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Effects of dietary protein-energy level on the survival, growth and body composition of tinfoil barb, *Barbonymus schwanenfeldii* fry

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Abstract

An 8-week feeding trial was conducted to study the effects of dietary protein-energy level on the survival, growth, and body composition of tinfoil barb, *Barbonymus schwanenfeldii* fry. Fry (0.70±0.02 g) were randomly stocked in 60 L glass aquaria at 25 fish per aquarium. Six test diets were formulated to contain 40, 45, and 50 % protein with 17 and 18 kJ g^{-1} gross energy. Each diet was randomly assigned to three replicate aquaria, and fish were fed twice a day until satiation. Fish survival was not affected by the dietary protein, energy level, and their interaction. The results showed that higher dietary energy did not have significant effects on weight gain, specific growth rate, and feed efficiency of tinfoil barb. However, dietary protein had significant effects on fish growth and feed efficiency, while its interaction with energy had significant effect on growth. All dietary fed groups did not have histopathological changes in the liver and intestine. Fish fed 50% protein and 17 kJ g^{-1} gross energy showed significantly highest ($p<0.05$) specific growth rate compared to fish fed with 40% protein and 17-18 kJ g^{-1} gross energy.

Keywords: Tinfoil barb, *Barbonymus schwanenfeldii*, Protein, Energy, Growth performance, Histology

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Introduction

The demand for fish has increased steadily over the years because it offers high quality of protein, fatty acids, minerals and vitamins for human and animal consumption (Zhang *et al*., 2020; Yamamoto *et al*., 2021). Fortunately, the aquaculture sector has continued to expand globally especially in Asia to meet the increasing demand while the capture fisheries have reached their maximal limits (Tacon., 2020). In aquaculture, feed cost makes up more than 50% of the total operational cost (Liu *et al*., 2021). Thus, it is crucial to reduce feed costs while providing optimal nutrients to the fish for best feed utilization. Feed utilization and growth of aquatic animals can be maintained by balancing protein and energy in the diet (Zheng *et al*., 2020). Protein is costly but remains very important for fish growth (Zhang *et al*., 2017; Zheng *et al*., 2020). However, locally available plant proteins can be viable alternatives for fishmeal and fish oil replacement in extruded diets (Ljubojevic *et al*., 2015; Cruz *et al*., 2018). Meanwhile, fish tends to regulate feed intake to meet its energy requirements.

Sustainable aquaculture is highly dependent on the success of providing the optimal nutrient requirements for the maximal growth and best health of fish. Besides growth and health, a proper balance of protein and energy in diets will give better feed efficiency, enhance protein utilization, reduce excessive lipid and glycogen in somatic tissues and liver, and improve the water quality of the environment (Wang *et al*., 2013). Increasing dietary energy has been found to spare and optimize protein and limit ammonia excretion in several species (Kim *et al*., 2012) by utilizing non-protein energy sources (Shoaib *et al*., 2020; Deng *et al*., 2011; Satpathy *et al*., 2003). Lipid as protein-sparing is well established in nutrition, especially in aquaculture (Kim *et al*., 2012). Jiang *et al*. (2015) stated that adequate energy provided from the dietary lipid could decrease the use of protein as an energy source. Other than sources of energy, lipid provides essential fatty acids (EFA) for growth and development (Ahmad, 2008).

Tinfoil barb (*Barbonymus schwanenfeldii*), is an omnivorous carp that has long been an important species in the global multibillion-dollar ornamental fish industry. It was previously sidelined as a food fish but is now accepted as a common protein source with a highly nutritious meat (Jaffar *et al*., 2019; Karim *et al*., 2021). This species can be found in many Asian countries such as Malaysia, Brunei, Cambodia, Indonesia, Laos, Thailand, Singapore, and Vietnam (Sabarudin *et al*., 2017; Dahruddin *et al*., 2021; Nafees *et al*., 2022). Tinfoil barb is economically suitable for aquaculture despite its slow growth (Eslamloo *et al*., 2017; Dewantoro *et al*., 2018). Despite its commercial importance in both ornamental fish and aquaculture industry, no commercial formulated feed is available for tinfoil barb due to a lack of studies on its nutrient requirements. In this study, the dietary effects of dietary protein-energy level on the growth performance of tinfoil barb were evaluated.

