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EFFECTS OF LYSINE AND METHIONINE SUPPLEMENTATION AND COST-EFFECTIVENESS IN PRODUCTION OF NILE TILAPIA DIETS (OREOCHROMIS NILOTICUS) IN WESTERN KENYA

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Abstract
The proximate composition of local feed ingredient is limited by unbalanced dietary amino acid contents, thereby increasing de-amination and ammonia levels in water. This study formulated experimental diets and balanced the Essential Amino Acids (EAA) to enhance the feed nutritive value for culture of *Oreochromis niloticus*. Four diets comprising methionine+lysine and lysine supplemented at 5.1 g kg^{-1}, 2.7 g kg^{-1} to non-EEAs supplemented and commercial diets at the University of Eldoret Fish Farm were tested. Growth performance was conducted in hapas suspended in earthen pond 150 m^2 in a randomized design for 105 days. There were significant variations in temperature (24 to 26°C), Dissolved oxygen (4.8 to 6.2 mg L^{-1}) and pH (7.2-7.6) but within optimal range for tilapia. The diets provided about 17.17 MJ kg^{-1} with 22.9% digestible Crude Protein and 8.03% ash content. Lysine supplemented Diet 2 induced highest mean final weight of 156.05±1.74 g, 2.4 Specific Growth Rate, 1.42 Feed Conversion Ratio and 2.68 Protein Efficiency Ratio. A high profit index (2.286±0.07) at low incidence cost (0.437±0.05) was observed in Diet 2.The study reports reduced production cost by supplementing plant proteins with limiting amino acids hence increasing nutritive value of aquafeeds.

Introduction
Tilapia farming poses a substantial challenge to fish farming in Sub-Sahara Africa despite the huge technological leaps achieved worldwide on improved strains. According to Craig and Helfrich (2002), high cost of fish feeds, comprising of over 50% of the production costs limits tilapia production and sustenance. Sustainability and success of aquaculture depends on type of feed used and management. Success of intensive fish culture depends to a large extent on adequate information on nutrient requirements, especially dietary protein, which is the most expensive component in artificial diets (Tacon et al., 2009; 2011).

Tilapia feed cost depends dietary protein level, the source, and type of ingredients derived from plant or animal sources. Fish meal is very expensive thus increasing the cost of fish production. It competes with man for food and for use in aquaculture feeds. The demand and growth of tilapia farming has resulted in the expansion of nutrient requirement data and improvements in feed formulations (NRC, 1993). Selection of a protein ingredient is not limited to only assessing the crude protein levels of feed ingredients but also involves in-depth knowledge in their amino acid profile and bioavailability.

Commercially available ingredients have significant amounts of anti-nutritional factors and digestibility of the proteins is highly variable. It has been generally noted that nutrient are lost during feed manufacturing and improper storage of aqua feeds in low-technology production systems. Fish growth and survival can be compromised by poorly digested feed that can deteriorate water quality. Pellet type and manufacturing process also influence water quality and digestion efficiency of fish species under culture.

Standard nutrient requirement cannot be applied to practical feed formulation since it is not adequate for high density commercial rearing situations. Many commercial feeds for tilapia contain lower protein levels (17–25 %) which are considerably below recommended levels to reduce the cost of production (NRC, 1993). Unbalanced dietary amino acid contents could result in an increased de-amination and can increase ammonia levels released into the water (Hasan *et al.*, 2007). Information about proximate composition of locally feed ingredient for farm made fish feed is usually limited and not reliable. Thus farmers depend only on the existing information about the feed composition given by different fish feeds manufacturers. There has however been several attempts to profile the nutritional value of different feed ingredients from different agro-ecological and geographical locations in the world (Hasan *et al*., 2007; Tacon *et al*., 2009; 2011).

Fish diets can be improved considerably by inclusion of Essential Amino Acids (EAAs) and EAA supplements whenever they become limiting for fish growth. It is also important to compose and process balanced and biologically available levels of EAAs that meet the targeted species nutrient requirements (Nunes *et al*., 2014). Appropriate dietary Methionine and Lysine levels improve the use of other Essential Amino Acids because they have the ability to reduce the oxidation rate of other amino acids (NRC, 2011). Profiles on amino acid contents of fish feed ingredients provide valuable information necessary for formulating diets that support maximum growth of the fish under various cultural techniques. Commercial manufactures usually produce feeds in bulk, leaving small-scale fish farmers with the option of buying large quantities of expensive feed, which often goes to waste (Pandey, 2013). Small quantities of fish feed required for experimental purposes can be easily made in the laboratory or on-farm, with particular ingredients of known nutritional quality, especially the EAAs.

