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Effect of Replacing Fish Meal by Soybean-Blood Meal Mixture on Performance of Tilapia (Oreochromis Niloticus)

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Abstract:

Commercial fish feeds are highly expensive and unaffordable to majority of fish famers, usually fish meal which is scarce and expensive is used in compounding fish feeds. A fourteen-week study was done to determine growth response and gross margin in Nile tilapia fish fed on soybean-blood meal mixture as an alternative protein source to fish meal in formulated rations. Five treatment diets were formulated with each containing 30% crude protein using maize flour, wheat bran, sunflower oil, mineral-vitamins premix and soybean-blood meal mixture ingredients to substitute fish meal at five levels. Diet 1 was positive control with 0% soybean-blood meal mixture, Diet 2 with 25% fish meal substitution with soybean-blood meal mixture, Diet 3 with 50%, Diet 4 with 75% and Diet 5 (100% fish meal substitution) was negative control diet with soybean-blood meal as the main protein source. Three hundred healthy sex reversed (males) Nile tilapia fingerlings weighing (0.7±0.1g) were selected and randomly assigned into fifteen aquaria with twenty fingerlings in each. The five dietary treatments were fed to fish in triplicates at 5% of live weight for fourteen weeks. Ten fish were sampled from each aquarium every two weeks and weighed. The weight recorded was used in calculation of growth response parameters and gross margin. Fish group fed on Diet 4 grew bigger and heavier (53.2g)(p<0.05) compared to Diet 3 (47.8g), Diet 2 (44.6g), Diet 1 (40.5g) and Diet 5 (32.9g). Gross margin analysis based on feeds showed Diet 4 had highest (p<0.05) gross margin (Ksh. 253.14) followed by Diet 3 (Ksh. 196.28). Gross margin for Diet 2 (Ksh. 153.31) and Diet 5 (Ksh. 149.56) were statistically similar (p>0.05) while Diet 1 (110.71) recorded the lowest. Based on the results, soybean-blood meal mixture can be used to substitute fish meal up to 75% in formulated diets to get good returns from Nile tilapia fish farming.

Keywords: Growth response, fish meal substitution, soybean-blood meal

1. Introduction

Aquaculture is growing at a very high rate compared to other segments of food production theworld over, with current production standing at 76.6 million tonnes (FAO, 2017). The rapid development is attributed to increase in consumption of fish and its products with fish farming currently accounting for over 45 percent of all consumed sea food and is projected to reach 75% in 20 years (Zaki et al., 2016). Tilapia is among major species of fish farmed because of their high growth rate, ability to feed on different varieties of feeds, disease resistance and adaptation to tropical and semi tropical climate (Osama et al., 2008). These excellent attributes has led to tremendous growth in tilapia farming at 12% annual growth and is reported as the third most cultured fish group in the world after salmonids andcarps (Ogello et al., 2014). In developing countries, it has been farmed in many communities in rural areas mainly to provide protein which is most deficient nutrient among them (Liti et al., 2005; Sumi et al., 2014). In Kenya it is the most preferred fish species and accounts for 90% of total production of all farmed fish (Mbugua, 2008). Munguti et al., (2014) reported that Nile tilapia fish forms 75% of total farmed fish species in Kenya. Same trend was reported in fingerlings production where Nile tilapia fingerlings recorded highest figures of fingerlings produced in hatcheries in the country (Nyonje et al., 2018).

The rapid development in fish farming sector calls for quality and cost effective complete commercial fish feeds for sustainability in the sector. Despite increase in demand for commercial feeds to enhance fish farming, the feeds are highly expensive and identified as the major hindranceto rapid development in the sector (SDF, 2016; KNBS, 2017). Fish require high levels of protein in their diets and usually fish meal is used in large amounts as the main source of protein in compounding fish feeds (Agbebi et al., 2009). Prices of fish meal have doubled in recent years due to competition by human and for use in other livestock commercial feeds and dwindling supply from capture fishery resources available. hence its use in compounding fish feeds makes feeds highly expensive and unaffordable to majority of fish farmers (FAO, 2013). Search for cheaper protein sources as alternative to fish meal is the trend to equal the pace and sustainability in fish farming sector (Abou et al., 2013). Soybean meal is a by-production after oil extraction and is less expensive available nutritious plant origin protein feed. Blood is considered as a waste in many abattoirs, slaughter houses and slaughter slabs and contributes topollution of environment, therefore can be collected and processed locally into blood meal to be used in compounding fish feeds. The two protein feed ingredients are available in Kenya in adequate amounts and can be used in formulating less expensive fish feeds (Magondu et al., 2016). Combination of blood meal and soybean meal can provide less expensive alternative protein source to create economic space for fish farming sector to thrive. The study was geared towards establishing effects of fish meal substitution with soybean-blood meal mixture in formulated diets on growth response and gross margin in Nile tilapia fish.

