# Review of: "The Growth Performance of Nile Tilapia (Oreochromis Niloticus) Fed Low-Cost Fish Feeds Formulated From Fish By-Products, Fishery By-Catch and Pig Blood-Meal"

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Potential competing interests: No potential competing interests to declare.

The Growth Performance of Nile Tilapia (Oreochromis Niloticus) Fed Low-Cost Fish Feeds Formulated From Fish By-Products, Fishery By-Catch and Pig Blood-Meal

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Declarations

### Abstract

A study was carried out to investigate the growth performance and nutrient utilization of *Oreochromis niloticus* fingerlings fed seven fish diets of 35% crude protein formulated by replacing fishmeal as a protein source at 0%, 50% and 100% using fish by-products, fishery by-catch, and pig blood meal. Twenty fingerlings average weight 00.6  $\pm$  0.02g were stocked per aquarium and replicated three times. Feeds with 35% crude protein were administered at 5% body weight twice a day.[DO1] [DO2] Growth performance of fish was highest in fish fed with 100% by-catch. Significant differences (p<sup>c</sup>0.05) in growth performance of *Oreochromis niloticus* subjected to different feed treatments were reported. Fish fed with 100% by-catch and 100% by-products diets showed higher growth performance than those fed other feed types. This study showed that the inclusion of by-catch and by-products at 100% could replace fishmeal in Nile tilapia diets. In terms of cost, 100% blood meal was the cheapest followed by 100% by-catch and 50% by-catch respectively and the commercial fish feed was the most expensive. Locally available low-cost fishery by-catch, fishery by-products and pig blood meal can be used to replace the expensive commercial feed.[DO3]

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#### Introduction

Aquaculture is an important source of food, nutrition, income and livelihoods for many people around the world (FAO 2022). The growth of aquaculture production in the last two decades has boosted the average consumption of fish and fish products at a global level. Nearly half of the fish being consumed by humans worldwide are produced by aquaculture (FAO 2022). In 2012, aquaculture production overtook global beef production in quantity for the first time (Larsen and Roney 2013). This development in global fish supply from aquaculture has increased the demand for fish feed resources, especially for high quality protein and high-quality lipid feed resources such as fish meal and fish oil (Naylor et al. 2009). Feed cost is now the major constraint in aquaculture and the price of fishmeal has been estimated to account for between 60-80% of the cost of intensive aquaculture operations (Rana et al. 2009). The price of fishmeal has increased greatly in the last two decades due to the rising demand of farmed fish products (Hardy 2010), thereby threatening the viability of small-scale aquaculture enterprises. For many small-scale fish farmers in Zimbabwe, the cost of commercial feed is often too high.

For fish farming to be successful, there is need to provide suitable and cost-effective fish feed (Delgedo and Minot 2003). This, in turn leads to the search for alternative highly nutritious feed ingredients in aqua-feeds (Hardy 2010). A substantial amount of research is already underway, testing potential protein sources that can replace fishmeal in tilapia diets. Oreochromis niloticus is naturally accustomed to eating plant-based feeds since it isprimarily herbivorous, with aquatic macrophytes, algae, and diatoms generally comprising >90% of its diet. Selection of ingredients for the formulation of fish feed is very crucial and these ingredients should be cheap and available in the local market (Zamal et al. 2009). Among cheaper proteins are plant proteins, single cell proteins, earthworm meal, insects, blood meal, fishery by-catch and fishery by-products which have high lipid contents. Several studies on alternative sources of cheap protein have been conducted (e.g. Li et al. 2004; Herring et al. 2005; Aladetohun and Sogbesan 2013; Ghaly et al. 2013; Tran et al. 2015; Rustad 2016). Despite some interesting findings, commercializing these feed formulations may be a challenge due to limitations on the quality of the feed. In order to successfully introduce semiintensive fish pond management, alternative feeds can be developed from locally available resources such as fishery bycatch, fishery by-products and pig blood meal which are affordable and easily available. Industrial processing of fish and capture fisheries generates huge quantities of by-products and by-catch. Fish by-products are largely constituted of viscera which have similar nutritional value to that of fish meal which is currently used in commercial aquafeeds (Ju et al. 2013).

Although the aquaculture industry is still a growing sector in Zimbabwe, it is worth noting that Zimbabwe is one of the top ten fish farming countries within the Sub-Saharan Africa (FAO 2022). Currently, the estimated total production from fish farming in Zimbabwe is at 10 600 tons and this is mainly from Lake Harvest Ltd. Growth and expansion of the aquaculture industry in Zimbabwe is mainly limited by the cost of fish feed. However, with the continued decline in capture fisheries

production, there is need to improve and boost aquaculture production so as to meet the ever-increasing demand for fish and fish products. The rapid growth of human world population, increase in food prices, improved distribution channels, rising incomes, urbanization and search for a cheaper source of protein has also increased the demand for fish consumption (FAO 2022). In Zimbabwe, the demand for fish and fish products has also increased putting more pressure on the capture fisheries.