Feed formulators are now adopting modern and environmentally-sound formulation techniques based on nutrient value, on supplementation with crystalline EAAs and on animal nutrient requirements. Commercial feed formulations are intended to meet nutritional requirement with quality product at cheaper prices depending on type of fish species grown. In commercial aquaculture production feed costs can be reduced by developing proper feed management and husbandry strategies to improve fish growth. Best Management Practices (BMPs) in fish husbandry involve proper stocking densities, nutrient ratios, aeration and water exchange to reduce metabolites that can deteriorate water quality. Plant proteins that are cheap and locally available are used to supplement animal protein at lower cost. Feeds consisting of soybean, wheat and corn meal, canola meal, extruded pea seed meal, supplemented with methionine have been used for formulation of diets for carps, tilapia and catfish without influencing growth performance (Tacon and Metian, 2008).

Homemade feed is useful when specific diets are needed to improve fish growth performance. The nutritional requirements for protein, lipids and energy for optimum growth of specific fish species are necessary for formulating a balanced diet. Lysine and methionine are essential amino acids that cannot be synthesized by the body but are obtained from the diet. Several studies have been conducted on the Amino Acid Nutrition requirements, composition, supplementation and balanced ratios of protein in aqua feeds. (Ketola 1982; NRC 1993; 2011; Ahmadr 2008; Munguti *et al*., 2012).

Materials and methods

Study area

The study was conducted at University of Eldoret fish farm from June to September 2015 in twelve hapas of capacity $1m^3$ suspended in an earthen pond of 150 m^2. The hapas for the experiment were made of foul resistant synthetic netting of mesh size 1.5 mm and were closed from all sides except the top.

Study design

Four diets were tested for the experiment in triplicate for each treatment in a randomized design. The stocking density was of twenty fingerlings per hapa. The experimental ponds 150 m^2 were limed at the rate of $500g^{-2}$ with Agricultural lime ($CaCO_3$) and fertilized at a rate of $3gm^{-2}$ and $2g^{-2}$ with urea and di-ammonium phosphate (DAP) to facilitate primary productivity in the experimental pond.

Acquisition of experimental fingerlings, feed ingredients and feed preparation

Monosex male *O. niloticus* fingerlings averaging 1.25 g were obtained from Sagana National Aquaculture Research and Development Centre (NARDC) and transported to the study area under well oxygenated condition in plastic bags. The fingerlings were acclimatized in holding tanks for two weeks prior the experimental stocking in hapas suspended in ponds. The feed ingredients consisted of freshwater shrimp (*Caridina nilotica*), Cottonseed cake, wheat bran, fish oil, and vitamin/mineral premix. Diet 1 was supplemented with both lysine and methionine at 5.1g $kg^{-1}$and 2.7g kg^{-1} respectively (Santiago and Lovell, 1988). Diet 2 supplemented with lysine only at 5.1g kg^{-1}, Diet 3 did not receive any EAAs supplement; Diet 4 was a commercial fish feed with 32% crude protein content. The three experimental diets were formulated at 30% crude protein at the University of Eldoret fish farm of which Diet 3 was used as a control (Table1).

Dry ingredients were passed through a sieve (0.6 mm diameter hole) before mixing into the diets. The ingredients were weighed and ground to small particle size (approximately 250 μm) and thoroughly mixed with water to obtain a 30% moisture level and partially cooked. Practical diets were prepared using warm water to reduce disintegration and leaching of nutrients in water. Oil, Vitamins, and minerals mixture were added to the diets. Fish oil was included in diet formulations to serve as source of lipids and to enhance feed floatability. The diet was dried for 8 h in the open air and broken to appropriate size of 1.5 to 2mm crumbles. Feeds were offered daily in hapas by broadcasting method at 10:00 am and 4:00 pm. Initial feeding was offered at 10% of body weight adjusted to 5% and to 3% body weight, respectively. Feeding rations were adjusted on monthly basis.