Materials and methods

Experimental animals, acclimation and experimental set up

Tinfoil barb fry were supplied by the Bukit Tinggi Aquaculture Extension Centre, Bentong, Pahang, Malaysia. The fish were acclimatized in a 1 tonne fiberglass tank for two weeks and fed a commercial tilapia diet (Dindings, 32% crude protein). After the acclimation period, 450 fish (0.70±0.02 g) were randomly and equally stocked into glass aquaria (25 fish aquarium^{-1}) filled with 60 L water. Each aquarium was fitted with an individual recirculation system composed of a mechanical filter, a biofilter and a water pump. Adequate aeration was also provided through individual air stones. Each aquarium top was covered with a net to prevent the fish from jumping out. The feeding trial was conducted at the Wet Laboratory, Department of Aquaculture, Faculty of Agriculture, Universiti Putra Malaysia for eight weeks.

Test diets and feeding

Six test diets which utilized menhaden fishmeal and soybean meal as protein sources was formulated to contain 40, 45, and 50 % crude protein with 17 and 18 kJ g^{-1} gross energy (Table 1). The diets were processed using a single screw extruder and 2 mm Ø die (Brabender KE 19, Germany). The pellets were oven-dried at 50°C for about 12 h. The test diets were coarsely ground into 1 mm crumbles and packed into air-tight containers with silica gel. Test diets were randomly assigned to the aquaria in triplicates. Fish were fed twice a day (0800h and 1700h) until satiation. Any dead fish was recorded and removed.

Sampling

At the end of the 8-week feeding trial, all fish were counted and the body weight of fish was recorded. Simultaneously three fish were sampled randomly from each tank, anesthetized, sacrificed and dissected for liver and viscera to evaluate the hepatosomatic index (HSI) and viscerosomatic index (VSI). The intestine and liver were then immediately preserved in 10% buffered formalin for histological analysis. All remaining fish were also collected from each tank for the whole body analysis.

Water quality

Water quality parameters such as water temperature, dissolved oxygen, pH, and total were measured using a multiprobe meter (YSI 556 MPS, USA), and 50% of the culture water was changed every week. Mean water temperature, pH, and dissolved oxygen during the trial were 27.6±1.2°C, 6.7±0.6, and 5.9±0.5 mg L^{-1}, respectively, while total ammonia ranged from 0.03-1.13 mg L^{-1}.

Proximate analysis

All diets and whole fish carcass (initial and final) were analyzed according to the respective AOAC (1990) methods for crude protein, lipid, crude fibre, ash, moisture, and gross energy. Moisture was analyzed by oven-drying (60°C) until constant weight.

Table 1: Feed ingredients and proximate composition (% as fed basis) of the experimental diets.

Ingredient	**17 kJ g^{-1} GE**			**18 kJ g^{-1} GE**		
	40% Protein	**45% Protein**	**50% Protein**	**40% Protein**	**45% Protein**	**50% Protein**
Fishmeal[a]	15.89	32.28	52.71	22.39	38.79	55.18
Soybean meal	64.34	49.28	27.74	53.87	38.81	23.75
Tapioca starch	15	15	15	15	15	15
Crude palm oil	2.78	1.44	1	6.74	5.4	4.07
Vitamin premix[b]	1	1	1	1	1	1
Mineral premix[c]	1	1	1	1	1	1
Cellulose[d]	0	0	1.56	0	0	0
Proximate Composition						
Moisture	11.78±0.11	11.20±0.11	10.36±0.11	11.47±0.11	11.35±0.11	11.93±0.11
Crude protein	41.64±0.39	47.18±0.39	52.41±0.39	42.32±0.39	47.18±0.39	51.90±0.39
Crude lipid	5.90±0.22	5.80±0.22	6.82±0.22	10.85±0.22	10.43±0.22	9.66±0.22
Crude fiber	2.57±0.05	2.38±0.05	2.10±0.05	2.37±0.05	2.14±0.05	1.94±0.05
Nitrogen free extract	31.02±0.38	23.42±0.38	16.30±0.38	24.76±0.38	19.47±0.38	13.41±0.38
Ash	7.09±0.27	10.03±0.27	12.00±0.27	9.43±0.27	9.66±0.27	11.16±0.27
Gross energy (kJ g^{-1})	17.19±0.03	17.27±0.03	17.32±0.03	18.23±0.03	18.27±0.03	18.28±0.03