In this study, we demonstrate that using cheap and locally available fish products is perhaps one of the most realistic options. The hypothesis is that these feed formulations will adequately meet the protein requirements for fish just as commercial feeds but at substantially lower cost. We therefore evaluated the suitability of by-catch, by-product meals (guts and skins) and pig blood meal at different inclusion levels in diets for *O. niloticus*. *Oreochromis niloticus* is the main species that is farmed successfully in Zimbabwe. It is favoured because of its fast growth rate, high resistance to diseases, ability to survive at low oxygen levels and ability to feed on wide range of foods (Azaza et al. 2010). In addition, the species can also tolerate higher dietary fibre and carbohydrate concentrations than most other cultured fish species (El-Sayed and Teshima 1992). In this experiment, growth, survival, feed intake, protein efficiency ratio and feed conversion ratio were measured in fish fed with various formulated diets.

#### Study Area

Feed preparation was done at the University Lake Kariba Research Station (ULKRS) in Kariba, Mashonaland west province, Zimbabwe. Kariba is found within the Agro-ecological region V and the mean annual rainfall is below 650 mm. Temperature averages are between 20°C and 27°C. The feeding experiment was done at the University of Zimbabwe Department of Biological Sciences, in Harare, Zimbabwe. Harare is in Agro-ecological region IIA. This area is situated in the Highveld plateau of Zimbabwe with subtropical highland climate. The average annual temperature is 17.95 °C and the average annual rainfall is about 825 mm. The experiment was carried out in a greenhouse so as to maintain warm temperatures during the day[DO4].

## Materials and Methods

#### Experimental setup and design

The experiment was done in a completely randomized design with eight treatments. It was carried out using 30 litre rectangular aquariums placed in a greenhouse to maintain constant temperatures[DO5]. These tanks were aerated throughout the whole study with aquarium air-pumps (Tetratec APS 150, Germany) to replenish the amount of dissolved oxygen in the water. A total of 160 *O. niloticus* fingerlings were used in this study with a stocking rate of 20 fish per aquarium. The average initial body weight of the fingerlings used in this study was  $0.6 \pm 0.02g$ . These fingerlings were sourced from Lake Harvest Aquaculture Company in Kariba, Zimbabwe. Before the start of the experiment, the fingerlings were left to acclimatize in aquariums for one week. After the acclimatization period, the fingerlings were selected randomly and weighed before they were stocked in aquarium tanks. The fish were conditioned to the rearing environment for 48 hours without feeding prior to the commencement of the experiment. Siphoning a portion of water from each aquarium was done every day for excreta removal and then replaced with an equal volume of water. Feed was fed to the fish in the

form of pellets at 5 % body weight twice a day The experiment was done for a period of 11 weeks. to ascertain whether there were any statistical differences in growth performance of fish fed formulated feeds and commercial feed.[DO6]

## Water Source and Water Quality Parameters

Borehole water from the University of Zimbabwe main campus was used in this study. For the purpose of controlling disease-producing organisms, the borehole water in this experiment was mixed with municipal chlorine treated water. The water was then aged for 24 hours in 20-litre buckets with vigorous aeration in order to remove chlorine from the water. The water in the experimental aquariums was maintained with the same condition for the whole period. Water quality parameters namely turbidity and total suspended solids were measured using the Portable Data logging Spectrophotometer Hach DR / 2010. Conductivity, Dissolved Oxygen, pH, Salinity, and Total dissolved Solids were measured using handheld pH/ Oxygen/ Conductivity Meters equipped with a CellOx 325 Oxygen Sensor (WTW), A SenTix 20 probe (WTW) and a Conductivity sensor (WTW). Analysis was done on filtered samples to measure ammonia nitrogen, nitrate nitrogen (NO<sub>3</sub>-N) and orthophosphate phosphorus (PO<sub>4</sub>-P) according to Bartram and Ballance (1996).

#### Collection of fishery by-products, fishery by-catch and blood meal

The fishery by-products (skins and guts of *O. niloticus)* were supplied by Lake Harvest (Zimbabwe). Fishery by-catch was collected from the fishing co-operatives around Lake Kariba, Zimbabwe. The three species of fish used in the feed formulation are *Synodontis zambezensis*, *Clarias gariepinus* and *Hippopotamyrus discorynchus*. Dry pig blood meal was purchased from Colcom Pvt. Ltd.