Determination of nutrient composition of practical fish feeds

Feed ingredients were subjected to proximate analysis before diet formulation to determine the crude protein, moisture content, lipid, crude fibre, ash, and digestible carbohydrate. Experimental diets were formulated using Winfeed Ver. 2.8 software. The chemical compositions of the formulated diets was determined following AOAC (1990) procedures: dry matter, by drying in an oven at 105 ˚C for 8 hours; crude fat, by Soxhlet extraction with ether; crude ash, by incineration in a muffle furnace at 580 ˚C for 8 hour; crude protein (N× 6 .25), by the Kjeldahl method after acid digestion.

Sampling for growth of Nile tilapia and water quality parameters

Total weights of fish sample were taken fortnightly using a ruler and 0.01g sensitive weighing balance. A ruler was used to measure the Total Length (TL) in centimetres (cm) while the weighing balance was used to measure weight in grams (g). Water quality parameters measured included dissolved oxygen, temperature and pH. Dissolved oxygen, temperature and pH were determined daily at the surface, in the middle and pond bottom. Dissolved oxygen was measured using an Oxymeter (YSI 200), pH was measured with a glass electrode-pH meter.

Statistical analysis

The water quality parameters were compared through the growth period using one way Analysis of Variance (ANOVA) to test the effects of the diets and to determine whether there were any significant differences among the means. Growth parameters and nutrient utilization were calculated using the following formulae:

$$Body\ Weight\ Gain\ (BWG) = \frac{[Final\ weight\ (g) - Initial\ weight\ (g)]}{Initial\ weight\ (g)}$$

$$Daily\ Weight\ Gain\ (DWG) = \frac{[Final\ weight\ (g) - Initial\ weight\ (g)]}{Time\ interval\ (days)}$$

$$Specific\ Growth\ Rate\ (SGR) \frac{[Ln\ (Final\ weight\ (g)) - Ln\ (Initial\ weight\ (g))]}{Time\ interval\ (days)}$$

$$Food\ Conversion\ Ratio\ (FCR) = \frac{Weight\ of\ dry\ feed\ (g)}{[Final\ weight\ (g)] - [Initial\ weight\ (g)]}$$

$$Protein\ Efficiency\ Ratio\ (PER) = \frac{[Final\ weight\ (g)] - [Initial\ weight\ (g)]}{Protein\ consumed\ (g)}$$

Economic analysis of the commercial and experimental diets
A simple economic analysis was used to assess the cost effectiveness of diets used in the feed trial. The cost of feed was calculated using market prices, taking into consideration the cost of feed and transport, assuming all other operating costs remained constant (cost of fingerlings, hapas and labour). Indices for economic evaluation included:

$$Incidental\ Costs = \frac{Cost\ of\ feeds\ (KES)}{Weight\ of\ fish\ produced\ (g)}$$

and,

$$Profit\ Index = \frac{Weight\ of\ fish\ produced\ (g)}{Cost\ of\ feeds\ (KES)}$$

Results

Proximate composition of practical diet before amino acid supplementation
The practical diets were formulated at 30% crude protein content using cheap and locally available feed ingredients. The commercial diet was indicated to constitute 32% crude protein content. According to the practical diet formulation in this experiment, the following nutritional values were calculated based on the ingredient profiles from tropical areas on as-is basis of dry feedstuff. The diet formulation provided a Gross Energy of 17.17 Mj kg^{-1} based on 5.65 Kcal g^{-1} for protein 9.45 Kcal $g^{-1}$for Fat, 4.1 Kcal $g^{-1}$for Carbohydrate (with 1 Kcal to 4.184 Kj) (Table 2).

The proximate composition of experimental and commercial diet after essential amino acid supplementation (Table 3). Proximate analysis for diet composition on the four tested diet indicated a significance difference at $p<0.05$ in crude protein, crude lipid and ash content. Laboratory proximate analysis after supplementation indicated a slight increase in the crude protein levels (32.3%) and (32.1%) in Diet 1 and 2.The non-supplemented control Diet 3 at 30.3% and the commercial Diet 4 indicated reduced crude protein level of 28.9%.

Growth in weight of Nile tilapia
There was a general increase in body weight of all the experimental fish fed on various diets during the study. However, the growth performance varied significantly at $p<0.05$ for different test diets (Fig. 1). The growth of monosex *O. niloticus* on the four test diets exhibited superior performance on Diet 2 which had lysine supplement, as compared to the commercial Diet 4 at the least end. The methionine/lysine and control (Diet 1 and 3) had an average growth performance.

