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PAPER • **OPEN ACCESS**

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To cite this article: B. Hasan *et al* 2021 *IOP Conf. Ser.: Earth Environ. Sci.* **695** 012054

IOP Conf. Series: Earth and Environmental Science **695** (2021) 012054 doi:10.1088/1755-1315/695/1/012054

Utilization of Fermented Salted Marine Trashfish Meal for Replacing Fishmeal in Nemurus Catfish (*Hemibagrus nemurus*) Diets

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Abstract. This research was carried out to determine the substitution values of fermented salted marine trash fish (FSMTF) for dietary conventional fishmeal (CFM) on growth of nemurus catfish. Four isoproteic (34% crude protein) and isocaloric (3.15 kcal DE g^{-1}) diets were formulated. One diet was a control diet (C) in which 44% CFM was included and no FSMTF. Other three diets were test diets in which CFM was substituted with FSMTF, 50% (FSMTF-50), 75% (FSMTF-75) and 100% (FSMTF-100). A commercial diet (COM) which composed of 31 % crude protein and 2.90 kcal DE g^{-1} was also tested as a comparison. Nemurus catfish, 47±3.94 g in weight were distributed into triplicate net cages (2x2x1.5 m), 40 fishes per m^3 water, and fed experimental diets two times daily for two months. Total substitution of fishmeal with FSMTF (FSMTF-100) in the fish diet did not affect fish growth (weight gain, specific growth rate, survival rate), feed efficiency ratio, protein efficiency ratio and protein utilization as compared to control diet. Total substitution of CFM with FSMTF gave better feed efficiency ratio, protein efficiency ratio and protein utilization than commercial diet. FSMTF therefore can completely replace CFM in the diet of nemurus catfish.

1. Introduction

An availability of high quality and cheap protein feed ingredients is a key to successful fish aquaculture, especially for nemurus catfish since the fish need higher protein level in its diet, 34-42% [1, 2, 3, 4, 5, 6]. Nemurus catfish is usually fed manufactured feed which composes of higher fishmeal. Fishmeal, a conventional protein source in the catfish diet [1, 3] is costly and scarcely available, therefore, replacement of dietary fishmeal with local ingredients needs to afford for efficient and sustainable aquaculture production.

Soybean meal which is usually included in the fish diet as a protein source, its price now is expensive, less economical for fish feed. Other protein sources, such as meize, palm kernel meal, rubber seed and cotton seed often contains anti nutrition such as higher fiber, enzyme inhibitor, saponin, lectine, tannin, pytic acid, gossypol which give negative effect to the growth of fish [7] Animal waste products such as poultry viscera and feather meal contain high protein but low digestible by fish [8, 9 10]. Animal by products are also considered unsafe as feed ingredient since they can transmite diseases to human [11]. Substitution fishmeal with animal and plan ingredients often give negative effect on body composition [12, 13, 14, 11], and flesh sensory quality [15, 16,1 7].

Trash fish from marine bycatch is a potential feed stuff for replacing conventional fishmeal in aquaculture diets as it containes high protein, 50 - 60% dry weight [4] [5] [6]; and its production is abundant which is estimated more than 3,33 milion tones per year or 4-19 times as high as the total trawler and trawler like catches in Indonesia [18]. As the production of marine by catch is scattered in small amount in remote fishing grounds, while availability of ice and chilling fasilities are limited, thus the marine by catch is usually utilized in form of salted products [4] [5] [6]. However, when the highly salted product (15-20%) is included in the diets, the diets will be low acceptable by fish.

Our previous study revieled that boiling could reduce salt concentration of bycach to 9-12% [4, 6]; and the product could replace konventional fishmeal 50-75% in the fish diet. However, boiling and drying is considered impractical and requires high production cost which is not economical for the producers or farmers. Our feeding trial on nemurus catfish indicated that feed diets containing fermented silage of added molases could improve palatability of the diet as compared to fishmeal diet [19] [2]. In this study, salted marine bycatch fermented with molases is included in the nemurus catfish diet replacing fishmeal; and the substitution effect is evaluated on growth, feed conversion and protein utilization.

