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FOREWORD

Aquaculture development in Zambia has had to undergo various positive aspects and credit goes to all the relevant officers and the key stakeholders for their passion to see to it that aquaculture is implemented and appreciated for its job creation, wealth creation, nutritional needs and sustenance and poverty reduction. Information needed to promote and sustain this development is generated at great cost and commitment and the production of this 4th Research and Technical Report is a feat worth appreciating.

The National Aquaculture Research and Development Centre (NARDC) took it upon itself to produce and distribute the first three of such publications and as such must be commended. There have been varied reactions to these publications and the positive ones have led to the production of the copy you are now holding in your hands. A special editorial team was constituted to produce this report that gives a glimpse of what aquaculture researchers are involved in. I am sure that they are going to be proud producers of this report.

It must be pointed that not all the reports received from the Aquaculture Researchers could be included in this edition and the Editorial team had difficulties accepting most of them. However a special recognition and appreciation goes to all the Researchers who contributed their works for inclusion in this report. An earnest appeal goes to those Researchers who have not made any effort to submit their work for consideration by the editorial team to do so quickly. We hope to receive back for consideration, for future publications, those reports that were sent back for more information and clarifications. There is still room for more reports to come in especially from those Research stations that are still not sure of what to submit for publication. The longer you hold on to your findings, the more harm you are doing to our stakeholders who are eagerly waiting for the dissemination of professional information.

An earnest appeal to all the readers is that you kindly make your contributions and reactions known in good time so that the Editorial team can give you a good product all the time. Suggestions on how to improve on the quality of these reports are most welcome and must be sent to the National Aquaculture Research and Development Centre (NARDC) who are the secretariat.

A special word of appreciation goes to Dr. Abila of the Programme for Luapula Agriculture and Rural Development (PLARD) for logistical and technical support towards the production of this report. The same goes to Misamfu Aquaculture Research Station (Kasama) for ably hosting the Editorial team and to all the Editorial team members for their commitment.

I would like to call upon all the stakeholders to sponsor future publications or to indeed help in information dissemination in one way or the other. Our vision is to produce an Extension manual that will have simplified extracts from this research and technical report. This Extension manual will be in English and in recognized vernacular languages of Zambia.

Let me also take this opportunity to thank and acknowledge the Government of the Republic of Zambia for providing funds to carry out programmes in all our Aquaculture Research Stations under the auspices of the Department of Fisheries in the Ministry of Agriculture and Livestock.

Last and not the least, let me thank all the readers of this report for the time taken to read and appreciate our efforts. Your positive and constructive criticism will be most welcome.

Let us go fish farming.

John Mwango
Acting Deputy Director – Aquaculture
Department of Fisheries

INTRODUCTION

Aquaculture is a dynamic sub-sector of Zambia's economy ably guided by the principles and themes as outlined in the National Aquaculture Strategy and National Aquaculture Development Plan. Aquaculture production currently stands at about 13 000 MT and this production is likely to increase owing to the different production system being promoted and used all over Zambia and the evident active participation of the private sector. This growth is welcome and it must be met with ecologically sustainable fish production practices. Pertinent issues requiring immediate attention include among others; water and air pollution, biotechnology, biosecurity and human and fish health. Climate change effects on the environment and on humans and fish must be well understood and mitigation measures for any identified negative effects quickly documented and disseminated for implementation.

For Aquaculture development to be sustainable it needs to adopt an ecosystem approach that holistically considers all the facets of Aquaculture as an important and unique food production system. The ever growing human population needs to be fed and fed it must be with nutritionally balanced diets. The demand for clean Aquaculture products by well informed clients is on the rise and this therefore calls for interventions that will produce fish to meet these aspirations. Ultimately, this calls for some serious re-thinking and more innovative and more investigative approaches by the researchers in generating credible information as appropriate. Time has come to have researchers who are visionary and awake to the needs of the industry - Researchers who are passionate and proactive in their dealings, who appreciate and acknowledge that research is expensive and that resources spent on research are supposed to be well utilized. The results of research are supposed to be clear and appreciated by all the stakeholders through timely elaboration and dissemination. The tools for information dissemination are many and must be fully exploited for the maximum benefit of end users.

Research offices at NARDC

Experimental semi – concrete ponds at NARDC

FEEDING RATIONS OF *OREOCHROMIS TANGANICAE* (GÜNTHER, 1894) GROWN IN SEMI – CONCRETE PONDS BASED ON GULLAND AND HOLT PLOT

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Abstract
An experiment was conducted in semi – concrete ponds for 150 days at the National Aquaculture Research and Development Centre (NARDC), Kitwe, Zambia. The aim of the experiment was to determine the optimal feeding ration of *Oreochromis tanganicae* raised in semi – concrete ponds based on Gulland and Holt plot. Three feeding rates (1% (T1), 3% (T2) and 5% (T3)) of the fish live weight were assigned to the ponds as treatments. The fish raised on 3% showed the highest K and L_{150}(mm). This was followed by the fish fed at 5% of the total body live weight. The Growth index (Ø) for T2 showed the highest index of 2.38. Significant differences ($P< 0.05$) were observed in size variation with T1 having the greatest. The slopes of the weight – length relationships were significant different ($P < 0.05$) among the treatments (T1 = 2.818, T2 = 2.945 and T3 = 3.001). However, they were close to the factor of 3 showing that the fish in all the treatments exhibited isometric growth. The GMR was highest in T1and was significant ($P < 0.05$). The results of this experiment show that feeding *O. tanganicae* at 3% in semi – concrete ponds would be optimal biologically. However, economically, 1% feeding ration is ideal if fish is sold on weight basis. The use of Gulland and Holt plot in this experiment shows that the method can easily be applied in other aquaculture experiments.

Key words: Feeding ration, *Oreochromis tanganicae*, Gulland and Holt, growth index, gross margin ratio.

Introduction

In aquaculture production traits in particular weight gain are indispensable as the motive for a farmer is fast growth and profit. Therefore, in aquaculture research comparison of time for the fish to reach a desirable market size (length or weight), growth rates (example; specific growth rate) or production at the end of the growing period is the normal procedure (Pauly *et al*., 1993). However, feed in this case becomes very important. But how much feed should be given to fish in the grow-out facilities such as fish ponds has been the topic of discussions among aquaculturists. Feed accounts for up to 60% of the total operational cost of producing fish (Virk & Saxena, 2003; Madalla, 2008).Therefore, the optimal feeding strategies would minimise feed wastage, reduce size variation and increase efficiency (Tekinay, 1999; Kubitza & Lovshin, 1999; Lee *et al*., 2000; Dwyer *et al*., 2002).

