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PROOF RESEARCH PAPER

Dietary Energy Requirement of *Pangasianodon hypophthalmus* (Sauvage, 1878) Juveniles Reared at Two Temperatures

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Abstract

A 60-days feeding trial was conducted to study the dietary energy requirement of *Pangasianodon hypophthalmus* (Sauvage1878) juveniles reared either at ambient (24.5°C) or higher temperature (32°C) and fed with one of the four diets having dietary energy content of 342 Kcal. 100 g^{-1}(T1), 379Kcal. 100 g^{-1}(T2), 428.5 Kcal. 100 g^{-1}(T3) and 448.6 Kcal. 100 g^{-1}(T4). Juveniles of *P. hypophthalmus* with amean weight 4.27±0.12 to 4.40±0.10 g was stocked in 24 uniform size plastic rectangular tanks in triplicates with 10 fish per tank following a 2x4 factorial design. Growth performance and nutrient utilization of *P. hypophthalmus* were higher at the dietary energy content of 428.5 Kcal. 100 g^{-1}(T3) at both the temperatures. The body composition of *P. hypophthalmus* did not differ significantly ($p>0.05$) as the energy content of the diet increase. However, the tissue lipid content was significantly varied ($p<0.05$) at different temperature and energy levels. Based on the second order polynomial regression equation the dietary energy requirement of *P. hypophthalmus* was recorded to be 410 Kcal. 100 $g^{-1}$and 402.5 Kcal. 100 g^{-1} at 32°C and at ambient temperature(24.5°C), respectively.

Keywords: *Pangasianodon hypophthalmus*; Dietary Energy; Temperature; Growth; Polynomial regression.

Introduction

Pangasianodon farming is one of the fastest growing types of aquaculture in the world. Since its introduction in Indian aquaculture, ithas achieved an impressive success as a commercial aquaculture species (Lakra and Singh 2010; Singh and Lakra 2012). Energy is the most important constituents of the diet as feed intake in fish is influenced by the available dietary energy content, because fish meet their energy requirements by eating or foraging (Lee and Putnam 1973; Jobling and Wandsvik 1983; Kaushik and Luquet 1984). The energy requirements of fish depends on the species, water temperature and physiological stage of their development (Guillaume *et al*. 2001). As it is evident from various findings that dietary energy level of the feed regulates the feed intake of fish and thus, it affects the growth performances, protein efficiency ratio, body lipid accumulation, water quality of pond and hence determines the financial profit from the fish culture (Lovell1998). Therefore, it is necessary that the energy content of the diet must be adjusted to ensure the desired intake of all required nutrients for optimal growth performance and nutrient utilization.

Temperature is the most important abiotic factor which directly influences the fish growth performance, carcass composition and energy requirement of fish (Brett and Groves 1979; Corey *et al*. 1983; Russell *et al*. 1996; Sun *et al*. 1999, 2000, 2006; Person-Le Ruyet *et al*. 2004,Bureau & Hua 2008; Helland *et al*. 2010; Amin *et al*. 2014).Since fish is a poikilothermic animal,their growth performances, nutrient utilization, feed conversion and other physio-metabolic functions are influnced with temperature (Houlihan *et al*. 1993; Britz *et al*.1997; Jobling 1997; Azevedo *et al*. 1998).A rise in water temperature increases the metabolic rate of aquatic organisms and therefore their energy requirement. In India, culture of this fish is propagating in many parts of the country having different temperature range that varies from 20°C to more than 35°C. Looking into this the present study on *Pangasianodon hypophthalmus*, growth performances are very much required to explore the optimization of the nutrient requirement of this fish in different thermal acclimation. The present study will be exploring with the following objectives a) To determine the optimum dietary energy requirement of *Pangasianodon hypophthalmus* reared in different thermal acclimation and b) To assess the growth performances of *Pangasianodon hypophthalmus* reared in different thermal acclimation.