2. Materials and Methods

The experiment was carried outat the hatchery of Fisheries and Aquatic Sciences Department, University of Eldoret. It issituated approximately 9 Kilometres along the Eldoret – Ziwa road in Eldoret – Kenya, altitude 2180 m above sea level, Latitude 0° 57' 02N'' and Longitude 35° 31'42E'' with average temperature of 23°C, Rainfall 900-1100mm per year and humidity range from 45% to 55% (Jaetzold and Schmidt, 2008). The area is in Uasin Gish County where main agricultural activities are wheat crop, maize crop, dairy cattle and fish farming.

2.1. Diet Formulation and Processing

Cattle blood was collected from Chwele Slaughter House and boiled at 100°C immediately in a container for 15 minutes. It was then drained, clots crushed by hand, dried in the sun for two days and milled into blood meal usinga hammer mill. Other feed ingredients were purchased from local animal feed shop in the market. These feed ingredients (maize grain, wheat bran, soybean meal and Rastrineobola argentae commonly known as "Omena" in Kenya in dry form) were ground separately using a hammer mill. Samples from each were taken to laboratory for proximate nutrient and amino acid analysis at Kenya Industrial Research and Development Institute and Fletcher Scientific Solutions Laboratories. Milled soybean meal and processed blood meal were first mixed at a ratio of one part soybean meal in two parts of blood meal in a mixer to produce soybean-blood meal mixture; its crude protein was calculated before being used to substitute fish meal for protein balancing. Based on the laboratory results, five treatment diets were formulated each with 30% crude protein using maize flour, wheat bran, sunflower oil, mineral-vitamin premix and soybean-blood meal mixture (1:2) to substitute fish meal at 0, 25, 50, 75 and 100% designated as Diet 1, Diet 2, Diet 3, Diet 4 and Diet 5, respectively (Table 1). Test feed ingredients provided 23% crude protein while the remaining 7% was from maize flour and wheat bran. Sunflower oil was added to boost oil content and energy levels to meet the requirement. The milled feed ingredients were weighed in proportions indicated in the formulas and mixed thoroughly in a mixer for three minutes. 25% clean water mixed with sunflower oil and mineral-vitamin premix was added and mixed again for three minutes to produce dough which was put in a pellet making machine to produce 3.5mm diameter floating pellets. Pellets were dried in the sun to a moisture content of less than 12% and packaged in air tight bags.

Feed Ingredients (%)	Formulated Dietary Treatments (% Fish Meal Substitution)					
	0	25	50	75	100	
Fish meal	38.27	28.7	19.14	9.57	0	
Soybean-blood meal	0	8.26	16.52	24.77	33.03	
Maize flour	20.73	21.84	22.94	23.96	24.97	
Wheat bran	39	39	39	39	39	
Sunflower oil	1	1.2	1.4	1.7	2	
Mineral-vit premix	1	1	1	1	1	
Total (Kg)	100	100	100	100	100	

Table 1: Diets Formulated at Five Levels of Fish Meal Substitution with Soybean-Blood Meal Mixture (0, 25, 50, 75 and 100%)

Nutrient	Formulated Dietary Treatments (% Fish Meal Substitution)						
Composition	0	25	50	75	100		
Dry matter	89.85±0.05 ^e	90.10±0.03 ^d	90.91±0.3 ^a	90.63±0.03 ^b	90.35±0.03°		
Crude	30.23±0.01 ^e	30.30±0.01 ^d	30.38±0.02 ^c	30.43±0.02b	30.5±0.02ª		
protein							
Crude fiber	3.95±0.02 ^e	4.13±0.02 ^d	4.32±0.02 ^c	4.51±0.02 ^b	4.7±0.02ª		
Ether	6.89±0.01 ^a	6.39±0.01b	5.9±0.01°	5.49±0.01 ^d	5.1±0.01e		
extracts							
Ash	9.52±0.01ª	8.12±0.01b	6.78±0.05 ^c	5.32±0.02 ^d	3.92±0.02 ^e		
NFE	39.25±0.09 ^e	41.16±0.04 ^d	43.54±0.03 ^c	44.87±0.04b	46.14±0.04 ^a		