In continuous experiments, ANOVA or T – test are frequently used to estimate the responses. However, these tests exhibit low power tests even when variances are low since two or three replicates are used (Searcy – Bernal, 1994; Shearer, 2000). They also equal feeding experiments that involve dose response to discrete rather than continuous. Furthermore, they require that fish has the same size at the start of the experiment. They further ignore the information collected between the start and termination of the experiment.

Growth in fish is not linear but asymptotic or sigmoid if length and weight are used respectively in analysing the growth. The von Bertalanffy Growth Function (VBGF), $Wt = W\infty*(1 - e^{-k(t - t_0)})^m$ or $L_t = L\infty*(1 - e^{-k(t - t_0)})$ where Wt and Lt = weight or length at time t, K = growth component, $W\infty$ and $L\infty$ = asymptotic weight and length the fish would reach if was grow indefinitely, t = age of the fish in days/months/years, t_0 = theoretical (generally negative) age of fish at age zero size and m = exponent of length - weight relationship but is equal to 3 in case of isometric growth, has been utilised as the best mathematical model in describing fish growth in fisheries research (Pauly *et al*., 1993). The VBGF has become more popular in aquaculture research too (Pauly *et al*., 1988; Springborn *et al*., 1992; Pauly *et al*., 1993; De Graaf & Prein, 2005). Hopkins (1992) proposed the use of VGBF as a routine function in growth estimates rather than the absolute or percent growth rates commonly used. Pauly & Munro (1984) introduced the growth index called the Phi-prime: (Ø) $=\log_{10}K + (2/3)\log_{10}W_\infty$and $\log_{10}K + 2\log_{10}L_\infty$= curvature parameter which is the slope on the changes of weight over time-mean weight regression, W_∞ and L_∞ as defined above. Aquaculture researchers can estimate the VBGF growth parameters K , L_∞ and W_∞ by the method termed the Gulland and Holt Plot (Gulland and Holt, 1959; Pauly *et al*., 1993).However, this method solely depends on length measurements.

Despite the numerous advantages of using the VBGF, few aquaculture researchers have used its parameters in growth experiments probably due to its requirement to routinely sample the fish and to further calculate the K and W_∞. This paper investigates the optimal feeding rate of *Oreochromis tanganicae* in obtaining the maximum growth rates using the multiple regression based on the Gulland and Holt plot.

Materials and methods

The experiment was conducted at National Aquaculture Research and Development Centre (NARDC), Kitwe, Zambia for one hundred and fifty days (150) days in semi – concrete ponds (concrete side wall but earth at the bottom). The ponds ($250m^2$) were stocked with *O. tanganicae* (39.301±22.914g; Mean ± standard deviation) at $1.3fish/m^2$.

Before stocking, a sack of chicken manure weighing 50kg was put in each pond ($0.2kg/m^2$). Forty fish were randomly sampled, weighed and measured for weight and length (standard length, SL and total length, TL) according to Skelton (2001). Monthly, thirty fish were seined, weighed and their length measured. At termination, all the ponds were drained and allowed the fish to congregate on the sump before being scooped, counted and batch weighed.

Isonitrogenous (30%) and isocaloric (4.02 kcal/g) diet was prepared using WinFeed 2.8 package after proximate analyses of ingredients (soya bean cake and maize bran) as shown Table 1. The ingredients were ground and mixed thoroughly to achieve a homogenous sample. Water (5 – 10%) was then added before taking the mixture to a pellet making machine (BSW 330) attached with a 3.2mm metal die. The feed was then spread on the sacks and sundried for two days. Upon drying, the feed was put in the plastics bags and put in a cooler set at 8^oC till used.

The fish were provided with rations of 1% (T1), 3% (T2) and 5% (T3) live body weight using the feed formulated as shown in Table 1. Feeding was done twice daily at 10:00hours and 15:00hours excluding Sundays. Adjustment of the amounts of the feed was done every month after sampling. Fish mortality was monitored daily. The fish that died were not replaced.

Table 1: Formulation and proximate composition of diets (%) used to feed *O. tanganicae*

Ingredients	Amount (%)
Soya bean cake (%)	57.3
Maize bran (%)	29.5
SoyaGold oil (%)	11.2
[1]Vitamin premix (%)	1
[2]DCP (%)	1
Proximate analysis	
Moisture (%)	18.6
Crude protein (%)	30
Crude Fat (%)	20
Crude ash (%)	3.8
Carbohydrates (%)	33.4
Crude fibre (%)	5.7
Gross Energy (Kcal/g)	4.320
P/E (g protein/Kcal)	0.069

Water temperature, Dissolved Oxygen (DO), electrical conductivity and pH were collected three times a week using YSI professional Plus water checker.

Data analysis

The VBGF parameters (K, L_∞ and W_∞**)** were calculated as follows:

K = slope of the regression of mean length in time interval and growth rate (GR), where;

$$GR = \frac{Changeinlength(mm)orweight\ (g)}{Changeintime}$$

$L_\infty = a/K$

Where;

K as defined above

a = intercept on the regression described above.

The calculated L_∞ and K were then used to approximate standard length (mm) of fish at 150 days in the equation;

$$L_{150\ days} = L\infty*(1 - e^{-k(t - t_0)})$$

The growth index (Ø) was calculated the by equation;

$$Ø = \log_{10}K + 2\log_{10}L_\infty$$

The length component was calculated from the length – weight relationship of

$w = aL^b$

but Linearised into logW = loga + blogL (Pauly *et al*., 1993) to obtain the allometric size – length relationship with linearity confirmed by polynomial regression.

Where;

a = intercept of the standard length and weight relationship

b = slope of the standard length and weight relationship

w = weight (g)

L = length (mm)

The correlation coefficient (r) and slopes of the regression lines for each treatment were calculated by first splitting the data into feeding rates (treatments) and then running a correlation between SL (mm) and weight (g). To determine if the r (s) were significant ($P< 0.05$), they were transformed into Z scores by the use of the z value table and used in the calculation of observed Z scores (Z_{obs}) from the equation according to Pallant (2001).

$$Z_{obs} = (Z_{t1} - Z_{t2}) / \sqrt{\frac{1}{N1-3} + \frac{1}{N2-3}}$$

Where;
Zobs = Calculated Z scores from the observed data
Zti = Z score for T1, T2 and T3
N = Number of observations

A slope (ß) between the treatments was calculated by running the linear regression of each treatment separately and resultant slopes used to calculate the Student's t – test using the formula below:

$t = (ß_1 - ß_2)/(S_{ß1-ß2})$

where;

$S_{ß1-ß2}$ = standard error of the difference between the two slopes = $\sqrt{Sß1^2 + Sß2^2}$

Where; Sß1 = standard error of slope 1 and Sß2 = standard error of slope 2

Comparisons were then made between T1 and T2, T1 and T3, and T2 and T3. Any Z score or t that fell outside ±1.96 and $df_{N1+N2-4}$(P = 0.025) respectively was considered significant thus the pair was considered significant ($P< 0.05$).