Materials and Methods

The experiment was conducted at the wet laboratory of CIFE, Mumbai over a period of 60 days. Subsequently, the laboratory work was carried out in Fish Nutrition, Biochemistry and Physiology laboratory of CIFE. Experimental fishes were procured from a commercial farm from Kolkata. The fishes were transported to the wet laboratory in polyethylene bags.To ameliorate the handling stress during transportation the fishes were given a mild salt treatment (4 ppm) and vitamin C treatment (4 tablet per thousand liter of water) the next day. The stock was acclimatized under aerated conditions for a period of 15 days at two temperatures (12 tanks at 32°C and 12 tanks at ambient temperature). Acclimation of fishes those who are acclimatized at 32°C was carried out by electrical heaters (fully submersible automatic aquarium heater, 300W, RS electrical heater; Zhongshan Risheng, China). The water temperature of the tank was increased at the rate of 1°C per day over ambient temperature till the fishes were acclimated at 32°C. Fishes were acclimated at this temperature for a further 15 days before the start of the experiment. During acclimation, fish were fed a basal diet containing 30% crude protein.Animals used for the experiment were juveniles of *Pangasianodon hypophthalmus*, with an average weight ranging from 4.2 g to 4.6 g (4.27±0.12 to 4.40±0.10). The setup consisted of 24 uniform size plastic rectangular tanks (80 cm × 57 cm × 42 cm, 150 L capacity) covered with perforated lids. Two hundred forty (240) fish were randomly and equally distributed and stocked into experimental tanks with a 2x4 factorial design in triplicates.The total volume of the water in each tub was maintained at 100 L throughout the experimental period. Round the clock aeration was provided. The aeration pipe in each tub was provided with aeration withcontrol to provide the air pressure uniformly in all the tanks.

During the feeding trial fishes were fed to satiation twice daily at 9:00 am in the morning and 5:00 pm in the evening. Water temperature in the tanks was measured twice daily and ranged from 22-28°C at ambient temperature (24.35 ± 0.21°C, mean ± SE) and 32-33°C at high temperature (32.2 ± 0.06°C, mean ± SE).Similarly, other parameters like DO, pH, free CO_2, hardness, Ammonia, Nitrite and Nitrate were also estimated periodically as per APHA method (APHA 1998) to keep the water quality optimum for the sustained culture of fish.

Diet Preparation

Purified ingredients such as casein (vitamin free), gelatin, dextrin, starch, cellulose, carboxymethyl cellulose (CMC), Butylated hydroxytoluene (BHT), cod liver oil, vitamin mix and mineral mixture, were taken for feed formulation (Table1). All the ingredients were weighed properly as per the requirement and were kept in a plastic container. Gelatin crystals were mixed in luke warm water so as to form a jelly, which mixes easily with the other ingredients. The required mixed ingredients were then mixed with the gelatin jelly to form a dough with the addition of the necessary quantity of water. When the dough was formed, the dough was then transferred to an aluminum container, which was

Table 1. Composition of purified experimental diets

Ingredients (%)	T1	T2	T3	T4
Casein	35	35	35	35
Gelatin	3	3	3	3
Starch	13	20	28	30
Dextrin	2	4	8	10.9
Cellulose	30.88	21.88	9.88	4.98
CMC	4	4	4	4
Cod liver oil	10.1	10.1	10.1	10.1
BHT	0.02	0.02	0.02	0.02
Vitamin- mineral mix	2	2	2	2
Total	100	100	100	100
Proximate Composition				
Moisture %	9.02±0.25	9.81±0.06	10.49±0.31	10.56±0.21
CP%	37.92±0.02	37.83±0.12	37.47±0.30	37.88±0.05
EE%	9.87±0.05	9.83±0.09	9.76±0.39	9.87±0.05
Ash%	2.64±0.03	2.75±0.05	2.92±0.32	2.68±0.05
GE (Kcal. $100g^{-1}$)	466.61±1.96	467.90±0.83	467.11±1.87	466.42±1.74
DE (Kcal. $100g^{-1}$)	342.0	379.1	428.5	448.6

