	2.83	3.18	3.53	0.71

*EAA values were calculated on the basis of reference value given in NRC (1983)
**According NRC (1983)

Growth Performance of Fish

The result of the growth trial showed that diet 4 containing 40% protein produced the best growth response among the experimental diets, but it was not significantly different (P>0.05) from that of diet 5 and 6. Diet 1 (25% protein) resulted in the poorest growth. Weight gain of fish in diets 2 and 3 were significantly lower (P<0.05) than those in diets 4, 5 and 6 respectively. The specific growth rates (SGR) of fish ranged between 1.65 to 3.57 with diets 4, 5 and 6 producing higher (P<0.05) SGR while diet 1 producing the lowest SGR value (Table 3). There was no significant difference (P>0.05) between the SGR values of diets 2 and 3.

The mean food conversion ratio (FCR) of different experimental diets ranged between 1.95 and 5.73. Diet 4 resulted in the lowest i.e. the best FCR while diet 1 resulted in the highest i.e. the worst FCR. The PER values ranged between 0.64 and 1.27 with diet 1 producing the lowest and diet 4 producing the highest PER values. However, the PER value in diets 4 and 5 were not significantly different (P>0.05). Similarly, there was no significant differences between the PER values diets 3 and 6. The mean apparent net protein utilization (ANPU) values ranged between 9.76 and 19.80%. Like the PER values, diet 4 with 40% protein level showed significantly the highest ANPU value. No mortality was observed in fish fed diets 3, 4, 5 and 6 while diets 1 and 2 showed a mortality of 25.0 and 16.6% respectively.

Proximate Carcass Composition

There was a marked increase in protein and lipid content of fish. The final carcass moisture content ranged between 72.30% and 74.59% (Table 5). Diets 1 and 2 had higher (P<0.05) carcass moisture content. However, there was no significant difference between the moisture contents of diets 2 & 3 and 4, 5 & 6 respectively. The carcass protein content of fish in different diets ranged between 14.50% and 15.48%. Fish fed diet 4 resulted in the highest (P<0.05) carcass protein content. The carcass lipid content of fish in different treatments ranged from 3.61% to 5.20%. Fish fed diet 4 resulted in the highest carcass lipid content but it was not significantly different (P>0.05) from those in diets 5 and 6. Again, there was no significant difference between the carcass lipid content of fish fed diets 3 and 6. The carcass ash

Table 5. Carcass proximate composition of fish samples at the start and end of the experiment (% fresh matter basis)

Components	Initial (all fish)	Diets					
		1	2	3	4	5	6
Moisture	77.29	74.59^{a}±0.51	74.01^{ab}±0.19	73.44^{b}±0.08	72.30^{c}±0.31	72.46^{c}±0.48	72.69^{c}±0.04
Protein	13.25	14.50^{f}±0.19	14.76^{e}±0.17	14.99^{d}±0.24	15.48^{a}±0.09	15.32^{b}±0.14	15.10^{c}±0.23
Lipid	2.86	3.61^{d}±0.12	3.98^{c}±0.12	4.52^{b}±0.12	5.20^{a}±0.11	5.15^{a}±0.12	5.02^{ab}±0.14
Ash	6.10	6.75^{a}±0.09	6.69^{a}±0.13	6.41^{ab} 0.15	6.10^{b}±0.14	6.25^{b}±0.11	6.30^{ab}±0.17

* Values are mean±standard deviation.

**Values in the same row having same superscripts are not significantly different (P>0.05).

content of the fish varied from 6.10 to 6.75%.

Discussion

Feed consumption, growth and feed efficiency of fish are normally governed by a few environmental factors (Fry, 1971). Environmental parameters exert an immense influence on the maintenance of a healthy aquatic environment and production of food organism. In the present study the water quality parameters were found to be within the acceptable level for fish culture. Recommendation for water quality usually specifies DO greater than 3 mg/L for growth of channel catfish (Weeks and Ogburn, 1973). In the present study DO levels were maintained at near saturation by artificial aeration, the range of DO was 6.5-8.4 mg/L. Ammonia concentration is also an important growth affecting factor. Total ammonia fluctuated within the range of 0.10 to 0.21 mg/L and was independent of dietary treatments. This is probably due to the denitrification of ammonia by bacteria grown in the biological filters used. The circum-neutral pH or slightly alkaline pH is more suitable for fish culture. According to Swingle (1967) and Boyd (2000) pH ranging from 6.5 to 9.0 is suitable for pond fish culture. The pH values obtained in the present study (6.9 to 8.6) fell within this suitable range.