Simple gross margin analysis was performed to determine the cost effectiveness of feeding rates. It was assumed that all other operating costs remained constant and only the variable cost of feed was used in calculations. The final weights were assumed to be the harvest weight of fish.

General Linear Model (univariate analysis procedure) was performed to determine the differences in final mean weight, length, total cost (TC), total revenue (TR) and gross margin ratio (GMR) among feeding rations means which were deemed significant at $P< 0.05$. Differences were separated by Duncan's Multiple Range Test (DMRT) (Duncan, 1955).

Statistical Package for Social Scientist (SPSS) 15.0 (SPSS Inc) and Stata 12.0 (StataCorp) softwares were used in analyzing the data. Microsoft excel was used in the production of figures and graphs. Untransformed data are presented to facilitate interpretation.

Results

The results of the VBGF parameters are shown in table 2. The T2 feeding rate showed the highest K in absolute terms and this was followed by the T3 feeding rate. However, the L_∞ (mm) was lowest in T2 but was highest at 150 days of growing *O. tanganicae*. The growth index (Ø) was highest in T2 too.

Table 2: K and L values according to the treatment

Treatment	ß coefficient (K)	Intercept (a)	r^2	L_∞(mm)	L_{150} (mm)	*Growth index (Ø)
T1	0.002	0.529	0.88	330.625	85.692	2.340
T2	0.007	1.245	0.63	185.821	120.795	2.383
T3	0.004	0.784	0.85	211.892	95.603s	2.254

*$\log_{10}K + 2\log_{10}L_\infty$

Table 3: Regression parameters of the length – weight relationship

Treatment	ß coefficient	SE	Intercept (a)	r^2	*R*
T1	2.818^a	0.047	4.009	0.917	0.958^a
T2	2.945^b	0.036	4.272	0.954	0.977^b
T3	3.001^c	0.038	4.401	0.951	0.975^b

The correlation coefficients differed among the feeding rates with the T1 being significant smaller ($P < 0.05$) than the other two treatments. The slopes of the treatments differed significantly ($P < 0.05$) among the treatments (Table 3).

The final mean weight (FMW) and final mean standard length (FMSL) and final mean total length (FMTL) of the fish differed among the treatments with the T2 having the highest mean weight although this was significant different ($P < 0.05$) from the T3. Economic analysis showed that the feeding cost increased with the feeding rates while total revenue was significantly highest on T2 although this was not significantly different ($P > 0.05$) from T3. The gross margin differed amount the treatments with T2 having the highest Gross margin (GM) but was insignificant different ($P > 0.05$) from that of T1 (Table 4). Gross margin ratio (GMR) decreased with an increasing feeding rates.

The Tl had the highest GMR followed by T2. However, the yield and total revenue (TR) for the 5% were the highest followed by the T2 (Table 4).

Water temperature did not differ significantly ($P < 0.05$) among the treatments. Treatments T1 and T3 feeding rates did not differ ($P < 0.05$) in pH. There were no significant differences ($P < 0.05$) in electrical conductivity and dissolved oxygen in treatments T2and T3 (Table 5).

Table 4: Production and economic analysis of *O. tanganicae* used in the experiment (mean±SE)

Water parameter	**Treatment**		
	T1	T2	T3
FMW (g)	157.900±2.955[a]	176.233±4.384[b]	174.267±4.406[b]
FMSL (mm)	156.233±1.453[a]	161.983±1.376[b]	159.267±1.453[b]
FMTL (mm)	195.750±1.373[a]	201.817±1.993[b]	200.667±1.926[b]
[1]TR (KR)	1.90±0.04[a]	2.12±0.05[b]	2.09±0.05[b]
[2]TC (KR)	0.08±0.01[a]	0.24±0.01[b]	0.43±0.01[c]
[3]GM (KR)	1.81±0.04[b]	1.88±0.41[b]	1.66±0.05[a]
[4]GMR	0.954±0.001[c]	0.885±0.003[b]	0.787±0.006[a]
*Total batch weight (kg)	43.5	46	53
*Yield (kg/ha)	870	920	1, 060
*[5]Survival (%)	73.44	71.41	73.44

*No statistical significance given as the data used was pooled.
[1]TR = P*Y where; P = price of fish/kg and Y = total weight (kg); KR5.00 = $1.00; KR = Zambian Kwacha rebased
[2]Amount of feed fed*unit cost of feed
[3]GM = TR – TC
[4]GMR = GM/TR
[5][(Number of fish at the end of the experiment – Number of dead fish)/ (Number of fish at the start of the experiment)]*100

Table 5: Selected water quality parameters (mean ± SE)

Water parameter	**Treatment**		
	T1	T2	T3
Water Temperature (^oC)	24.594±0.283[a]	24.879±0.264[a]	24.915±0.272[a]
pH	8.72±0.05[a]	9.06±0.05[b]	8.69±0.05[a]
Electrical Conductivity (µmho/cm)	154.778±2.529[a]	182.323±4.155[b]	189.703±4.025[b]
Dissolved Oxygen (mg/L)	7.633±0.217[a]	9.621±0.288[b]	7.633±0.217[b]

Discussion

A higher L or K results in higher net yield and can be used to predict yield under different management regimes in *Oreochromis niloticus* (De Graaf & Prein, 2005). In the current study the highest K was observed in T2 although this was not significant different ($P > 0.05$) from T3. Using ANOVA on the final mean weight, the results were similar. This shows that feeding *O. tanganicae* at 3% of their body weight would produce similar *O. tanganicae* fed at 5% feeding rate. Chowdhury (2011) found that feeding *Oreochromis niloticus* at 3 % of their body weight between 80 – 115g but 1.2% for fish larger than 260g would maximise growth. In this experiment the feeding rate was fixed throughout the 150 days experimental period. In this study the 3% produced the fastest growing fish as can be attested by the highest K. The treatment also showed to have the largest fish at harvest although this was not significant different ($P > 0.05$) from that of T3. High mortality may have attributed to the fastest growth and largest mean weight at harvest in T2 as the biomass reduced and this allowed fish having extra feed. However, T3 had the higher feeding rate justifying the similar final mean weight at harvest. Aydim *et al* (2011) found that feeding juvenile Black Sea Turbot to satiation was sufficient for maximal growth than feeding at 1%. However, there was no attempt to study the most cost effective method.

