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Partial Replacement of Fish Meal by A Mixture of Lupin (*Lupinus albus* L.) and Grass Pea (*Lathyrus sativus* L.) Meal in the Diets of Nile Tilapia (*Oreochromis niloticus* L.) Fingerlings in a Recirculating Aquaculture System

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Abstract

This study evaluated effects of fishmeal replacement with a mixture of lupin and grass pea meal (LGM) at inclusion levels of 0, 25, 50, and 75% as potential plant protein sources in the diets of Nile tilapia fingerlings on growth performance, feed utilization, and carcass composition. Experimental diets were formulated to be iso-nitrogenous (36g 100 g^{-1}), iso-lipidic (10g 100 g^{-1}) and iso-energetic (18 KJ g^{-1}), and fed at 6-10% of their body weight day^{-1}. Total of 276 tilapia fingerlings with average initial body weight of 1.37±0.06 were stocked into 4 treatments using completely randomized design. Results revealed replacement of 25, 50, and 75% of fishmeal by LGM meals did not significantly (P>0.05) affect most of growth and feed utilization parameters. However, fish fed with 25% LGM meal revealed enhanced final body weight and improved feed intake. Compared to control diet, increasing LGM meals in diets significantly increased hepatosomatic index. Replacement of fishmeal by LGM meal has also resulted in comparable carcass composition. In general, there are promising nutritional justifications for replacing fishmeal with a mixture of lupin and grass pea protein sources in diets of Nile tilapia considering growth parameters, feed utilization, and carcass composition up to 75% level.

Introduction

Aquaculture is among the fastest-growing food production systems globally, producing nearly half (47%) of the total world fish production (FAO, 2018) which depends heavily on aquafeed input. It has become responsible for the continuing impressive growth in the supply of fish in contrast to the relatively static growth of capture fishery. The rapid growth of the aquaculture sector results from the progressive intensification of using quality aqua-feeds and production systems (FAO, 2006). However, aqua-feed is the major determinant factor for up to 60% of the total aquaculture production costs in semi-intensive and intensive types of systems (El-Sayed, 2004; Dorothy *et al.*, 2018). Fish meal have been considered as a highly preferred ingredient for formulating aqua-feeds through feed production mainly depending on many protein and energy ingredient sources (El-Saidy and Gaber, 2003; Khalil, 2012). However, the limited source of feed ingredients, such as fish meal, fish oil and soybean meal are also the main reason for increase in the price of aqua-feed. Furthermore, an increasing human demand for nutrients derived from those ingredients could reduce the contribution of those feed ingredients towards satisfying the demand for sustainable aqua-feed production. Thus, there is a serious concern about the long term access and availability of these feed ingredients for use in aqua-feed production (Kumar *et al.*, 2013). The necessity to

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search and identify alternative sources of protein to formulate and develop low-cost feed ingredients based on sustainable and renewable feed sources for subsistence and commercial scale fish farmers is very indispensable (Madalla, 2008; FAO, 2010). Besides, it's also important to consider that the selected feed ingredients should not compete with the accustomed food security interests (Tacon *et al.*, 2001; Madalla, 2008). Replacing fish meal with plant-based protein ingredients show that plant proteins can be successfully used in fish feed instead of fish meal for about 30 to 50% (Francis *et al*., 2001).

Plant proteins are not only less risky, but they are also abundant offering a more suitable option as alternative protein sources in fish feeds (Naylor *et al.*, 2009; Yalew *et al.*, 2019) although they often contain anti-nutritional factors (ANFs), which can affect growth performance and fish health. For these reasons, Lupin (*Lupinus albus*) and grass pea (*Lathyrus sativus*) are expected to be one of the potential plant species to be a cheap protein source for fish feed production in the future if anti-nutritional factors and toxicants are well detoxified.

Lupin and grass pea are plant species which are widely distributed in many subtropical and tropical countries including Ethiopia. Lupin and grass pea have a nutritional quality comparable with most legumes as they contain similar nutritional quality in the proportion of protein, fat, minerals, and vitamins (Caruso, 2015; Lambein *et al.*, 2019). Therefore, it is expected to be an excellent feed ingredient in the future to replace fish meal and soybean meal. Despite the fact that a high content of protein with an excellent amino acid profile and good proportion of carbohydrates, the levels of toxic substance and anti-nutritional substances like phytate, lectins, protease inhibitors, haemagglutinins, saponins, alkaloids, glycosides, oxalic acids and lathyrogens) restricts its use in animal feed production (Viola *et al.*, 1988; Francis *et al.*, 2001). The purpose of this research was therefore, to evaluate the effect of partial replacement of fish meal by a mixture of lupin and grass pea-meal (LGM) on the growth performance, feed utilization, and carcass proximate composition of Nile tilapia fingerlings and contribute to developing sustainable aquaculture feed resources.

Materials and Methods

Culture Facility and Growth Trial

The study was conducted at the Center for Aquaponics and Recirculating Aquaculture System of the Department of Zoological Sciences in the College of Natural and Computational Sciences, Addis Ababa University. The fingerlings of Chamo strain Nile tilapia were produced in the hatchery unit within the center and fingerlings were stocked an average weight of 1.34±0.03 gram with 23 fish per aquarium (60 liters each) in a closed recirculation system. They were acclimatized to the experimental conditions for two weeks while being fed with the control diet. During the feeding trial, fish were hand-fed to apparent satiation three times a day (8:00, 12:00, and 16:00) for 10 weeks. The amount of feed was adjusted every week according to the new mean fish weight in each treatment. The recirculation system was supplied with continuous flow (2 L min^{-1}) of physically, biologically and UV light filtered and aerated water from a sump tank heated at 27.78±0.12°C and subjected to the natural 12h of light and 12h of dark period. The study was conducted with three experimental diets and a control diet, fed in triplicates using a completely randomized design. In the system, water quality parameters measured during the experiment averaged (±SD): pH, 7.85±0.08; ammonia, 0.02±0.002 mg L^{-1}; DO, 5.78±0.08 mg L^{-1}; nitrate, 0.39±0.035 mg L^{-1}; nitrite, 0.21±0.11 mg L^{-1} and they were within acceptable ranges for tilapia culture (Table 2).

Experimental Design and Diet Preparation

The study utilized a completely randomized design with four diets formulated for the experiment, each being iso-nitrogenous (36% protein), iso-lipidic (10% lipids), and iso-energetic (18 KJ g^{-1}) based on Nile tilapia requirements (Table 3). The control diet was formulated with a fish meal protein and this was replaced at different levels (25, 50, and 75%) with a mixture of lupin and grass pea-meal (LGM) (mixed 1:1 ratio). The diets were formulated on a fed basis, and fish meal was a main dietary protein source, and wheat and corn grains were used as a main carbohydrate source in the experiment. Experimental diets were prepared by supplementing the basal formulated diet with different levels of mixture of lupin and grass pea meal (Table 1). The experimental diets (lupin and grass pea grains) were soaked for 48 hrs with a continual water change, dried, roasted, dehulled, ground, sieved and then mixed in a 1:1 ratio before being incorporated into the feed formulation in order to minimize astringent effect (antinutritional factors). Diets were prepared by wet extrusion using a meat mincer (TJ22 model). A poultry grade vitamin/mineral premix (AMINOVIT) at 34.4 g Kg^{-1} and binder carboxymethyl cellulose, high viscosity at 20 g Kg^{-1} were added. The proportion of the feed ingredients in the experimental diets was prepared using WinFeed version 2.8 feed formulation packages after proximate analysis of each ingredient was done as presented in Table 3. Feed ingredients and prepared experimental diets were separately analyzed for a proximate composition to check the nutritional quality.