All values are Mean ± SE, obtained from three replicates. CP (%) =Crude Protein; EE (%) =Ether Extract; GE=Gross Energy; DE= Digestible Energy; CMC=Carboxy methyl cellulose ;BHT=Butylated hydroxy toluene; SE=Standard Error.
Composition of Vitamin- mineral mix (PREMIX PLUS) (quantity. kg^{-1})
Vitamin A (55,00,000 IU); Vitamin D_3 (11,00,000 IU); Vitamin B_2 (2,000 mg); Vitamin E (750 mg); Vitamin K (1,000 mg); Vitamin B_6 (1,000 mg); Vitamin B_{12} (6 mcg); Calcium Pantothenate (2,500 mg); Nicotinamide (10 g); Choline Chloride (150 g); Mn (27,000 mg); I (1,000 mg); Fe (7,500 mg); Zn (5,000 mg); Cu (2,000 mg); Co (450) L-lysine(10g);DL-Methionine(10g);Selenium(125mg).

then placed in a pressure cooker for cooking/ steaming for half an hour. The pressure cooker was then removed from the flame and kept aside for cooling. The steamed dough was taken out and was cooled further. When the steamed dough was completely cooled, the calculated concentration of the oils, vitamins and minerals mixture were incorporated in it and mixed well. After incorporation of these elements, the dough was mixed properly and was pressed through a semi- automatic pelletizer(Uniextrude-S.B.Panchal and company,Mumbai,India) to get uniform sized pellets, which were spread on a sheet of paper and were initially sun dried. After that the feed was transferred to trays and were kept in a hot air oven overnight for complete drying at 50- 60 °C. After drying the pellets were packed in polythene bags and were sealed airtight and were labeled according to the treatments.

Fish Sampling and Proximate Analysis

At the end of feeding trial fishes were fasted for the 24 hr. and then weighed for calculating growth performance and nutrient utilization parameters like weight gain (%), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), whole body weight of 6 fishes per replicates.

Prior to chemical analysis, 6 fish from every replicate were taken and killed and were dried in hot air oven (100 °C±2) for 24 hr .The test diets and fish were ground into fine powder form with a pestle and mortar. The moisture content, crude protein, ether extract and ash content in the test diets and fish were analyzed following AOAC method (AOAC 1995).The gross energy content of test diets and fish were measured using a Parr oxygen bomb calorimeter (Parr- 6772, calorimetric thermometer, USA).

Calculation

Weight gain (%), specific growth rate (SGR), feed conversion ratio (FCR) and protein efficiency ratio (PER) of the cultured fishes were calculated as below:

$$\text{Weight gain (\%)} = \frac{\text{Final weight of fish} - \text{Initial weight of fish}}{\text{Initial weight of fish}} \times 100$$

$$\text{SGR} = \frac{\text{Log}_e\ \text{Final weight} - \text{Log}_e\ \text{Initial weight}}{\text{Number of days}} \times 100$$

$$\text{FCR} = \frac{\text{Feed given (Dry Weight basis)}}{\text{Body Weight Gain (Wet Weight basis)}}$$

$$\text{PER} = \frac{\text{Net Weight Gain (Wet Weight basis)}}{\text{Protein Fed}}$$

Statistical Analysis

The differences in the WG (%), SGR, FCR, PER and body composition (Moisture, Crude Protein, Ether extract, Ash) and gross energy of the test diets and fish were examined following 2×4 factorial ANOVA. Duncan’s multiple range test ($P<0.05$) was performed to examine the differences in the above variable among the different treatments. Energy requirement of *P. hypophthalmus*, juveniles was estimated by using second order polynomial regression (Zeitoun *et al*. 1976) model.

Results

Average Body Weight

The body weight of the experimental groups was recorded at the start and end of the experiment as shown in Table 2. The highest growth was observed in the T3 group at both the temperature i.e., at 32°C and at ambient temperature. Similarly, the lowest growth was observed in the T1 group at both temperatures.