The overall growth performance parameters (final mean weight, body weight gain and Specific growth rate and feed conversion ratio) are presented in Table 4. There was a significant difference in all the experimental diets fed to *O. niloticus* during the culture period. Diet 2 induced the highest mean weight of 156.05±1.741g within 15 weeks whereas the commercial Diet 4 induced the least mean weight gain of 94.083 ±3.064g. Both Lysine and Methionine supplemented Diet1 indicated a better mean final weight of 125±1.681g while the control diet had 114±1.917g.

All the other performance indicators; Body Wight Gain (BWG), Daily Weight Gain (DWG), Food Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) showed that the lysine supplemented Diet 2 was better than all the other diets. Protein utilization was assessed through protein dependent parameters indicating a significance difference at $p<0.05$. The better PER values were obtained in the supplemented Diets 2 and Diet 1 while lower PER values were obtained in non-supplemented Diet 3 and commercial Diet 4 as shown in Table 4.

Carcass composition of Nile tilapia fed on experimental and commercial diets

There was a significant difference the crude protein, lipid and ash contents between the supplemented and non-supplemented diets at p<0.05. There was no significant difference in the moisture content of all the carcasses fed on different diets. Supplemented diet 1 and 2 indicated crude protein content of (64.21±0.20[a]) and (64.12±0.18[a]) respectively. The Crude Protein and lipid contents in whole fish body were affected by dietary protein level. Crude lipid content was relatively lower in the supplemented diet 1 (10.371±0.21[a]) and Diet 2 (10.346±0.27[a]) as compared to higher crude lipid contents in commercial (14.358±0.23[c]) and non-supplemented control diet (12.704±0.26[b]). The control Diet 3 recorded a relatively lower ash content (13.375±0.93[b]) compared to the supplemented diets as shown in Table 5.

Economic analysis of commercial and experimental diets

The Simple economic analysis of test diets exhibited a significant variation at P<0.05 for different test diets. Simple economic analysis indicated that higher profit index was achieved in the supplemented Diet 2 (2.973±0.07[a]) followed by Diet 1 at a low incidence costs as compared to Diet 4 with low profit index (1.052±0.04[d]) at a high incidence cost (0.951±0.14[d]) as shown in Table 6. The control Diet 3 had an average profit index of 2.272±0.06[c]. The cost per kilogram of purchasing commercial diet was expensive with lower yield in terms of final mean weight gain compared to the formulated practical diet where the ingredients were locally purchased, formulated and supplemented (Table 6).

Effect of water quality on growth performance of Nile tilapia

For temperature, there were significant variations with time ($F_{0.05,15,80}$ = 4.27; p-value<0.00005). Since the p-value of the F-test is less than 0.05, there is a statistically significant difference between the mean temperatures from one level of Days to another at the 95.0% confidence level. Temperature maintained at a range of 24-26°C in all the sampling weeks as shown in Figure 2. The dissolved oxygen (DO) concentration did not show any significant variation with time ($F_{0.05, 15, 80}$ = 1.31; p-value=0.217). Since the p-value of the F-test is greater than or equal to 0.05, there was no statistically significant difference between the mean DO from one level of time to another at the 95.0% confidence level. Dissolved oxygen levels were at a range of 4.8-6 mg L^{-1} (Fig. 2). The water pH was statistically significant in time ($F_{0.05, 15, 80}$ = 7.63; p-value<0.00005). Since the p-value of the F-test is less than 0.05, there is a statistically significant difference between the mean pH from one level of days to another at the 95.0% confidence level. The pH range was 7.2-7.6 throughout the study (Fig. 2). The range of these critical water quality parameters were within suitable range for tilapia culture.

Discussion

Mean Weight Gain (MWG) was statistically (p<0.05) different among all the diets, Fingerlings of *O. niloticus* could be raised to about 156 g with any of the diets used and that the slight variations could be due to chance. The final average weight for Lysine supplemented Diet 2 (156±1.738g) was higher than both lysine and methionine supplemented Diet 1 (125±1.681g) because methionine is limiting in plant proteins but can be partially spared by non-essential amino acids cysteine. Wilson (2002) estimated that cysteine can spare 40–60% of methionine in the diets for various fishes. Dietary lysine supplementation is reported to advantages on weight gain feed conversion, nitrogen retention and reduction in body lipid contents (Marcouli *et al.*, 2006). This is concurrent with the results of the present study reporting better Feed Conversion Ratio, Specific Growth Rate, Daily Weight Gain and Protein Efficiency Ratio. Salama *et al.* (2013) found out that increasing levels of lysine and methionine + cysteine in the diet improved the protein efficiency ratio, and the Feed Conversion Ratio (FCR). The final average weights for non-supplemented and commercial Diet 4 (94±3.064g) had least growth performance among the tested diets because of the dietary content and were deficient of the limiting EAAs.

