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OPTIMAL DIETARY LIPID LEVEL FOR THE GROWTH OF TIRE TRACK EEL (*MASTACEMBELUS FAVUS*) FINGERLING

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ABSTRACT

A growth experiment was conducted to determine the optimal dietary lipid level of tire track eel (Mastacembelus favus) fingerlings. Five iso-energetic 20 kJ/g and iso-nitrogenous 43% purified diets containing 1.5% - 13.5% lipid (squid oil/vegetable oil) in 3% increments were fed for 90 days to triplicate groups of tire track eel in a recirculated filtered rearing system. The results showed that weight gains were higher in the fish fed the 7.5% and 13.5% lipid diets, followed by the 4.5% lipid diet and lowest in fish fed the 1.5% lipid diet. Protein efficiency ratio (PER) in fish generally showed the same pattern of weight gains. Feed conversion ratios (FCR) were better (P < 0.05) for fish fed the lipid diets over 7.5% lipid than those fed 4.5% and 1.5% lipid diets. Body lipid content was highest (P < 0.05) in fish fed the 13.5% lipid diet, followed by those fed the 10.5% and 7.5% lipid diets, and lowest in fish fed the 4.5% and 1.5% lipid diets. The trend of the body indices was not definite regarding to dietary lipid content.
Second-order regression of weight gains on concentrations of dietary lipid containing 43% protein indicated that the optimal dietary lipid level resulting in maximal growth of tire track eel fingerling was about 8.5%. Based on the results, 7.5% of dietary lipid containing 43% protein appears to meet the minimal requirement for this species.

1. INTRODUCTION

Tire track eel (*Mastacembelus favus*) is a species with high economic value due to its delicious meat quality (Gupta &Banerjee, 2016). Thus, there is an increasing consumers' demand which, in turn, results in a need to expand commercial farming of this species. To extend the farming scale of this species, designed feed containing all the essential nutrients needed to keep the fish healthy and growing is necessary. The fish need to be fed diets producing energy and essential nutrients for their maintenance, movement, normal metabolic functions, and growth.

In recent years, there have been several studies on biological characteristics (Trieu, 2009; Trung, 2010), maturation and artificial reproduction (Loan, Anh, Vinh, Tung, Linh, Ngoc and cs., 2010; Trung, Anh and Thanh, 2010; Diem, 2009) as well as genetic diversity (Jamaluddin, So, Tam, Ahmad, Grudpan, Md Sah and cs., 2021; Jamaluddin, So, Tam, Ahmad, Grudpan, Page and cs., 2019; Jamsari, Nam, Tam and Siti-Azizah, 2014) of this species conducted. Few studies have observed the nutritional requirements of tire track eel. In that limited range, the protein requirements of tire track eel fingerlings have been studied (Nhi, Ngoc, Lan, Hang, Hanh, Van Ty and cs., 2022), and the ability to use black soldier fly and cricket meal as the protein sources in feed fish (Nhi, Ngoc, Hang, Hanh and Lan, 2023, 2024), but there have been no studies on the lipid requirements of this species conducted. To produce cost-effective fish diets for commercial farming of this species, the research on lipid requirement is necessary because it is known to affect the dietary protein requirement of fish. Although protein is an important and expensive dietary component, the dietary protein requirement of fish is affected by the content of other nutrients such as lipid and carbohydrate (Wang, Han, Wang and Ma, 2013). In fish diets, lipids, also known as fats, play an important physiological role as sources of metabolic energy for somatic growth. They are also the source of essential fatty acids and fat-soluble nutrients required for the formation of cell membranes, as well as for normal growth and development of fish (Zhou, Feng, Li, Ji and Gisbert, 2022). In carnivorous fish, when adding lipids to feed, they will affect the growth as well as the efficiency of protein use of animals, because fat can share energy with protein in the feed (Watanabe, 1982; Manam, 2023; NRC, 2011). Several studies have demonstrated the protein sparing effect of lipids in fish. For example, in rainbow trout (*Oncorhynchus mykiss*), at the optimum protein/fat ratios of 35:15–20, dietary proteins could be reduced from 48% to 35% with no loss in weight gains (Takeuchi, Yokoyama, Watanabe and Ogino, 1978). Furthermore, in gilthead sea bream (*Sparus aurata*) there was evidence of a protein-sparing effect of dietary lipid, as evidenced by a reduction of dietary protein from 52 to 46%, and an increase of dietary lipid from 9 to 15% in diets (Vergara, Robainà, Izquierdo and De La Higuera, 1996). Similar results have been observed by (Martino, Cyrino, Portz and Trugo, 2002) in surubim (*Pseudoplatystoma coruscans*) using diets with different lipid levels. Fat is considered a top concern because it is a component nutrient with high energy levels and high digestibility values. Thus, it is often added to food for many fish species, especially carnivorous fish, e.g. tire track eels which are often low in the digestion of carbohydrate. To design a diet with essential and balanced nutrients (not only in proteins and energy but also in lipids), there is a need to research on lipid requirements of this species. Therefore, this study was conducted to determine optimal dietary lipid levels for the growth of tire track eel fingerling and the fish chemical composition to develop optimal feed formulations for completing commercial farming processes.

2. MATERIALS AND METHODS

2.1 Experimental animal and conditions

The experiment was carried out over 90 days on an experimental farm at An Giang University, Long Xuyen city, An Giang province, Vietnam, in compliance with the laws, policies, as well as University and national guidelines for the care and use of experimental animals. Tire track eel (*Mastacembelus favus*) fingerlings were selected to have a relatively uniform size (around 3g/fish) and packaged in sealed 0.5 m^3 plastic bags filled with oxygen saturated water in a density of 1 kg fish per bag. These bags were transported by car from a hatchery in Thoai Son, An Giang province, Vietnam to the research station. On arrival, all fish were placed in a solution of 3% NaCl for 5 min to eliminate any risks of parasite infections. The fish were then kept in two tanks (3 m^3) for 2 weeks to allow them to acclimatize to the indoor conditions before allocated to the experimental tanks.