Estimation of Growth Performance and Production Parameters

Fish samples from each treatment were collected, including the control treatment, and euthanized by using concentrated clove oil to minimize fish stress and

Table 1. Composition of feed ingredients in the diets (g Kg^{-1} as-fed) fed to Nile tilapia fingerlings in the replacement of fish meal with a mixture of lupin and grass pea-meal.

Feed ingredients	Control diet	LGM25	LGM50	LGM75
Fish meal	323.00	242.25	161.50	80.75
Wheat grain	120.00	120.00	120.00	120.00
Corn grain	109.80	109.80	109.80	109.80
Soybean grain	362.80	362.80	362.80	362.80
LGM	0	80.75	161.50	242.25
CMC	20.00	20.00	20.00	20.00
VMP	34.40	34.40	34.40	34.40
SBO	30.00	30.00	30.00	30.00

Table 2. Some representative water quality parameters of recirculating aquaculture system.

	Treatments			
Parameters	Control	LGM25	LGM50	LGM75
Temperature (°C)	27.9±0.140^a	27.7±0.127^a	27.7±0.12^a	27.8±0.108^a
DO (mg L^{-1})	5.83±0.072^a	5.80±0.081^a	5.77±0.079^a	5.71±0.083^a
pH	7.81±0.084^a	7.89±0.072^a	7.83±0.077^a	7.87±0.096^a
NH_3 (mg L^{-1})	0.02±0.002^a	0.02±0.002^a	0.02±0.002^a	0.02±0.002^a
NO_3^- (mg L^{-1})	0.390±0.031^a	0.410±0.031^a	0.380±0.021^a	0.384±0.016^a

Table 3. Proximate composition (g Kg^{-1}), energy content (KJ g^{-1}) and prices (Birr Kg^{-1}) of individual feed ingredients used in the study

Components (g Kg^{-1})	Control	LGM25	LGM50	LGM75
DM	905.8	902.6	903.7	910.7
CP	376.0	366.4	362.6	358.3
CL	117.3	109.6	120.1	113.3
CF	46.5	53.8	67.5	79.5
Ash	46.4	47.4	45.0	44.7
NFE	319.6	325.4	308.5	314.9
GE (KJ g^{-1})	19.14	18.71	18.74	18.48

treat ethically. Fish samples were weighed by electronic balance and measured by measuring table to collect the required data and returned in to the respective glass aquaria. Following the aforementioned protocol, all fish were weighed and measured every week and growth performance and production parameters were calculated following the procedures described in (Kaushik and de Oliva Teles, 1985):

$$\text{Average initial weight (IBW, g)} = \frac{\text{Sum of individual weight at the beginning}}{\text{Total number of fish}} \quad (1)$$

$$\text{Average final weight (FBW, g)} = \frac{\text{Sum of individual weight at the end}}{\text{Total number of fish}} \quad (2)$$

$$\text{Weight gain (WG, \%)} = \frac{(\text{FBW}-\text{IBW})}{\text{IBW}} \text{ x } 100 \quad (3)$$

$$\text{Specific growth rate (SGR, \% per day)} = \frac{\ln(\text{FBW}) - \ln(\text{IBW})}{\text{Culturing days}} \text{X}100 \quad (4)$$

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Total weight of dry feed given}}{\text{Total live weight gain by fish}} \quad (5)$$

$$\text{Survival rate (SR, \%)} = \frac{\text{Number of fish harvested}}{\text{Number of fish stocked}} \text{ x } 100 \quad (6)$$

$$\text{Fulton's condition factor (FCF)} = (Wt/L^3) \text{ x } 100 \quad (7)$$

Where Wt = body weight and L= length of the fish. Feed intake (FI) was determined by adding the total amount of feed consumed by the fingerlings in each treatment for the whole experimental period. It was extrapolated by using the weekly feed ration adjustment of each treatment where fingerlings were fed at 10% of their body weight for the first four weeks and then reduced to 6% of their body weight based on weekly weight gain.

Body Proximate Composition of Fish

At the end of the experiment, whole body proximate, energy content, and hepatosomatic index (HSI) were determined. Ten fish were randomly sampled from each aquarium during harvest for the final carcass composition analysis. Fish were killed by immersing into an overdose of clove of solution and dried in an oven at 60 °C until a constant weight was achieved. The dried fish carcasses were ground in order to analyze proximate composition. Analysis for crude fat (lipid), crude protein, moisture and ash of experimental diets as well as fish carcass were done using standard methods of (AOAC, 2002) procedures: dry matter (130°C to constant weight), ash by the gravimetric method (combusted at 550°C to constant weight), crude protein (Nx6.25) by the Kjeldahl method, and crude lipid extracted by the Soxhlet extraction method. All analyses were performed in triplicate, and ingredients and proximate composition of experimental diets presented in Table 4 and expressed as a percentage of dry weight basis. Simultaneously, fish samples from each treatment were collected, including the control treatment, and euthanized by using concentrated clove oil to kill the fish ethically, dissected and livers were removed, weighed and used to estimate HIS (El-Sayed, 1990).

$$\text{HSI} = \frac{\text{Liver weight}}{\text{Bodyweight}} \text{ x } 100\% \quad (8)$$

Statistical Analysis

The results are expressed as Mean ± SD by using SPSS version 20, and figures were produced using Sigma plot version 11.0. The differences among treatments were analyzed using One-Way Analysis of Variance (ANOVA) and significant mean differences were compared using Tukey's Multiple Comparison Test. The statistical significance was set at $P<0.05$, and SPSS for Windows version 20 (SPSS, Michigan Avenue, Chicago, IL, USA) was used.

Results

Physicochemical Parameters

The physicochemical parameters such as temperature, pH, DO, NH_3, and NO_3^- recorded in the culture system are shown in Table 1. During the whole experimental period water temperature ranged from 25.4-29.8°C, DO from 5.24-6.23 mg L^{-1}, pH from 7.22-8.16, and NH_3 and NO_3^- exhibited considerably low concentrations as stated in Table 1. There were no significant differences in physicochemical parameters among treatments during the whole experimental period, indicating that the experimental diets did not have a detrimental effect on the surrounding water quality where the cultured Nile tilapia fingerlings had been stocked. These physicochemical parameters are expressed as mean values ± SD as presented in Table 1. Thus, the growth performance and feed utilization of Nile tilapia fingerlings were not significantly affected by the physicochemical parameters of the culture system and all fish were in normal condition.

Proximate Composition and Energy Contents of Feed Ingredients

As stated in Table (3) lupin and grass pea meals contained good protein content (38.08% and 25.58%, respectively), high-fat content (12.29.% for lupin), and allowable ash content, while nitrogen-free extract recorded were with moderate to higher value (39.73% to 60.33%).