Diet 4 had the least growth performance because of dietary content, poor digestibility and more of uneaten feeds were found in the hapas at the end of the study. Davies and Wareham (1988) observed that Essential amino acids (EAAs) imbalance could reduce efficiency in growth and feed utilization in plant protein diets. Mai *et al*. (2006a) also reported that PER values were reduced when juvenile Japanese sea bass fed lysine deficient diets, while growth response and diet utilization were improved with supplementation of crystalline lysine. Murai *et al*. (1982) also reported improved growth of fish after amino acid supplementation. The lower FCR

1.42 and 1.21 obtained in Diet1 and 2 indicated better feed utilization and are economical for commercial tilapia culture and were contributed by supplementation with limiting Lysine and Methionine. Lysine and methionine are essential amino acids that cannot be synthesized in the body but obtained from protein in the diet; improving fish appetite and fillet yield (Barrows and Hardy, 2001; Furuya and Sakaguti, 2006). The FCR 1.61 and 1.80 obtained in commercial and non-supplemented diet had relatively higher FCR meaning a larger quantity of the diet and a lot of money is needed to produce a unit of flesh.

Therefore there has been a worldwide increasing effort to supplement many fish diets with EAAs and other additives in order to improve diet quality and enhance growth (Ketola, 1983; Ruscoe *et al*.,. 2005: Tacon and Metian, 2008; Nunes *et al*., 2014). Commercial supplemented amino acids are effective and are consequently available to different fish species; shrimp, rainbow trout, tilapia, grass carp, and catfish (Liu *et al*., 2010; Sangsue *et al*., 2010; Wang *et al*., 2010).Proximate analysis indicated a slight increase in the supplemented Diets (32.3% and 32.1%) compared to non-supplemented 30.2% and commercial Diet (28.9%). The slight increase in crude protein level was attributed by supplementation with crystalline Lysine and Methionine hence improving the nutritive value of the diet and better growth performance. Commercial diet 4 showed reduced crude protein level of 28.9% and this could be linked to the dietary protein content that exhibited least growth among the tested diets. The experimental diet consisted of 0.05-0.054 (gkg^{-1}) crude lipid equivalent to 5-5.4% dietary content that is within the recommended levels. According to Lim *et al.* (2011), the optimum dietary lipid requirement for tilapia is 5 to 12%

The economic benefits of using the test diets lies in the fact that the cost of feed (in terms of Incidence Cost (IC) in raising *O. niloticus* to an average weight of 156g was lowest for fish fed on Diet 2 and Diet 1 (Lysine, both Lysine and methionine supplemented diets). Therefore it could be cheaper to raise fingerlings of *O. niloticus* when fed on Diet 2. The cost per kilogram of Diet 4 was higher than Diets 1, 2 and 3.The higher IC for fish fed on diet 4 shows higher cost of the feed with low biomass produced. Therefore, farm made diets supplemented with Lysine and methionine would be more economical to raise fingerlings *O. niloticus* because it is relatively cheaper. Mai *et al*., (2006b) reported similar findings on supplementation of Lysine to plant protein-based diets to enhance cost-effective reduction of dietary crude protein without affecting fish growth performance. Opiyo *et al*., (2014) reported a cost benefit analysis results for the on-farm formulated feed, to be economically viable for semi-intensive system rearing of *O. niloticus* in the earthen ponds.

Most commercial tilapia feeds in East Africa region have achieved poor performance due to a number of reasons (Munguti *et al*., 2012) while there has been a worldwide increasing effort to supplement many fish diets with EAAs and other additives in order to improve diet quality and enhance growth (Ketola, 1983; Ruscoe *et al*., 2005; Tacon and Metian, 2008; Nunes *et al*., 2014). The cost, availability, and quality of tilapia fish feed still poses a challenge to many small-scale farmers in East Africa.