The experiment was arranged in a completely randomized design with 15 tanks (0.5m^3/tank) with five treatments, three replications (*n* = 3). Five diets were formulated with different lipid levels: 1.5%, 4.5%, 7.5%, 10.5% and 13.5%, with the iso-energy of 20 kJ/g and iso-protein of 43%. Fish were reared in composite tanks at the density of 100 fish/m^3 for three months. The tanks were set up with an aeration system and provided with substrates for shelter.

2.2 Feed ingredients and formula

Five iso-nitrogenous (43% crude protein), iso-energetic (20 kJ/g) diets were produced with different lipid levels: 1.5%, 4.5%, 7.5%, 10.5%, and 13.5% (Table 2). Diet recipes are given in Table 1. Table 2 is the chemical composition and level of selected essential amino acid and fatty acids given. The ingredients were obtained from local markets, Long Xuyen City, Vietnam. All ingredients were thoroughly mixed and then pelleted by using an electronic meat grinder (Quoc Hung company, Long Xuyen city, Vietnam) with pellet diameter and length in the range of 1 to 2 mm. All diets were sun-dried for two days, and then weighed and stored in sealed plastic bags in small portions at 5 °C until use.

Table 1. Ingredients compositions (g kg^{-1} DM) of diets with different lipid levels for tire track eels.

Ingredient composition (%)	**Lipid levels**				
	1.5%	**4.5%**	**7.5%**	**10.5%**	**13.5%**
Fish meal	310	315	320	323	330
Soybean meal	300	305	310	316	316
Wheat flour	328.8	289	250	211	175
Squid oil	1.2	21	30	50	69
Soybean oil	0	10	30	40	50
Earthworm liquid	20	20	20	20	20
Premix (Vitamin-minaral)[a]	20	20	20	20	20
CMC[b]	20	20	20	20	20

[a] *Vitamin and mineral premix content per kg: vitamin A 4,000,000 UI; vitamin D3 800,000 UI; vitamin E 8,500 UI; vitamin K3 750 UI; vitamin B1 375 UI; vitamin C 8,750 UI; vitamin B2 1,600 mg; vitamin B6 750 mg; folic acid 200 mg; vitamin B12 3,000 µg; biotin 20,000 µg; methionine 2,500 mg; Mn, Zn, Mg, K and Na 10 mg.* [b] *Carboxymethyl cellulose, imported from Korea.*

Table 2. Chemical compositions and amino acid content (g/kg DM) of diets with different lipid levels for tire track eels.

	Lipid levels				
	1.5%	**4.5%**	**7.5%**	**10.5%**	**13.5%**
Chemical composition					
Crude protein	439.9	430.3	430.7	432.4	431.0
Crude lipid	15.2	45.0	75.4	105.1	135.5
NFE	405.7	373.7	342.3	311.3	280.8

Crude fibre	21.1	21.2	21.3	21.4	21.3
Gross energy (kJ/g)	18.2	18.9	19.5	20.2	20.1
Essential amino acids					
Arginine	581.1	582.6	582.1	580.2	574.3
Alanine	218.0	240.1	260.2	235.3	206.3
Isoleucine	172.8	185.5	196.1	196.2	192.3
Leucine	265.4	297.8	328.1	308.9	285.6
Lysine	230.0	251.3	270.6	252.3	229.9
Methionine	58.3	70.0	79.6	65.1	46.6
Phenylalanine	180.3	203.5	224.7	209.6	190.4
Threonine	212.9	188.3	161.6	153.3	141.0
Tryptophan	42.0	44.0	44.0	49.5	51.0
Valin	191.0	214.7	236.3	219.4	198.4
Cystine	175.0	197.0	216.9	183.2	145.4
Tyrosine	108.1	121.2	132.2	123.3	110.4
Essential fatty acids					
DHA[a]	0.4	0.3	0.6	1.4	1.4
EPA[b]	0.01	0.01	0.01	0.1	0.1
CLA[c]	4.2	2.9	8.7	18.8	23.4
GLA[d]	0.3	0.01	0.6	1.2	1.1

[a] *DHA: Cis- 4,7,10,13,16,19-Docosahexaenoic acid.*

[b] *EPA: cis-5,8,11,14,17-Elcosapentaenoic acid.*

[c] *CLA: Linoleic acid.*

[d] *GLA: y-Linolenic acid (C18:3).*

2.3 Fish management

The tire track eels were fed to satiation (approximately 10% of BW) manually three times per day (between 7:00 – 9:00, 12:00 – 13:00, and between 18:00 – 19:00) by hand. Each feeding was closely monitored in each tank, the feed residue was collected, and the feeding rate was adjusted depending on the feed consumed on the previous day. The amount of feed used was recorded during the experiment and used to calculate true feed intake. The water source was municipal tap water, de-chlorinated by aeration for 24 h before use. Around 30% of the water was replaced daily. The tanks were housed in an open hall structure, with steel net for walls, i.e. the tanks were exposed to natural light, being very close to 12:12 h light/dark of this latitude. Water environmental factors (e.g., temperature, pH, O_2 and TAN and NO_2^-) were daily monitored and controlled. The temperature in the experimental treatments ranged from 26.5 to 29.8 ^oC, pH from 7.5 to 8.1, dissolved oxygen from 5.8 to 6.5 mg/L, total protein (TAN) 0.003 – 0.01 mg/L, and NO_2^- from 0.1 to 0.3 mg/L were all within the appropriate limits for the normal growth and development of tire track eels (Boyd & Pillai, 1985).