The slopes of the weight – length relationships were significant different ($P < 0.05$) among the treatments. The correlation coefficients of the treatments were significant different ($P< 0.05$) too. However, they were close to factor 3 showing that the fish in all the treatments exhibited isometric growth. Variation of fish in weight was greatest in the least feeding rate used since it had the least coefficient of determination. The quantity of available feed is one of the most important factors impacting size variation in and interactions among fish during culture. This can be manifested in the creation of hierarchy in groups of cultured fish with dominant and subordinate individuals that may

have a range of consequences (Kozlowski *et al.*, 2012**)**. The fish under this experiment exhibit shoaling behavior, therefore, weaker ones would be deprived of food.

In aquaculture, the GMR is the ratio of gross profit of the business to its revenue. It is also called gross profit margin. It is a profitability ratio measuring what proportion of revenue is converted into gross profit which is the revenue from the cost of fish sold. Higher values indicate that more money is earned per unit amount of revenue which is favourable because more profit will be available to cover non-production costs. The GMR was highest in T1and was significant ($P < 0.05$) from T1. However, T3 was significantly lower ($P< 0.05$). Therefore, 1% gave more money for any revenue collected. It is also means that 1% feeding rate has a higher mark-up price on fish sold. This shows that over feeding may just impose unnecessary cost that may not yield any returns.

The results of this experiment show that feeding *O. tanganicae* at 3% in semi – concrete ponds would be optimal biologically. However, economically, 1% feeding ration is ideal if fish is sold on weight basis. The use of Gulland and Holt plot is this experiment shows that the method can easily be utilised in other aquaculture experiments.

Acknowledgement
The authors would like to thank the Government of the Republic of Zambia (GRZ) through the Ministry of Agriculture and Livestock (MAL), Department of Fisheries and NARDC for funding and provision of experimental facilities. Gratitude also goes to all members of staff at NARDC who helped in any way or another.

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The Director of Fisheries Mr. Patrick Ngalande (right), Chief Aquaculture Officer Mr. Mulenga Musonda (c) and Deputy Director (Aquaculture) Mr. John Mwango at NARDC on 16th December 2013.

Mr. Chad Kancheya and Mr. Given Kujila constructing a Tilapia incubating facilicity at Fiyongoli Aquaculture Research Station, Mansa.

USE OF MORINGA OLEIFERA LEAVES AS AN ADDITIVE IN FEEDS OF *OREOCHROMIS MACROCHIR, OREOCHROMIS ANDERSONII* AND *OREOCHROMIS NILOTICUS*

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Abstract

Increasing levels of powered moringa leaves (5%, 10% and 15%) in the diets of *Oreochromis niloticus*, *O. andersonii* and *O. macrochir* were assigned randomly to hapas in triplicates. In *O. niloticus and O. andersonii the* highest final mean weight and Specific Growth Rate (SGR) did not differ (P >0.05) among the treatments. The mean body weight in *O. macrochir* was least in the 15% diet and was significant (P < 0.05) from the other treatments. Similarly the SGR was least in the 15% diet of *O. macrochir* and was significantly different (P< 0.05) from the other treatments. The condition factor (K) was significantly different (P< 0.05) in *O. niloticus* but not in *O. andersonii* and *O. macrochir*. Second order polynomial regression of mean body weight and moringa percent level in diet revealed a strong negative relationship. *Oreochromis niloticus*($Y = 0.0007x^2 - 0.0505x + 7.548$, *O. andersonii*($Y = 0.0036x^2 - 0.0894x + 5.0752$) and *O. macrochir* ($Y = -0.029x^2 + 0.129x + 12.158$) with an optimal inclusion value of 2.22%. This may indicate that tolerance of moringa varies according to species. The results have revealed that increasing moringa levels in the diet does not result in corresponding somatic growth of fish. A follow up study with anti nutritional factors deactivated in moringa and cost of moringa feed in relation to fish growth must be conducted.

Key words

Additive, protein, moringa, growth performance

Introduction

Aquaculture has been noted to be one of the fastest growing food-producing systems in the world (Ahmed and Lorica, 2002). In the last three decades (1980–2010), world food fish production of aquaculture has expanded by almost 12 times, at an average annual rate of 8.8 percent (FAO, 2012). Despite this increase the high cost of fish feeds especially in developing countries has slowed the development of aquaculture. The high cost and fluctuating quality as well as the uncertain availability of fish meal have led to the need to identify alternative protein sources for fish feed formulation (Richter *et al*., 2003).

As noted by El – Sayed (2006) nutrition is one of the most expensive components in the aquaculture industry where it can represent about 50% of total operation costs. In Zambia, most fish farmers depend on agricultural bran such as maize bran and fertilization which limits their production and consequently economic returns (Kefi, 2008) since their nutrient composition is low. The ingredients of feed from plant based and agriculture by-products commonly used in semi-intensive culture systems do not provide the high protein source found in animal source ingredients such as fish meal. Research has focused on finding locally available, cheap and unconventional protein source to replace animal protein sources like fish meal for use in fish feeds (Afuang *et al*., 2003).

There have been efforts made to replace protein in fish meal with plant sources either partially or totally. Moringa leaf has been studied as an alternative protein source in fish diet and seems to be a promising protein source (Yuangsoi and Masumoto, 2012). Tagwireyi *et al*(2008) and Richter *et al* (2003) have indicated that moringa is a promising protein source for inclusion in fish diets.

Moringa oleifera, a member of the Moringaceae family, is a fast growing plant widely available in the tropics and subtropics with several economically important uses for industry and medicine (Richter *et al*., 2003). The leaves, which are easy to grow and rich in proteins, vitamins, carotenoids, ascorbic acid and minerals are becoming widely used in projects fighting against malnutrition (Sauveur, 2010). However, no known documented trials have been conducted in Zambia to investigate its effect on fish growth. Therefore, this experiment aimed at investigating the effect of moringa in fish feed on the growth performance of *O. niloticus, O. andersonii and O. macrochir*.

Materials and Methods

Brooders of *O. andersonii* and *O. niloticus* were selected, sexed and conditioned separately for a

period of one month. They were then placed in four breeding hapas measuring 8mX2mX1m (Figure 1) stocked at 2 fish/m^2 and sex ratio of 1:2 (Male (182.75g ± 5.32g): Female (123.5g ±5.12g)) (mean ± SD) according to species. The fish were checked weekly for breeding by opening of the mouth to check for eggs. The brooders were fed with NARDC formulated isonitrogenous feed at 30% crude protein (CP) made from; fish meal, soya bean meal, maize bran, oil, binder, dicalcium phosphate and vitamins. The fish was fed at 3% live body weight twice daily. Water temperature, Dissolved Oxygen (DO) and conductivity were measured daily using a water checker (YSI professional plus) during the breeding period.