Proximate Composition and Energy Contents of Experimental Diets

The experimental diets were generally formulated and prepared to be almost iso-nitrogenous (36% protein), iso-lipidic (10% lipids), and iso-energetic (18 KJ g^{-1}). Any differences in the performances of the cultured Nile tilapia fingerlings that received such diets could be attributed to the quality of the feed, feeding value of the experimental ingredients and level of replacement. The proximate composition and energy contents of the experimental diets including the control diet are presented in Table 6. The CP contents of diets ranged (358.3- 376.0 g Kg^{-1}), CL (109.6-120.1 g Kg^{-1}), CF (46.5-79.5 g Kg^{-1}), ash (44.7-47.4 g Kg^{-1}), NFE (308.5-325.4 g Kg^{-1}). The GE content of experimental diets varied little within a narrow range (18.71-19.14 KJ g^{-1}). The CF content in diet 3 (LGM75) (79.5g Kg^{-1}) was higher almost double to the CF content of control diet (46.5 g Kg^{-1}) and it was the highest CF content found among the rest of experimental diets from the nutritional viewpoint. All experimental diets were accepted readily by the stocked Nile tilapia fingerlings. Feed was given in hand moistened expeller-like strands to improve palatability and fingerlings were fed actively in all experimental groups and within approximately 15-20 minutes, all feeds had been consumed from the aquaria. No abnormal Nile tilapia or disease symptoms were observed during the experimental feeding trial. No health-related behavioral issues were observed in response to the experimental diets.

Growth Performance and Production Parameters

The growth and feed utilization responses of Nile tilapia fingerlings fed lupin and grass pea mixture meal at different fish meal replacement levels are presented in Table 5. The highest final weight gain was recorded in LGM25, and improved feed intake was recorded in all experimental diets as compared to the control diet. However, there were no significant differences ($P<0.05$) in IBW, %WG, SGR, SR, FCR, and FCF. No significant feed-related mortality was observed during the whole experimental period. However, the survival rate of the fish that fed on control and LGM50 resulted in a little lower than the fish groups fed on the rest of the experimental diets. The recorded mortality of fish was clear since continuous signs of sexual competition behavior for courtship were observed at the end of the ninth and tenth weeks. The results of the present study revealed that all experimental diets were well accepted by tilapia fingerlings. This implies that the different experimental feed ingredients did not negatively affect the palatability of the diets, and diets were actively eaten by fingerlings.

Body Proximate Composition

The whole-body proximate composition of all experimental fish samples including a control group is presented in Table 6. The moisture content of carcasses increased little with the increased replacement of fish meal at higher levels (LGM75) protein, however, fish fed the control diet, LGM25 and LGM50 had not shown

Table 4: Proximate composition and energy contents of experimental diets fed to Nile tilapia

Components (g Kg^{-1})	Control	LGM25	LGM50	LGM75
DM	905.8	902.6	903.7	910.7
CP	376.0	366.4	362.6	358.3
CL	117.3	109.6	120.1	113.3
CF	46.5	53.8	67.5	79.5
Ash	46.4	47.4	45.0	44.7
NFE	319.6	325.4	308.5	314.9
GE (KJ g^{-1})	19.14	18.71	18.74	18.48

Table 5: Growth performance and feed utilization of Nile tilapia fingerlings fed different experimental diets

Parameters	Experimental diets			
	Control	LGM25	LGM50	LGM75
IBW (g)	1.34±0.03^{a}	1.41±0.10^{a}	1.35±0.04^{a}	1.37±0.08^{a}
FBW (g)	9.03±0.045^{a}	9.49±0.052^{b}	9.11±0.129^{a}	8.92±0.035^{a}
WG (%)	573.38±18.76^{a}	583.85±42.21^{a}	576.24±27.52^{a}	551.09±47.61^{a}
SGR (%)	2.72±0.04^{a}	2.73±0.10^{a}	2.73±0.06^{a}	2.62±0.09^{a}
SR %)	99.52±0.48^{a}	100±0.00^{a}	99.52±0.48^{a}	100±0.00^{a}
FCR	2.30±0.17^{a}	2.37±0.23^{a}	2.33±0.19^{a}	2.50±0.26^{a}
FI (g $fish^{-1}$)	17.65±0.20^{a}	19.15±0.16^{b}	18.07± 0.38^{ab}	18.87±0.31^{b}
FCF	2.69±0.012^{a}	2.73±0.029^{a}	2.62±0.038^{a}	2.66±0.012^{a}

Table 6: Whole body proximate composition (% dry weight), gross energy and HSI of Nile tilapia fingerlings fed the different experimental diets

	Experimental Diets			
Components	Control	LGM25	LGM50	LGM75
MC	21.63±0.18^{a}	21.76±0.12^{a}	21.87±0.25^{a}	23.43±0.34^{b}
CP	57.33±0.13^{a}	57.35±0.05^{a}	57.13±0.23^{a}	56.95±0.02^{a}
CL	18.42±0.08^{a}	18.49±0.10^{a}	18.40±0.09^{a}	17.30±0.08^{b}
Ash	13.12±0.07^{a}	12.96±0.09^{a}	13.05±0.15^{a}	13.13±0.09^{a}
GE	20.91±0.04^{a}	20.93±0.04^{a}	20.86±0.06^{a}	20.38±0.03^{b}
HSI	2.70±0.08^{a}	2.88±0.09^{ab}	3.16±0.17^{b}	3.23±0.09^{b}

significant differences (P>0.05) on moisture content. The crude lipid and gross energy contents of whole-body proximate were significantly affected (P<0.05) by the replacement of lupin and grass pea mixture meal in Nile tilapia fingerlings fed LGM75 diet as compared to fish fed on a control diet. The crude protein and ash contents of all whole-body proximate were not significantly different. Highest HSI value was recorded from fish fed with LGM50 and LGM75 diets.

Discussion

Growth Performance and Production Parameters

The result of the present study indicated the potential of lupin and grass pea mixture meal (LGM) as a major protein source replacing fish meal in the diets of Nile tilapia fed at higher levels with success. Fish fed with mixtures of lupin and grass pea meal resulted in enhanced body weight gain (Figure 1). This might be associated to reduced exposure to ANFs in the diet responsible for suppressing feed intake, palatability and digestibility resulted from the combined preprocessing techniques in the feed preparation. Similarly, combining several plant protein sources with inherent ANFs can reduce the exposure of fish to those ANFs due to lower inclusion levels of each of the individual protein sources in the combined mixture (Borgeson, 2005). This is in agreement with Francis *et al.* (2001) who reported that mixing plant proteins may also lead to interactions between various ANFs or with other components in the diets resulting in the reduction of their deleterious effects. For instance, Fish *et al.* (1991) reported that interaction between tannins and lectins removed the inhibitory action of tannins on digestive enzyme amylase, and interactions between tannins and cyanogenic glycosides reduced the deleterious effects of cyanogenic glycosides (Goldstein and Spencer, 1985).

Besides, result of the present study agrees with the findings of (Yones, 2010) who reported replacement of fish meal with a mixture of extruded lupin kernel meal (*Lupinus albus*) at 25, 50, and 60% inclusion levels in diets of red hybrid tilapia that found best growth performance. Similarly, the results of the present study confirmed the previous work of Zhang *et al.* (2012) who reported a better growth performance of Nile tilapia fed on a mixture of lupin and soy protein concentrate than fish groups fed on a fish meal-based diet. The current

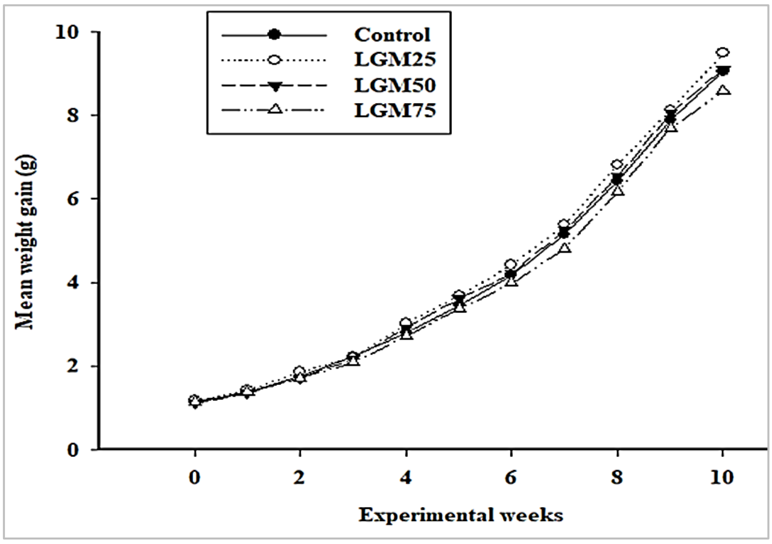

Figure 1. Growth response of Nile tilapia fingerlings fed LGM diets for 10 weeks.