2.4 Measurements and calculations

All fish were anesthetized to measure weight and length before and after the experiment.

At the end of the experiment, five fish from each tank were sampled randomly and sacrificed by lethal anesthesia using ethylene glycol monophenyl ether at 0.5 mg L^{-1}. The liver and intestine were dissected out and weighed/measured. Hepato-somatic index (HSI, %) and intestinal quotient (Qi) index were then determined following procedures described by De Silva, Gunasekera, Gooley and Ingram (2001), Moreira, Silveira, Teixeira, Moreira, Moura and Farias (2012) and Da, Lundh and Lindberg (2012) and using the equations:

HSI (%) = [100× (Liver weight (g)/Body weight (g))]

Qi = Intestine length/Total length of the individual

Survival rate (SR), weight gain (WG), specific growth rate (SGR), daily weight gain (DWG), feed conversion ratio (FCR), protein efficiency ratio (PER), lipid efficiency ratio (LER) and lipid retention (LR) were calculated using the equations (H. Y. N. Nguyen, T. L. Trinh, K. Baruah, T. Lundh and A. Kiessling, 2021; Tan, Sun, Tan, Liu, Luo and Wei, 2018):

SR (%) = (Total number of fish harvest/Total number of fish cultured) x 100

WG = Final Wt - Initial Wt

SGR (%/day) = [(ln final Wt – ln initial Wt)/days] x 100

DWG (g/day) = (Final Wt – Initial Wt)/days

FCR = Total feed intake (g)/Total wet weight gain (g)

PER = Total wet weight gain (g)/Protein intake (g)

LER = Total wet weight gain (g)/Lipid intake (g)

LR (%) = [(Lipid in final fish – Lipid in initial fish)/Lipid intake] x 100

2.5 Chemical analysis

The samples of raw materials, feed and experimental fish, before allocating to the experiments and after harvesting, were frozen (-20 ^oC) until analysis. Chemical compositions (e.g., moisture, crude protein, crude lipid, fatty acid, minerals, crude fibre, crude energy and amino acid) of these samples were analysed in triplicate and measured in dry mass according to AOAC (2000) as cited in N. Y. H. Nguyen, L. T. Trinh, K. Baruah, T. Lundh and A. Kiessling (2021). Specifically, the dry matter was determined by drying in an oven at 105 ^oC until constant weight. Ash content was determined by incinerating the samples at 560 ^oC for 4 hours (until constant weight). Crude protein was calculated as 6.25 x % N analysed by the Kjeldahl method. Crude lipid was determined by Soxhlet extraction without acid hydrolysis. Crude fibre content was analysed using acid-base digestion. Nitrogen-free extract (NFE) was calculated as NFE (%) = 100 – (% protein + % lipid + % fibre + % ash). Gross energy content (kcal/kg) was calculated or measured by Parr Calorimeter. Amino acid content was determined by high-performance liquid chromatography using the method previously described by Vázquez-Ortiz, Caire, Higuera-Ciapara and Hernández (1995).

The procedures for analysis of the fatty acid (FA) in diets were performed according to the method described by Metcalfe, Schmitz and Pelka (1966) with modification. FA methyl esters were separated and quantified using HP6890 gas chromatograph equipment with a fused silica capillary column (007-CW) and a flame ionization detector. The column temperature was programmed to increase from 150 to 200 °C at a rate of 15 °C min^{-1}, and from 200 to 250 °C at a rate of 2 °C min^{-1}. The injector and detector temperatures were 250 °C.

2.6 Statistical analysis

Average values were calculated in Microsoft Excel. Means between treatment groups were compared based on ANOVA and Duncan's tests with a significant level of 0.05 using Minitab version 16.0. Lipid requirement was determined by the quadratic curve method (Zeitoun, Ullrey, Magee, Gill and Bergen, 1976), which analysed the correlation between DWG and lipid contents of fish diets with the equation y = ax2 + bx + c (where y is growth and x is the lipid content of the diet). This equation determined a y max point and its corresponding x max value. The x max value is the lipid content for the fish to achieve maximum growth.

3. RESULTS

3.1 Growth of tire track eels with different lipid levels

There was no difference in the initial weight of fish between treatment groups ($p > 0.05$). Thus, the initial weight of fish had no effect on the growth after the experimental period (Table 3).

Table 3. Growth of tire track eels fed experimental diets with different lipid levels after a 90-day treatment period.

Criteria	Treatments					SEM	P-value
	1.5%	4.5%	7.5%	10.5%	13.5%		
Initial weight (g)	3.07	3.04	3.04	3.05	2.99	0.08	0.98
Initial length (cm)	10.80	10.73	10.87	10.80	10.80	0.13	0.97
Final weight (g)	7.77^{c}	8.61^{bc}	10.24^{a}	9.70^{ab}	9.01^{abc}	0.36	0.002
Final length (cm)	13.18^{b}	13.55^{b}	15.09^{a}	14.88^{a}	14.80^{a}	0.21	0.001
Weight gain (g)	4.70^{c}	5.57^{bc}	7.20^{a}	6.65^{ab}	6.02^{abc}	0.3	0.001
DWG (g/day)	0.052^{c}	0.062^{bc}	0.08^{a}	0.074^{ab}	0.067^{abc}	0.003	0.003
SGR (%/day)	1.03^{b}	1.16^{ab}	1.35^{a}	1.28^{a}	1.23^{ab}	0.05	0.007

DWG: daily weight gain; SGR: Specific growth rate.

SEM = Standard error of the mean.

Means with different superscript letters within rows are significantly different ($p<0.05$).