[a] Menhaden fish meal (62% crude protein)

[b] Vitamin premix (g kg-1 premix): ascorbic acid, 45; myo-inositol, 5; choline chloride, 75; niacin, 4.5; riboflavin, 1; pyridoxine, 1; thiamin mononitrate, 0.9; Ca-pantothenate, 3; retinyl acetate, 0.6; cholecalciferol, 0.08; vitamin K menadione, 1.7; a-tocopheryl acetate (500 IU g-1), 8; biotin, 0.02; folic acid, 0.1; vitamin B12, 0.001; cellulose, 845.1.

[c] Mineral premix (g kg-1 premix): KCl, 90; KI, 0.04; Ca(H2PO4).H2O, 500; NaCl, 40; CuSO4.5H2O, 3; ZnSO4.7H2O, 4; CoSO4, 0.02; FeSO47H2O, 20; MnSO4.H2O, 3; CaCO3, 215; MgOH, 124; Na2SeO3, 0.03; NaF, 1.

[d] Cellulose: Sigma Aldrich Co., St. Louis, MO, USA

After an acid digestion, the crude protein was evaluated using the Kjeldahl method (Foss KjeltecTM 2300 analyzer Unit Foss Tecator, Sweden). Crude lipid was determined by Soxhlet ether extraction (Foss Soxtec™ 8000 System, Sweden) and ash analysis determined by using a muffle furnace (Carbolite CWF 1100, England) which samples were combusted at 600°C for 5 h. Gross energy was determined using a bom calorimeter (Leco AC 350, USA).

Histological analysis

After 24 h, extracted livers and intestines preserved in 10% buffered formalin were then preserved in 70% ethanol until tissue processing. An automatic tissue processor (Leica TP1020, USA) was used for the tissue processing. Fish tissue sections were placed into tissue cassettes for dehydration and later embedded in paraffin blocks subsequently cut (5 µ thickness) and mounted on glass slides. Each tissue sample was then dewaxed and stained with hematoxylin and eosin solution using standard paraffin-embedding procedure. After the embedding process, slides were examined under a microscope (Leica DM750, Germany) supported with Dino-Eye Microscope Eyepiece Camera (Dino-lite AM7025X, USA).

Statistical analysis

After a homogeneity test, data on survival, growth performance, and feed utilization were analyzed by two-way analysis of variance (ANOVA) analysis using IBM SPSS Statistic 20 (SPSS Inc., USA). Differences among means were analyzed using Tukey test at $p<0.05$. All percentage data were arcsine transformed prior to further analysis.

Results

Table 2 shows survival, growth, feed intake and efficiency, and body indices of tinfoil barb fry fed various protein-energy diets. Significant interactions between protein and energy levels were found in weight gain, SGR, and VSI of tinfoil barb fry. All parameters were not affected by dietary energy level while dietary protein affected almost all parameters except survival, CF, and HSI. Best growth performance and feed efficiency were found when fish were fed 50% protein and 17 kJ g^{-1} gross energy but these values were not significantly different from those fed 45-50 % protein at 18 kJ g^{-1} gross energy or those fed 45% protein at 17 kJ g^{-1} gross energy.

Interaction of protein and energy had no significant effect on the fish body composition. Dietary protein level had no significant effects on whole body composition of tinfoil barb except ash, while energy had only significant effects on body lipid and energy (Table 3). Body lipid and energy were significantly higher ($p<0.05$) at the higher dietary energy while body ash was higher with the highest dietary protein.

Nutrient retention of tinfoil barb is shown in Table 4. No significant effects of protein, energy, and interaction were observed on the nutrient retention except for lipid where the retention was significantly higher ($p<0.05$) at the lower dietary energy.

Figure 1 showed the liver sections of tinfoil barb fry fed various protein-energy diets. The vacuole areas of liver sections were quantified by the image analysis software and showed no significant differences in tinfoil barb fry liver from various protein-energy diets.