Figure 1: Breeding hapas

Upon breeding, the fry were scooped from hapas and placed in already fertilized concrete ponds (12.5m^2) according to species. The pond was fertilized using chicken manure at a rate of 0.06kg/m^2. The fry were raised to 3g using NARDC formulated feed 40% CP (ingredients; fish meal, soya bean meal, maize bran, oil, binder, dicalcium phosphate and vitamins). The ponds were covered with big meshed net on top to prevent predatory birds (figure 2).

Figure 2: Raising of fry in concrete tanks

Water temperature; Dissolved Oxygen (DO) and conductivity were collected as described above.

A semi-concrete pond (750m^2) was drained and sundried before erecting hapas (1m x1m x 1m). The hapas were fastened to the ground using bamboo sticks to prevent the escape of fish. Four triplicates treatments were used and assigned in a complete randomized design (CRD). The treatments used were:

Treatment 1(T1) = Control (0%), Treatment 2 (T2) = 5%, Treatment 3 (T3) = 10 % and Treatment 4 (T4) = 15 % of moringa inclusion to the feed. Twenty (20) juvenile fish of *O. niloticus* (3.9±1.1g; mean ± SE) and *O. andersonii* (2.5± 0.5g; mean ± SE) respectively were stocked in each hapa.

The experiment at Solwezi Aquaculture Research Station (SARS) was conducted in 9 hapas measuring 2mx2mx1m.

Figure 3: Hapas set in a prepared pond at NARDC (Left) and at SARS (right)

Similarly brooders of *O. macrochir* were selected and placed in semi-concrete ponds at NARDC.

Upon breeding the fry of average weight of 0.5g were transported to SARS. The fry were then raised in a concrete pond fertilized with chicken manure applied at a rate of 0.06kg/m^2. Upon growing to 3g the fish were stocked in hapas measuring 2mx2mx1m placed in a 176m^2.

Feeding for both experiments was done at 5% body weight twice daily at 09:00 hours and 14:00 hours adjusted fortnightly upon sampling. The feed was formulated into pellets of smaller sizes. The diet formulations were as follows:

Table 1: Diet formulation (DM %) of the four experimental diets

Ingredients	T1 Control	T2 5%diet	T3 10%diet	T4 15% diet
Maize meal	59.95	56.08	52.3	48.53
Moringa leaf meal	0	5	10	15
Fish meal	38.65	37.42	36.2	34.97
Soya gold oil	0.5	0.5	0.5	0.5
Mineral mix	0.5	0.5	0.5	0.5
Vitamin mix	0.5	0.5	0.5	0.5
Total	100	100	100	100

The fish were sedated using whole crude cloves before measurements were taken at a rate of 1g/5L of water. Measuring of fish weight, Standard Length (SL) and Total Length (TL) were done fortnightly using a measuring scale and measuring board. The data was entered in field data sheets. The following parameters were monitored and calculated:

Mortality rate (%) = (Number died/Number stocked)*100

Specific Growth Rate (SGR%/day) = ((lnW_f – lnW_i)/t) *100 where;

> lnW_f is the natural logarithm of final body weight, $lnWi$ is the natural logarithm of initial body weight of the fish and t is final time of experiment in days (De Silva and Anderson, 1995).

Condition factor (K)

$W= L^3$. Where W is the weight (g) of fish, L is the Total length (mm) of fish.

Dry matter and ash content analysis

A sample of 20 fish was collected from each treatment for dry matter and ash content analysis for the experiment conducted at NARDC. The sampled fish were weighed individually, dried and incinerated in an electric furnace (Avantec) for 24hrs at 105°C and 550°C to determine dry matter and ash content respectively. Each fish was labeled properly before placing in the electric oven.

Dry matter (DM) was calculated using the following formula:

DM (%) = (weight of sample after drying (g)/ weight of sample before drying (g))*100

Ash content was calculated using the following formula:

Ash (%) = (weight of ash from sample (g)/weight of sample (g))*100 (AOAC, 2002).

Statistical analysis

The data was analyzed using a Statistical Package for Social Scientists (SPSS) version 15.0. General linear model, univariate analysis procedure, was conducted to determine if significant differences existed at $P < 0.05$ and if so Duncan's Multiple Range test (Duncan, 1955) was performed to separate the differences. In order to obtain the optimum moringa inclusion levels in the diet, Polynomial regression was used to determine the best regression model between the mean body weight of fish and moringa inclusion level. Microsoft excel 2007 was used to generate the graphs.

Results

Growth performance

The final mean weight of *O. niloticus* in the 65 days experimental period was not significantly different ($P > 0.05$) among the four diets (Control, 5% diet, 10% diet and 15%). However, the 5% moringa diet had the highest final mean body weight of 7.596g±0.254followed by control diet (7.454g±0.346), and then 15% diet (7.051g±0.311) and 10% diet (6.835g±0.192) was the least. The SGR was equally not significantly different among the four diets but was higher in the control followed by the 5% then 10% and 15% diet was least. The condition factor (K) of the fish was significantly different in the fish fed on 10% diet compared to the remaining diets (Table 2).

Table 2: The growth parameters of *O. niloticus* fed on four different diets for 65 days (means ± standard error (SE))

	TRT 1	TRT 2	TRT 3	TRT 4
Initial mean weight (g)	4.285±0.0670[a]	4.557±0.642[b]	4.292±0.638[a]	4.183±0.089[a]
Final mean weight (g)	7.454±0.346[a]	7.596±0.254[a]	6.835±0.192[a]	7.051±0.311[a]
SGR (%)	0.830±0.177[a]	0.770±0.789[a]	0.689±0.661[a]	0.687±0.130[a]
Condition factor (K)	4.127±0.070[b]	4.004±0.060[b]	3.872±0.584[a]	4.078±0.060[b]
DM%	22.930±0.518[a]	23.920±1.316[a]	22.215±0.481[a]	22.215±0.602[a]
ASH%	5.149±0.517[a]	4.704±0.614 [a]	4.912±0.445[a]	5.068±0.382[a]
*Mortality (%)	1.2	8.3	10.0	46.7

Values within columns that have the same superscripts are not significantly different (P>0.05).
**No statistical analysis was done as determination was on collective fish samples*