Table 2: Proximate Nutrients Composition (%) of the Formulated Treatment Diets with Five Levels of Fish Meal Substitution (0, 25, 50, 75 and 100%) with Soybean-Blood Meal Mixture Values Presented as Mean ± SE and Values with Different Superscripts in the Same Row are Different Statistically (P<0.05)

2.2. Experimental Design

Fifteen aquaria (fish holding units) measuring 60 x 40 x 40 cm were cleaned with clean water and a disinfectant, dried in the sun and installed on a wooden stand in the fish hatchery. Each aquarium was filled with clean water, fitted to aerator (8 nozzles, CM@320w) through air stones and plastic tubule and a thermostat (AC-W23y) set at 28°C temperature. Dissolved oxygen (DO) and pH were measured twice a week using digital DO meter (YSI 550A USA) and digital pH meter (pHTestr20), respectively. Five hundred healthy sex reversed Nile tilapia (males) fingerlings were purchased from JEWLET Farm in Homa Bay County- Kenya and transported to the study site where they were acclimatized for fourteen days, three hundred fingerlings (0.7±0.1g) were selected and randomly assigned to five groups with twenty in each aquarium. The five formulated experimental diets were fed in triplicate to the five groups of fingerlings under Completely Randomized Design (CRD).

2.3. Feeding and Data Collection

Experimental fish were fed three times a day at 1000hrs, 1300hrs and 1600hrs at 5% of total average live bodyweight for the entire experimental period. Faeces at the bottom of the aquaria were syphoned every day at 0900hrs using plastic horse pipe by gravity and topped up with fresh clean water for the duration of the experiment. Fish sampling for various measures was done fortnightly and feed provided was adjusted basing on weight gain. At every sampling 10 fish were sampled from each aquarium using scoop net and put in a container with small amount of fresh clean water to reduce stress in handling. The sampled fish were weighed in grams using a weigh balance and length measured in centimetres using a ruler, after taking measurement the fish were returned to their respective aquaria. The amount of feeds fed was recorded throughout the entire experiment time. Weight in grams was used in calculation of growth response parameters as indicated below.

ADG- Average daily gain in gram per day

$$ADG = \frac{\text{Final body weight - Initial body weight}}{\text{Experiment time}}$$
Specific growth rate (SGR)
$$SGR = \left\{ \frac{\text{Log final body weight - Log initial body weight}}{\text{Experimental period}} \right\} X \ 100$$
Relative growth rate (RGR%) = $\frac{\text{Initial average fish weight x 100}}{\text{Final average fish weight}}$

Feed conversion ratio (FCR)FCR = $\frac{\text{Feed intake in grams}}{\text{Weight gain in grams}}$

2.4. Gross Margin

The cost of feeds was considered in this study for gross margin evaluation and other costs were assumed to be constant. The market prices of the feed ingredients were used in calculation of feed cost (Table 2). Gross margin was calculated by subtracting the cost of formulated feeds from estimated value of harvested fish.

- Gross margin = QP Cost of feed
- Q- Is the quantity of harvested fish in Kilograms
- P- Is the prevailing market price of fish per Kilogram in Kenya shillings (Ksh)

Price Per Kilogram (KSH.)
250
15
70
17
30
140
250

Table 3: Price of Feed Ingredients Used in Formulating Experimental Diets

2.5. Data Analysis

Analysis of Variance (ANOVA) was carried out on growth response parameters and gross margin analysis data to establish significant differences at p \leq 0.05 level of significance using SPSS statistical computer software version 22. Where there were differences, mean separation was performed using LSD (least significant differences) at p \leq 0.05 significance level. General Linear Model for the experiment was

 $Y_{ij} = \mu + T_i + \epsilon_{ij}$

Y_{ij}= Total observation on jth fish and ith treatment

 μ = the overall population mean

T_i = the effect due to soybean-blood meal mixture level (0%, 25%, 50%, 75% and 100%)

 ϵ_{ij} = is the error term

3. Results

3.1. Water Quality Parameters

Water temperature in the aquaria was maintained by the thermostats at $28 \pm 1^{\circ}$ C. Dissolved oxygen (DO) varied from 8.5mg/L to 8.7mg/L, this was supplied and maintained by the aerators connected to the aquaria. The pH ranged from 7.05 to 7.1 (Table 4). All measured water parameters were statistically similar (p>0.05).