After a 90-day treatment period, the growth rate of tire track eels tended to increase with an increase in the lipid contents of the feed from 1.5 to 7.5% fat and started to decrease when the lipid levels were higher (Table 3). DWG and SGR of tire track eels in the 1.5% lipid containing diet were the lowest and significantly different from the rest diets. DWG ranged from 0.052 g day^{-1} to 0.08 g day^{-1}, reaching the highest in the 7.5% lipid treatment group, and the difference was not statistically significant ($p > 0.05$) compared to the 10.5% and 13.5% lipid treatment groups, and the difference was significant ($p < 0.05$) compared to the remaining groups (Table 3).

SGR ranged from 1.03 to 1.35%/day and was highest in the 7.5% lipid treatment group, which was significantly different ($p < 0.05$) from that of the 1.5% lipid treatment group. There was no significant difference in SGR between the 7.5% lipid treatment group and the remaining groups (Table 3).

3.2 Optimal lipid levels in the feed formulation for tire track eel fingerlings

Optimal lipid requirements of tire track eel fingerlings were determined by using the regression correlation between the DWG of tire track eels and the lipid contents of the diets. The correlation equation $y = -0.0005x^2 + 0.0083x + 0.0389$ and the correlation coefficient $R^2 = 0.89$ showed a relatively close correlation between the lipid content of the feed and the DWG of the fish. Thus, the optimal lipid content in the feed to help the fish achieve the best growth was 8.5% (Figure 1).

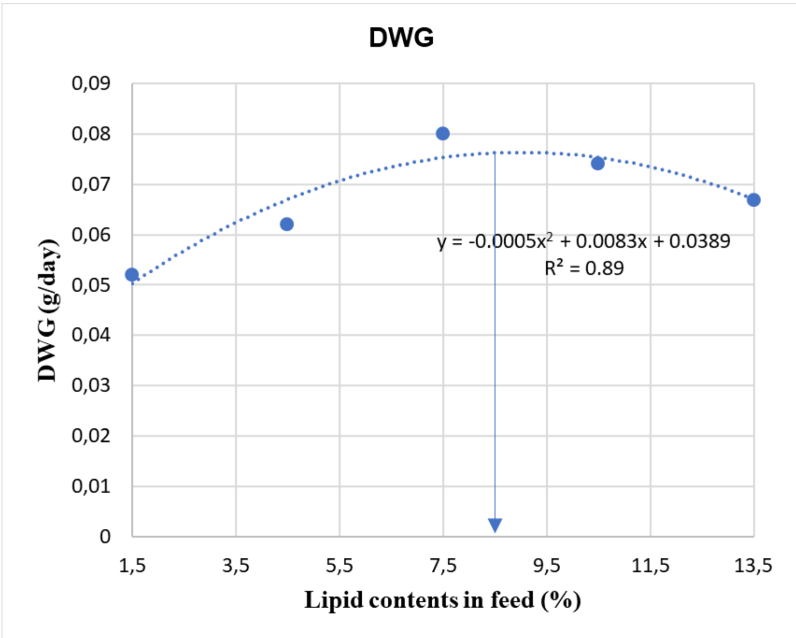

Figure 1. Correlation between the lipid contents in feed and daily weight gains of tire track eel fingerlings. Each point represents the mean of three groups/replicates of fish.

3.3 Survival rates and feed efficiency

The survival rates of tire track eels after the 90-day treatment period ranged from 79.3% - 88%, which was no significant difference ($p > 0.05$) between the treatment diets with different lipid content. Hence, experimental diets with different lipid content did not affect the survival rates of tire track eels (Table 4).

Table 4. Survival rates and feed efficiency of tire track eels fed experimental diets with different lipid levels after the 90-day treatment period

	Treatments						
	1.5%	**4.5%**	**7.5%**	**10.5%**	**13.5%**	**SEM**	**P-value**
Survival rates (%)	82	79.3	88	79.3	79.3	4.5	0.611
FCR	1.91[a]	1.70[ab]	1.2[c]	1.43[bc]	1.48[bc]	0.06	0.001

PER	1.19^{c}	1.38^{bc}	1.93^{a}	1.62^{b}	1.58^{b}	0.06	0.001
LER	34.47^{a}	13.15^{b}	11.03^{b}	6.65^{c}	5.02^{c}	0.49	0.001
LR	207^{a}	79.9^{b}	66.1^{bc}	41.6^{cd}	36.8^{d}	5.58	0.001

PER: Protein efficiency ratio; PER: Protein efficiency ratio, LER: lipid efficiency ratio; LR: lipid retention.

SEM = Standard error of the mean.

Means with different superscript letters within rows are significantly different (p<0.05).

The feed conversion ratio (FCR) of tire track eel fingerlings was highest in the treatment containing 1.5% lipid (1.95), and it was significantly different ($p < 0.05$) from those of experimental diets containing 7.5%, 10.5% and 13.5% lipid. FCR was lowest in the treatment diets containing 7.5% lipid, and there was no significant difference ($p > 0.05$) in FCRs between the diets of 7.5%, 10.5% and 13.5% lipid (Table 4).

Protein efficiency ratios (PERs) of the fish ranged from 1.19 to 1.93, reaching the highest level in the diet containing 7.5% lipid, which was significantly different ($p < 0.05$) from those of the remaining groups. However, the PER was lowest in the 1.5% lipid group and not significantly different ($p > 0.05$) from the 4.5% lipid group (Table 4).

Lipid efficiency ratios (LERs) and lipid retentions (LRs) of the fish gradually decreased as the lipid contents in the feed increased. LERs and LRs decreased from the diets containing 1.5% to 13.5% lipids. Specifically, LER and LR were the highest (34.47% and 207%, respectively) in the 1.5% lipid content group compared to the rest of the treatment groups (Table 4).

