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CONTENTS

GROWTH PERFORMANCE OF THREE NILE TILAPIA (*OREOCHROMIS NILOTICUS* L, 1758) POPULATIONS FED ON TWO FORMULATED DIETS IN CONCRETE POND, SEBETA, ETHIOPIA

Abelneh Yimer[1], Adamneh Dagne[1] and Zenebe Tadesse[1]

Abstract

Two iso-nitrogenous (29 % protein) diets were formulated and their effect on the growth performance of O.niloticus in unfertilized concrete ponds was evaluated. One of the formulated diets contained vitamin and mineral premix while the other did not. There were significant differences in growth rates of O.niloticus and in feed conversion ratios on the two diets. Fish fed on feed with premix grew faster ($p < 0.05$) than those without premix. The reason for better growth performance of fish fed on the feed with premix can be explained by better metabolic activities, better health and a balanced diet as compared to the fish fed on feed with no premix. Among the three fish populations, fish from Lake Chamo showed better growth performance than fish from either Lake Tana or Lake Hashenge , and the growth performance of Lake Tana fish was the lowest.

Key words/phrases: *Oreochromis niloticus*, minerals, premix, vitamins

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Introduction

The Nile tilapia (*Oreochromis niloticus*) is one of the most studied species among other tilapia species because of its fast growth and ability to feed wide variety of food items, both artificial and natural (Halver and Hardy, 2002; El-Sayed, 2006; Liti *et al*., 2006; Carmen and Geoff, 2007; Royes and Chapman, 2008). *O. niloticus* is at a lower trophic level in its feeding habit, and has an ability to transfer energy from lower food chain to the higher energy level. Moreover, it has flexible feeding habit and has the ability to use more diversified feed resources. The feeding habit preference of *O. niloticus* is also related to the life stage of the fish. Tilapia larvae feed on zooplankton, juvenile of *O. niloticus* feed on phytoplankton and zooplankton, and adult Nile tilapias feed on small invertebrate and other artificial feeds (El-Sayed, 2006). *O. niloticus* is also most widely distributed and commercially important species, accounting for more than 60% of the total catch in Ethiopia (MoARD, 2008). It is also widely farmed in different aquaculture systems for subsistence or commercial purposes in most semi-intensive culture systems (El-Sayed, 2006).

Feeding fish is the main activity in aquaculture production systems and accounts for 40-50% of the total production costs in semi-intensive culture systems (Craig and Helfrich, 2002; FAO, 2006). In order to develop aquaculture production system it is necessary to provide cheaper and locally available feedstuff to reduce expenses. Industrial and agricultural by-products have been used as sources of suitable and cheap feed (FAO, 2006; Munguti *et al*., 2006, Kassahun Asaminew *et al*., 2012). However, the production of fish increases by improving feed quality and by maintaining the health of fish through supplementing formulated protein rich feed stuff with vitamins and minerals (Lim and Webster, 2006).

Vitamins and minerals are needed in small amounts for normal growth, better health and metabolic activities (Carmen and Geoff, 2007). In Ethiopia, only limited studies have been conducted on the effect of fish feeds based on agro-industrial byproducts on the growth performance of fish (Kassahun Asaminew *et al*., 2012; Zenebe Tadesse *et al*., 2012). However, all these attempts failed to include vitamins and minerals in the formulated fish feed. The present work was initiated in order to evaluate the growth performance of three *O. niloticus* populations by feeding two locally formulated feeds.

Materials and methods

The study was conducted at National Fisheries and other Aquatic Life Research Center, Sebeta, Ethiopia (8°55.076'N; 38°38.161'E), located 24 km South-west of Addis Ababa, the capital city, at an altitude of 2240 m above sea level. Two iso-nitrogenous (29 %) diets were formulated from brewery waste, noug (*Guizotia abyssinica*) cake and wheat bran. The formulated feed was fed in pelleted form using 2 mm mesh size sieve for the first two months and 3 mm mesh size sieve for the later months. Amount of inclusions and proximate compositions of the ingredients of the two tested feed types are shown on table 1.

Table 1: Ingredient and proximate composition (dry weight basis) of locally available feeds.

	Test diets	
Ingredient	**With premix**	**Without premix**
Brewery waste	35	40
Noug cake	54	51
Wheat bran	9	9
Premix	2	-
Total	100	100
Proximate composition (g per 100g)		
Dietary protein levels	29	29
Lipid	4.8	5
Crude fiber	22	22

One of the formulated diets was supplemented with commercial (Deutsche Vilomix GmbH) vitamin and mineral premix at 2% inclusion level (Chapman, 1992). The premix is a composition of different vitamins and minerals (Table 2).