Little or no information on the nutritional requirement of climbing perch is available in the literature. Considering the importance of this fish for aquaculture, it is essential to know the optimum protein requirement for formulating balanced diet for its intensive culture. The dietary protein requirement of various cultured species have been investigated by various authors (Chareontesprasit and Jiwyam, 1996; Zhang *et al*., 2000; Ng *et al.*, 2001; Hossain *et al*., 2002; Kim *et al*., 2003; Giri *et al.*, 2003 and Hossain *et al*., 2005). The dietary protein requirements of fish are about 2 to 3 times higher than that of mammal (Pandian, 1987). He pointed out that most workers have chosen juvenile fish, fixed feeding rate and used diet with limiting protein sparing action.

The minimum level of dietary protein which produced maximum growth in *A. testudineus* in the present study was estimated to be approximately 40% on the basis of weight gain, specific growth rate and feed conversion ratio. A similar dose response dietary protein requirement has been reported for Gulsha, *M. cavasius* (Hosssain *et al.*, 2005), Mahseer, *Tor putitura* (Hossain *et al.*, 2002), Malaysian freshwater catfish, *Mystus nemurus* (Khan *et al*., 1993), tilapia, *Tilapia mossambicus* (Jauncey, 1982), milk fish, *Chanos chanos* (Lim *et al.*, 1979) and rainbow trout, *Salmo gairdneri* (Satia, 1974).

The present study indicated that 40% dietary protein is the optimal level for maximum growth of *A. testudineus*. This level is comparable to the protein requirement reported for Gulsha, *M. cavasius* 40% (Hossain *et al.*, 2005), Mahseer *Tor putitura* 40% (Hossain *et al*., 2002), Indian catfish *Clarias batrachus* 37-40% (Singh, 1994), Catfish *C. batrachus* 40% (Khan and Jafri, 1990). However, this level is higher than that of 30% reported for *A. testudineus* (Chareontesprasit and Jiwyam, 1996) and certain other catfish species, particularly *C. batrachus* fry, 30% (Chuapoehuk, 1987) and *Pangasius sutchi*, 30% (Aizam *et al*., 1980).

In the present study, weight gain of *A. testudineus* increased at increasing protein levels of (25-40%) and decreased when the protein level further increased to 45-50%. This typical growth response to changing dietary protein level in iso-caloric diets has been observed in many other species irrespective if rearing strategies (Jauncey, 1982; Khan *et al.*, 1993; Vergara *et al*., 1996; Hossain *et al.*, 2005). This decrease in weight gain at protein levels above the optimum may be due to a reduction in available energy for growth because of inadequate non-protein energy necessary to deaminate and excrete excess absolute amino acids as described by Cho *et al*. (1985) and Vergara *et al*. (1996).

It has been postulated that the decrease in growth response at protein levels above the optimum may be due to the reduction in dietary energy available for growth as extra energy is required to deaminate and excrete the excess amino acids absorbed (Jauncey, 1982). Lim *et al.* (1979) also reported that the slightly lower weight gain of milkfish *Chanos chanos* (Forskal) fed diets with protein levels above the optimal could be due to insufficient non-protein energy in the diet. It has been suggested that high levels of proteins and low amounts of non-protein energy may be toxic to channel catfish, *Ictalurus punctatus* (Prather and Lovell, 1973).

Another cause of low growth rate at low dietary protein levels might be the content of low metabolic energy in experimental diets. Although diets were formulated on an iso-caloric basis, when metabolizable energy is considered, the experimental diets used in the present study may not be strictly iso-caloric. The experimental diets with higher protein and lower carbohydrate mixture may contain more metabolizable energy than those with lower protein and higher dextrin as reported by Moore *et al.* (1988). Although there was no mortality of fish in the present study fed diets 3, 4, 5 and 6 containing higher protein levels (35-60%), the mortality in diets 1 and 2 might be related to the lower level of dietary protein (25% and 30% respectively) which could not fulfill the nutritional requirement of the fish for their optimal growth.