Figure 2 shows the intestinal sections of tinfoil barb fry at the end of the feeding trial. The parameter of intestines and length of villi were normal in all treatments. The histological parameters of intestine diameter, lumen diameter, intestine wall width, villi height, and villi width were estimated (Table 5). Intestine wall width was significantly higher ($p<0.05$) at the higher energy level. However, no treatment differences were noted in the histological cross-section of the intestine after eight weeks when fish were fed with different dietary protein and energy diets.

Discussion

Dietary protein to energy ratio plays a vital role in fish nutrition as it affects fish growth performance and feed efficiency (Yang *et al*., 2016). Apart from protein, an adequate amount of energy needs to be precisely determined to balance fish growth (Zhang *et al*., 2017). In addition, excess energy may affect the feed consumption of the fish (Mohanta *et al*., 2009).

Table 2: Growth performance and feed utilization of tinfoil barb fry fed with various protein-energy diets for 8 weeks.

Protein (%)	Gross Energy (kJ g^{-1})	Survival (%)	Final weight (g)	WG (%)	SGR (% d^{-1})	DFI (% d^{-1})	FCR	PER	CF	HSI	VSI
40	17	96	2.66^{c}	293.58^{b}	2.47^{b}	3.73	1.96	1.34	1.44	1.22	10.15^{a}
45	17	98.67	3.55^{abc}	412.56^{ab}	2.97^{ab}	3.90	1.36	1.85	1.46	1.34	10.11^{a}
50	17	98.67	4.30^{a}	526.99^{a}	3.34^{a}	4.74	1.32	1.90	1.42	1.18	9.63^{a}
40	18	100	3.12^{bc}	345.58^{b}	2.71^{b}	3.96	1.63	1.53	1.49	1.41	10.12^{a}
45	18	100	3.59^{ab}	404.35^{ab}	2.93^{ab}	4.13	1.45	1.73	1.48	1.39	10.02^{ab}
50	18	98.67	3.28^{bc}	395.27^{ab}	2.90^{ab}	4.42	1.57	1.60	1.43	1.36	10.02^{ab}
Pooled SE		1.89	0.19	30.35	0.12	0.23	0.15	0.11	0.04	0.06	0.09
Mean Protein (%)											
40		98.00	2.89^{B}	319.58^{B}	2.59^{B}	3.80^{B}	1.80^{A}	1.44^{B}	1.47	1.31	10.14^{A}
45		99.33	3.57^{A}	408.46^{B}	2.95^{A}	4.02^{AB}	1.41^{AB}	1.79^{A}	1.47	1.37	10.06^{A}
50		98.67	3.79^{A}	461.13^{A}	3.12^{A}	4.58^{A}	1.44^{B}	1.75^{A}	1.43	1.27	9.82^{B}
Mean Energy (kJ g^{-1})											
17		97.78	3.50	411.05	2.93	4.13	1.55	1.70	1.44	1.25	9.96
18		99.56	3.33	381.73	2.85	4.17	1.55	1.62	1.47	1.39	10.05
Probability											
Protein		0.78	0.001	0.002	0.003	0.017	0.037	0.013	0.39	0.29	0.009
Energy		0.27	0.30	0.26	0.45	0.82	0.96	0.42	0.39	0.01	0.23
Protein*Energy		0.57	0.006	0.03	0.049	0.40	0.16	0.11	0.81	0.40	0.035

Weight gain (WG) = $\frac{\text{Final Weight-Initial Weight}}{\text{Initial Weight}} \times 100$, Specific growth rate (SGR) = $\frac{\ln \text{Final Weight-} \ln \text{Initial Weight}}{\text{Days}} \times 100$, Daily feed intake (DFI) = $\frac{\text{Total Feed}}{[0.5(\text{Initial Weight+Final Weight}) \times \text{Days}]} \times 100$,

Feed conversion ratio (FCR)= $\frac{\text{Total Feed}}{\text{Final Weight-Initial Weight}}$, Protein efficiency ratio (PER) = $\frac{\text{Final Weight-Initial Weight}}{\text{Total Protein Fed}}$,

Condition factor (CF) $= \frac{\text{Body weightt}}{(\text{Total length})3} \text{ X } 100$, HSI $==\frac{\text{Liver Weight}}{\text{Total body weight}} \text{ X } 100$,

VSI $= \frac{\text{Viscera weight}}{\text{Total body weight}} \text{ X } 100$

Means (n = 3) within a column and followed by a same letter are not significantly different ($p>0.05$).