2. Materials and Methods

2.1. Formulation and Analyses

High quality conventional fishmeal (CFM), salted marine trash fish, tofu by product, rice bran, palm kernel, meize by product meal were purchased from local feed supplier. Salted marine trash fish composed of various fish species was ground in meat grinder and fermented with 10% molases for 3 days; and the fermented product was oven dried at 55°C until about 12% in moisture. All inggredients were analyzed for proximate composition. Four diets, a control and 3 test diets, were designed to compose of 34% crude protein and 3.15 kcal digestible energy (DE) g^{1}. For control diet, 44% CFM was included and no FSMTF. In the test diets, CFM was substituted by FSMTF, 50%, 75% and 100% respectively. A commercial diet (COM) composed of 31% crude protein and 2.90 kcal DE g^{-1} was also tested for a comparison in the feeding experiment.

All formulated diets were thoroughly mixed; and the mixtures were made for sinking pellets (2–3 mm in diameter) using pelleting machine. The pelleted diets were oven dried at 55°C to about 12% in moisture; and before fed to the fish, they were kept at chilling temperatur (5-7°C). Proximate, amino acid and salt concentration of the pelleted diets were determined by AOAC method [20]. Moisture determination was made after the sample was dried in oven-dryer at 105°C to a constant weight. Ash analysis was conducted after the sample was combusted in muffle furnace at 550°C for 5 hours. Crude protein analysis was performed by micro-Kjeldahl method, and the value was calculated as Nx6.25. Crude lipid determination was conducted after the sample was extracted in petroleum ether using Soxhlet apparatus. Amino acids analysis was carried out by HPLC (Waters, USA) using Pico-tag procedure developed by [21]. The amino acid determination was made after the samples were hydrolyzed under nitrogen in 6N HCl at 110°C for 24 hours. The salt concentration was analyzed by incinerating the sample at 550°C and titrated with $AgNO_3$ 0.1 N.

Water stability evaluation of the pelleted diets was carried out in accordance with the procedure recommended by [22]; [23] [19] [4] [5] [6]. Fifty gram samples (triplicate) of the formulated pelleted diets were weighed in the screen and slowly submerged in a 50 L aquarium which was filled with distilled water at room temperature for 10 and 30 minutes. The screen were taken out and dried in oven-dryer at 105°C for 2 hours. The samples were reweighed after cooling in a desiccator. The stability of the pelleted diets in the water was calculated as the loss in weight after reweighing; and the values were indicated as percent loss of dry matter (% LDM).

2.2. Fish Sample and Growing Condition

Nemurus catfish weighing 47±3.94 g each was collected from a local hatchery in Sungai Paku, Riau. The fish was adapted to the feeding condition for one week; and during adaptation, the fish was fed a commercial diet. Feeding trial was conducted in triplicate 2x2x1.5 m net cages which were constructed in 30x30m earthen pond and continuously flowed with surface water at flowing rate of 2.5 L min^{-1}. Forty fish were randomly distributed into each of 15 cages with triplicates per dietary treatment.

2.3. Feeding and Growth Analysis

Before feeding trial, all fish was weighed and 10 fish was randomly collected for body protein and amino acid composition. During feeding trial, the fish was fed the diets ad libitum at 07:30 AM and 4:30 PM for two months. At the end of the feeding trial, all fish was reweighed, and 15 fish per cage was randomly collected for body protein analysis. Growth, feed conversion and protein utilization were determined by the following formula:

Weight gain (WG, g) = (Final body weight – Initial body weight)
Specific growth rate (SGR, % day^{-1}) = 100 x (Ln final body weight - Ln initial body weight)/days
Survival rate (SR) (%) = 100 x (Final fish number / Initial fish number)
Feed consumption (FC, g fish^{-1}) = Total amount of feed consumed throughout the experiment /fish
Feed efficiency ratio (FER) = Weight gain (g)/Feed intake (g)
Protein efficiency ratio (PER) = Weight gain (g)/ Protein intake (g)
Protein utilization (PU, %) = [(Final body weight x Final body protein)-(Initial body weight x Initial body protein)]/ [(Total protein intake (g) x 100)]

2.4. Data Analysis

The data analysis was conducted by One-way Analysis of Variance (ANOVA) using SPSS for Windows software, version 17 [24]. The difference among the treatments was determined by the Least Significant Different test at 95% significance level (P>0.05).

3. Results and Discussions

3.1. Ingredients and diet composition

Feed ingredients formulated in the diets are presented in Table 1. FSMTF was characterized by lower protein and ash; and higher lipid, NFE and salt (NaCl) than CFM. Tofu by-product, meize by-product, tapioca by-product, palm kernel and rice bran were lower in both protein and lipid but higher in NFE.