The protein requirements of fish can sometimes be overestimated when practical diets are used because factors such as digestibility and amino acid composition of dietary proteins are not always given adequate consideration. The calculated essential amino acid (EAA) composition of the experimental diets in the present study showed that none of the diets were deficient in any of the EAAs compared to the requirement pattern for channel catfish *Ictalurus pantatus* (Table 4) as reported in NRC (1983). Although there was no apparent deficiency of EAAs in experimental diets some fish fed diets 1 and 2 showed nutritional deficiency symptoms like tail and fin rot and eventual mortality (Table 4).

There was a distinct trend for FCR to decrease with increasing protein levels upto a certain levels similar to the trend observed in Tilapia (Juancey, 1982) and *Puntius gonionotus* (Wee and Ngamsnae, 1987). The PER in the present study followed more or less similar trend of FCR and ranged from 0.64 to 1.27 with highest value in diet 4 containing 40% protein. The PER values obtained in the present study also indicated that the optimum dietary protein requirement is 40%. Below and beyond this the PER of *A. testudineus* was significantly reduced. The decreasing trend of PER was also observed with snakehead, *Channa micropeltes* (Wee and Tacon, 1982), grass carp (Dabrowski, 1977), *P. gonoinotus* (Wee and Ngamsnae, 1987), and white sturgeon, *Acipenser transmontanus* (Moore *et al.*, 1988).

There was increasing trend of ANPU values with increasing dietary protein concentration but at over the optimal concentration it started to decrease. Similar trends were also reported by Jauncey (1982) in *Tilapia mosambicus*, Wee and Ngamsnae (1987) in *P. gonionotus,* Hossain *et al*. (2002) in *Tor putitora* and Hossain *et al.* (2005) in *M. cavasius.* Steffens (1981) also reported that raising the dietary protein level improves the growth rate and food conversion but reduces PER and protein productive values (PPV) in *Cyprinus carpio*. The dietary protein contents showed an obvious positive relation with carcass protein with a corresponding negative relation with fish body moisture contents which was reported earlier (Jauncey, 1982; Khan *et al.*, 1993).

The carcass protein content of *A. testudius* in the present study tended to increase with increasing dietary protein levels of 25- 40% and were significantly ($P<0.05$) higher than that obtained with fish fed 50% dietary protein. Khan *et al*. (1993) reported that the whole body protein of *M. nemeurus* increased significantly with increasing dietary protein levels up to 42% and then decreased when fish were fed with higher protein levels. Works on other fish species have found a similar increase in body protein content with increasing dietary protein (Cowey *et al.*, 1972; Jauncey, 1982). In the contrary other researchers have reported no significant changes in fish body protein content when fish were fed with various levels of dietary protein (Winfree and Stickney, 1981; Moore *et al.*, 1988). Shearer (1994) pointed out that both endogenous factors such as fish size, sex as well as exogenous factors such as diet composition and culture environment influence the proximate composition of fish. This may partly explain the lack of agreement concerning the influence of various levels of dietary protein on the protein content of the fish body.

Fish fed higher dietary protein showed a higher carcass lipid (Table 5). A similar trend for crude lipid has been reported by Murai *et al.* (1985) in *Cyprinus carpio* and Hossain *et al.* (2005) in *M. cavasius*. This observation suggested that the protein: energy ratio of the diets was different because a higher dietary protein energy ratio usually results in higher body lipids in fishes (Garling and Wilson, 1976).

The importance of protein level in relation to the energy levels of the diets in fish is well recognized. The results of the present study showed that 40% protein with 4.37 Kcal/g gross energy is the best protein energy ratio (92.63) based on the growth and feed utilization of *A. testudineus*. This may be due to the fact that this protein (40%) in diet 4 was optimum to promote growth and energy supplied by this diet was adequate for maintenance and growth. Hossain *et al.* (2005) observed best growth performance and feed utilization in *M. cavasius* fed a fish meal based diet with a similar protein energy ratio of 91.29. Ng *et al.* (2001) recommended a 44% dietary protein with a protein energy ratio of about 83.68 (mg protein/kcal gross energy) for maximum growth of *M. nemurus.*

Based on the weight gain, SGR, feed and protein efficiency of *A. testudineus* observed in the present study, a diet with 40% protein with a protein to energy ratio of 92.63 is recommended for maximum growth of *A. testudineus* under experimental condition used in this study.

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