Table 3: Whole body proximate composition (% wet weight basis) of tinfoil barb fry fed with various protein-energy diets for 8 weeks

Protein (%)	Gross Energy (kJ g^{-1})	Moisture	Protein	Lipid	Fibre	Ash	NFE	Energy (kJ g^{-1})
Initial		76.70	13.03	4.38	0.27	2.91	2.72	4.99
40	17	69.56	17.99	9.31	0.22	2.28	0.64	7.56
45	17	67.18	19.75	9.75	0.29	2.54	0.50	8.05
50	17	67.55	17.76	9.68	0.08	2.97	1.97	7.91
40	18	65.35	19.12	11.81	0.30	2.56	0.86	9.01
45	18	66.18	17.68	11.04	0.14	2.66	2.30	8.76
50	18	65.78	18.37	11.39	0.26	2.94	1.27	8.72

Protein (%)	Gross Energy (kJ g^{-1})	Moisture	Protein	Lipid	Fibre	Ash	NFE	Energy (kJ g^{-1})
Pooled SE		1.80	0.86	0.59	0.13	0.16	0.82	0.45
Mean Protein (%)								
40		67.46	18.55	10.56	0.26	2.42^{B}	0.75	8.29
45		66.68	18.72	10.40	0.22	2.60^{AB}	1.40	8.40
50		66.67	18.06	10.53	0.17	2.96^{A}	1.62	8.32
Mean Energy (kJ g^{-1})								
17		68.10	18.50	9.58^{B}	0.20	2.60	1.04	7.84^{B}
18		65.77	18.39	11.41^{A}	0.23	2.72	1.48	8.83^{A}
Probability								
Protein		0.88	0.74	0.96	0.79	0.014	0.56	0.96
Energy		0.14	0.88	0.003	0.77	0.35	0.52	0.019
Protein*Energy		0.66	0.18	0.59	0.47	0.63	0.34	0.68

Means (n = 3) within a column and followed by a same letter are not significantly different (*p*>0.05).
NFE calculated as 100% - (moisture + protein + lipid + ash + crude fiber)

Table 4: Nutrient retention (%) of tinfoil barb fry fed with various protein-energy diets for 8 weeks.

Protein (%)	Gross Energy (kJ g^{-1})	Protein	Lipid	Carbo-hydrate	Energy
40	17	25.29	99.52	0.39	26.02
45	17	33.73	141.86	0.59	37.89
50	17	27.16	119.40	6.63	37.47
40	18	30.35	79.34	1.19	36.44
45	18	27.56	84.33	5.99	38.99
50	18	22.31	119.40	3.30	33.27
Pooled SEM		2.60	11.27	2.93	3.54
Mean Protein (%)					
40		27.82	89.43	0.79	31.23
45		30.65	113.10	3.29	38.44
50		24.74	100.55	4.97	35.37
Mean Energy (kJ g^{-1})					
17		28.73	120.26^{A}	2.54	33.79
18		26.74	81.79^{B}	3.49	36.23
Probability					
Protein		0.12	0.15	0.39	0.17
Energy		0.37	0.001	0.70	0.42
Protein*Energy		0.10	0.29	0.36	0.16

Means (n=3) within a column and followed by a same letter are not significantly different (*p*>0.05).

Table 5: Effects of dietary protein and energy in the morphometric parameters of distal intestine of tinfoil barb fry

Protein (%)	Gross Energy (KJ g^{-1})	Vacuole Area/ Total Area (%)	Villi Height (mm)	Villi Width (mm)	Intestine Wall Width (mm)	Intestine diameter (mm)	Lumen diameter (mm)
40	17	44.25	0.21	0.06	0.03	0.91	0.43
45	17	48.47	0.25	0.08	0.05	1.11	0.51
50	17	43.07	0.24	0.07	0.04	1.18	0.53
40	18	47.07	0.29	0.07	0.06	1.07	0.50