3.4 Body indies and chemical composition of tire track eels

The results obtained for body indices are shown in Table 5. Hepato-somatic index (HSI) was significant difference ($p<0.05$) between the treatments, with the highest HSI in the treatment group with 7.5% lipid (2.12%) and the lowest in the treatment group with 1.5% lipid (1.5%). However, intestinal quotient (Qi) of tire track eels did not differ significantly ($p>0.05$) between treatments.

Table 5. Body indices of tire track eels fed diets with different lipid level.

	Treatments						
	1,5%	4,5%	7,5%	10,5%	13,5%	SEM	P- value
HSI, %	1.50^{b}	1.65^{ab}	2.12^{a}	1.76^{ab}	1.88^{ab}	0.138	0.027
Qi	0.27	0.31	0.27	0.30	0.32	0.021	0.289

HSI: Hepato-somatic index = [100× (Liver weight (g)/Body weight (g))].

Qi: intestinal quotient = Intestine length/Total length of the individual.

SEM = Standard error of the mean.

Means with different superscript letters within rows are significantly different (p<0.05).

The chemical composition of tire track eels before and after the experiment is shown in Table 6. The moisture content (water in the body) of tire track eels tended to increase at the end of the experiment. The body water content of fish was lowest in the 1.5% lipid treatment group (74.77%), and there was no significant difference ($p > 0.05$) between all treatment groups. In contrast to moisture content, the lipid content of tire track eels after the experiment seemed to increase gradually with the elevation of lipid content in the feed. The lipid content of fish in the 13.5% lipid group was the highest (5.86%) and significantly differed ($p < 0.05$) from the 1.5% lipid group (4.8%) but was not significantly different ($p > 0.05$) from those of the remaining treatment groups. The protein and ash contents of the fish after the experiment were little changed as the protein content ranged from 13% to 15.1% and the ash content ranged from 2.28% to 2.63%, and there was no significant difference ($p > 0.05$) in the protein and ash contents between diets containing different lipid levels (Table 6).

Table 6. Proximate compositions of the whole body of tire track eels (% fresh weight) fed diets with different lipid level.

Treatments (Lipid levels)	Moisture	Protein	Lipid	Ash
Fish before experiment	81.97	11.45	2.98	1.86
1.5%	74.77	15.1	4.80b	2.63
4.5%	75.85	14.55	5.07ab	2.60
7.5%	78.18	13.13	5.31ab	2.47
10.5%	78.37	13.00	5.22ab	2.28
13.5%	76.98	13.75	5.86a	2.33
SEM	1.10	0.62	0.180	0.15
P- value	0.178	0.152	0.022	0.428

SEM = Standard error of the mean.

Means with different superscript letters within columns are significantly different ($p<0.05$).

4. DISCUSSION

In carnivorous fish species, increasing lipid levels in diets has been indicated to be an effective method not only to improve growth rates, feed efficiency and protein utilization, but also to decrease nitrogen waste outputs as well as feed costs and providing adequate amount of lipid in fish diets to reduce protein inclusion without compromising growth is the nutritional approach for protein sparing effect (López, Torres, Durazo, Drawbridge and Bureau, 2006).

Dietary lipid is an important energy source for fish and the capacity in utilising dietary lipid varies among fish species (Wang, Han, Wang and Ma, 2013). In the present study, effects of increasing dietary lipid levels were observed on growth, SGR, FCR and PER of tire track eel fingerlings. The growth response data showed that the maximum growth was obtained at 7.5% lipid with the iso-energy of 20 kJ/g and iso-protein of 43%. In tire track eels, this is the first study determining lipid requirements at fingerlings. These findings are similar to results reported for other freshwater fish species, like Asian red-tailed catfish, Hemibagrus wyckioides (Deng, Zhang, Sun, Zhang and Mi, 2021), striped catfish, Pangasianodon hypophthalmus (Sivaramakrishnan, Sahu, Jain, Muralidhar, Saravanan, Ferosekhan and cs., 2017), hydrid tilapia, Oreochromis niloticus x Oreochromis aureus (Chou &Shiau, 1996; Gan, Li, Feng, Gong, Huang and Li, 2009), Chinese longsnout catfish, Leiocassis longirostris and gibel carp, Carassius auratus gibelio (Pei, Xie, Lei, Zhu and Yang, 2004), red-tailed catfish, Hemibagrus wyckioides (Hung, Binh, Thanh Truc, Tham and Ngoc Tran, 2017). In contrast, elevating the dietary lipid levels from 65 to 125 g/kg could not induce protein sparing action in golden pompano (Trachinotus ovatus) as the fish prefer to utilise the diets containing higher contents of protein, rather than the diets with higher content of lipid (Wang, Han, Wang and Ma, 2013). However, increased dietary lipid to more than 7.5% in 43% protein diet did not improve weight gains, SGR, FCR and PER of tire track eel fingerlings. This can be explained that high dietary lipid levels may cause limited ability to digest and absorb high amounts of lipid, a reduction in feed intake, excess lipid accumulation in liver and other visceral organs, or creation of dietary or metabolic imbalances which, in turn, depress growth (López, Torres, Durazo, Drawbridge and Bureau, 2006).