Weight Gain (%)

The body weight gain was expressed in percentage to nullify the initial variation in body weight and is presented in Table 3. The two-way ANOVA analysis showed that temperatures and energy levels affected the WG (%) significantly ($p<0.05$) and there was a significant interaction between temperatures and energy levels when the diet with a different energy level fed at 32°C and at ambient temperature. The fish show the highest

Table 2. Average Body Weight (g) of *P. hypophthalmus* of different experimental group fed with different experimental diet at two temperatures

Temperature	Treatments	Initial Body Weight (g)	Final Body Weight (g)
32°C	T1	4.37±0.03	8.80±0.10
	T2	4.37±0.03	9.43±0.05
	T3	4.37±0.05	9.80±0.10
	T4	4.40 ±0.10	9.33±0.12
Ambient	T1	4.57±0.05	7.50±0.10
	T2	4.27±0.12	8.27±0.05
	T3	4.27±0.12	8.43±0.15
	T4	4.33±0.12	7.60±0.17

All values are Mean ± SE, obtained from three replicates.

Table 3. Growth performance and feed utilization of *P. hypophthalmus* Juveniles fed different experimental diets at two temperatures

Dietary energy (Kcal. 100 g^{-1})	Temperature	WG %	SGR %	FCR	PER
342.0	32°C	101.22^{c}	1.17^{c}	2.18^{c}	1.21^{c}
	Ambient	64.24^{f}	0.83^{f}	2.92^{a}	0.90^{e}
379.1	32°C	116.05^{b}	1.28^{b}	2.00^{cd}	1.32^{ab}
	Ambient	93.83^{d}	1.10^{d}	2.12^{cd}	1.25^{bc}
428.5	32°C	124.47^{a}	1.35^{a}	1.96^{d}	1.34^{ab}
	Ambient	97.69^{cd}	1.14^{cd}	1.94^{d}	1.35^{a}
448.6	32°C	112.16^{b}	1.25^{b}	2.00^{cd}	1.31^{ab}
	Ambient	75.43^{e}	0.94^{e}	2.61^{b}	1.01^{d}
SEM		2.08	0.02	0.06	0.03
ANOVA Table			p-value		
Dietary Energy		S	S	S	S
Temperature		S	S	S	S
Dietary Energy× Temperature		**S**	**S**	**S**	**S**

All values are Mean ± SEM, obtained from three replicates. Values in the same column with different superscript letters are significantly different (P< 0.05). SEM=Standard Error of Mean; ANOVA=Analysis of Variance; S= Significant.
WG (%) =Weight Gain; SGR (%)

weight gain (%) with the diet containing the digestible energy level 428.5 Kcal. 100 g^{-1}dietat 32°C as well as at ambient temperature. Similarly, the lowest weight gain (%) was observed with the diet containing the digestible energy level 342.0 Kcal. 100 g^{-1}diet at 32°C as well as at ambient temperature.

Specific Growth Rate (SGR)

The SGR of the different experimental groups is given in Table 3. The two-way ANOVA analysis showed that temperatures and energy levels affected the SGR %, significantly and there was a significant interaction between temperatures and energy levels when the diet with a different energy levels fed at 32°C and at ambient temperature. The fish show the highest weight gain (%) with the diet containing the digestible energy level 428.5 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature. Similarly, the lowest weight gain (%) was observed with the diet containing the digestible energy level 342.0 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature.

Feed Conversion Ratio (FCR)

The FCR values of the different experimental groups are presented in Table 3. The two-way ANOVA analysis showed that temperatures and energy levels affected the FCR significantly and there was a significant interaction between temperatures and energy levels when the diet with a different energy level fed at 32°C and at ambient temperature. The lowest FCR was observed with the diet containing the digestible energy level 428.5 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature, whereas it was highest at the diet containing the digestible energy level 342.0 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature.

Protein Efficiency Ratio (PER)

The PER of different treatment groups is given in Table 3. The two-way ANOVA analysis showed that temperatures and energy levels affected the PER significantly and there was significant interaction between temperatures and energy levels when the diet with a different energy levels fed at 32°C and at ambient temperature. The fish show the highest PER with the diet containing the digestible energy level 428.5 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature. Similarly, the lowest PER was observed with the diet containing the digestible energy level 342.0 Kcal. 100 g^{-1}diet at 32 °C as well as at ambient temperature.