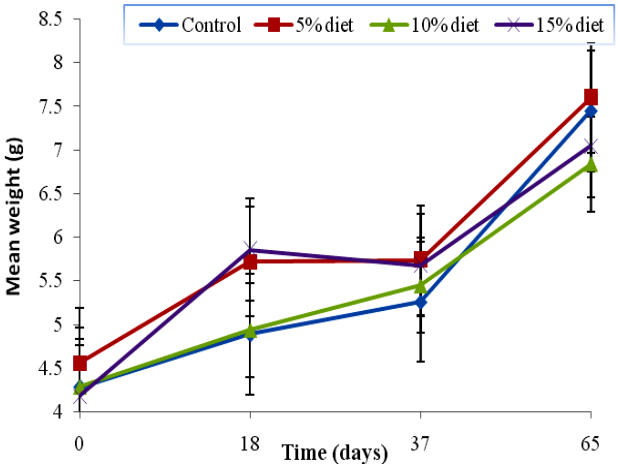

Figure 4: Growth trend of *O. niloticus* feeding on different diets during the 65 days experimental period

The growth trend of *O. andersonii* and *O. niloticus* reveals a steady increase in body weight of fish in all the treatments. However, of interest is treatment 4 which reveals better performance unto day 37 of the experiment when the growth trend reduces (Figure 4 and Figure 6). In *O. macrochir* however, the growth trend is lowest in treatment 4 (Figure 8)

A second order polynomial regression described the best relationship between the mean body eight (g) and the moringa percent level inclusion ($Y= 0.0007x^2 - 0.0505x + 7.548$) (Figure 5).

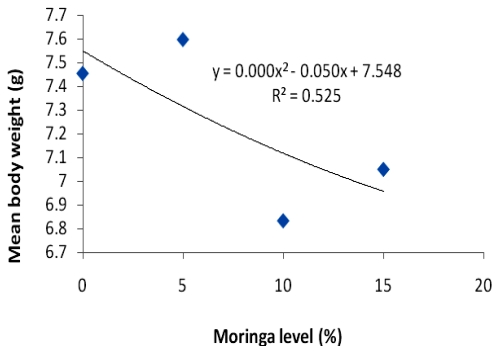

Figure 5: The relationship between the level of moringa inclusion and the mean body weight (g) for *O. niloticus*

In *O. andersonii* the highest final mean weight of fish was in fish fed on the control diet (5.098g±0.180) and it was significantly different (P<0.05) from the weight of fish feeding on other diets. It was followed by the 5% diet (4.650g±0.100), 10% (4.611g±0.147) and 15% diet was least (4.524g±0.143). The SGR and condition factor (K) were all not significantly different (P > 0.05) among the treatments. The Dry matter percentage of *O. andersonii* was higher in the 5% diet followed by the control diet. It was followed by the 15% diet and the 10% diet was least. However, there were no significant differences (P>0.05) among the treatments. On the other hand, percentage ash content was highest in the control diet, followed by 5% diet, then 10% diet and 15% diet was least. The percentage ash content in the control diet was significantly different from the rest of the diets (Table 3).

Table 3: The growth parameters of *O. andersonii* fed on four different diets for 65 days (means ± SE)

	TRT 1	TRT 2	TRT 3	TRT 4
Initial mean weight (g)	2.490±0.064[a]	2.352±0.567[a]	2.320±0.596[a]	2.273±0.049[a]
Final mean weight (g)	5.098±0.180[b]	4.650±0.100[a]	4.611±0.147[a]	4.524±0.143[a]
SGR (%)	1.085±0.035[a]	1.055±0.099[a]	1.150±0.171[a]	1.043±0.110[a]
Condition factor (K)	4.225±0.0718 [a]	4.339±0.825 [a]	4.508±0.967 [a]	4.473±0.071 [a]
DM%	23.934±0.256 [a]	24.430±0.949 [a]	22.357±0.812 [a]	22.416±0.358 [a]
Ash%	4.794±0.195[a]	4.242±0.259 [a]	4.156±0.243 [a]	4.483±0.294 [a]
*Mortality (%)	21.7	20	36.7	13.3

Values within columns that have the same superscripts are not significantly different (P>0.05).
**No statistical analysis was done as determination was on collective fish samples*

A second order polynomial regression described the best relationship between the mean body eight (g) and the moringa percent level inclusion ($Y= 0.0036x^2 - 0.0894x + 5.0752$) (Figure 7).

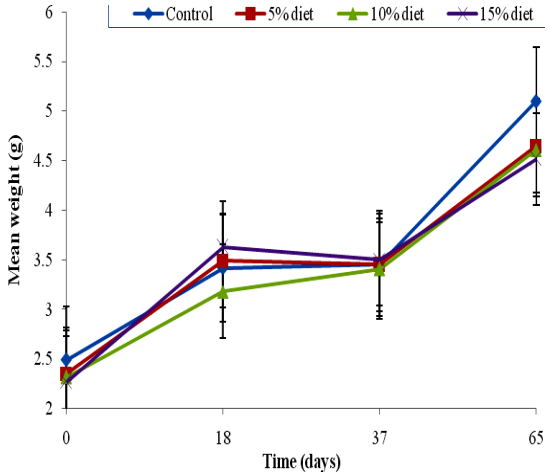

Figure 6: Growth trends of *O. andersonii* feeding on different diets during the 65 days experimental period

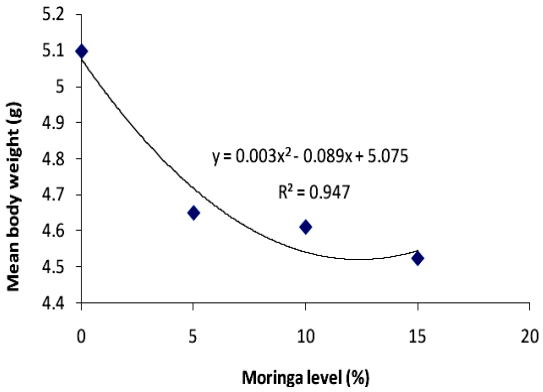

Figure 7: The relationship between the level of moringa inclusion and the mean body weight (g) for *O. andersonii*

In the experiment conducted at Solwezi Aquaculture Research Station on *O. macrochir*, results have indicated that there were no significant difference (P>0.05) in the mean body weight of fish fed on control, 5%, and 10% diets. The 15% diet was significantly different (P < 0.05) from the other three diets. However, the fish in the control diet had slightly higher mean body weight of fish followed by the 5% diet and 10% diet was least. Fifteen percent (15%) moringa level recorded the lowest SGR and this was significant different (P<0.05) from the other moringa levels. The condition factor showed no significant difference (P > 0.05) among all the treatments (Table 4).