finding also agrees with the study reported by Fournier *et al.* (2004) who found that replacing fish meal up to 50% by plant protein mixtures of lupin, wheat and corn gluten meal in the diets of juvenile turbot (*Psetta Maxima*) had not significantly affected growth responses. Similarly, a study conducted by El-Saidy and Gaber (2003) found that partial or complete replacement of fish meal by mixtures of plant protein sources such as soybean, cottonseed, sunflower and linseed meals (all in equal proportion of 25%) in the diets of Nile tilapia did not exhibit significant differences in growth performances as compared to a control diet. Furthermore, Al-Thobaiti *et al.* (2018) evaluated the replacement of fish meal with mixtures of corn gluten meal, wheat gluten meal, and bagasse kenna meal on growth performance of Nile tilapia fingerlings. They found that fish groups fed with 20% of fish meal replaced diet by plant protein mixtures revealed enhanced growth performance and improved feed utilization efficiency as compared to 100% fish meal based diet.

The Specific growth rate (SGR) which is described as the percentage increase in weight of fish per day resulted in no significant variation among treatments (Table 5) and a similar trend was observed with the other growth and production parameters. The growth parameter results of the present study revealed enhanced SGR values as compared to several reports done on Nile tilapia fed with mixture and individual feed ingredients. This might be attributed to the compensatory effects of mixing plant proteins that improve the amino acid profile, bioavailability, and palatability of diets. This is supported by the findings of Geremew (2015) who reported better growth performance of Nile tilapia fingerlings fed with mixtures of different oil seed cakes (niger seed cake and linseed cake) than fish fed with individual feed ingredients. Moreover, the improved growth performance recorded in the present study might be associated with the preprocessing techniques applied to remove ANFs and their effects on LGM feed ingredients. For instance, dehulling of legume grains reduce saponins and tannin content and positively affects the appearance, digestibility and palatability of diets. It also improves the crude protein content of feed ingredients by (10-23%), decrease crude fiber content by approximately (76-83%), and increases mineral bioavailability (Brenes *et al.*, 2003; Khan *et al.*, 2013; Ertop *et al.*, 2018). Similarly, soaking and roasting of LGM feed ingredients remove alkaloids, water-soluble phytates, trypsin inhibitors, and thermo-labile ANFs such as haemagglutinins (lectins) thus maximizing their potential utilization as aquafeed ingredients (Akande and Fabiyi, 2010). Furthermore, the enhanced growth responses could be attributed to a high concentration of L-homoarginine amino acid in the LGM diets that could be a substrate for arginase, since *Lathyrus* seeds are the only dietary source of this natural amino acid (Pentyala and Rao, 1999; Lambein *et al.*, 2019). Besides that, this improved growth could be attributed to the controlled and optimum physicochemical parameters of culture system for a healthy growth of Nile tilapia (El-Sherif *et al.*, 2009; Begum *et al.*, 2014).

The SGR result of the present study agrees with the findings of (Soltan *et al.*, 2008; Khan *et al.*, 2013) and Geremew (2015) who reported SGR values ranging from 2.39 to 2.73, 2.60 to 2.73 and 2.08 to 2.76 for Nile tilapia in a controlled environment in the replacement and inclusion of a mixture of cottonseed, sunflower, canola, sesame; linseed meal, rice polish, and oilseed cakes into the basal diet, respectively. However, the result of the present study is in contrast with the findings of Sudaryono *et al.* (1999) where replacement of lupin kernel meal in the diets of *P. monodon* containing 40% CP replaced at (25, 50, 75, and 100%) of fish meal protein with an equivalent amount of lupin protein resulted in a progressive decline of weight performance for those replaced over 25% of fish meal.

Unlike, to the present study, Fournier *et al.* (2004) reported that replacing fish meal by plant protein mixtures of lupin, wheat and corn gluten meal in the diets of juvenile turbot (*Psetta maxima*) at highest replacement levels (75 and 100%) significantly reduced growth performance. Besides, Goda *et al.* (2007) also reported lower SGR ranged from 1.73 to 1.97 in the diets of Nile tilapia fed with mixtures of different plant proteins. Furthermore, Yalew *et al.* (2019) reported very low SGR ranged from 1.68 to 2.02 in the replacement of fish meal by sweet lupin meal at inclusion levels of 75% and 100% in the diets of juvenile catfish. The contradiction between the current and the aforementioned results could be attributed to identified or unidentified ANFs in the experimental diets which could reduce FI and adverse FCR (Sklan *et al.*, 2004; Soltan *et al.*, 2008). The ANFs that are dominantly abundant in lupin and grass pea grains such as phytic acid, lectins, trypsin inhibitors, protease inhibitors, alkaloids, saponins, and oligosaccharides could increase the delay of diet retention in stomach affecting FI through feedback on satiety signals (Khan *et al.*, 2013). Aquafeeds made of lupin, grass pea and other pulses contain phenolic compounds such as tannin that may reduce palatability and reduce protein digestibility. Besides, phytic acid in both grains negatively affect the utilization of minerals which can be seen by its ability to bind up 75% of all phosphorus, chelating and trivalent metals such as Ca^{2+}, $Mg^{2+,}$ and Fe^{3+} into compounds not easily absorbed in the intestine and inhibiting digestive enzymes (NRC, 1998).

Feed conversion ratio (FCR) which is commonly used as indicator of quality of feed was not significantly different among treatments (Table 5). The FCR values of the present study ranging from 2.30 to 2.50 indicated that 2.3 to 2.5g of feed was used to produce one gram fish flesh. According to Stickney (1979), 1.5g of feed is recommended to produce one gram of live weight in aquaculture. This high FCR value of the present study might be associated to the quality of experimental diets, dietary protein and energy content (Awad *et al.*, 2012; Khan *et al.*, 2013) and relatively higher CF content that affects feed utilization efficiency and adverse FCR (Soltan *et al.*, 2008). Similarly, the increased FCR values of the present study could be attributed to variations in fish strain (Guimarães *et al.*, 2008), size and life stage of the stock (Ridha, 2007). Besides that, the stocking density of the present experiment could affect the FCR values since culturing at a confined environment results in increased energy expenditure because of increased agonistic behavior (Diana *et al.*, 2004). However, the FCR value of the present study agrees with the findings of El-Saidy and Gaber (2003) 2.03 to 2.17, Khan *et al.* (2013) 2.23 to 2.31, Agbo *et al.* (2015) 2.07 to 3.17, Al-Thobaiti *et al.* (2018) 2.33 to 3.0 in the diets of Nile tilapia fed with mixtures of different plant protein sources. Besides, the result of present study is in agreement with the study done by Yones (2010) who reported that the FCR values of hybrid Nile tilapia fed with a replaced fish meal at (25 and 50%) of basal diet by lupin kernel meal had not shown significant variation as compared to the control diet. Unlike the present study, high FCR values (low feed conversion efficiencies) have been also reported by other studies. For instance, Yalew *et al.* (2019) reported very high FCR values ranging from 2.70 to 8.92 for catfish fingerlings that fed sweet lupin meal in the replacement of fish meal at 0, 50, 75, and 100% inclusion levels in a controlled condition. Moreover, Workagegn *et al.* (2014) also reported FCR values ranging from 3.23 to 3.38 in the diets of Nile tilapia fingerlings fed with mixtures of different plant proteins. This variation could be associated with the preprocessing techniques used in the present study that may efficiently reduce the ANFs like saponins, phytic acids, tannin content, trypsin inhibitors and alkaloids which can affect the digestibility, palatability, and improve feed utilization efficiency (Brenes *et al.*, 2003; Ertop *et al.*, 2018).