Table 2: Vitamin and Mineral Composition of the premix (per kg of premix).

vitamins	content	vitamins	content	minerals	content
A	4000 000 IU	Nicotinic acid	30 000 mg	Iron	25 000 mg
E	40 000 mg	ca-pantothenate	10 000 mg	Copper	1200 mg
D3	400 000 IU	p-amino benzoic	20 000 mg	Manganese	4000 mg
B1	3000 mg	folic acid	1000 mg	Zinc	6000 mg
B2	4000 mg	Biotin	300 000 µg	Iodine	400 mg
B6	3000 mg	inositol	50 000 mg	selenium	20 mg
B12	8000 µg	C	50 000 mg	Cobalt	200 mg
K3	1200 mg				

Proximate analysis of the feeds was carried out as described in AOAC (1990) in triplicates. Protein content of the diets was determined by using micro-Kjeldhal method; percent fat was measured by using ether extraction method, while crude fiber content was known by acid-alkali digestion method.

Partitioned concrete ponds ($25m^2$) were dried and limed for two weeks and filled with water before the experiment was carried out. After two weeks of conditioning of the ponds, *O. niloticus* fingerlings with mean weight range of 13-17 g were stocked with a stocking density of 2 fish m^{-2} in triplicates. The duration of the experiment was from November 2013 to October 2014. The sources of the parents of the experimental fish were from Lakes: Hashenge, Tana and Chamo. After collecting the parent fish from each lake, they were allowed to produce first and second generations at Sebeta research center. Second generation of the fish was utilized for the experiment. The fish were fed at 5 % of their body weight daily (half of the feed at 10:00 am of the day and the remaining half at 4:00 pm). A sample of 50 % of the fish stocked was measured every month using seine netto collect data on length-weight increment. Feed was adjusted based on the monthly weight gain. At the end of the experiment, all fish were counted and length-weight data were taken. Specific growth rate (SGR), feed conversion ration (FCR) and survival rate were calculated using the following formulae (Ridha, 2006):

SGR (% day^{-1}) = $(\frac{\ln Wf - \ln Wi}{dt})x100$, Where: - Wf and Wi are the final and initial body weight of the fish, respectively.

- dt is the time interval in days during the study period

FCR = $\frac{FI(g)}{Wg(g)}$, Where, FI is amount of feed intake (g) in dry weight basis , Wg is Weight gain in gram (g)

Survival rate (%) = $(\frac{NSF - NDF}{NSF})x100$, Where NSF and NDF are the No. of stocked and dead fish during the study period, respectively, and the fish weight gain was calculated by the difference between the final weight and the initial weight.

Water temperature, dissolved oxygen (Do), pH and conductivity of the experimental ponds were measured three times a day (morning, noon and afternoon) using multi-line probe. Water transparency was measured using a black and white Secchi-disk. To get information on the natural food conditions of the experimental ponds, chlorophyll A was analyzed using photo-spectrometer. Nutrients like phosphorus and ammonia were also analyzed in the laboratory following a standard procedure. Plankton samples were also taken by using plankton nets (30um for phytoplankton and 50 um for zooplanktons) and identified by using binocular microscope.

Statistical Analysis

Two-way ANOVA was used to test for the presence of significant differences among the feedstuff, and Duncan's multiple range test was used to differentiate the level of significance of the means at $p<0.05$. All data were analyzed by using the SPSS version18 method.

Results and discussion

Physico-chemical Parameters

The mean temperature of the experimental units during the beginning of the experiment in November/December was 16.8 ^{0}C in the morning and 20.4 ^{0}C in late afternoon. Low water temperature is expected during these months as the regular cool season usually occurs from October to December in most parts of Ethiopia. However, there was an increasing trend in the water temperature after December where the minimum and maximum were 20.4 ^{0}C and 24 ^{0}C, respectively. Temperature of the water body has significant impact on chemical and biological features of the aquatic system (Table 3).

According to Kassaye Balkew (2012), at lower water temperature or below the critical level, fish could stop feeding and would even die. The metabolic activity and physiological functions of aquatic animals (e.g. feed utilization, feed conversion, growth rates) can be affected by the water temperature (Halver and Hardy, 2002; Azaza *et al.,* 2008; Kassaye Balkew, 2012; Zenebe Tadesse *et al*., 2012). This was evident from our result where fish growth rate has increased as the water temperature rises (Fig. 1).

Table 3: Physico-chemical values (mean $\pm$ SD) of the experimental ponds.