The formulation and proximate composition of the diets were presented in Table 2. The compositions of the formulated diets by analysis showed that the dry matter, crude protein, lipid, and energy of the FSMTF diets (FSMTF-50, FSMTF-75 and FSMTF-100) were similar to control diet, except ash which was lower and salt which was higher in FSMTF diets than control diet. Dietary salt concentration increased as the higher FSMTF in the diets; and the highest concentration was shown by FSMTF-100, 4.74%. This value was still within the tolerable level in the nemurus catfish diet as the level of reducing growth was > 5.22% [6]. The A/E ratio which was used as an indicator for amino acid balance in the fish diets [19, 4, 6] was also similar between FSMTF diets and control diet (Table 3).

Tabel 1. Feed ingredients and proximate composition

Ingredients	Proximat composition (%)				
	Protein	lipid	Ash	NFE*	NaCl
Fishmeal (FM)	62.18	9.99	18.74	9.09	2.76
Fermented salted marine trash fish (FSMTF)	59.68	13.57	12.90	13.86	11.03
Tafu by product meal	28.27	4.09	2.41	65.25	-
Rice bran	12.32	9.84	11.06	66.77	-
Meize by product meal	8.02	3.03	2.01	87.04	-
Tapioca by product meal	5.60	5.20	4.40	84.80	-
Palm kernel by product meal	14.19	9.6	3.6	72.61	-

*NFE (100-moisture-ash-protein-lipid)

Table 2 Diet formulation and proximate composition

Ingredients	Diet (%)				
	C[a]	FSMTF-50	FSMTF -75	FSMTF -100	COM[b]
Conventional fishmeal (CFM)	44	22	11	0	-
Salt fermented marine trash Fish (FSMTF)	0	22	33	44	-
Tafu by-product meal	16	16	16	18	-
Rice bran	24	24	24	23	-
Meize by-product meal	10	10	10	10	-
Tapioka by-product meal	2	2	2	2	-
Palm kernel by product meal	2	2	2	2	-
Palm oil	1.5	1	1.5	0.5	-
Vitamin and Mineral mix	0.50	0.50	0.50	0.50	-
Proximate composition by analysis (% dry matter)					
Dry matter	88.11	87.90	88.08	87.71	90.39
Protein	34.12	34.09	33.98	34.04	31.79
Lipid	15.97	16.97	17.13	17.46	6.71
Ash	5.65	3.06	2.21	1.35	8.62
NFE[c]	44.26	45.88	46.68	47.15	52.88
Energy (kcal DE/100g)	3.10	3.13	3.12	3.15	2.94
Salt (NaCl, %)	1.32	2.42	3.65	4.74	-

[a]C=Control; [b]COM= Commercial diet

Table 3 A/E ratio[a] (% sample) of experimental diets and fish

Essential amino acids	C	SFMTF-50	SFMTF- 75	SFMTF-100	*Nemurus catfish*
Histidine	9.45	9.09	9.13	11.09	7.27
Arginine	9.83	9.36	10.58	9.73	6.88
Threonine	7.55	7.21	8.44	6.44	9.73
Alanine	6.90	7.44	8.20	8.66	6.98
Valine	6.37	7.79	6.43	6.17	7.75
Methionine	8.10	8.71	7.59	7.84	7.07
Isoleucine	10.45	10.95	10.09	10.12	12.26
Leucine	16.72	16.10	17.27	16.10	18.05
Phenylalanine	10.62	10.29	9.09	10.28	9.37
Lysine	14.01	13.05	13.18	13.57	14.65
Tryptophane	ND	ND	ND	ND	ND[b]

[a]A/E= % Individual essential amino acid/Total essential amino acids
[b]ND=Not determined

3.2. Pelleted diets stability in the water

The pelleted diet stability in the water which was determined by percentage loss of dry matter (LDM) after emerging 10 and 30 minutes in the water was shown in Table 4. There was no difference in LDM value between FSMTF diets and FM diet (p>0.05), indicating that the inclusion of FSMTF in the diets did not significantly influence the stability of the pelleted diets in the water. LDM values of the C and FSMTF pelleted diets were 9.37-9.48% and 5.47-5.53% respectively for 10 and 11.68-11.76% for 30 minutes. These values were very stable as a minimum LDM value for catfish feed was <10% for 5 minutes [22, 23].