The dose–response experiments with increasing lipid contents are accepted in principle as a method for determining optimal dietary lipid level of grouper, Epinephelus malabaricus (Lin &Shiau, 2003), white seabass, Atractoscion nobilis (López, Torres, Durazo, Drawbridge and Bureau, 2006), large yellow croaker, Pseudosciaena crocea (Ai, Zhao, Mai, Xu, Tan, Ma and cs., 2008), Pangasianodon hypophthalmus (Sivaramakrishnan and cs., 2017), tilapia (Paul, Sardar, Sahu, Jana, Deo, Harikrishna and cs., 2022). In the present study, weight gains of tire track eels fed lower 7.5% lipid diet decreased significantly, thus the relationship between fish growth and dietary lipid level is best expressed by a second order regression curve (Fig.1) and the maxima of the curve was obtained at about 8.5% lipid. Note that weight gain of the fish in the 7.5% lipid group was not significantly different from those in the 10.5% and 13.5% lipid groups (Table 3) suggesting that 7.5% dietary lipid may meet the minimum lipid requirement for tire trạck eels. The present study showed that the estimated optimal dietary lipid level of tire track eels was 8.5 % based on the daily weight gain. This agrees well with some previous studies on large yellow croaker, Pseudosciaena crocea (Ai and cs., 2008), Solea senegalensis (S Morais, Narciso, Dores and Pousao-Ferreira, 2004; Sofia Morais, Torten, Nixon, Lutzky, Conceição, Dinis and cs., 2006), Dicentrarchus labrax (Infante &Cahu, 1999), Sparus aurata (Salhi, Hernández-Cruz, Bessonart, Izquierdo and Fernández-Palacios, 1999) and Pangasianodon hypophthalmus (Sivaramakrishnan and cs., 2017).

The experimental diets with different lipid contents did not affect the survival rates of tire track eels. Similar results were found in other studies such as on white seabass (Atractoscion nobilis) fingerlings fed four levels of lipid (15.5%, 18%, 19.5% and 21.5%) at one level of protein (61% crude protein) for six weeks (López, Torres, Durazo, Drawbridge and Bureau, 2006), on larvae red drum Sciaenops ocellatus with cod liver oil and soy lecithin as lipid sources (Buchet, Infante and Cahu, 2000). showed that the growth increased significantly with increasing dietary lipid, and survival was independent of dietary lipid. The differences were probably due to fish species, diet types, experimental conditions, etc.

The hepatosomatic index (HSI) increased more with feeding high lipid diets 7.5% than low lipid diets (1.5%) in the fish (Table 5). There was a significant difference found in HSI with different dietary lipid levels in tire track eel, the trend was not definite with regards to dietary lipid content. The finding on is similar to the observation found in striped bass (Millikin, 1983), Atlantic salmon (Hemre &Sandnes,

1999) and gilthead seabream (Santinha, Medale, Corraze and Gomes, 1999). However, a significant difference in HSI was found directly proportional to the increase in dietary lipid level in Pangasianodon hypophthalmus (Sivaramakrishnan and cs., 2017) and Pacific blue fin tuna juvenile (Biswas, Ji, Biswas, Seoka, Kim, Kawasaki and cs., 2009) and inversely ratio to the increase in dietary lipid level in European seabass (Ballestrazzi &Lanari, 1996).

Protein efficiency ratios (PERs) of tire track eel ranged from 1.19 to 1.93, reaching the highest level in the diet containing 7.5% lipid and lowest in the 1.5% lipid group (Table 4). Protein efficiency ratio improved steadily with the dietary lipid content up to 7.5%, and then reached a plateau. Similarly, protein retention increased gradually with the dietary lipid content up to 7.5 %. Conversely, lipid retention decreased linearly with increasing dietary lipid content, but no significant difference was found among juveniles fed diets with 7.5 %, and 10.5 % lipid which was fix well with previous researches on Asian red-tailed catfish (Hemibagrus wyckioides) (Deng, Zhang, Sun, Zhang and Mi, 2021).

The increase in dietary lipid level usually resulted in the total body fat deposition in fish (Deng, Zhang, Sun, Zhang and Mi, 2021; Paul and cs., 2022; Sivaramakrishnan and cs., 2017). Similar result was observed in this study, the whole-body lipid content of tire track eel increased with increasing dietary lipid level.

5. CONCLUSION

The optimal lipid level in the dietary containing 43% protein of tire track eel fingerling (size of 3 g/fish) was 8.5%. The suitable lipid level (8.5%) in diet containing 43% protein improved the feed conversion ratio in tire track eel. The body lipid content of the treated fish tended to rise with the increase of the lipid content of the experimental feed. The body indices, moisture, protein and ash contents in the body of the treated fish did not clearly show a relationship with the lipid content in the feed. The survival rates of tire track eels in this study were not affected by the lipid content in the feed. Future researches are still needed to determine the optimum protein/energy ratio in diets for this species.

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REFERENCES

Ai, Q., Zhao, J., Mai, K., Xu, W., Tan, B., Ma, H., & Liufu, Z. (2008). *Optimal dietary lipid level for large yellow croaker (Pseudosciaena crocea) larvae*. Aquaculture Nutrition, 14(6), 515-522.

AOAC. (2000). *Official methods of analysis of AOAC International (17th ed.).* In). AOAC International, Gaithersburg, MD, USA (2000).

Ballestrazzi, R., & Lanari, D. (1996). *Growth, body composition and nutrient retention efficiency of growing sea bass (Dicentrarchus labrax L.)* fed fish oil or fatty acid Ca salts. Aquaculture, 139(1-2), 101-108.

Biswas, B. K., Ji, S.-C., Biswas, A. K., Seoka, M., Kim, Y.-S., Kawasaki, K.-i., & Takii, K. (2009). *Dietary protein and lipid requirements for the Pacific bluefin tuna Thunnus orientalis juvenile. Aquaculture, 288(1-2), 114-119.*

Boyd, C. E., & Pillai, V. (1985). *Water quality management in aquaculture*. CMFRI special Publication, 22, 1-44.

Buchet, V., Infante, J. Z., & Cahu, C. (2000). *Effect of lipid level in a compound diet on the* *development of red drum (Sciaenops ocellatus) larvae*. Aquaculture, 184(3-4), 339-347.

Chou, B.-S., & Shiau, S.-Y. (1996). *Optimal dietary lipid level for growth of juvenile hybrid tilapia, Oreochromis niloticus x Oreochromis aureus*. Aquaculture, 143(2), 185-195.