Protein (%)	**Gross Energy (KJ g^{-1})**	**Vacuole Area/ Total Area (%)**	**Villi Height (mm)**	**Villi Width (mm)**	**Intestine Wall Width (mm)**	**Intestine diameter (mm)**	**Lumen diameter (mm)**
45	18	47.35	0.27	0.07	0.06	1.15	0.49
50	18	47.01	0.23	0.07	0.05	1.30	0.60
Pooled SEM		1.15	0.02	0.01	0.00	0.14	0.07
Mean Protein (%)							
40		45.66	0.25	0.06	0.99	0.99	0.47
45		47.91	0.26	0.08	1.13	1.13	0.50
50		45.04	0.24	0.07	1.24	1.24	0.56
Mean Energy (kJ g^{-1})							
17		45.26	0.23	0.07	1.07^{B}	1.07	0.49
18		47.14	0.26	0.07	1.17^{A}	1.17	0.53
Probability							
Protein		0.07	0.66	0.32	0.18	0.26	0.43
Energy		0.07	0.20	0.78	0.004	0.38	0.51
Protein*Energy		0.11	0.19	0.54	0.25	0.90	0.74

Means (n=3) within a column and followed by a same letter are not significantly different ($p>0.05$).

In the present study, there was a significant interaction between protein and energy on the growth performance of tinfoil barb fry. Weight gain and SGR of fish fed with 50% with 17 kJ g^{-1} GE were significantly higher than other fish groups except those fed with 45% protein at 17 kJ g^{-1} GE and 45-50 % protein at 18 kJ g^{-1} GE. SGR of tinfoil barb in this study was within the range 1.48-1.72% % d^{-1}) reported by (Nafees *et al*., 2022) and higher than those reported for Malaysian Mahseer (Misieng *et al*., 2011).

Higher dietary energy slightly decreased growth performance of tinfoil barb despite a slightly higher feed intake. Similar patterns had been reported in silver barb, *Barbonymus gonionotus* (Mohanta *et al*., 2009), *Channa argus* (Sagada *et al*., 2017), hybrid snakehead *Channa maculata* ♀ × *Channa argus* ♂ (Zhang *et al*., 2017), *Pseudobagrus ussuriensis* (Wang *et al*., 2013) and *Rhamdia quelen* (Meyer and Fracalossi, 2004). In contrast, high energy diet improves the growth performance of *Clarias gariepinus* (Ali and Jauncey, 2016) and flounder *Paralichthys olivaceus* (Lee *et al*., 2000). These findings illustrated that the protein and gross energy ratio are species-specific, and the best protein and gross energy levels for tinfoil barb were 50% and 17 kJ g^{-1}, respectively. In addition, when dietary 50% crude protein was lowered to 45% at 17 kJ $g^{-1,}$ a substantial decrease by approximately 30% in weight gain was observed even though it was not significantly different. This suggested that tinfoil barb fry required a minimum of 45% crude protein in its diet. Dewantoro *et al*. (2018) demonstrated that tinfoil barb can still have a high survival (100%) but with a very poor growth when fed 25% protein. In the present study, all dietary treatments with 18 kJ g^{-1} GE showed a reduction in weight gain numerically. This indicated that a high lipid content may impair the growth performance of tinfoil barb. A dietary lipid of more than 8% suppresses the growth of tinfoil barb (Sulaiman *et al*., 2020) and hybrid lemon fin barb (Ismail *et al*., 2013) while lipid of more than 10% reduces the growth among Malaysian Mahseer (Ramezani-Fard *et al*., 2012).

In the present study, the survival of the barb was not significantly affected by the dietary protein and energy levels. Similar reports have been made by Yan *et al*. (2017) and Kim and Lee (2005) on juvenile loach, *Misgurnus anguillicaudatus* and bagrid catfish, *Pseudobagrus fulvidraco,* respectively. Daily feed intake and FCR were also not affected by both dietary protein, energy and interaction. Nevertheless, similar trends were found in freshwater angelfish, *Pterophyllum scalare* (Alexandre *et al*., 2009) and *Siniperca scherzeri* (Sankian *et al*., 2017). Regardless of energy level, the highest DFI and the best FCR were noted in dietary 50% protein but these were not significantly different for those fed 45% protein.

Inadequate dietary energy level in feed will lead to the catabolization of protein for energy rather than growth. However, excess of energy and low protein will reduce feed intake, which will badly affect fish growth (Wang *et al*., 2013). In this study, PER was not affected by the interaction between dietary protein and energy. Generally, the better the protein quality, the better is the PER value (Liu *et al*., 2021). PER of tinfoil barb showed significant differences with varying protein levels. When fish fed with a higher protein, PER improved. This trend can also be found in Japanese eel, *Anguilla japonica* (Okorie *et al*., 2007) and juvenile rockfish, *Sebastes schlegeli* (Cho *et al*., 2015).