#### Microbiological sample collection and analysis

Fish by-products, by-catch, pig blood and the formulated feed were analysed for total coliforms and *Escherichia coli*. Aseptically, fish by-products (skins and guts), by-catch, pig blood were individually transferred into sterile plastic bags and immediately put in ice for preservation purposes. Commercial feed samples and formulated feed were taken using sterile scoops and held in sterile bags on ice until examined. Samples were separately ground into smaller pieces using a blender that had been disinfected. A sample was taken from each and was stored in standard stomacher bag that was sterile and containing distilled water. The mixture was homogenised and blended in a stomacher for 60 seconds. All samples from fish by-products, by-catch, pig blood meal and feed were inoculated on MacConkey broth and Brilliant green bile broth designed for recovery of total coliforms and *Escherichia coli* respectively.

#### Feed ingredients

Mineral and vitamin premix were added to the feed. Samples of fish feed ingredients were collected and analysed for DM[DO7], Moisture, Crude fibre, ether extract, Ash, Nitrogen free Extract and Protein Content according to AOAC (2000).

#### Diet Formulation, Preparation and Composition

Seven iso-nitrogenous diets of 35% crude protein for *O. niloticus* were formulated using the Animal Feed Optimization Software (AFOS). The ingredients were used to replace soya bean meal in the formulated diets as follows: 100%

Fishmeal (diet 1), 50% blood meal (diet 2), 100% blood meal (diet 3), 50% by-products (diet 4), 100% by-products (diet 5), 50% by-catch (diet 6) and 100% by-catch (diet 7). A certified commercial diet which was bought from a local fish feed producing company was used as the [DO8] positive control diet.

The fishery by-product meal and fishery by-catch meal were prepared by drying the them for 3 days at 60°C in a circulating-air oven. The dried ingredients were ground to a fine powder, homogenized, and immediately used in the feed preparation. The feeds were prepared by mixing the macro-ingredients first which included fish meal/ blood meal/ by-products meal/ by-catch meal with soybean meal, maize meal and wheat bran before adding oi[DO9] I. Micro-ingredients which included di-calcium phosphate, ascorbic acid and fish oil were then added each at 1.5%, 0.5% and 1%, respectively. The binder [DO10] was added at an inclusion rate of 2% to increase pellet firmness, improve stability in water and also as a way of reducing the amount of fines produced during handling and processing.

Pellets were made by adding 350ml of water per kg of the diet and the mixture was stirred to form a stiff paste with a plastic consistency when compressed. The dough was passed through a pellet making machine made by SBK Engineering (2014). The pellets were allowed to dry away from direct sunlight until they were firm. The dry pellets were stored in a well-ventilated room.

## Proximate Analysis

Samples of the formulated fish feeds were analysed for DM, Moisture, Crude fibre, ether extract, Ash, Nitrogen free Extract and Protein Content according to AOAC (2000).

## Determination of Energy Value

The energy value of the diets was determined by multiplying the protein content by 4, carbohydrate content by 4 and fat content by 9 (AOAC, 2000)

Energy Value = (Crude Protein x 4) + (Total Carbohydrate x 4) + (Crude Fat x 9)

#### Measurement of Biological Parameters

Fish growth was monitored through weekly sampling. A digital scale was used to weigh each fish during the experiment. At the end of the study, fish were harvested and weighed to obtain a final mean body weight. The growth performance parameters calculated include total weight gain (TWG), specific growth rate (SGR) (%/day), feed conversion ratio (FCR), protein efficiency ratio (PER), Feed Intake (FI) and percentage survival rate (%SR).

The following formulae were use;

- Mean Weight Gain (MWG) = Final Mean Weight Initial Mean Weight
- Specific Growth Rate (% SGR) = In (Mean Final Weight) In (Mean Initial Weight) g x 100/Time (days)
- Total Feed Intake (FI) g = Total dry feed fed (g/fish)
- Feed Conversion Ratio (FCR) = Weight of Feed Fed/ Weight gain of fish
- Protein Efficiency Ratio (PER) = (% Protein in the diet x weight of diet consumed)/100

• Percentage Survival Rate (% SR) = (Total number of fish – Total number of Dead fish / Total number of fish) x 100

#### Cost Benefit Analysis

The economic performance of each diet was evaluated using the partial enterprises budgets. The variable costs included the costs of the ingredients that were used to formulate the diets. These included the market costs of soya meal, fish meal, blood meal, fish skins, fish guts, by-catch, maize meal, wheat bran, binder, ascorbic acid, fish oil and calcium.

#### Statistical analysis

All data were captured and recorded in Microsoft excel for subsequent analysis. The data did not meet assumptions for parametric tests (e.g. Normality, Homogeneity of variance). As a result, all data on proximate analysis, growth performance, feed intake, feed conversion ratio and protein efficiency ratio were analysed using the Kruskal-Wallis ANOVA test. Mann-Whitney pairwise comparisons were used to determine if there were significant differences between treatments.