Table 4: The growth parameters of *O. macrochir* fed on four different diets for 61 days (means ± SE)

Treatment	**T1**	**T2**	**T3**	**T4**
Initial mean body weight (g)	2.650±0.0621[a]	2.566±0.078[a]	2.575±0.079[a]	2.625±0.078[a]
Final mean body weight (g)	12.400±0.340[b]	11.350±0.416[b]	11.275±0.416[b]	7.325±0.416[a]
Initial Condition Factor (K)	3.989±0.181[a]	3.951±0.204[a]	3.848±0.175[a]	4.006±0.151[a]
Final Condition Factor (K)	3.152±0.028[a]	3.098±0.0514[a]	3.163±0.036[a]	3.047±0.055[a]
Specific growth rate (%)	2.523±0.060 [b]	2.358±0.133[b]	2.410±0.091[b]	1.659±0.081[a]

Values within columns that have the same superscripts are not significantly different (P>0.05).

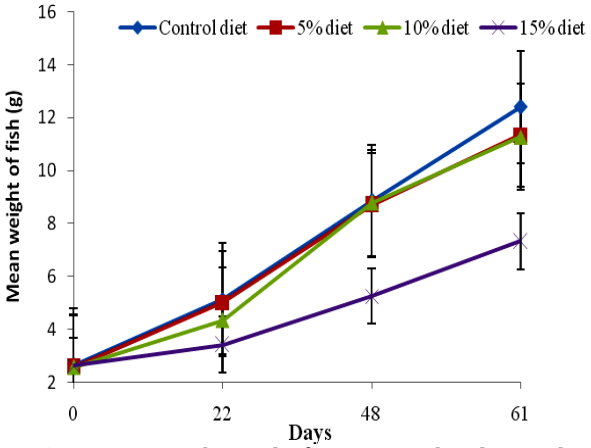

Figure 8: Growth trend of *O. macrochir* during the 61 days growing season.

A second order polynomial regression described the best relationship between the mean body weight (g) and the moringa percent level inclusion Y= -$0.029x^2$ + 0.129x + 12.158. A differential equation shows that the maximum mean weight gain would occur at a moringa level of 2.22% (Figure 9).

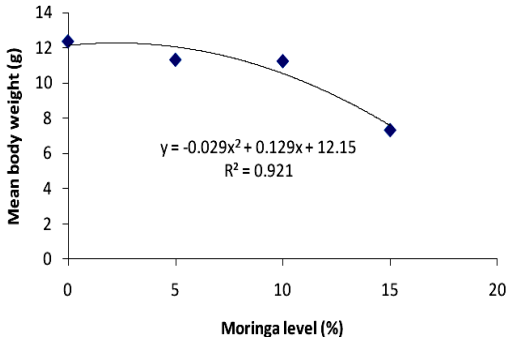

Figure 9: The relationship between the level of moringa inclusion and the mean body weight (g) for *O. macrochir*.

Water quality

The water quality measurements in all treatments of *O. andersonii* did not differ significantly (P > 0.05) (Table 5). Equally in treatments of *O. niloticus* there were no significant differences (P > 0.05) in the water quality measurements.

Table 5: The means (± SE) for selected water quality parameters for the four different treatments in *O. niloticus*.

Parameter	TRT 1	TRT 2	TRT 3	TRT 4
Temp (°C)	27.784±0.273	27.791±0.274	27.777±0.273	27.783±0.273
DO (mg/L)	7.917±0.419	7.921±0.422	7.937±0.420	7.903±0.431
Cond (µS/cm)	207.467±5.664	208.378±5.974	203.100±5.650	204.571±5.665

Table 6: The means (± SE) for selected water quality parameters for the four different treatments in *O. andersonii*.

Parameter	TRT 1	TRT 2	TRT 3	TRT 4
Temp (°C)	27.863±0.260	27.857±0.260	27.867±0.260	27.852±0.261
DO (mg/L)	8.457±0.404	8.482±0.403	8.510±0.401	8.509±0.404
Cond (µS/cm)	208.652±6.258	211.532±6.284	212.109±6.285	216.709±6.019

Discussion

In this study the growth performance of *O. niloticus* was not significantly different (P > 0.05) among the treatment. In *O. andersonii* the fish fed on the control diet without moringa showed the highest growth performance (P < 0.05). In *O. macrochir* however, growth performance of the fish was significantly lower in fish fed on a diet with 15% moringa inclusion. The optimal inclusion levels of moringa in *O. macrochir* were at 2.22%. In all species there was a negative relationship between the mean body weight of fish and the percent moringa level. Increase in moringa level resulted in a lower final body weight of fish.

The results mean that increasing moringa levels in the diet does not result in corresponding somatic growth of fish. Similar results were observed by Richter *et al* (2003) in hepato-somatic index and percentage of moringa leaves in diets fed to *O. niloticus*. This observation could be attributed to some anti-nutritional factors such as phenolics, saponin and phytic acids in the moringa leaves as reported by Richter et al (2003). The de-activation of these anti-nutritional factors might result in higher inclusion of moringa leaf meal in the fish diet. A study by Tagwireyi et al (2008) discovered that steamed heated moringa leaf meal substituted 10% of dietary protein in Nile tilapia fry without significant reduction in growth performance.

The variances in the response of the fish species used in the experiment to moringa may indicate species specific tolerance to the plant. However, this requires further studies to prove the utilization

of the plant according to fish species. In addition, the cost of moringa in relation to fish growth requires determination.

Moringa oleifera is a promising plant based protein source in fish feed. However, higher inclusion of the plant in fish diets results in depressed growth. The acceptance of the plant seems to vary among different species. However it is recommended that for higher inclusion of moringa in fish feed a follow study should be conducted in which the anti-nutritional factors in the moringa leaf should be de-activated. Furthermore, the cost of the moringa incorporated feed should be determined in relation to fish growth. Another study should be conducted to compare the performance of fish fed on moringa leaf and soya bean.

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Research tanks at Solwezi Aquaculture Research station

Research Offices at Chipata Aquaculture Research Station

IS FORTIFICATION OF METHIONINE NECESSARY IN *OREOCHROMIS ANDERSONII* SOYA BEAN (GLYCINE MAX) BASED FEEDS (CASTELNUE, 1861)?