The condition factor (K) indicates the level of energy reserve and fish health (Nayak *et al*., 2018). The present study showed no interaction between dietary protein and energy on K value of tinfoil barb. Similar trends had been reported in fancy carp, *Cyprinus carpio* var. *koi* (Choi *et al*., 2015) and northern snakehead fish *Channa argus* (Sagada *et al*., 2017). Meanwhile, HSI and VSI can be indicators of nutrient utilization and fish health (Shoaib *et al*., 2020) and are primarily influenced by body lipid deposition (Sagada *et al*., 2017). If the lipid in the carcass body is higher, it will cause the fat deposition in fish and affect the flesh quality. It is important to balance the content of protein and lipid in diets in order to offer the best of flesh quality of fish with high protein and low in fat (Wang *et al*., 2013). In this study, VSI and HSI increased when dietary energy level was increased similar to trends found in *C. argus* (Sagada *et al*., 2017) and *Silurus meridionalis* (Liu *et al*., 2013).

The final body composition of fish is influenced by the nutrient composition of its feed (Ishak *et al*., 2016; Yamamoto *et al*., 2020). In this study, the interaction of dietary protein and energy did not affect the whole-body nutrient deposition of tinfoil barb except for body lipid and ash, and energy. Significantly higher body lipid and energy were observed when the fish was fed a higher energy diet while body ash was significantly lower at the lowest dietary protein level. Similar trends were reported in *Megalobrama amblycephala* (Li *et al*., 2010), *Silurus asotus* (Kim *et al*., 2012), *Pseudobagrus ussuriensis* (Wang *et al*., 2013) and *Barbonymus gonionotus* (Nayak *et al*., 2018). In contrast, body compositions of *Siniperca scherzeri* (Sankian *et al*., 2017) and Japanese eel, *Anguilla japonica* (Okorie *et al*., 2007) were not affected by dietary protein-energy level.

Karalazos *et al*. (2011) demonstrated positive effects of protein sparing when an increase of the dietary lipid content improves protein retention. However, the interaction of dietary protein and energy did not affect the protein, lipid, carbohydrate and energy retention of tinfoil barb. The retention percentages were quite similar to the ranges reported by (Nafees *et al*., 2022). A recent study with largemouth bass, *Micropterus salmoides* (Li *et al*., 2020) showed similar results where protein and energy retention are not affected. However, a higher lipid retention occurs when the fish are fed higher energy diets. Similar results have also been found in Pacific threadfin, *Polydactylus sexfilis* (Deng *et al*., 2011), and blackspot seabream, *Pagellus bogaraveo* (Figueiredo-Silva *et al*., 2010).

Pathology alteration is one of the histology part to determine the response of animals to a nutritional source or treatment (Hu *et al*., 2013). The absorption efficiency of digested nutrients can be assessed through the histology of fish intestine (Martínez-Llorens *et al*., 2021). The abnormalities of intestine structure are important to be understood as it can affect the nutrient absorption. A decrease in the absorption can be due to poor nutrient utilization that leads to an increased FCR (Refaey *et al*., 2018). However, in this study, intestine remained normal for all treatments when fish fed with different levels of protein and energy. Similar results had been reported in a recent study with shi drum, *Umbrina cirrose* (Kokou *et al*., 2019). In the present study, liver remained normal for all fish groups. In contrast, Kowalska *et al*. (2011) found larger lipid vacuoles in the liver of pikeperch, *Sander lucioperca* fed with high lipid diets. Meanwhile, the liver of largemouth bass, *Micropterus salmoides* has smaller lipid vacuoles when fed with 50% protein and 7.5-10.0 % lipids (Li *et al*., 2020). In this study, the growth performance decreased with high energy levels.

Conclusion

The best growth of tinfoil barb was achieved when fed 50% dietary protein at 17 kJ g^{-1} GE. The results also indicated that its minimum dietary protein requirement of 45% at 17-18 kJ g^{-1} GE.

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Conflict of interest

The authors report no declarations of interest.

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