One possible solution to this feed challenge is for fish farmers to formulate their own feeds in the farm so as to guarantee the desired quality. However, on-farm formulated tilapia feeds may also not solve the feed scarcity and feed quality problems due to the nutritionally unbalanced composition. This experimental study has shown that the feed formulation, processing and nutritional balancing can be achieved through supplementing the diets with EAAs, as these are often the limiting factors in tilapia feed performance. The study has shown that nutritional balancing of tilapia fish feeds can provide up to 17.17 Mj kg^{-1} of energy to fish and at the same time produce high protein efficiency ratio of up to 2.7. The FCR values of 1.44 to 1.24 obtained in this study are close to the ones reported in the literature for extruded diets (1.88; 2.1; 1.5 and 1.7 kg feed for each kg gain in weight).

Conclusion

The study concludes that Amino acids profile is key guide in selection and use of feedstuffs in the production of processed tilapia feeds. Practical feeds for tilapia can be formulated using locally available protein sources at the farm level and supplementing the diets with EAA to produce a nutritionally balance diet for tilapia. Only limiting EAAs require supplementation in the on-farm diet formulations. Supplementing practical tilapia diets with lysine at a rate of 5.1g per kg^{-1} improves the growth performance of monosex *O. niloticus* in hapas.

On-farm practical diets of tilapia can produce superior Daily Weight Gain, Specific Growth Rate, Feed Conversion Ratio, and Protein Efficiency Ratio as compared to commercial feeds in the market. On the other hand proper pond fertilization provides natural food that improve the essential amino acid deficiency in plant based diets. The supply of amino acids from natural food may be an economically attractive strategy of supplementing limiting amino acids in tilapia diets.

The experimentation and testing of the diets have shown that the technology can be implemented by farmers at the farm level with minimal investment in a hammer mill and pelletizing equipment. Farmers can now purchase EAAs and supplement their formulated on-farm diets with recommended quantities. About 600 farmers in Uasin Gishu, 300 farmers in Vihiga County, and 400 farmers in Kakamega counties stand to benefit from this initiative through the University of Eldoret extension initiatives.

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Table 1. Fish feed ingredients and essential amino acid supplementation

Ingredient	Diet 1	Diet 2	Diet 3
Wheat bran	48.0	48.0	48.0
C. nilotica	30.0	30.0	30.0
Cotton seed meal	18.0	18.0	18.0
Fish oil	2.0	2.0	2.0
Trace mineral	1.0	1.0	1.0
Lysine%	1.96% + Supplement 5.1 g kg^{-1}	1.96% + Supplement 5.1 g kg^{-1}	No supplement
Methionine%	+ Supplement 2.7 g kg^{-1}	No supplement	No supplement

.Table 2. Nutrient profile estimate based on diet formulation and feed ingredients database of feed formulator in percentages

Parameter	Nutritive value	Parameter	Nutritive value
DM	90.72	Cholesterol	0.12
Ash (mg kg^{-1})	8.03	Astaxanthin	804.05
Gross Energy (Mj kg^{-1})	17.17	Arginine	2.53
Digestible Energy (Mj kg^{-1})	9.67	Histidine	0.81
Crude Protein	30.15	Isoleucine	1.04
Digestible Crude Protein	22.91	Leucine	2.24
Lipid	5.53	Lysine	1.96
Fibre	6.87	Methionine	0.95
LOA (18:2n-6)	0.72	M+C	2.27
LNA (18:3n-3)	0.03	Phenylalanine	1.57
ARA (20:4n-6)	0.02	P+T	2.62
EPA (20:5n-3)	0.64	Threonine	1.21
DHA (22:6n-3)	0.40	Tryptophan	0.30
Total n-3	1.07	Valine	1.34
Total n-6	0.75	Ca	1.44
n3:n6	1.43	Available P	1.14
Total phospholipids	1.77		

Table 3. Proximate composition of commercial and experimental diets after amino acid supplementation

Composition (g kg^{-1})	Diets			
	Diet1	Diet 2	Diet 3	Diet 4
Dry Matter	90.6	90.6	90.5	90.3
Moisture content	0.38	0.48	0.415	0.47
Crude Protein	32. 3	32.1	30.3	28.9
Crude Lipid	0.054	0.052	0.05	0.036
Ash Content	7.07	7.06	7.07	8.02

Table 4. Growth performance parameters of O. niloticus over 105 days on the four test diets

Parameter	Diet 1	Diet 2	Diet 3	Diet 4
Initial average weight (g)	1.22 ±0.018	1.25±0.019	1.24±0.021	1.25±0.021
Final average weight (g)	125.05±1.68[a]	156.12±1.74[b]	114.03±1.92[c]	94±3.064[d]
Weight of dry feed (g)	250.00	250.00	250.00	250.00
BWG	123.12.±0.02[a]	154.67±0.01[b]	112.26±0.01[c]	92.03±0.01[d]
DWG	0.265±0.05[a]	0.274±0.03[b]	0.247±0.04[c]	0.236±0.05[d]
SGR	2.34±0.01	2.40±0.01	2.25±0.01	2.28±0.01
FCR	1.42	1.21	1.61	1.80
PER	2.60	2.68	2.42	2.32