The feed intake (FI) results of the present study ranged from 17.65 to 19.15 g fish^{-1}. Fish groups fed on all experimental diets revealed improved FI values as compared to control group. The slight tendency towards a higher FI as dietary LGM content increased can be explained as a compensatory intake to meet demands for protein to maintain maximum growth. This is supported by the previous findings reported by Higuera *et al.* (1988) in rainbow trout, Yones (2005) in sea bream, and Espe *et al.* (2007) in Atlantic salmon, which had shown a similar trend in feed utilization. Interestingly, feeding with lupin and grass pea mixture meal led to higher FI compared to the control diet, and this might also be attributed to the actual nutritional composition of feed ingredients, and the size and life stage of stock affect FI values (Abdel-Tawwab *et al.*, 2010). Moreover, the energy content of the control diet in the present study is higher than others, thus fish fed on it might not consume a high amount of feed as compared to others that resulted in reduced FI values.

The result of the present study is consistent with the findings of Awad *et al.* (2012) who reported higher FI in rainbow trout as compared to the control diet, which might be due to the mixture of feed ingredients used (lupin, mango, and stinging nettle) had better nutritional composition. The result of the present study confirmed the feed intake ranging from 38.1 to 39.3 reported by Zhang *et al.* (2012) where an increasing tendency with increasing fish meal replacement by mixtures of lupin and pea proteins in the diets of rainbow trout at different inclusion levels. The FI results of the present study varied from the results reported by several authors who found a decreasing trend of FI with increasing the replacement levels of fish meal by mixtures of different plant protein in the diets of Nile tilapia. For instance, Soltan *et al.* (2008) reported a decreased FI trend ranged from 36.12 to 41.50 g fish^{-1} in the diets of Nile tilapia fingerlings where a fish meal was replaced by a mixture of cottonseed, sunflower, canola, sesame, and linseed meals up to 75%. Similarly, the results of the current study deviate from the findings of El-Saidy and Gaber (2003) who reported FI ranging from

30.2 to 38.4 g fish^{-1} in the diets of Nile tilapia fingerlings fed on fish meal replaced by mixtures of 25% for each (soybean, cottonseed, and sunflower meals) at 25, 50, 75, and 100% inclusion levels. In contrast to the aforementioned FI reports, the results of the present study showed a slight tendency toward higher FI as dietary LGM content increased. Besides, Borgeson (2005) reported improved feed utilization and enhanced growth response of Nile tilapia fed with a mixture of soybean meal, canola, whole peas and whole flax meal at inclusion levels of 0, 33, 67, and 100%.

In the case of survival, no mortality concerning the effect of experimental diets was recorded. The relatively lower survival rate of 99.52% was recorded in fish groups fed on control and LGM50 and the survival rate of LGM25 and LGM75 was 100%. However, there was no significant (P>0.05) difference in survival rate among treatments. The insignificant reduction in survival rate obtained was due to the mortality rate recorded in both groups during the ninth and tenth weeks of the experimental period and some observed that cultured fish were showing sexual competition for courtship. As a result, lots of eggs were found while samples were taken and dissected during hepatosomatic index analysis in the laboratory. Generally, the survival rate was successful which implies LGM meal can be incorporated into the diets of Nile tilapia by replacing fishmeal up to 75% without posing any survival problem.

Whole Body Proximate Composition

The moisture content among treatments of Nile tilapia fingerlings in the present study is presented is in Table 6. The percentage range of moisture content in the present study is within the acceptable moisture level required for fish commonly reported to be about 60-80% by different authors. For instance, the moisture content of the current study is close to the results of El-Saidy and Gaber (2003) and Geremew (2015) who reported moisture content ranging from 75.45±1.10 to 77.81±0.90 and 76.4±0.10 to 77.48±0.18 for Nile tilapia fed with mixtures of different plant proteins, respectively. Moisture content of the present study closely agrees with the findings of Gebreanenia (2018) who reported moisture content ranging from 77.8±0.23 to 78.1±0.06 in the growth performance and proximate composition of three Nile tilapia strains in a controlled condition.

Percentage crude protein (CP) content of Nile tilapia fingerlings fed on LGM protein in the present study ranged from 56.96 to 57.35% (Table 6) and no significant variation was obtained in all treatments. The percentage CP content of the present study is in the range of permissible protein limit in dry weight basis for fish fillets and fisheries products. This might be associated to the dietary protein levels of feed ingredients and experimental diets. This is supported by the findings of Hafedh (1999) who pointed out that carcass composition of fish is significantly affected by the dietary protein level. Fish fed on a high-protein diet resulted in a higher percentage of protein than fish fed on low protein diets. The percentage CP of the present study agrees with the CP content reported by Montoya-Mejía *et al.* (2017) ranging from 57.5±0.1 to 60.0±0.8 for Nile tilapia fed on experimental diet prepared from mixtures of extruded bean, extruded chickpea meal, coconut paste, jatropha curcas meal, and chickpea meal in a controlled environment. Similarly, the CP content of the present study closely agrees with the previous works done by other different researchers. For instance, Awad *et al.* (2012) and Mzengereza (2015) have reported CP content ranging from 56.28±1.68 to 62.45±6.74 and 61.88±0.25 to 63.21±0.03 for rainbow trout and tilapia fed with mixtures of mango, lupin, stinging nettle; sweet potato, cassava, banana, and maize proteins as dietary supplements, and as main protein sources, respectively.

However, the CP content of the present study is lower than reports of others. For instance, Soltan *et al.* (2008) reported CP ranged from 66.11 to 68.58% in the diets of Nile tilapia fed with diets prepared from a mixture of plant proteins in the replacement of fish meal at inclusion levels of 15 to 100% of the basal diet in a controlled environment. Hassaan *et al.* (2018) have also reported CP content ranging from 61.75±1.17 to 65.31±1.12 for Nile tilapia grown in controlled condition. The variation in the CP content of the present study and formerly reported findings could be attributed to differences in the quality of experimental diets and protein content of feed ingredients used that affect directly the protein content of fish fillet. This is supported by the findings of Glencross *et al.* (2004) who reported similar results where rainbow trout fed with a mixture of plant proteins consisted of 12.5% yellow lupin meal resulted in increased CP content as compared to other diets, and body proximate composition of fish depends on the composition of the feed since it affects directly the protein content of fish fillet.