Water Quality Parameter	Aquaria for Dietary Treatments (% Fish Meal Substitution)					
	0	25	50	75	100	
DO (mg/L)	8.7 ± 0.12 ^a	8.6 ± 0.10 ^a	8.6 ± 0.10 ^a	8.5 ± 0.09 ^a	8.5 ± 0.09 ^a	
рН	7.1± 0.04ª	7.06 ± 0.03 ^a	7.07 ± 0.03 ^a	7.05 ± 0.02 ^a	7.06 ± 0.02 ^a	
Temperature °C	28.3 ± 0.16 ^a	28.3 ± 0.16 ^a	28.1 ± 0.18 ^a	28.01 ± 0.18 ^a	28.2 ± 0.19 ^a	

Table 4: Water Quality Parameters, Dissolved Oxygen (DO) Ph and Temperature Values Presented as Mean ± SE and Values with Different Superscripts in the Same Row are Different Statistically (P<0.05)

3.2. Growth Response Parameters

Fish group fed on Diet 4 had highest (p<0.05) final average weight (53.2g) followed by Diet 3 (47.8g), Diet 2 (44.6g), Diet 1 (40.5g) and the least mean weight was recorded in Diet 5 (32.9g). Highest average daily gain (p<0.05) was recorded in Diet 4 (0.54g/day) compared to Diet 3 (0.48g/day), Diet 2 (0.45g/day), Diet 1 (0.41g/day) and Diet 5 (0.33g/day), respectively. Diet 4 was found to have highest (p<0.05) specific growth rate (SGR) of 1.91% while Diet 3 (1.87%), Diet 2 (1.83%), Diet 1 (1.79%) and Diet 5 had the least (1.71%). Slight difference (p<0.05) was realized in relative growth rate (RGR) with Diet 4 (98.7%) having the highest and Diet 5 having the least (97.9%). Feed conversion ratio (FCR) followed the same trend with Diet 4 (1.1) recording the highest (p<0.05) and Diet 5 (1.8) the lowest. Survival rates were high and statistically similar (p>0.05) for all the five diets (Table 5).

Growth	Dietary Treatments (% Fish Meal Substitution)						
Parameter	0	25	50	75	100		
Initial weight	0.71 ± 0.011 ^a	0.71 ± 0.010 ^a	0.70 ± 0.011ª	0.71 ± 0.010 ^a	0.70 ± 0.011a		
(g)							
Final weight	40.53 ± 0.17^{d}	44.60 ± 0.22 ^c	47.80 ± 0.30b	53.20 ± 0.34^{a}	32.90 ± 0.18 ^e		
(g)							
ADG (g/day)	0.41 ± 0.03^{d}	$0.45 \pm 0.00^{\circ}$	0.48 ± 0.00^{b}	0.54 ± 0.03^{a}	0.33 ± 0.00^{e}		
SGR (%)	1.79 ± 0.03^{d}	1.83 ± 0.03 ^c	1.87 ± 0.00^{b}	1.91 ± 0.00 ^a	1.71 ± 0.03 ^e		
RGR (%)	98.2 ± 0.03^{d}	$98.4 \pm 0.00^{\circ}$	98.5 ± 0.00^{b}	98.7 ± 0.00 ^a	$97.9 \pm 0.00^{\rm e}$		
FCR	1.4 ± 0.03^{d}	1.3 ± 0.03 ^c	1.2 ± 0.03^{b}	1.1 ± 0.03 ^a	1.8 ± 0.00^{e}		
Survival Rate	95 ± 2.9 ^a	93 ± 1.7 ^a	95 ± 2.9 ^a	97 ± 2.9 ^a	92 ± 1.7 ^a		
(%)							

 Table 5: Growth Parameters of Nile Tilapia Fed on Formulated Diets Values Presented as

 Mean ± SE and Values with Different Superscripts in the Same Row are Different

 Statistically While Same Superscripts Indicate Insignificant Difference (P<0.05)</td>