#### Results

#### Proximate Composition of individual feed ingredients

In this study, fish skins had the highest crude protein (74.2%), followed by fishmeal (70.5%) while maize meal had the lowest crude protein of 7.5% (Table 1). Fish guts had the highest fat content 33.8% while by products and by catch had 22.8% and 16.79% respectively. Crude fibre was highest in wheat bran 11.2%, followed by fish meal (6.5%), and fishery by-products with 0.6%. Ash was highest in by-catch (15.6%) followed by fishmeal (12.9%) while blood meal and by-product meal had 11.1% and 3.6% respectively. Maize meal had the lowest ash content (1.4%). Dry matter was highest in soya meal (95.6%), followed by fish skins (94.8%) and wheat bran (88.8%). The nitrogen extract free (NFE) was highest in maize meal (80.1%), followed by soya bean meal (52.0%), and fish skins with 1.9%.

#### Proximate Composition of experimental diets

Table 2 shows the nutrient composition of experimental diets. Fat was highest in 100% by-products diet (28.0%), followed by 50% by-products diet (27.7%) with the fish meal diet having the lowest fat content of 8.3%. Hundred percent fish meal and 50% by-catch diets had the highest amount of crude fibre (5.2%), followed by 50% by-products diet (3.3%), while the 100% by-catch diet had the least amount of crude fibre (0.6%). Ash was highest in 100% by-catch diet (18.9%) and least in the 50% by-catch diet (4%). The dry matter of the diets ranged between 90.8% and 93.3%. The control diet had highest dry matter of 93.3% with a low moisture content of 6.7% while the 100% blood meal diet had the lowest dry matter of 9.2%. Nitrogen free extracts (NFE) was highest in the 100% fish meal diet (40.3%) and was least in the 50% by-product diet (16.6 %).

#### Energy levels in the diets

The highest energy values were found in 100% by-products diet (22.21KJ/g) and the least energy value was obtained in

100% blood meal diet (14.29 KJ/g) (Table 3).

#### Water quality parameters

Table 4 shows the weekly water quality parameters. Temperature ranged from  $16-32^{\circ}$ C. The mean values for dissolved oxygen ranged from 5.5 to 6.7 mg/l. pH in this study ranged from 7.5 to 8.3 while ammonia ranged between 0.077 - 0.103 mg/l in the treatments.

## Total Feed Intake and Feed Utilization

There were overall significant differences (p<0.05) in total feed intake of fish fed the experimental diets (Table 5). The highest total feed intake on formulated diets were observed in fish fed 100% by-catch diet and the control diet (commercial feed), both with a total feed intake of 8.2g (Table 5). The lowest total feed intake (6.6g) was observed in 100% fish meal diet.

The best FCR was obtained from fish fed 100% by-catch (4.9), followed by 100% by-products (5.0), and the least FCR was for fish on the 100% fish meal (6.2). There were significant differences (p<0.05) in FCR of fish that were fed experimental diets.

There were significant differences (p<0.05) in protein efficiency ratio in fish that were fed the experimental diets. The 100% by-catch diet had the highest protein efficiency ratio (2.8), followed by 50% by-products (2.7), while the 100% fish meal diet performed poorly (2.3). The 100% by-catch diet was the best diet and was not significantly different ( $p^{\circ}0.05$ ) from the commercial control diet which had a PER of 2.9. PER increased with a decrease in crude fibre, fat and nitrogen free extract content in the formulated feeds.

#### Growth performance of fish

Table 6 shows growth performance of *O. niloticus* fish fed from the experimental diets. There was a significant difference  $(p^{\circ}0.05)$  in growth performance of fish fed from different experimental diets (Table 6). Fish that were fed 100% by-catch diet attained more weight compared to all other formulated diets with a mean weight gain of 1.70 ± 0.42g, while fish on the commercial control diet had a mean weight gain of 1.78 ± 0.21g (Table 6). Notably, the control diet was not statistically different from the 100% by-catch and 100% by-products diets in terms of weight gain (Table 6).

Specific growth rate of the fish that were fed experimental diets were significantly different (p<0.05). 100% by-catch had the highest specific growth rate of 1.23% per day followed by 100% by-product diet. Hundred percent fishmeal had the least specific growth rate of 0.92%. There was no significant difference (p>0.05) between the 100% by-catch diet and the control diet (commercial diet) which had a specific growth rate of 1.26%. Both the control diet and the 100% by-catch diet were significantly different (p<0.05) from all the formulated diets.

The growth curves of *O.niloticus* for the whole experimental period are illustrated in Figure 1. The separation of the different treatments became apparent during the second week of the experimental period. At the end of the first week, growth of fish were almost the same but after eleven weeks of culture, the growth curves for all the treatments had not

levelled.