The crude lipid (CL) content of cultured fish recorded in the present study ranged from 17.30±0.08 to 18.49±0.10. Fish group fed LGM75 protein had a relatively lower percentage of CL content as compared to the control diet, and there was a significant difference (P<0.05) in CL content among fish groups fed with the different experimental diets. This lower CL content of fish groups fed on LGM75 might be attributed to high moisture content of fish carcass and composition of experimental diets. Yildirim *et al.* (2003) have reported an inverse correlation between fat and moisture content and it is common among fish species. Similarly, FAO (2010) has pointed out an inverse relation between moisture content and CL contents in fish fillet, and feed composition significantly affects CL and fatty acid profiles of fish flesh (Tadesse, 2010). The CL content obtained in the present study is in the range of CL content of 17.50±0.23 to 17.65±0.47 and 15.5±0.00 to 22.1±0.6 reported for Nile tilapia fed with fermented sunflower meal, and chickpea meal, chickpea, and Jatropha curcas meal (Mzengereza, 2015; Montoya-

Mejía *et al.*, 2017) in the replacement of fish meal. Furthermore, the percentage of CL of the present study is closely in agreement with CL contents ranged from 22.69 to 27.2% reported by Hassaan *et al.* (2018) in the diets of tilapia rendalli fed with a mixture of different plant proteins using glass aquaria.

However, CL content of the present study is lower than the reports of Awad *et al.* (2012) who reported higher CL contents of rainbow trout fed with plant protein mixture of lupin, mango and stinging nettle as dietary supplement ranging from 27.77±2.11 to 35.28±0.43. Unlike results of the present study, Geremew (2015) and Yones *et al.* (2019) have also reported low CL contents ranging from 5.13 to 7.81% for juvenile Nile tilapia in the replacement of fish meal by mixtures of different plant proteins at different inclusion levels. The variation in the CL content of the present study and previous works might be attributed to the lower moisture content of the carcasses, and the inclusion a of higher dietary protein content in the experimental diet (Hafedh, 1999).

The percentage of ash content of the present study is presented in Table 6. The increased ash content in the present study might be due to increased dietary levels of plant proteins, indicating sufficient amounts of dietary minerals in diets and its absorption. This increased ash content of carcasses is supported by the reports of Zhang *et al.* (2012) and Hassaan *et al.* (2018) that showed the same trend of ash contents in fish carcasses. Furthermore, the increased ash content of the present study might be attributed to processing techniques applied to remove different ANFs that otherwise reduces the availability of several minerals such as calcium, magnesium, zinc, iron, and phosphorus in the fish diets and severely affect the liver function.

Ash content of the present study follows the ash (11.23 to 15.12%) content reported by (Soltan *et al.*, 2008) for Nile tilapia fingerlings cultured in glass aquaria in the replacement of fish meal with a mixture of cottonseed, sunflower, canola, linseed and sesame meals at inclusion levels of 15 to 100%. Besides, several authors have reported similar ash content of fish carcasses. For example, Hassaan *et al.* (2018) have reported ash content ranged from 15.05±0.56 to 16.05±0.52 for Nile tilapia in a controlled environment. Azaza *et al.* (2009) have also pointed out similar ash content ranging from 14.80±0.11 to 16.51±0.49 for Nile tilapia in the replacement of fish meal by a mixture of plant proteins and black soldier fly maggot meal at 0, 25, 50, 75, and 100% inclusion levels. However, Zhang *et al.* (2012) have reported higher ash content ranging from 20 to 23.3% for rainbow trout fed with a mixture of lupin and pea protein concentrates in a controlled environment. The obtained ash content of the present study is higher than the ash content reported by other authors. Geremew (2015) have reported ash content ranging from 4.17±0.07 to 4.28±0.11 and 4.09±0.08 to 4.23±0.09 in dietary inclusions of linseed cake and Niger seed cake meals on growth performance and feed utilization of juvenile Nile tilapia, respectively. Furthermore, Goda *et al.* (2007) and Yones *et al.* (2019) have also found lower ash content values ranging from 5.0±0.2 to 6.73±0.53 in the inclusion of mixtures of wheat bran, corn and sorghum and full-fat soybean meal in the diets of Nile tilapia fingerlings.

The HSI result of the current study ranged from 2.7±0.08 to 3.23±0.09. In the present study fish groups fed LGM75 and LGM50 had shown higher HSI as compared to fish groups fed on control and LGM25 diets and an increasing trend was observed with increasing the replacement of fish meal by LGM protein at higher levels (Table 6). This increasing tendency might be associated with the storage of glycogen or lipid and is most likely associated with the higher feed intake and high energy content in the experimental diets (Zhang *et al.*, 2012). Hemre *et al.* (2001) pointed out that excessive digestible carbohydrates in fish diet may produce negative effects in glycogen accumulation, liver morphology and function that indicate the nutritional and physiological status of fish. These effects may have a great influence on suppressing the immune system and increasing susceptibility to infectious diseases Abarra *et al.* (2017). However, the HSI result of the present study agrees with the reports of several researchers and it was within the acceptable range for healthy Nile tilapia. For instance, Geremew (2015) reported HSI values ranged from 2.26±0.76 to 3.23±0.25 in the diets of Nile tilapia fingerlings fed with Nigerseed cake meal at inclusion levels of (0, 20 and 40%) of a basal diet. Similarly, Velasquez *et al.* (2016) reported HSI ranged from 1.92±0.22 to 3.13±0.13. Furthermore, Montoya-Mejía *et al.* (2017) have also reported HSI values ranged from 2.1±0.4 to 2.9±0.9.

Conclusions

The results of the current experiment demonstrated that Nile tilapia diets formulated with variable levels of LGM meal as a replacement for a more expensive fish meal protein had no discernable negative effects on all evaluated growth performance and feed utilization parameters and no feed-related mortality was observed. There were no significant differences in growth performance in response to the experimental diets**.** This might be due to a compensatory-effect of both grains which could lead to reducing ANFs and improved palatability and amino acid profile because of mixing feed ingredients. Evaluation of nutritional composition of LGM to replace fish meal at inclusion levels of 0, 25, 50, and 75% in the diets of Nile tilapia fingerlings indicated that it is possible to be used up to 75% with no adverse effect on the whole-body proximate composition. The results of the present study also suggested that replacing fish meal at inclusion levels of 0, 25, 50, and 75% with LGM meal diet as a protein source had no adverse effect on the HSI and hematological parameters of Nile tilapia fingerlings. However, an increasing tendency of HSI in the increased replacement of fish meal at LGM50 and LGM75 was recorded, and interestingly enhanced final body weight and improved feed utilization efficiency of tilapia fingerlings fed on LGM25 was obtained. This increased HSI might be associated with higher FI and the high energy content in the feed that resulted in increased storage of glycogen or lipid in the liver. Since the experimental diets used in the present study were formulated and prepared based on the proximate composition of feed ingredients, studies focused on evaluating Nile tilapia diets based on the amino acid and fatty acid profiles of mixtures of lupin and grass pea meal are recommended as future study. Besides, exploring sophisticated methods that reduce ANFs, maximizing the nutritional profile and increased bioavailability of nutrients may further step-forward the success of complete fish meal replacement with LGM diets. Moreover, extensive research is recommended to see the effect of complete replacement of the expensive and less available fish meal protein in the diets of Nile tilapia on the digestibility, supplement of prebiotic, immune response, intestinal histology, and stress-related factors may also be the focus for further succeeding studies and assure its best potential as a replacement for fish meal.

Ethical Statement

Analysis of all hematological parameters and experimental protocols have been approved by the Animal Protection Index (API) 2020, and under the general surveillance of Ethiopian Criminal Code No. 414/2004 that bans inhumane treatments being inflicted on animals in public places.

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Author Contribution

Tsegay Fisseha: Conceptualization, writing-review and editing, visualization and writing-original draft, formal analysis, investigation, data curation and methodology.