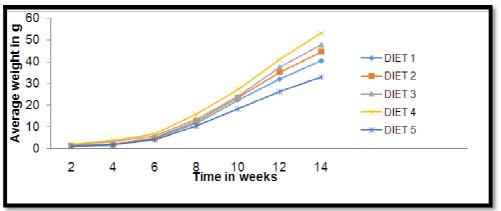


Figure 1: Growth Curves of Nile Tilapia Fish Fed Formulated Fish Meal Substitution with Soybean-Blood Meal Mixture Diets

3.3. Gross Margin Analysis

Diet 1 had highest (p<0.05) feed cost per kilogram (Ksh. 112.42) and total feed cost (Ksh. 120.29) which reduced with fish meal substitution levels with soybean-blood meal mixture (Table 6). Harvested weight and value of harvested fish increased (p<0.05) with fish meal substitution levels from 0 to 75% but dropped drastically in Diet 5 (100% fish meal substitution). Gross margin for Diet 4 (Ksh. 253.14) was highest (p<0.05) followed by Diet 3 (Ksh. 196.28). Gross margin for Diet 2 (Ksh. 153.33) and Diet 5 (Ksh. 149.56) were statistically similar (p>0.05) while Diet 1 recorded the lowest gross margin (Ksh. 110.71).

Parameter	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5
Feed cost/Kg (Ksh.)	112.42	91.86	71.33	50.88	30.43
Feed fed (Kg)	1.07 ± 0.03	1.05 ± 0.02	1.07 ± 0.03	1.09 ± 0.02	1.03 ± 0.02
Total feed cost (Ksh.)	120.29±3.9 ^a	96.45±1.8 ^b	76.34±2.5 ^c	55.46±1.0 ^d	31.34±0.6 ^e
Harvest weight (Kg)	0.770±0.02 ^d	0.833±0.01°	0.909±0.03b	1.03±0.02 ^a	0.603±0.01 ^e
Fish price/Kg (Ksh)	300	300	300	300	300
Value of fish (Ksh)	231.0±6.7d	249.77±4.1°	272.60±8.7b	308.60±5.6ª	180.9±3.5 ^e
Gross margin (Ksh)	110.71±2.9 ^d	153.31±2.3 ^c	196.28±6.2b	253.14±4.6 ^a	149.56±2.8 ^c

Table 6: Gross Margin Analysis for All Formulated Values Presented As Mean ± SE and Values with Different Superscripts in the Same Row are Different Statistically (P<0.05). Ksh. 105 = \$1

4. Discussion

Growth improved significantly with increase in fish meal substitution levels with soybean-blood meal mixture up to 75% (Diet 4) which recorded the highest growth rate and dropped drastically at 100% (Diet 5) with total fish meal replacement. This could be as a result of presence of important nutrients specifically digestible essential amino acids and lipids in soybean meal, fish meal protein and blood meal which formed a blend that met nutrient requirement of fish. For instance soybean meal contain linoleic fatty acids which when fortified with other nutrients especially from animal protein feed ingredients enhance Nile tilapia fish growth compared to fish meal alone which has arachdonic fatty acids (Takeuchi et al., 1983). Soybean meal was reported to have good amino acid profile close to fish meal and is highly digestible with apparent protein digestibility (APD) of 96% reported in Tilapia rendalli (Mothwa et al., 2013). Blood meal was also reported to have high dry matter digestibility compared to fish meal inLabao rohita fingerlings, apparent protein digestibility of 80% in Oncorhynchus mykiss (El-Haroun and Breau, 2007; Hussain et al., 2011).

High digestibility of soybean meal protein and blood meal may have also contributed to increase in growth rate with increase in fish meal substitution levels with soybean-blood meal mixture up to 75%. Results concur with reported 75% fish meal substitution level with blood meal without affecting growth performance in Mozambique tilapia by researchers (Agbebi et al., 2009; Bekibele et al., 2013). Kirimi et al., (2016) in their study reported that blood meal substituted fish meal up to 50% in Nile tilapia fish when blood was subjected to cooking at 100°C for 45 minutes. Exposure of blood to high temperatures for a long period of time during blood meal processing denatures proteins and reduces their bioavailability (Batterham et al., 1996). This may be the cause of lower growth rate with increase in fish meal substitution levels with blood meal than results reported in this study. It was noted that mixing plant protein feeds with animal protein feed ingredients enhances quality of compound complete fish feeds (Huet, 1994; Sadiku and Jauncey, 1995)