 Figure 1. Growth (weight gain) of Oreochromis niloticus raised in aquariums on different
 formulated diets for 11

 weeks
 Figure 1. Growth (weight gain) of Oreochromis niloticus raised in aquariums on different

## Cost benefit analysis of experimental diets

Feed cost decreased as the replacement levels of blood meal and by-catch increased. The 100% blood meal diet was the cheapest (US\$0.53 cents/kg), followed by 100% by-catch costing US\$0.61 cents/kg and 50% blood meal which cost US\$0.64 cents/kg (Table 7). The price for 100% by-products was high, at US\$0.93 cents. Commercial diet was the most expensive and it costed US\$1.70 /kg.

## Discussion

The results for the proximate analysis of the individual feed ingredients revealed that fish skins had the highest protein content. Several studies have also shown that crude protein of fishery by-products such as skin, frames, guts, bones, heads and tails contain high amounts of protein and high levels of essential amino acids (Venugopal et al. 2008). The findings of this research are in agreement with other studies since the crude protein content of fishery by-products ranged from 33.27 – 74.20% [DO11] in this study. The fish skin is known to be very rich in protein which ranges from 15-75% (Arnesen and Gildberg 2006; Jung et al. 2006). Fish guts had a high fat content and this was due to peritoneal fats and this is confirmed in other studies that found catla visceral waste to range from 12.80 - 68.21% (Bhaskar et al. 2008). Cultured fish use proteins and lipids primarily for energy needs and carbohydrates secondarily. Lipid levels for best fish growth is between 9-17% fat (Kim and Mendis 2012), High fat levels affect quality, flavour, storage and cause fat deposition (Cowey 1993). Fat content of commercial feed and 100% by-catch were within the required range for best fish growth. Findings of this study are in agreement with Kim and Mendis (2012) since commercial feed with a fat content of 14.24% yielded highest growth followed by by-catch with 14.24% fat. High lipid content in fishery diets is an indication that proper storage at room temperature is essential since this will slow down rancidity. The high ash content in fishery bycatch meal is mostly contributed by bones and these include head, fin, frame and tail (Batista et al., 2010). In other studies fishery by-products such as visceral waste, cod head (Arnesen and Gildberg 2006) and catfish frame (Amiza et al. 2011) showed relatively high ash content (2.50, 7.08 and 6.8%).

The main objective of this study was to determine if selected fish meal replacements could be used to formulate low-cost fish feeds as a means of addressing the unsustainable cost of commercial feeds. Growth, feeding rates and FCR are the three major variables for assessing fish productivity in an aquaculture enterprise. The relationship between these variables should be understood since this is fundamental in optimizing fish production (Nadir et al. 2007). Results showed that diets formulated with 100% by-catch and 100% by-products can effectively replace commercial feed as well as commercial fish meal, in fish feed formulation.

The results showed that feed intake was highest in fish fed commercial control diet, and where soyabean meal was replaced by 100% by-catch and 100% by-products and this could be due to the fact that these feeds contain dried fish guts and other viscera, which were reported to improve the free amino acids of the diets (Tabinda et al. 2013). The

presence free amino acids are known to improve the flavour and taste of these diets, thus a higher feed intake by fish fed these diets (Gupta et al. 2013). Gupta et al (2013) and Giri et al. (2010) also found that the replacement of fish meal could be partially done using dried fish guts without negatively affecting the growth and feed conversion ratio. These have similar nutritional value like that of fish meal which is currently used in aqua feeds (Chotikachinda et al. 2013; Ju et al. 2013). Fish viscera also contain well balanced proteins, highly digestible, significant quantities of long chain lipids, unsaturated n-3 fatty acids including taurine and other growth factors. Many studies have revealed satisfactory feed utilization responses and growth by replacing fishmeal with viscera in the diets of *O. niloticus* (Hernandez et al. 2014; Mammauag and Ragaza 2016). However, the fish in this experiment were not feeding well because of sometimes lower than average temperatures and this resulted in lower average mean weight gain. Feed intake depends on biotic and abiotic factors such as size of fish, temperature and the quality of the feed. It was also observed that total feed intake also increased with an increase in ash content. Ash content is an indication that the feed has minerals that are necessary in the diet for aquaculture species. High ash above 20% cannot be utilized well by *O. niloticus* (Azaza et al. 2010).

The best FCR which range from 1.5 to 2 are obtained at an optimum temperature of  $3\theta$ C for *O.niloticus* (Craig 2009; Delong et al. 2009). A lower Feed Conversion Ratio (FCR) indicates better utilization of the fish feed. The FCRs in this study were higher than 2 and this may be due to the fact that the experiment was done in winter. The fish did not feed well because the temperatures were lower ranging from 16 [DO12] –  $32^{0}$ C. *Oreochromis niloticus* feed normally at temperatures  $25 - 30^{0}$ C. These findings are in agreement with El-Saidy and Gaber (2004) who also reported that high fibre content can reduce the digestibility of ingredients in the diet leading to low palatability of feed and poor growth of fish. 100% fish meal diet had the highest crude fibre of 5.26% and was the least in terms of growth performance. In this study 100% by-catch had the lowest crude fibre and showed the highest growth rate.