Dietary Energy requirement for growth of *P.hypophthalmus*

The dietary energy requirement for optimum growth of *P.hypophthalmus*was calculated by using second order polynomial regression equation i.e.

$$y = -0.0001x^2 + 0.0805x - 15.022 \text{ (at ambient Temperature)}$$

$$y = -0.00004x^2 + 0.0325x - 5.2989 \text{ (at 32°C)}$$

Based on the above polynomial regression equation the Dietary Energy requirement for optimum growth of *P.hypophthalmus* was calculated and it was found to be 402.5Kcal. 100 g^{-1} (Figure 1) and 410 Kcal. 100 g^{-1}(Figure 2) at ambient temperature and at 32°C respectively.

Proximate Composition of Fish

Data pertaining to the carcass composition of all

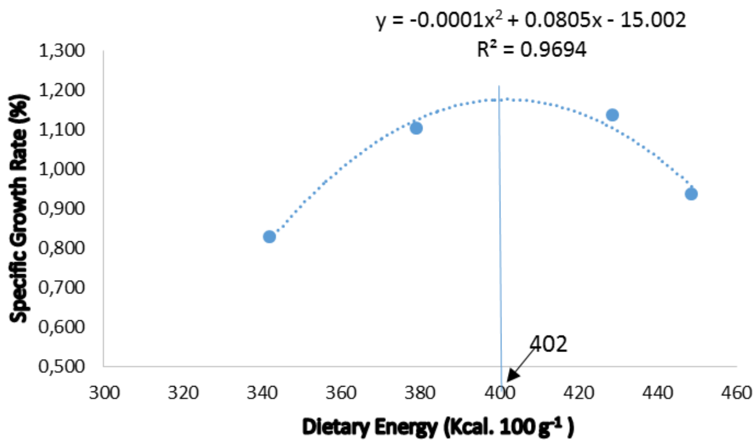

Figure 1. Dietary Energy (DE) requirement of *P. hypophthalmus* at ambient temperature.

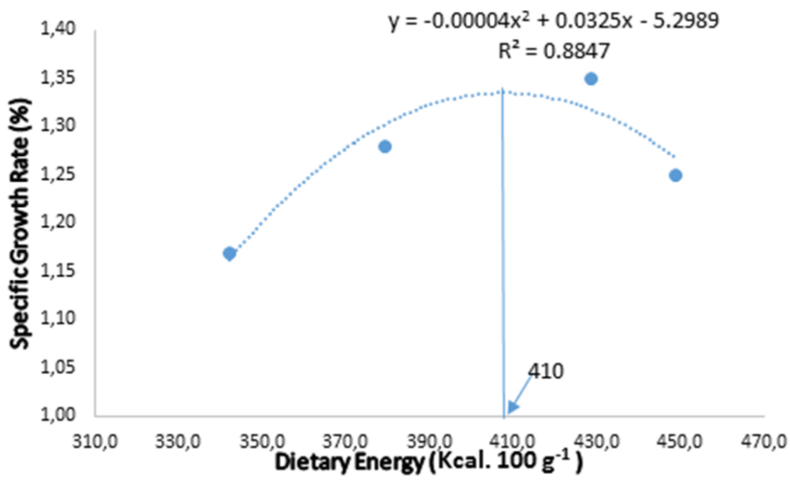

Figure 2. Dietary Energy (DE) requirement of *P. hypophthalmus* at 32°C.

the experimental animals in terms of moisture, ash, crude protein, ether extract, and gross energy content of carcass tissue at the end of the experiment are given in Table 4.The two-way ANOVA analysis showed that the carcass moisture(%),ash (%),CP (%) and GE content of *P.hypophthalmus*do not vary significantly ($p>0.05$) when fed the diet containing different level of dietary energy. Similarly, theinteraction of Dietary Energy and Temperature do not significantly affect the carcass composition as shown in Table 3. Moisture (%), CP (%), EE (%) and GE content of *P.hypophthalmus* varies significantly ($p<0.05$) whereas Ash (%) do not vary significantly ($p>0.05$) at two temperatures as shown in Table 4.