Total fish meal substitution with soybean-blood meal mixture resulted to drastic drop in growth performance. Nogueira et al., (2012) and Kirimi et al., (2016) also observed drop in growth performance with total fish meal substitution with blood meal in sea bream and Nile tilapia fish respectively. Poor growth performance reported may have been caused by imbalanced essential amino acids in the formulated diets as soybean meal protein and blood meal feed ingredients are known to have low levels of amino acid methionine (NRC, 1993). Crude fiber in total fish meal replacement diet was highest due to increased amount of soybean meal which had high amount of fiber. De Silva and Anderson, (1995) recommended crude fiber content range from 8% to 12% in tilapia diets and also reported that high fiber content in fish diets interfere with digestion of feeds leading to poor growth performance and increased feed conversion ratio. Though total fish meal replacement formulated diet had less thanrecommended amount of crude fiber, it may have played part in lowering digestibility of feeds hence poor growth performance and high feed conversion ratio realized in the study. There could be animal growth factors in fish meal that are not present in soybean meal protein and blood meal feeds that when fish meal is not part of the fish diet causes sharp drop in growth performance. Growth curves indicated low growth rate at the initial stage up to week six after which there was an exponential growth rate for all treatment diets (Figure 1). This may be associated with acclimatization to new feed types. High growth rate of tilapia fish is realized at temperatures ranging from 28°C to 36°C, pHfrom 7 to 9 and dissolved oxygen at concentration greater than 3mg/L (Ross, 2000; Boyd, 2004; FAO, 2012). The water parameters were within recommended range for optimum biochemical reactions in digestion and metabolic utilization of feeds to enhance growth rate.

Survival rate was high ranging from 92% in Diet 5 to 97.7% in Diet 4 and statistically similar (p>0.05) for all diets which were slightly lower than results of 98% reported by Kirimi et al., (2016) when Nile tilapia of initial weight (12g) were fed on fish meal substitution with blood meal formulated diets. They also reported high and statistically similar survival rate in their study where fish meal was substituted at 0, 50 and 100% with blood meal. Results were lower in this study because the fingerlings were younger and smaller (0.7g) initial weight which are more susceptible to stress than bigger fish. The high rate of survival may be as a result of proper management and handling practices that minimized fish stress. Water pH was maintained by syphoningfaeces at the bottom of aquaria every day and refilling with fresh clean water. Dissolved oxygen was supplied and maintained by water aeration while thermostats maintained optimal water temperatures. During sampling fish were scooped and placed in a container with some amount of fresh clean water. Eyo and Olatunde, (1999) reported high mortality in fish group fed on high blood meal levels in their study. It was not clearly understood as to what caused high mortality in fish. Results reported agree with findings by Agbebi et al., (2009) in their study in which blood meal completely replaced fish meal in juvenile cat fish (Claria gariepinus). 100% survival rate was reported in an experiment in which blood meal totally substituted fish meal in Nile tilapia fingerlings (Aladetohun and Sogbesan, 2013).

Gross margin analysis of the feeds used showed high returns in Diet 4 and low returns realized in Diet 1. It was established that 75% fish meal substitution with soybean-blood meal mixture in compounding complete fish diets is more profitable than to use fish meal as a single protein feed ingredient in Nile tilapia farming. Kirimi et al., (2016) reported high incidence cost in fish meal based formulated diets, low incidence cost and high profit index in blood meal formulated diets in Nile tilapia. Blood is considered a waste in abattoirs, slaughter houses and slaughter slabs in Kenya and often leads to pollution of the environment. It was collected free of charge, some little money was paid to people who assisted in blood collection. Soybean meal was less expensive than fish meal therefore combination of the two resulted to low cost formulated feeds hence contributing to high gross margin realized.

5. Conclusion

Combination of soybean meal protein and blood meal feed ingredients as alternative protein source to fish meal in formulated diets enhances growth of Nile tilapia. However, based on results in this study it was noted that fish meal substitution with soybean-blood meal mixture up to 75% provides good results and total substitution drastically reduces growth. 75% fish meal substitution level with soybean-blood meal mixture has high gross margin and therefore more profitable than use of fish meal as a single protein ingredient in formulating diets for Nile tilapia fish.

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