Akewake Geremew: Data curation, formal analysis, investigation, methodology and supervision

Tadesse Fetahi: Fund acquisition, project administration, resources, supervision, writing review and editing

Conflict of Interest

The author(s) declare that they have no known competing financial or non-financial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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References

Abarra, S. T., Velasquez, S. F., Guzman, K. D. D., Felipe, J. L. F., Tayamen, M. M., and Ragaza, J. A. J. A. r. (2017). Replacement of fishmeal with processed meal from knife fish Chitala ornata in diets of juvenile Nile tilapia *Oreochromis niloticus* 5: 76-83.

Abdel-Tawwab, M., Ahmad, M. H., Khattab, Y. A., and Shalaby, A. M. J. A. (2010). Effect of dietary protein level, initial body weight, and their interaction on the growth, feed utilization, and physiological alterations of Nile tilapia, *Oreochromis niloticus* (L.) 298 (3-4): 267-274.

Agbo, N., Madalla, N., Jauncey, K. J. J. o. S., and Technology (2015). Mixtures of oilseed meals as dietary protein sources in diets of juvenile Nile tilapia (*Oreochromis niloticus* L.) 35(3): 11-24.

Akande, K., and Fabiyi, E. (2010). Effect of processing methods on some antinutritional factors in legume seeds for poultry feeding.

Al-Thobaiti, A., Al-Ghanim, K., Ahmed, Z., Suliman, E., and Mahboob, S. J. B. J. o. B. (2018). Impact of replacing fish meal by a mixture of different plant protein sources on the growth performance in Nile Tilapia (*Oreochromis niloticus* L.) diets 78: 525-534.

AOAC (2002). Official methods of analysis 16th edition.

Awad, E., Austin, B., and Lyndon, A. J. J. o. A. S. (2012). Effect of dietary supplements on digestive enzymes and growth performance of rainbow trout (*Oncorhynchus mykiss*, Walbaum) 8(12): 858-864.

Azaza, M. S., Kammoun, W., Abdelmouleh, A., and Kraïem, M. M. J. A. I. (2009). Growth performance, feed utilization, and body composition of Nile tilapia (*Oreochromis niloticus* L.) fed with differently heated soybean-meal-based diets 17: 507-521.

Begum, A., Mondal, S., Ferdous, Z., Zafar, M., and Ali, M. J. I. J. A. F. S. (2014). Impact of water quality parameters on monosex tilapia (*Oreochromis niloticus*) production under pond condition 2(1): 14-21.

Borgeson, T. L. (2005). Effect of replacing fish meal with simple or complex mixtures of vegetable ingredients in diets fed to Nile tilapia *(Oreochromis niloticus)*. University of Saskatchewan.

Brenes, A. n., Viveros, A. n., Arija, I., Centeno, C., Pizarro, M., Bravo, C. J. A. F. S., and Technology (2003). The effect of citric acid and microbial phytase on mineral utilization in broiler chicks 110(1-4): 201-219.

Caruso (2015). Use of plant products as candidate fish meal substitutes: an emerging issue in aquaculture productions.

Diana, J. S., Yi Yang, Y. Y., and Lin, C. K. (2004). Stocking densities and fertilization regimes for Nile tilapia (*Oreochromis niloticus*) production in ponds with supplemental feeding.

Dorothy, M., Raman, S., Nautiyal, V., Singh, K., Yogananda, T., and Kamei, M. J. I. J. C. M. A. S. (2018). Use of potential plant leaves as ingredient in fish feed-a review 7(7): 112-125.

El-Sayed, A.-F. M. (2004). Protein nutrition of farmed tilapia: searching for unconventional sources.

El-Sayed, A.-F. M. J. A. (1990). Long-term evaluation of cotton seed meal as a protein source for Nile tilapia, *Oreochromis niloticus* (Linn.) 84(3-4): 315-320.

El-Sherif, M. S., El-Feky, A. M. I. J. I. j. o. A., and Biology (2009). Performance of Nile tilapia (*Oreochromis niloticus*) fingerlings. I. Effect of pH 11(3): 297-300.

El-Saidy, D. M., and Gaber, M. M. J. A. r. (2003). Replacement of fish meal with a mixture of different plant protein sources in juvenile Nile tilapia, *Oreochromis niloticus* (L.) diets 34(13): 1119-1127.

Ertop, M. H., Bektaş, M. J. F., and Health (2018). Enhancement of bioavailable micronutrients and reduction of antinutrients in foods with some processes 4(3): 159-165.

Espe, M., Lemme, A., Petri, A., and El-Mowafi, A. (2007). Assessment of lysine requirement for maximal protein accretion in Atlantic salmon using plant protein diets 263(1-4): 168-178.

FAO (2006). The state of world fisheries and aquaculture. 2006.

FAO (2010). The state of world fisheries and aquaculture 2010. Facing challenges and seizing opportunities.

FAO, F. J. C. B.-S. (2018). Agriculture Organization of the United Nations 2018 The state of world fisheries and aquaculture 2018-Meeting the sustainable development goals 3.

Fish, B. C., and Thompson, L. U. J. J. o. a., and chemistry, f. (1991). Lectin-tannin interactions and their influence on pancreatic amylase activity and starch digestibility 39(4): 727-731.

Fournier, V., Huelvan, C., and Desbruyeres, E. J. A. (2004). Incorporation of a mixture of plant feedstuffs as substitute for fish meal in diets of juvenile turbot (Psetta maxima) 236(1-4): 451-465.

Francis, G., Makkar, H. P., and Becker, K. J. A. (2001). Antinutritional factors present in plant-derived alternate fish feed ingredients and their effects in fish 199(3-4): 197-227.

Gebreanenia, G. (2018). Evaluation of growth performance and body proximate composition of three strains of *Oreochromis niloticus* L.,(1758) under greenhouse condition. Bahir Dar University.

Geremew, A. (2015). Nutritional evaluation of some Ethiopian oilseed cakes in the diets of juvenile Nile tilapia*, (Oreochromis niloticus* L.) Addis Ababa University.

Glencross, B., Evans, D., Hawkins, W., and Jones, B. J. A. (2004). Evaluation of dietary inclusion of yellow lupin (*Lupinus luteus*) kernel meal on the growth, feed utilisation and tissue histology of rainbow trout (*Oncorhynchus mykiss*) 235(1-4): 411-422.

Goda, A. M. A. S., Wafa, M., El-Haroun, E., and Kabir Chowdhury, M. J. A. r. (2007). Growth performance and feed utilization of Nile tilapia *Oreochromis niloticus* (Linnaeus, 1758) and tilapia galilae Sarotherodon galilaeus (Linnaeus, 1758) fingerlings fed plant protein-based diets 38(8): 827-837.

Goldstein, W. S., and Spencer, K. J. J. o. C. E. (1985). Inhibition of cyanogenesis by tannins 11: 847-858.

Guimarães, I. G., Pezzato, L. E., Barros, M. M., and Tachibana, L. J. J. o. t. W. A. S. (2008). Nutrient digestibility of cereal grain products and by-products in extruded diets for Nile tilapia 39(6): 781-789.

Hafedh, Y. A. J. A. r. (1999). Effects of dietary protein on growth and body composition of Nile tilapia, *(Oreochromis niloticus* L) 30(5): 385-393.

Hassaan, M. S., Soltan, M. A., Mohammady, E. Y., Elashry, M. A., El-Haroun, E. R., and Davies, S. J. J. A. (2018). Growth and physiological responses of Nile tilapia, (*Oreochromis niloticus*) fed dietary fermented sunflower meal inoculated with Saccharomyces cerevisiae and Bacillus subtilis 495: 592-601.