Parameters	With premix	Without premix
Temperature (^{o}C)	20.4 ± 0.09^{a}	20.0 ± 0.13^{a}
Oxygen (mg l^{-1})	12.7 ± 0.4^{a}	9.4 ± 0.37^{a}
pH	7 ± 0.10^{a}	6.8 ± 0.14^{a}
Conductivity(µs cm^{-1})	185.7 ± 1.98^{a}	182.9 ± 1.63^{a}
Ammonia- nitrogen (mg l^{-1})	0.12 ± 0.05^{a}	0.14 ± 0.01^{a}
Total phosphorus (mg l^{-1})	0.12 ± 0.01^{a}	0.13 ± 0.02^{a}
Secchi depth (cm)	46.9 ± 3.0^{a}	46.9 ± 3.0^{a}
Chlorophyll-a (µg L^{-1})	23.4 ± 3.9	18.2 ± 2

*Values with the same superscript in each row are not significantly different.

Temperature has also an indirect effect on the survival and growth of fish. According to Wetzel (2001), the solubility of dissolved oxygen (DO) which is essential to all forms of aquatic life depends on temperature, pressure and altitude. Dissolved oxygen (DO) concentration in the pond water at three different times of the day ranged between 6 and 14.5 mg/l. DO concentration was higher during November-December and started to decline when the temperature increases. Relatively, the DO concentration was higher in this study as compared to other similar studies (Liti *et al.*, 2005; Kassaye Balkew, 2012). This could be due to increased algal growth which had created low grazing pressure with fish utilizing commercial feeds.

The level of nutrients assessed was only for total phosphorus (TP) and ammonia-N. The reason behind measuring only these two nutrients was, TP is essential in the algal biomass production, which avails natural food to the fish, while the concentration of ammonia in water is important as it is toxic to aquatic life. Fish cannot survive when ammonia, in unionized form, is high in pond water (El-Shafai *et al*., 2004; Brook Lemma, 2008). In the present study, the concentration of TP and ammonia-N were 130 and 140 μg/l, respectively. Thus, the concentration of ammonia was within the range that cannot lead to toxicity. This could be due to the fact that the supplemental feed was delivered in a suspended net plate where the fish can utilize readily with no feed leftover to settle down at the bottom to form ammonia. The other probable reason could be the suspended net plate was on the surface of the well oxygenated part of the water column and hence ammonia formation was not fast.

Biological variables

Phytoplankton species composition

A total of 17 phytoplankton species of which six were green algae, four diatoms, four blue greens, two euglena and one dinophyta were identified during the study period (Table 4). The most frequently observed algal species were *Pediastrum*, *Haematococcus* and *Euglena* species.

Table 4. Phytolankton species identified during the study period (1-4 indicates frequency of occurrence of species: 1= rare, 2= sporadic, 3= common, 4= abundant)

Group	Species	Relative abundance
Chlorophyta	*Pediastrum*	4
	Haematococcus	3
	Zygnema	3
	Staurastrum	1
	Coelastrum	1
	Scenedesmus	3
Bacillariophyta	*Cymbella*	1
	Navicula	1
	Nitzschia	1
	Synedra	1
Cyanophyta	*Anabaena*	3
	Anabaenopsis	2
	Microcystis	1
	Oocystis	1
Dinophyta	*Peridinium*	1
Euglenophyta	*Euglena*	4
	Phacus	2

Source: Own survey

In aquaculture feed trial experiments, one has to consider the natural food conditions in the experimental ponds. *O. niloticus* is capable of using a wide range of food materials, such as plankton (phytoplankton and zooplankton) and also grows well on artificial feed. The phytoplankton community in the present study consisted of high number of Chlorophyta and Bacillariophyta species which can be utilized by the fish. The presence of diverse phytoplankton species in each experimental pond could be due to less consumption by the fish in preference of the supplemental feed. Probably, the transparency of the pond water Secchi depth greater than half of the pond water depth could favor most phytoplankton species to grow. The other reason for such diverse phytoplankton species could be the absence of large filter feeding cladocerans, such as, *Daphnia* species (Table 5) (Fernando, 1994; Sarma*et al*., 2005).

Zooplankton species composition

Zooplankton species identified from the experimental ponds belong to three groups: *Rotifera*, *Cladocera* and *Copepod* (Table 5). A total of 14 zooplankton species were identified where *Rotifers* being the species contributing to greater than 70% of the total zooplankton taxa. On the other hand, *Copepods* were represented only by one species and *Cladocerans* by two species.