Protein Efficiency Ratio (PER) was high in 100% by-catch and 100% by-products diets because crude fibre was low. As a result, these feeds had better digestibility and hence the proteins promoted growth. The PER results of this study were similar to those that were reported by Gupta et al. (2013). Workagegn et al. (2014) reported that high levels of dietary fibre level reduce growth performance, feed and protein utilization in fish. Fats in 100% by-catch and 100% by-products diets were also within the acceptable range to promote fish growth.

The mean initial weight indicated that the fish were of uniform size. The initial weights show that the grading procedure done prior to stocking in the aquariums was efficient. Rodney and Confred (2015) reported that in aquaculture, initial stocking size is an important factor which affects the culture period, feed conversion, and final weight. It is generally accepted that weight gain could be used as a parameter in evaluating different levels of nutrient intake (protein level) and thus the nutrient requirement of the fish. Mean final weight was highest in 100% by-catch followed by 100% by-products.

The control diet in this study was the commercial diet which was produced by a local company and is the one which is mostly being used by fish farmers in Zimbabwe. The control diet contained the same ingredients as those used in the formulated diets. It contains maize meal, wheat bran, soya meal, fish meal, blood meal, animal and fish products and also minerals, vitamins and binder. Fish that were fed 100% fish meal had the least mean weight gain. The highest specific

growth rate was observed in fish fed on diet with 100% by-catch, 100% by-products and the control diets due to the high protein content, the lower crude fibre content (which increased the digestibility of diets), and the lipid content which was within the acceptable range (12-19%) for the growth of *O. niloticus*. The 100% by-catch and control diets had the same amount of lipids (14.24%) and growth rate was lower for the 100% by-products diet and this could be attributed to the high lipid content (2N 8.09%). The 100% fish meal diet had low specific growth and this may be due to low fat content and high crude fibre content.

The aim of aquaculture business is to maximise profit. The most expensive formulated diet was 50% by-products while the cheapest was 100% blood. The price of the commercial diet was the highest compared to all the formulated diets. Fish by-products are readily available where fish processing is done and fish by products can be value added by using them to make fish feed. Wastes disposal from the various stages of the fish processing can lead to pollution of the environment and also attracting scavengers and spreading disease (Caruso 2015). This can be avoided by utilizing these fish processing waste by-products to make fish pellets thus making use of valuable resources that are often not considered of economic value. Further studies are required to do the amino acid profile of the ingredients and the diets, to improve the diets and to determine long term effects on growth performance of fish fed fishery by-catch and by-products under on-farm conditions.

## Tables

	PARAMETERS	%				
Feed Ingredients	Crude Protein	Fat	Crude Fibre	Ash	Dry Matter	N.F.E[DO13]
Fish guts	33.27 ± 1.63	33.84 ± 1.22	$2.65 \pm 0.04$	3.36 ± 0.11	93.71 ± 0.72	20.59 ± 1.51
Fish skins	74.20 ± 0.28	18.03 ± 0.14	$0.67 \pm 0.09$	5.12 ± 1.06	94.84 ± 0.69	1.98 ± 0.04
Blood meal	59.52 ± 0.75	1.69 ± 0.01	1.94 ± 0.22	11.11 ± 1.26	62.43 ± 2.23	25.74 ± 0.62
By-product	65.91 ± 1.68	22.84 ± 0.81	0.66 ± 0.07	3.60 ± 0.71	90.65 ± 2.06	7.64 ± 0.35
By-catch	63.55 ± 1.2	16.79 ± 0.62	2.11 ± 0.02	15.60 ± 0.57	94.08 ± 2.47	3.03 ± 0.73
Fish meal	70.58 ± 1.48	3.13 ± 0.20	6.54 ± 0.55	12.99 ± 0.02	91.12 ± 1.75	6.76 ± 0.51
Maize meal	7.55 ± 1.45	3.45 ± 0.29	1.47 ± 0.11	1.40 ± 0.28	94.00 ± 1.24	80.13 ± 1.58
SBM	31.79 ± 2.19	1.39 ± 0.23	5.18 ± 0.67	5.17 ± 0.38	95.60 ± 1.30	52.07 ± 1.93
Wheat bran	18.60 ± 1.41	30.18 ± 0.44	11.26 ± 1.61	5.30 ± 0.42	88.82 ± 0.97	23.48 ± 0.95