Hemre, G., Mommsen, T., and Krogdahl, A. J. A. N. (2001). Carbohydrates in fish nutrition: effects on growth, glucose metabolism and hepatic enzymes 7: 1-20.

Higuera, M., Garcia-Gallego, M., Sanz, A., Cardenete, G., Suarez, M., and Moyano, F. J. A. (1988). Evaluation of lupin seed meal as an alternative protein source in feeding of rainbow trout (*Salmo gairdneri*) 71(1-2): 37-50.

Kaushik, S. J., and de Oliva Teles, A. J. A. (1985). Effect of digestible energy on nitrogen and energy balance in rainbow trout 50(1-2): 89-101.

Khalil, M. H. S. K., Ma Hashem, S Mazumder, Sk (2012). Substitution of fish meal by meat and bone meal for the preparation of tilapia fry feed.

Khan, M., Siddique, M., and Zamal, H. (2013). Replacement of fish meal by plant protein sources in Nile tilapia (*Oreochromis niloticus*) diet: growth performance and utilization.

Kumar, V., Khalil, W., Weiler, U., Becker, K. J. J. o. a. p., and nutrition, a. (2013). Influences of incorporating detoxified Jatropha curcas kernel meal in common carp (*Cyprinus carpio* L.) diet on the expression of growth hormone-and insulin-like growth factor-1-encoding genes 97 (1): 97-108.

Lambein, F., Travella, S., Kuo, Y.-H., Van Montagu, M., and Heijde, M. J. P. (2019). Grass pea (*Lathyrus sativus* L.): orphan crop, nutraceutical or just plain food? 250: 821-838.

Madalla, N. A. (2008).

Mondal, K., Kaviraj, A., and Mukhopadhyay, P. K. J. I. J. o. A. S. (2012). Effects of partial replacement of fishmeal in the diet by mulberry leaf meal on growth performance and digestive enzyme activities of Indian minor carp Labeo bata 3(1): 72-83.

Montoya-Mejía, M., García-Ulloa, M., Hernández-Llamas, A., Nolasco-Soria, H., and Rodríguez-González, H. J. R. B. d. Z. (2017). Digestibility, growth, blood chemistry, and enzyme activity of juvenile *Oreochromis niloticus* fed isocaloric diets containing animal and plant byproducts 46: 873-882.

Mzengereza, K. (2015). *Nutritional evaluation of plant ingredients for diets of tilapia rendalli in Nkhatabay, Northern Malaŵi*. Mzuzu University.

Naylor, R. L., Hardy, R. W., Bureau, D. P., Chiu, A., Elliott, M., Farrell, A. P., Forster, I., Gatlin, D. M., Goldburg, R. J., and Hua, K. J. P. o. t. N. A. o. S. (2009). Feeding aquaculture in an era of finite resources 106 (36): 15103-15110.

NRC (1998). Nutrient requirements of swine.

Pentyala, J., and Rao, S. (1999). Sustained nitric oxide generation with L-homoarginine 3(3/4): 223-232.

Ridha, M. (2007). A Comparative Study on the Growth, Feed conversion and production of fry of improved and nonimproved strains of the Nile tilapia (*Oreochromis niloticus)*.

Sklan, D., Prag, T., and Lupatsch, I. J. A. R. (2004). Apparent digestibility coefficients of feed ingredients and their prediction in diets for tilapia *Oreochromis niloticus× Oreochromis aureus* (Teleostei, Cichlidae) 35(4): 358-364.

Soltan, M., Hanafy, M., and Wafa, M. J. G. V. (2008). Effect of replacing fish meal by a mixture of different plant protein sources in Nile tilapia (*Oreochromis niloticus* L.) diets 2(4): 157-164.

Stickney, R. R. (1979). *Principles of warmwater aquaculture*. John Wiley & Sons.

Sudaryono, A., And, E. T., and Evans, L. H. J. J. o. t. W. A. S. (1999). Replacement of soybean meal by lupin meal in practical diets for juvenile Penaeus monodon 30(1): 46-57.

Tacon, A., Forster, I. J. I. A.-D., and Guide, B. (2001). Global trends and challenges to aquaculture and aquafeed development in the new millennium: 4-25.

Tadesse, Z., Gebriel, A. W., Jovani, M., Tefera, F., and Degefu, F. J. E. J. o. B. S. (2012). Effect of supplementary feeding of agro-industrial byproducts on the growth performance of Nile tilapia (*Oreochromis niloticus* L.) in concrete ponds 11(1): 29-41.

Tadesse, Z. J. I. V. f. t. u. a. L. V. (2010). Diet composition impacts the fatty acid contents of Nile tilapia, *Oreochromis niloticus* L., in Ethiopian highland lakes 30(9): 1363-1368.

Velasquez, S. F., Chan, M. A., Abisado, R. G., Traifalgar, R. F. M., Tayamen, M. M., Maliwat, G. C. F., and Ragaza, J. A. J. J. o. a. p. (2016). Dietary Spirulina (*Arthrospira platensis*) replacement enhances performance of juvenile Nile tilapia (*Oreochromis niloticus*) 28: 1023-1030.

Viola, S., Arieli, Y., and Zohar, G. J. I. J. o. A.-B. (1988). Unusual feedstuffs (tapioca and lupin) as ingredients for carp and tilapia feeds in intensive culture 40(1): 29-&.

Wang, Y.-B., Li, J.-R., and Lin, J. J. A. (2008). Probiotics in aquaculture: challenges and outlook 281(1-4): 1-4.

Workagegn, K. B., Ababboa, E. D., Yimer, G. T., Amare, T. A. J. J. o. A. R., and Development (2014). Growth performance of the Nile tilapia (*Oreochromis niloticus* L.) fed different types of diets formulated from varieties of feed ingredients 5(3): 1-4.

Yalew, A., Getahun, A., Dejen, E. J. E. J. o. S., and Technology (2019). Effect of replacing fish meal by sweet lupin meal on growth performance of African catfish fingerlings, Clarias gariepinus (Burchell, 1822) 12(1): 1-17.

Yildirim, M., Lim, C., Wan, P. J., and Klesius, P. H. J. A. (2003). Growth performance and immune response of channel catfish (Ictalurus puctatus) fed diets containing graded levels of gossypol–acetic acid 219(1-4): 751-768.

Yones, A.-M. A. S., Eissa, I. A.-M., Ghobashy, M. A. E.-F., and Marzok, S. S. J. E. J. o. H. (2019). Effects of Different Dietary Carbohydrate Sources on Growth Performance and Liver Histology of Nile Tilapia (*Oreochromis niloticus*) Fingerlings 42(3): 599-607.

Yones, A. (2005). Incorporation of lupin seed meal as plant protein source in gil thead sea bream (Sparus aurata) diets. *Journal of Animal Poultry Production.* 30(11): 6553-6564.

Yones, A. J. A. J. B. S. (2010). Effect of lupin kernel meal as plant protein sources in diet s of red hybrid tilapia (*Oreochromis niloticus× O mossambicus*), on growt h performance and nutrients utilization 6: 1-16.

Zhang, Y., Øverland, M., Sørensen, M., Penn, M., Mydland, L. T., Shearer, K. D., and Storebakken, T. J. A. (2012). Optimal inclusion of lupin and pea protein concentrates in extruded diets for rainbow trout (*Oncorhynchus mykiss*) 344: 100-113.