Discussion

Weight gain (%) and SGR of *P. hypophthalmus*, juveniles increased with dietary energy up to dietary energy content of 428.5 Kcal. 100 g^{-1}(T3) and decreased subsequently when a further increase in the dietary energy level in the diet (Table. 3). Phillips (1972) found that increase in weight gain/ growth rate up to a certain energy level and decreasing thereafter may be the result of an increase in the energy requirement of fish to get rid of excess toxic nitrogenous waste production due to increased rate of amino acid breakdown as temperature increases. Similar results were also found in studies carried out in other fishes like *Channa striata* fingerling (Samantaray andMohanty1997); black catfish, *Rhamdia quelen*, fingerlings (Meyer and Fracalossi 2004; Salhi *et al.*2004), red drum (McGoogan and Gatlin 1999). A positive relationship between energy requirement and the temperature was found by several researcher, i.e., as water temperature increases, the energy requirement of fish also increases within an optimum temperature range of water and decreases thereafter (Jobling 1994;De Silva andAnderson 1995; Bailey andAlanärä 2006,Person-Le Ruyet *et al.* 2006, Katersky and Carter 2007), which very well supports our finding.The possible reason for increased energy requirement at high temperature is due to the increased metabolic rate (Katersky and Carter 2007).

In the present study, the maximum value of FCR

Table 4. Proximate composition of whole body of *P. hypophthalmus* (% dry matter basis)

Dietary energy (Kcal. 100 g^{-1})	Temperature	Moisture (%)	ASH (%)	CP (%)	EE (%)	GE (Kcal. 100 g^{-1})
342.0	32°C	76.05^{ab}	16.59	51.68^{a}	20.69^{a}	587.53^{c}
	Ambient	76.38^{ab}	14.30	41.20^{c}	15.69^{c}	546.59^{e}
379.1	32°C	75.17^{b}	15.43	53.19^{a}	18.21^{b}	538.26^{ab}
	Ambient	77.68^{a}	13.71	43.78^{bc}	15.69^{c}	515.32^{bc}
428.5	32°C	74.97^{b}	13.77	52.03^{a}	16.28^{c}	544.67^{ab}
	Ambient	75.93^{ab}	13.99	48.01^{ab}	13.33^{d}	495.59^{a}
448.6	32°C	76.25^{b}	16.09	52.86^{a}	13.96^{d}	558.44^{ab}
	Ambient	75.43^{ab}	16.57	48.23^{ab}	11.07^{e}	519.70^{d}
SEM		2.08	0.879	1.724	0.475	19.86
ANOVA Table				p-value		
Dietary Energy		NS	NS	NS	S	NS
Temperature		S	NS	S	S	S
Dietary Energy× Temperature		NS	NS	NS	NS	NS

All values are Mean ± SEM, obtained from three replicates. Values in the same column with different superscript letters are significantly different (P< 0.05).
CP (%) =Crude Protein; EE (%) = Ether Extract; GE=Gross Energy; SEM=Standard Error of Mean; S=Significant; NS=Not Significant.

was found in a diet with the lowest level of dietary energy i.e., 342Kcal. 100 g^{-1}while the lowest FCR was observed at the energy level 428.5 Kcal. 100 g^{-1}.Fishes consume feed to meet their nutrient and energy requirement. It is believed that fishes adjust their food consumption to meet the metabolic energy demand and once metabolic energy demand are fulfilled theexcess energy is used for the growth of fish (De Silva and Anderson 1995). Fish adjust their feed intake function of dietary energy content. Excess energy in the feed reduces feed utilization that results the lower amount of protein and other essential nutrients available to the fishes (NRC 1993). Similar results were also found in studies carried out in different fishes such as in juvenile cobia, *Rachycentron canadum* (Sun *et al*.2006); snakehead, *Channa striata* fingerling (Samantaray and Mohanty1997); black catfish, *Rhamdia quelen* fry (Salhi *et al.* 2004); which is supported by the present results.Temperature also have a marked influence on the better feed efficiency.It is generally found that fish are able to eat maximum level of feed at a given temperature condition when feed supply are unlimited (Amin *et al*. 2014). When ration is restricted the best feed effieciency is found at lower temperature but as feed supply is increased the best feed effieciency is found at higher temperature(Jobling 1997) which is in agreement with our results.Other probable explanation for improved feed efficiency at higher temperature might be the increase in feed intake of the fish with increased water temperature , which resulted in better growth of fish, leading to better FCR at higher temperature.