Rotifers account for three-fourth of the total zooplankton species identified during the study period. Absence of large Daphnia species could favor the diversity of *Rotifers*. On the other hand, such zooplankton species composition also reflects typical tropical aspects where *Rotifers* being the diverse taxa (Green, 1993; Fernando, 1994; Adamneh Dagne *et al*., 2008).

Table 5: List of Zooplankton species identified during the study period (1-4 indicates frequency of occurrence of species: 1 =rare, 2 = sporadic, 3 = common, 4 = abundant).

Group	Species	Relative Abundance
Copepods	Cyclopoid copepod	3
	Nauplii spp.	4
Cladoceran	*Moinamicrura spp.*	2
	Diaphanoso maexcisum	3
Rotifera	*Asplanchina spp.*	3
	Brachionus calyciflorus	4
	Brachionus plicatilis	4
	Brachionus caudatus	2
	Brachionus plicatilis	1
	Trichocerca spp.	3
	Hexarthra spp.	1
	Kera tellatropica	2
	Kera tellaquadrata	2
	Polyarthra spp.	1
	Lecane spp.	1

Growth performance of *Oreochromis niloticus* populations

The fish population that had consumed formulated feed with premix showed higher mean weight than fish which received formulated feed without premix ($p < 0.05$). Among the three fish populations, *O. niloticus from* Lake Chamo had shown better growth performance than fish population from either lakes Tana or Hashenge, while Tana fish had shown the lowest performance (Table 6). The Growth trends of *O. niloticus* fed with and without premix are presented in Figure 1.

Table 6: Growth performances of *O. niloticus* fed on premix and without premix dietary treatments.

Parameters	L. Hashenge *O.niloticus*		L. Tana *O.niloticus*		L. Chamo *O.niloticus*	
	Fed with premix	Fed without premix	Fed with premix	Fed without premix	Fed with premix	Fed without premix
Stocking weight (gm)	16.6±0.5^{a}	16.4±0.6^{a}	15±1.6^{a}	13±0.7^{a}	17.3±0.4^{a}	16.7±0.4^{a}
Harvesting weight (gm)	177.8±10^{a}	135.1±11.5^{b}	160.7±11^{ac}	147.1±11.8^{bd}	250.0±7.7^{e}	209.7±4.6^{f}
Weight gain (gm fish^{-1})	161.2	118.7	145.7	134.1	232.7	193.0
Average daily gain (gm day^{-1})	0.44	0.32	0.40	0.37	0.64	0.53
Specific growth rate (SGR)	0.65	0.57	0.65	0.66	0.73	0.69
Feed conversion ratio (FCR)	3.9	4.6	4.0	3.8	2.3	2.5

*Values with the same superscript in each row are not significantly different.

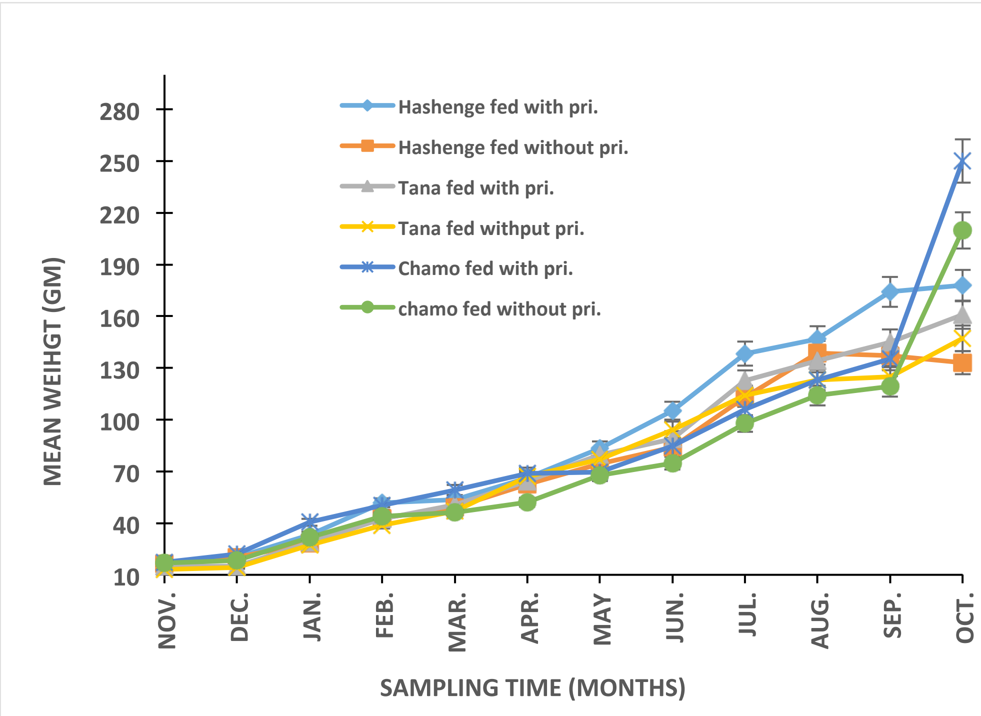

Figure 1. Growth curves of *O. niloticus* for the experimental ponds.