Values are expressed as mean± SE. The superscripted letters a, b, c, d indicate significant difference at p<0.05

Table 5. Carcass composition of O. niloticus fed on different diets

Composition (%)	Diets			
	Diet 1	Diet 2	Diet 3	Diet 4
Moisture content	7.77±0.12[a]	7.74±0.19[b]	7.69±0.14[b]	7.75±0.16[c]
Crude Protein	64.21±0.20[a]	64.12±0.18[a]	63.10±0.23[b]	58.9±0.19[c]
Crude Lipid	10.371±0.21[a]	10.346±0.27[a]	12.704±0.26[b]	14.358±0.23[c]
Ash Content	14.750±0.52[a]	15.125±0.66[a]	13.375±0.93[b]	14.042±0.55[c]

Values are expressed as mean± SE. Superscripted letters a, b, c and d indicate significant difference at p<0.05

Table 6. Economic analysis of Nile tilapia fed on different dietary treatment in hapas for 105 days

Diets	Cost/Kg feed (KES)	Harvested Biomass	Incidence Cost	Profit Index
Diet1	54.70	125.05±0.04[a]	0.437±0.05[a]	2.286±0.07[a]
Diet2	52.75	156.8±1.738[b]	0.336±0.09[b]	2.973±0.02[b]
Diet3	50.25	114.19±1.917[c]	0.440±0.11[c]	2.272±0.06[c]
Diet4	90.00	94.66±3.064[d]	0.951±0.14[d]	1.052±0.04[d]

Values are expressed as mean± SE in similar row with different superscript letters are significantly different (p<0.05).

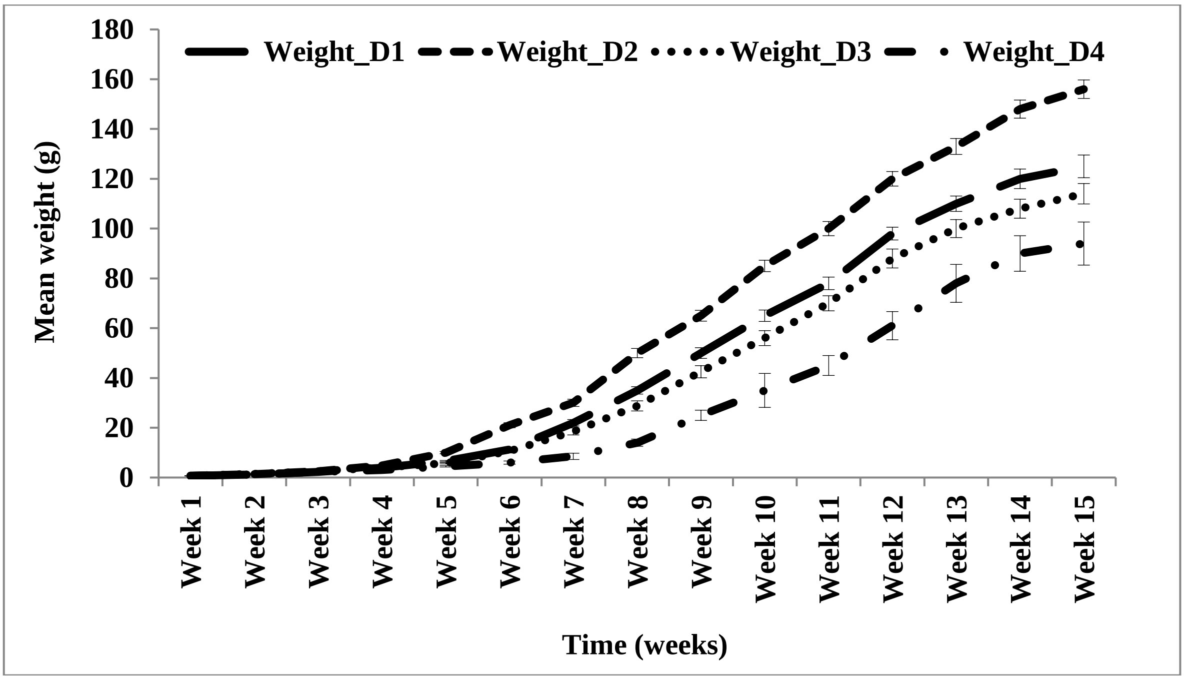

Figure 1. Growth perfomance of monosex O. niloticus over 105 days on the four test diets denoted as D1, D2, D3 and D4.