The quality and utilization of protein in a diet is evaluated by the protein efficiency ratio (PER). The PER of *P. hypophthalmus* in the present study increased linearly up to T3 (Dietary Energy level, 428.5 Kcal. 100 g^{-1}) and then again start decreasing when the energy content in the diet further increased. Similar results were supported by Lee *et al*. (2002); Steffens (1981) and De Silva *et al*. (1991) who find that an increase in dietary energy content of feed resulted in higher protein efficiency ratios which indicate a lower use of protein as an energy source when non-protein energy was increased in the diet.Temperature also has marked effect on PER of fish. In present study, it is observed that PER increases as temperature of the water increase, but it decreases again when the energy content of diet was maximum.Our finding is consistent with the finding Degani *et al*. (1989). The possible explanation for the above results is that excess energy in the feed may cause a lower nutrient utilization by the fish, due to lower feed intake (Bromley 1980; Metailler *et al*. 1981; Alsted and Jokumsen 1989; Chou and Shiau 1996).

In the present study, the dietary energy requirement of *P. hypophthalmus*, juveniles were optimized, using the “Polynomial regression” model. The polynomial regression analysis is used to establish the interrelationship of growth performance and essential nutrient intake as recommended by Zeitoun *et al.* (1976). Shearer (2000) reported that second order polynomial regression is much moreappropriate for estimation of optimal dietary nutrient level in dose-response experiments. He also emphasized that physiological responses of an organism to the increase of a limiting dietary nutrient are not broken at one particular point and hence non-linear model like Polynomial regression method is most appropriate to find out the optimal nutrient requirement of the animal. In the present study, the relationship between specific growth rate (SGR) and dietary energy content of the diet was established by using second order polynomial regression. Based on the second order polynomial regression equation the Dietary Energy requirement for optimum growth of *P.hypophthalmus* was found to be 402.5 Kcal. 100 g^{-}

[1](Figure 1) and 410 Kcal. 100 g^{-1}(Figure 2) at ambient temperature and at 32°C respectively. Energy requirement in the similar range was reported for catfish by Guillaume *et al*., (2001), *Pangasius hypophthalmus* by Hung *et al*. (2002), Channel Catfish by Gatlin *et al*. (1986), Rainbow Trout by Mustapha *et al.* (2012).

In the present study Moisture(%), Ash(%), CP (%) and gross energy content of the different treatment group containing different energy level and at the interaction of energy level and temperature did not vary significantly ($p>0.05$), however, all the above parameters were significantly varied at different temperature except the Ash (%). Similar reports were supported by Fang *et al*. (2010) in *Cynoglossus semilaevis* and Sun andChen (2014) in *Rachycentron canadum.*The ether extract (EE%) in fish carcass varies significantly in different temperature and at different energy level.However, dietary interaction of temperture and energy donot significantly affect the ether extract (EE%) of P. *hypophthalmus*. The possible reason for lower body lipid accumulation at higher temperature and high energy level as found in present study is that as water temperature increases maintenance energy requirement of fish increases but in response to higher temperture fish are unable to consume enough food (energy) to accumulate body lipid.Similar findings were supported by Desai and Singh (2009) in *Clarias batrachus* fry and by Phumee *et al*. (2009) in *Pangasius hypophthalmus* fry.

Conclusion

Increase in water temperature sinficantly affects the growth and nutrient utilization of *P. hypophthalmus* that in turn affects the energy requirement of fish.In present study the dietary energy requirement of *P. hypophthalmus* was found to be 410 Kcal. 100 $g^{-1}$and 402.5 Kcal. 100 g^{-1}at 32°C and at ambient temperature (24.5°C), respectively.Hence it can be concluded that increase in temperature affects the energy requirement of *P. hypophthalmus.*

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