Da, C. T., Lundh, T., & Lindberg, J. E. (2012). *Evaluation of local feed resources as alternatives to fish meal in terms of growth performance, feed utilisation and biological indices of striped catfish (Pangasianodon hypophthalmus) fingerlings*. Aquaculture, 364–365, 150-156.

De Silva, S., Gunasekera, R., Gooley, G., & Ingram, B. (2001). *Growth of Australian shortfin eel (Anguilla australis) elvers given different dietary protein and lipid levels*. Aquaculture Nutrition, 7(1), 53-57.

Diem, T. H. (2009). *Breeding for maturation and spawning of Tire track eels (Mastacembelus armatus)*. Bachelor thesis. Can Tho University. (in Vietnamese).

Deng, J., Zhang, X., Sun, Y., Zhang, L., & Mi, H. (2021). *Optimal dietary lipid requirement for juvenile Asian red-tailed catfish (Hemibagrus wyckioides)*. Aquaculture Reports, 20, 100666.

Gan, H., Li, J., Feng, G., Gong, Z., Huang, K., & Li, J. (2009). *Effects of different lipid levels on growth and haematological biochemistry in juvenile tilapia (Oreochromis niloticus× Oreochromis aureus)*. Journal of Shanghai Ocean University, 18(1), 35-41.

Gupta, S., & Banerjee, S. (2016). Food, *feeding habit and reproductive biology of tire-track spiny eel (Mastacembelus armatus)*: A review. Journal of Aquaculture Research and Development, 7(5), 429.

Hemre, & Sandnes. (1999). *Effect of dietary lipid level on muscle composition in Atlantic salmon Salmo salar*. Aquaculture Nutrition, 5(1), 9-16.

Hung, L., Binh, V., Thanh Truc, N., Tham, L., & Ngoc Tran, T. (2017). *Effects of dietary protein and lipid levels on growth, feed utilization and body composition in red-tailed catfish juveniles (Hemibagrus wyckioides, Chaux & Fang 1949)*. Aquaculture Nutrition, 23(2), 367-374.

Infante, J. L. Z., & Cahu, C. L. (1999). *High dietary lipid levels enhance digestive tract maturation and improve Dicentrarchus labrax larval development*. The Journal of Nutrition, 129(6), 1195-1200.

Jamaluddin, J. A. F., So, N., Tam, B. M., Ahmad, A., Grudpan, C., Md Sah, A. S. R., & Mohd Nor, S. A. (2021). *Genetic diversity of the tire track eel Mastacembelus favus in Southeast Asia inferred from microsatellite markers*. Ichthyological Research, 68, 529-540.

Jamaluddin, J. A. F., So, N., Tam, B. M., Ahmad, A., Grudpan, C., Page, L. M., Khaironizam, M., & Mohd Nor, S. A. (2019). *Genetic variation, demographic history and phylogeography of tire track eel, Mastacembelus favus (Synbranchiformes: Mastacembelidae) in Southeast Asia*. Hydrobiologia, 838(1), 163-182.

Jamsari, A. F. J., Nam, S., Tam, B. M., & Siti-Azizah, M. N. (2014). *Isolation and characterisation of microsatellite loci in the tire track eel, Mastacembelus favus and cross-species amplification. Conservation Genetics Resources*, 6(2), 477-479.

Lin, Y.-H., & Shiau, S.-Y. (2003). *Dietary lipid requirement of grouper, Epinephelus malabaricus, and effects on immune responses*. Aquaculture, 225(1-4), 243-250.

Loan, P. P., Anh, N. T., Vinh, V. H., Tung, L. T., Linh, L. V., Ngoc, T. K., Tinh, N. V. H., & Hong, T. T. (2010). *Developing an artificial seed production process for Tire track eels (Mastacembelus favus) in An Giang*. Scientific research project of An Giang University. (in Vietnamese), 70.

López, L. M., Torres, A. L., Durazo, E., Drawbridge, M., & Bureau, D. P. (2006). *Effects of lipid on growth and feed utilization of white seabass (Atractoscion nobilis) fingerlings*. Aquaculture, 253(1-4), 557-563.

Manam, V. K. (2023). *Fish feed nutrition and its management in aquaculture*.

Martino, R. C., Cyrino, J. E. P., Portz, L., & Trugo, L. C. (2002). *Effect of dietary lipid level on nutritional performance of the surubim, Pseudoplatystoma coruscans*. Aquaculture, 209(1-4), 209-218.

Metcalfe, L., Schmitz, A., & Pelka, J. (1966). *Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. Analytical chemistry*, 38(3), 514-515.

Millikin, M. R. (1983). *Interactive effects of dietary protein and lipid on growth and protein utilization of age-0 striped bass*. Transactions of the American Fisheries Society, 112(2A), 185-193.

Morais, S., Narciso, L., Dores, E., & Pousao-Ferreira, P. (2004). *Lipid enrichment for Senegalese sole (Solea senegalensis) larvae: effect on larval growth, survival and fatty acid profile*. Aquaculture International, 12, 281-298.

Morais, S., Torten, M., Nixon, O., Lutzky, S., Conceição, L. E., Dinis, M. T., Tandler, A., & Koven, W. (2006). *Food intake and absorption are affected by dietary lipid level and lipid source in seabream (Sparus aurata L.) larvae*. Journal of Experimental Marine Biology and Ecology, 331(1), 51-63.

Moreira, R. L., Silveira, L. P., Teixeira, E. G., Moreira, A. G. L., Moura, P. S. d., & Farias, W. R. L. (2012). *Growth and gastrointestinal indices in Nile tilapia fed with different diets*. Acta Scientiarum. Animal Sciences, 34, 223-229.