Table 1. Mean (± SD) proximate composition of feed individual ingredients

\*NFE = nitrogen free extract; SBM =Soy Bean Meal

	PARAMETER %	6				
FEED	Crude Protein Protein	Fat	Crude Fibre	Ash	Dry Matter	N.F.E
Fish meal 100%	35.69 ± 0.59	8.39 ± 1.00	5.26 ± 1.06	7.89 ± 0.50	91.57 ± 0.76	40.34 ± 1.10
Blood meal 50%	35.81 ± 0.43	24.66 ± 0.70	2.66 ± 0.35	6.94 ± 0.16	91.14 ± 1.42	21.07 ± 1.50
BM 100%	35.56 ± 0.78	21.74 ± 1.00	1.31 ± 0.35	5.95 ± 1.20	90.81 ± 0.73	26.25 ± 0.37
BP 50%	35.38 ± 1.08	27.72 ± 0.86	3.33 ± 0.93	8.50 ± 0.69	91.62 ± 0.75	16.69 ± 0.59
BP 100%	35.56 ± 0.83	28.09 ± 1.33	1.33 ± 0.17	8.97 ± 0.21	91.62 ± 0.75	33.92 ± 1.37
By-catch 50%	35.58 ± 0.78	12.88 ± 1.20	5.26 ± 0.47	4.00 ± 0.13	92.59 ± 2.07	34.87 ± 1.54
By-catch 100%	35.44 ± 0.96	14.24 ± 1.27	0.67 ± 0.28	18.95 ± 0.89	91.62 ± 0.73	22.32 ± 1.88
Commercial	35.99 ± 0.22	14.24 ± 1.13	1.33 ± 0.31	$7.62 \pm 0.55$	93.33 ± 2.53	25.14 ± 1.34

Table 2. Proximate composition (Mean  $\pm$  S.D.) of the experimental and control diets

FEED	ENERGY (KJ/g)
Fish meal 100%	15.84
Blood meal 50%	17.56
Blood meal 100%	14.29
By-products 50%	19.15
By-products 100%	22.21
By-catch 50%	16.63
By-catch 100%	15.42
Commercial	15.59

# Table 3. Energy values of diets

Parameters	Results	Acceptable Ranges
Temperature ( <sup>o</sup> C)	16 – 32	25 – 35
рН	7.5 - 8.3	6 - 9
Conductivity (µS/cm)	392 - 490	0-2000
Total dissolved solids (mg/l)	265 - 306	0 - 1500
Salinity (ppt)	Not detected	0-7
Dissolved Oxygen (mg/l)	4-5.5	4 - 7.5
Turbidity (NTU)	8 -20	0-5
Total suspended solids (mg/l)	1 – 15	0– 50
Ammonia (mg/l)	0.0772 - 0.1026	0-0.2
Reactive phosphorus (mg/l)	0.650 - 0.996	0 -1.5

**Table 4.** Water quality parameters measured (range based on averages)

Treatment	Feed Intake(g)	Feed Conversion ratio	Protein efficiency ratio
100% fish meal	6.68 ± 0.36 <sup>a</sup>	6.24 ± 1.22 <sup>a</sup>	$2.33 \pm 0.12^{a}$
50% blood meal	6.86 ± 0.54 <sup>a</sup>	5.10 ± 0.26 <sup>b</sup>	2.39 ± 0.19 <sup>a</sup>
100% blood meal	7.38 ± 0.37 <sup>b</sup>	5.18 ± 0.26 <sup>a</sup>	$2.58 \pm 0.14^{b}$
50% by-products	7.38 ± 0.37 <sup>b</sup>	5.86 ± 1.23 <sup>a</sup>	$2.79 \pm 0.29^{b}$
100% by-products	7.98 ± 0.49 <sup>b</sup>	$5.09 \pm 0.23^{b}$	2.77 ± 0.17 <sup>b</sup>
50% by-catch	7.45 ± 0.87 <sup>b</sup>	$5.63 \pm 0.49^{a}$	2.61.± 0.31 <sup>b</sup>
100% by-catch	8.20 ± 0.67 <sup>bc</sup>	$4.94 \pm 0.78^{b}$	$2.87 \pm 0.24^{bc}$
Commercial	8.28 ± 0.87 <sup>bc</sup>	$4.87 \pm 0.81^{b}$	2.90 ± 0.31 <sup>bc</sup>

**Table 5.** Mean (± SD) Feed intake, feed conversion ratio and protein efficiency ratio of O.

niloticus reared

in aquariums for 11 weeks

N.B. Values with different superscripts in the same column are significantly different (p<0.05)

N.B. Prices of feed ingredients based on retail prices and quoted in US dollar, FM = Fish meal, BM = blood meal, BP =
by-products, BC = by-catch

Table 7. Cost (US\$) per kilogram of experimental formulated fish feeds and commercial fish