The observed growth differences could be due to vitamins and mineral additives (premix), as they are known to contribute for higher metabolic activity, better appetite, protein digestion, lysine oxidation, better health and growth (Pillay and Kutty, 2005; Carmen and Geoff, 2007). The deficiency of such premixes in feed could result in impaired carbohydrate metabolism, poor appetite, muscle atrophy, low hemoglobin and poor growth (Halver and Hardy, 2002; Carmen and Geoff, 2007). Thus, fish on formulated feed regime without premix had poor growth performance. It was also observed that fish consuming feed with premix finish their feed faster than those fed without premix. In contrast to other studies, which were conducted in fertilized ponds (Foteder, 2004; Liti *et al*., 2005; El-Sayed, 2006; Ispır *et al*., 2011), a positive effect of the premix on growth performance was obtained in this study. This difference could be due to the use of unfertilized ponds in this study and also the culturing pond contained low biomass of natural food, as the experimental fish depended mainly on the supplementary feed.

With regard to growth rate of fish, two growth characteristics were observed (Fig. 1). Towards the beginning of the experiment, the growth curves of fish had overlapped, suggesting that the nutrient supply from the natural source was adequate for fish growth in both treatments. Thus, during this phase, there was no need to supplement micro-nutrients to the formulated feed because natural foods are rich in vitamins and minerals. Therefore, the natural food was able to adequately compensate for the inadequacies of micro-nutrients in the formulated feeds (Liti *et al*., 2005 and El-Sayed, 2006). Also in this phase, the growth performance of fish were slow, may be due to low water temperature (16.8 - 20.4 ^{0}C) of the ponds. Zenebe Tadesse *et al*. (2003) and Kassaye Balkew (2012) had indicated that water temperature below 20^{o}C can lead to reduced feeding of the fish.

During the second phase, from January to October, fish growth rate differences were observed among the dietary treatments. The growth rate of the fish was also faster during this phase, may be due to an increase in the water temperature (20.4 - 24 ^{0}C) which might have increased the metabolic activity of the fish. Hence, there is a need to incorporate premix (additives) to the feed formulation at this stage.

Among the three fish populations, *O. niloticus* from lake Chamo had shown better growth performance than either lakes Tana or Hashenge population. However, the growth rate of O. *niloticus* population from lake Ashenge was higher from May to August (Fig. 1). This might be due to environmental factor where the fish is adapted to live. Lake Hashenge is located at an altitude of 2443 m.a.s.l and the water temperature is in the range of 16-22^{o}C, which has similarity with the experimental site (16.8 – 19.5 ^{o}C). That may be the reason for the fish brought from this lake adapted well and grew better during the experimental period. From September to April, however, the growth rate of *O. niloticus* population from Lake Chamo had shown better performance than fish from other lakes. As Lake Chamo is one of the rift valley lakes and the annual mean temperature of the valley is more than 23 ^{o}C, which has almost similar water temperature with the experimental site (19-24 ^{o}C); the fish from that area might have found a favorable condition for growth during that period.

Under all circumstances, O. *niloticus* population from Lake Tana had shown the lowest growth performance. The reason might be that the fish from this lake could not feed actively during the culturing period as compared to others.

During the experimental period, gender hierarchy and size differences of fish were observed, which might be due to the use of mixed-sex in the trial. Most of the females were smaller in size and the males were relatively bigger. According to Adamneh Dagne *et al* (2013), the growth rate of mixed sex tilapia is lower as compared to male tilapia. This may be due to the fact that female *O. niloticus* spent much energy for egg production as compared to males and hence reflected on their size.

Conclusion

The present study had shown that supplementation of formulated agro-industrial by-products with vitamins and minerals resulted in better growth performance of *O. niloticus* in unfertilized ponds. The Chamo *O. niloticus* population showed better growth performance than either Tana or Hashenge. The Chamo *O. niloticus* population could be considered as potential fish to improve tilapia production at higher altitude and low water temperature conditions. Further studies should focus on testing the premix under different conditions, like in fertilized ponds, use of single-sex fish and under different agro-ecological conditions.

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