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Table 4 Diet stability values in the water (% loss of dry matter, LDM) for 10 and 30 minutes (triplicate data)

Pelleted Diets	Diet stability values (% LDM)	
	LDM-10 Minutes	LDM-30 Minutes
C	5.47±1.32[a]	11.76±1.33[a]
FSMB-50	5.52±1.44[a]	11.68±1.45[a]
FSMB -75	5.51±1.39[a]	11.71±1.35[a]
FSMB -100	5.53±1.48[a]	11.73±0.46[a]

Means in the same columns indicated by the same superscrips are not different (p>0.05)

3.3. Growth performance

There was neither difference in weight gain (WG), spesific growth rate (SGR), survival rate (SR) (Table 5) nor feed consumption (FC), protein consumption (PC), feed efficiency ratio (FER), protein efficiency ratio (PER) and protein utilization (PU) (Table 6) between fish fed FSMTF diets and C (p>0.05). FSMTF therefore could completely replaced CFM in the nemurus catfish diet. Weight gain and feed conversion and protein utilization usually highly correlated with nutrient quality of the feeds. In this study, protein, energy, amino acid profile and ratio of individual and total essential amino acid were similar between FSMTF and FM diet; so they gave similar effects on growth and feed conversion and protein utilization. Fermented salted marine trash fish in this study gave better weight gain, feed conversion and protein utilization as compared to the salted marine trash fish without fermentation [4, 5, 6]. The reason may be fermentation of added molases inprove the palatability of the salted trash fish diets by minimizing the salty taste of the salted diets. Increasing palatability of the diets formulated with molasses fermented ingredients [19, 2].

Compared to commercial diet (COM), FSMTF diet gave similar WG, SGR and SR values (p<0,05), but better FER, PER and PU values (p>0,05). Better feed efficiecy for FSMTF diets was probably due to higher protein and energy composition of FSMTF diets, which could fullfille optimum dietary protein and energy requirement for the fish, 34% protein and 3,25 kcal DE g^{-1} [1] as compared to COM (31% protein and 2.90 kcal DE g^{-1}).

Table 5 Growth performances

Growth performances					
DIETS	IW[a]	FW[b]	WG[c]	SGR[d]	SR[e]
C	49.75	91.46	41.71±8.88[a]	1.02±0.10[a]	100[a]
FSMTF-50	47.07	88.21	41.64±4.28[a]	1.05±0.05[a]	100[a]
FSMTF-75	41.29	82.67	41.67±2.97[a]	1.15±0.07[a]	100[a]
FSMTF-100	45.67	87.25	41.68±1.56[a]	1.08±0.12[a]	100[a]
COM	51.50	92.97	41.47±0.56[a]	0.98±0.01[a]	100[a]

[a]IW= Initial weight; [b]FW=Final weight; [c]WG=weight gain; [d]SGR=Specific growth rate;
[e]SR=Survival rate Means in the same columns indicated by the same superscripts are not different (P>0.05)

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Table 6. Feed utilization

DIET	Feed utilization				
	FC[a]	PC[b]	FER[c]	PER[d]	PU[e]
C	109.85±7.22[a]	37.48±2.46[a]	2.63±0.17[a]	1.12±0.15[a]	18.71±0.46[a]
FSMTF-50	110.49±5.07[a]	37.67±1.73[a]	2.77±0.14[a]	1.10±0.13[a]	18.05±3.11[a]
FSMTF-75	108.10±2.85[a]	36.73±1.21[a]	2.70±0.05[a]	1.13±0.06[a]	17.83±3.98[a]
FSMTF-100	108.73±3.86[a]	36.33±1.31[a]	2.68±0.03[a]	1.14±0.04[a]	17.89±0.59[a]
COM	117.57±2.69[b]	37.38±0.88[a]	2.84±0.03[b]	1.11±0.04[a]	14.90±1.25[b]

[a]FC=Feed consumption; [b]PC=Protein consumption; [c]FER=Feed efficiency ratio; [d]PER=Protein efficiency ratio; [e]PU=Protein utilization;
Means in the same columns indicated by the same superscrips are not different (P>0.05)

4. Conclusion
Total substitution of CFM with FSMTF in the nemurus catfish diets do not affect growth, feed conversion and protein utilization, therefore the FSMTF can completely replace CFM in the fish diets.

5. Acknowledgment
The authors wish to express their grateful acknowledgement to Universitas Riau for providing research grant.

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