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Treatment	Initial mean weight (g)	Final mean weight (g)	Mean weight gain (g)	Specific growth Rate (%)
100% fish meal	1.03 ± 0.03	2.11 ± 0.16 <sup>a</sup>	1.09 ± 0.18 <sup>a</sup>	$0.92 \pm 0.09^{a}$
50% blood meal	0.98 ± 0.08	2.34 ± 0.23 <sup>a</sup>	1.35 ± 0.18 <sup>a</sup>	1.12 ± 0.07 <sup>b</sup>
100% blood meal	1.03 ± 0.07	$2.45 \pm 0.22^{b}$	1.42 ± 0.14 <sup>b</sup>	1.13 ± 0.02 <sup>b</sup>
50% by-products	0.97 ± 0.12	2.39 ± 0.27 <sup>a</sup>	1.39 ± 0.20 <sup>b</sup>	1.15 ± 0.2 <sup>b</sup>
100% by-products	1.08 ± 0.05	$2.63 \pm 0.2^{b}$	1.56 ± 0.14 <sup>bc</sup>	1.15 ± 0.05 <sup>b</sup>
50% by-catch	1.06 ± 0.17	2.39 ± 0.27 <sup>a</sup>	1.33 ± 0.14 <sup>b</sup>	1.06 ± 0.12 <sup>b</sup>
100% by-catch	1.07 ± 0.11	2.77 ± 048 <sup>bc</sup>	1.70 ± 0.42 <sup>bc</sup>	1.23 ± 0.16 <sup>bc</sup>
Commercial	1.08 ± 0.10	2.87 ± 0.30 <sup>bc</sup>	1.78 ± 0.21 <sup>bc</sup>	1.26 ± 0.06 <sup>bc</sup>

Table 6. Initial, Final and mean weight (±SD) gain of O. niloticus reared in aquariums for [DO14] 11 weeks

N.B. Values with different superscripts in the same column are significantly different (p<0.05)

Ingredients	100% FM	50% BM	100% BM	50% BP	100% BP	50% BC	100% BC	Commercial
Soya meal	0.25	0.25	0.25	0.25	0.25	0.25	0.25	
Fish meal	0.42	0.22	-	0.21	-	0.21	-	
Blood meal	-	0.05	0.10	-	-	-	-	
Fish skins	-	-	-	0.13	0.25	-	-	
Fish guts	-	-	-	0.13	0.25	-	-	
By-catch	-	-	-	-	-	0.09	0.18	
Maize meal	0.03	0.03	0.03	0.03	0.03	0.03	0.03	
Wheat bran	0.02	0.02	0.02	0.02	0.02	0.02	0.02	
Binder	0.02	0.02	0.02	0.02	0.02	0.02	0.02	
Ascorbic Acid	0.01	0.01	0.01	0.01	0.01	0.01	0.01	
Fish oil	0.10	0.10	0.10	0.10	0.10	0.10	0.10	
Calcium	0.02	0.02	0.02	0.02	0.02	0.02	0.02	
Total Cost USD	0.87	0.64	0.53	0.99	0.93	0.73	0.61	1.70

feed

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[DO1]Fish of this size should have been fed at least 40% CP diet and at least 4 times, not only twice a day. Feeding rate should have been at least 15% of their body weight (20 would have been better. Of course, as the fry grew, the feeding ration should reduce. Explain clearly if you adjusted the feeding ration as the fish grew bigger

## [DO2]

[DO3]How diverse was the bycatch? Would you get the same relative proportion/ratio of species in weight terms when you collect the bycatch several times over? If not, the results of this study may not be repeatable especially if the species composition of the bycatch and thus its nutrition profile could change from time to time

[DO4]It is unlikely that temperature would remain warm even during the night in a greenhouse if the tanks are not equipped with aquarium heaters

## [DO5]See the last comment

[DO6]Fish of 0.6 grams should have been fed at least 4 times a day and this rate reduced to three times a day for the rest of the experimental period. It is advisable to feed twice a day only for tilapia of at least 60 grams

## [DO7]Write in full

[DO8]This part is not clear. Please show the treatments (actually, the seven formulations, with all ingredients used in a table.

[DO9]Show these in a table

[DO10]Name the binder

[DO11]This is a very wide range. The industry needs a product with clearly defined and fairly invariable nutritional values to ease feed formulation

[DO12]Looks like the fish were actually underfed, and the nutritional content (protein) of the diets was also sub-optimal

[DO13]Whereas the data in this table is useful and informative, the major purpose of analyzing proximate composition of experimental diets is to obtain information for use in formulation. Please add a table of the formulated feeds and how the ingredients were combined in each of the 7 diets to achieve isonitrogenous feeds. To really realize the 35% formulations, you shouldn't have analyzed only the alternative protein sources, but also the other ingredients used like maize, wheat bran and soybean meal

[DO14]In my opinion, this growth performance was not satisfactory