Nguyen, H. Y. N., Trinh, T. L., Baruah, K., Lundh, T., & Kiessling, A. (2021). *Growth and feed utilisation of Nile tilapia (Oreochromis niloticus) fed different protein levels in a clear-water or biofloc-RAS system*. Aquaculture, 536, 736404.

Nguyen, N. Y. H., Trinh, L. T., Baruah, K., Lundh, T., & Kiessling, A. (2021). *Growth and feed utilisation of Nile tilapia (Oreochromis niloticus) fed different protein levels in a clear-water or biofloc-RAS system*. Aquaculture, 536.

Nhi, N. H. Y., Ngoc, T. K., Hang, N. T. T., Hanh, N. T. B., & Lan, T. T. (2023). *Evaluation of the replacement of fishmeal with black soldier fly larvae meal in feed for tire track eel (Mastacembelus favus) fingerling*. Livestock Research for Rural Development, Volume 35(Article #93).

Nhi, N. H. Y., Ngoc, T. K., Hang, N. T. T., Hanh, N. T. B., & Lan, T. T. (2024). *Evaluate the replacement of fishmeal with cricket meal (Gryllus bimaculatus) in feed for tire track eel (Mastacembelus favus) fingerling*. Livestock Research for Rural Development, 36(2).

Nhi, N. H. Y., Ngoc, T. K., Lan, T. T., Hang, N. T. T., Hanh, N. T. B., Van Ty, D., Lundh, T., & Kiessling, A. (2022). *Dietary protein requirements of tire track eel (Mastacembelus favus Hora, 1923) fingerling*. Livestock Research for Rural Development, 34(September 26).

NRC. (2011). *Nutrient requirements of fish and shrimp*. Washington, D.C.: National Academies Press.

Paul, M., Sardar, P., Sahu, N. P., Jana, P., Deo, A. D., Harikrishna, V., Varghese, T., Shamna, N., Kumar, P., & Krishna, G. (2022). *Effect of dietary lipid level on growth performance, body composition, and physiometabolic responses of genetically improved farmed tilapia (GIFT) juveniles reared in inland ground saline water*. Aquaculture Nutrition, 2022.

Pei, Z., Xie, S., Lei, W., Zhu, X., & Yang, Y. (2004). *Comparative study on the effect of dietary lipid level on growth and feed utilization for gibel carp (Carassius auratus gibelio) and Chinese longsnout catfish (Leiocassis longirostris Günther).* Aquaculture Nutrition, 10(4), 209-216.

Salhi, M., Hernández-Cruz, C., Bessonart, M., Izquierdo, M., & Fernández-Palacios, H. (1999). *Effect of different dietary polar lipid levels and different n− 3 HUFA content in polar lipids on gut and liver histological structure of gilthead seabream (Sparus aurata) larvae*. Aquaculture, 179(1-4), 253-263.

Santinha, Medale, Corraze, & Gomes. (1999). *Effects of the dietary protein: lipid ratio on growth and nutrient utilization in gilthead seabream (Sparus aurata L.)*. Aquaculture Nutrition, 5(3), 147-156.

Sivaramakrishnan, T., Sahu, N., Jain, K., Muralidhar, A., Saravanan, K., Ferosekhan, S., Praveenraj, J., & Artheeswaran, N. (2017). *Optimum dietary lipid requirement of Pangasianodon hypophthalmus juveniles in relation to growth, fatty acid profile, body indices and digestive enzyme activity*. Aquaculture International, 25, 941-954.

Takeuchi, T., Yokoyama, M., Watanabe, T., & Ogino, C. (1978). *Optimum ratio of dietary energy to protein for rainbow trout*.

Tan, C., Sun, D., Tan, H., Liu, W., Luo, G., & Wei, X. (2018). *Effects of stocking density on growth, body composition, digestive enzyme levels and blood biochemical parameters of Anguilla marmorata in a recirculating aquaculture system*. Turkish Journal of Fisheries and Aquatic Sciences, 18(1), 9-16.

Trieu, N. V. (2009). *Study on biological characteristics and artificial reproduction tire track eel (Mastacembelus armatus Lacepade, 1800).* Scientific report on ministerial-level research topic. (in Vietnamese).

Trung, N. T. (2010). *Artificial propagation of tire track eel (Mastacembelus favus)*. Scientific and technological report. Can Tho Department of Science and Technology. (in Vietnamese).

Trung, N. T., Anh, N. T., & Thanh, N. Q. (2010). *Experiments of artificial propagation of tire track eel (Mastacembelus favus).* (in Vietnamese).

Vázquez-Ortiz, F., Caire, G., Higuera-Ciapara, I., & Hernández, G. (1995). *High performance liquid chromatographic determination of free amino acids in shrimp.* Journal of Liquid Chromatography & Related Technologies, 18(10), 2059-2068.

Vergara, J. M., Robainà, L., Izquierdo, M., & De La Higuera, M. (1996). Protein sparing effect of lipids in diets for fingerlings of gilthead sea bream. Fisheries Science, 62(4), 624-628.

Wang, F., Han, H., Wang, Y., & Ma, X. (2013). *Growth, feed utilization and body composition of juvenile golden pompano Trachinotus ovatus fed at different dietary* *protein and lipid levels.* Aquaculture Nutrition, 19(3), 360-367.

Watanabe, T. (1982). *Lipid nutrition in fish.* Comparative Biochemistry and Physiology Part B: Comparative Biochemistry, 73(1), 3-15.

Zeitoun, I. H., Ullrey, D. E., Magee, W. T., Gill, J. L., & Bergen, W. G. (1976). *Quantifying nutrient requirements of fish.* Journal of the Fisheries Board of Canada, 33(1), 167-172.

Zhou, J., Feng, P., Li, Y., Ji, H., & Gisbert, E. (2022). *Effects of dietary lipid levels on growth and gonad development of Onychostoma macrolepis broodfish.* Fishes, 7(5), 291.