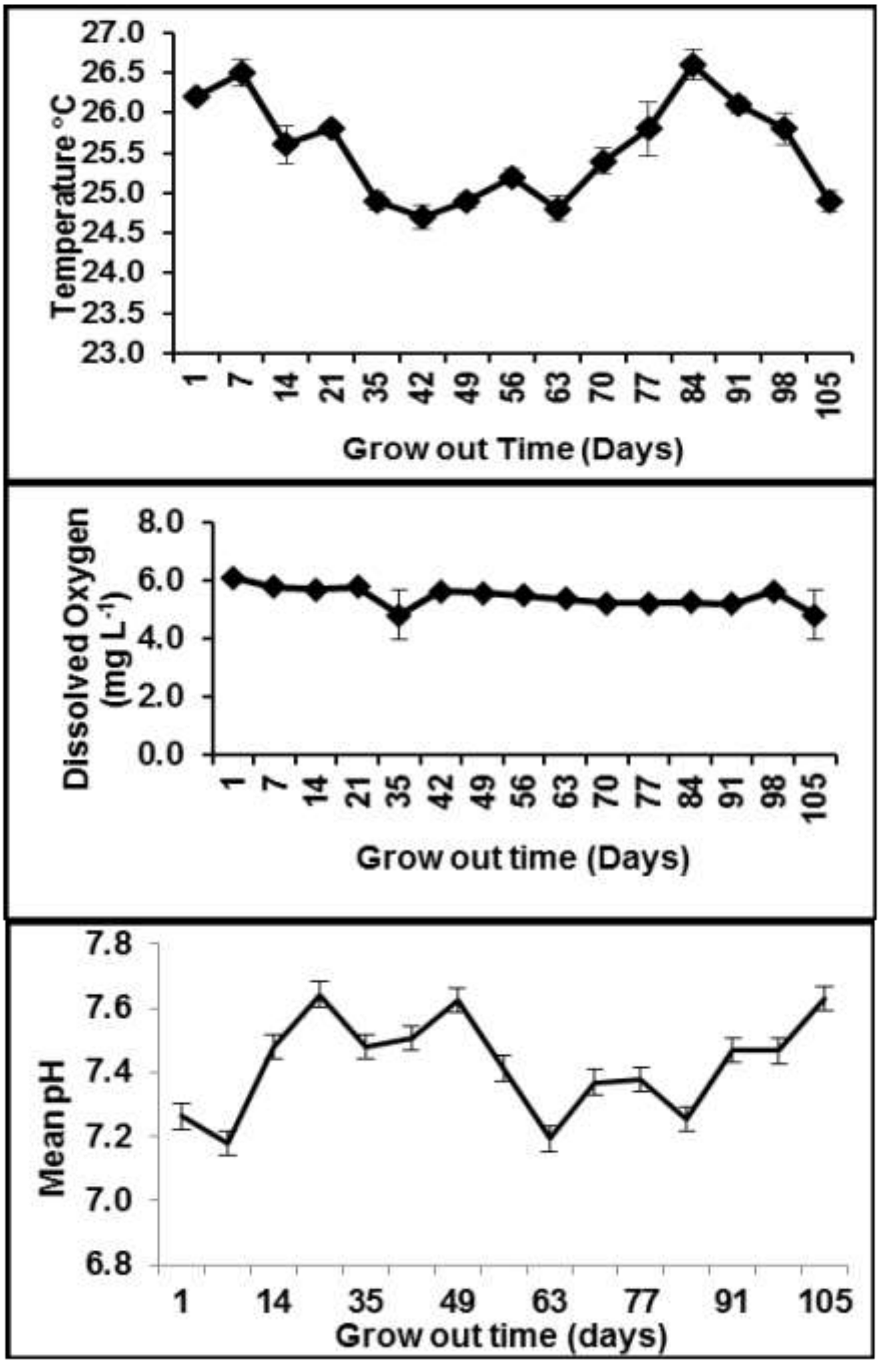

Figure 2. Water quality parameters of experimental pond for 105 days