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Abstract
An experiment was conducted to determine the optimal amount of methionine that can be incorporated in the soya bean based *O. andersonii* feeds. Three levels (2%, 4% and 6%) of methionine included in soya bean based feed (30% and 10% crude protein and crude lipid respectively) were tested on *Oreochromis andersonii* for seventy eight (78) days in hapas erected in semi – concrete pond (250m^2). Specific growth rate (SGR% day $^{-1}$), mean body weight gain (g) and feed conversion efficiency (FCE %) differed ($P < 0.05$) among the treatments although the 4% and 6% gave similar SGR (%), weight gain (g) and FCE (%). Methionine did not affect the reproduction of *O. andersonii*. However, females and larger sized fish appeared to mature earlier than males and smaller fish respectively. It is, therefore, recommended that soya bean based feed can be fortified with 4% methionine to improve the *O. andersonii* growth. However, there is need to conduct a similar study for a longer period of time and in aquaculture facilities used in the production of fish for commercial purposes. Furthermore an economic analysis is required to ascertain the economical utilization of methionine in fish feeds.

Key words: Methionine, *Oreochromis andersonii*, soya bean, growth and reproduction

Introduction

The use of soya bean has now generally been accepted as the best alternative to the use of fish meal in the livestock feed including fish. This is because soya bean has similar balanced amino acid (AA) profile although its methionine level is lower than that of fish meal. It is considered to meet requirements for some fish species (NRC, 1993; Deguara, 1998). It has also relatively high amounts of vitamins (thiamine, niacin, B – Complex and carotene) (Martin and Ruberte, 1980; Fafioye *et al*., 2005). Furthermore, soya bean is readily available on the market at a relatively lower price (Guillaume *et al*., 2001). If properly cooked, its low levels of cellulose and complex carbohydrates provide an additional advantage of using the plant in aquaculture nutrition. Osman *et al.* (2008) speculated low levels of both methionine and lysine in commercial fish feeds. Several authors have, therefore, recommended the fortification of soya bean based diet with synthetic methionine in order to balance the AAs. Methionine is a sulfur containing and an essential AA required by fish as well as terrestrial vertebrates for normal growth and metabolic functions. It is in the form of *S* – adenosyl – methionine which serves as a principal donor of methyl group, a major contributor to the whole body pool of one carbon units that are required for trans-methylations, the biosynthesis of choline, thymidine, polyamines and creatine (Alam*et al*., 2000; Mai *et al*., 2006; Bae *et al*., 2011).

Oreochromis andersonii is indigenous to Zambia and has been adopted as the fish species of choice for aquaculture. It is naturally in lagoons of the Upper and Middle Zambezi River, Kafue systems (Skelton, 2001) and Lake Kariba (Fish Base, 2013). Adults occupy deep water while juveniles remain inshore. It feeds on detritus and zooplankton although bigger individuals also take insects and other invertebrates. In aquaculture the requirement for soya bean protein and lipid has been studied. Kefi *et al* (press) found 30% crude protein to be ideal economically. Furthermore, Kefi *et al*. (2013) observed that the minimal lipid requirement was lower than 10% although the optimal lipid requirement was observed to be 14.9%.

Although there are similar studies that have been conducted in other fish species, there has been no published work on *O. andersonii*. For instance Osman *et al*. (2008) found 3.05% methionine of the crude protein to be optimal for *O. niloticus*. The experiment was conducted to determine the optimal methionine inclusion levels in soya bean based diet fed to *O. andersonii* juveniles.

Materials and method

The study to investigate the optimal methionine levels in soya bean based feed was carried out at National Aquaculture Research and Development Centre (NARDC), Kitwe for seventy eight days in hapas placed in a 250m^2 semi-concrete pond. *Oreochromis andersonii* was the fish species used in this study with methionine levels (2%, 4% and 6%) as treatments. The experimental pond was prepared by draining it, setting up of hapas (1 X 0.9 X 1m) and refilling with water to 60cm level. The treatments were allocated to hapas in a randomised block design in triplicates. Two hundred and fifty

O. andersonii (19.58±0.103; mean ± SD) were seined from 250m^2 semi – concrete pond and were taken to concrete tank (50m^2) for conditioning for 48 hours. Thereafter, 20 fish were randomly sampled and their morphometric measures (weight, standard length and total length) taken as described by Skelton (2001) before being placed in each hapa. The remaining fish were kept in the same concrete tank for possible replacements upon mortality that occurred within first fourteen days of the experiment.

Isonitrogenous (30%) soya bean based feed (soya bean 54.6%, Maize meal 43.4%, vitamin starter 1% and Dicalcuim Phosphate 1%) fortified with DL – methionine feed grade 99%(Sumitomo Chemical Limited, Japan) levels at 2%, 4 % and 6 %was prepared and fed to fish at 3% live body weight twice a day with an exception of Saturdays and Sundays.

Selected water quality parameters (dissolved oxygen, water temperature, conductivity and Chlorophyll *a*) were taken once daily three times in a week (Monday, Wednesday and Friday) at 10 hours. Mortality was also monitored every day throughout the study.

At the end of the experiment, all the fish were weighed and the length measured. Three fish were randomly selected from each treatment and analysed for dry matter and ash content according to the procedures of AOAC (2002).The ash was determined as total inorganic matter by incineration of the sample in an advantec electric furnace at 550^{o}C for 5 hours. The remaining inorganic material was reduced to their stable form, oxides or sulphates were considered as ash. Then the ash was calculated as follows:

$$Ash\ (\%) = \frac{weight\ of\ ash}{weight\ of\ original\ sample}\ x\ 100$$

Moisture was determined by drying samples in an advantec electric furnace maintained at 105^{o}C for 5 hours. The difference between the initial weight of the sample and that after drying was recorded and moisture calculated as follows:

$$Moisture\ (\%) = \frac{Difference\ between\ before\ and\ after\ drying\ (g)}{Weight\ of\ sample\ before\ drying}\ x\ 100$$

The remainder of the fish was then placed in buckets filled with 10% formalin for 10 days. After this period, the fish were dissected and the gonads recovered. Upon recovery, the maturity stage of the gonad was identified as described by Balarin (1983). The gonads were then weighed on sensitive analytical balance.

Statistical analysis

Growth of the fish was calculated following the equations; Weight gain (g) = Final fish weight (g) – Initial fish weight (g); Specific growth rate = 100(LnWT1 – LnWT0)/t where, Ln is natural logarithm, WT0 is the initial weight (g), WT1 is the final fish weight (g) and t is time in days. Feed convention efficiency (FCE%) was calculated as follows: 100(body weight gain/feed taken). Condition factor was calculated as follows: 100, 000W/L^3 where W is weight (g) while L is standard length (mm) of the fish (Williams, 2000).The gonadosomatic index (GSI) was calculated as follows: 100(weight (g) of gonads/weight (g) of fish).

General Linear Model (univariate analysis procedure) was performed to determine the differences among methionine levels means which were deemed significant at $P < 0.05$ for parametric data. Differences were separated by Duncan's Multiple Range Test (DMRT) (Duncan, 1955). Before analysis, parametric data were tested for normality using Shapiro – Wilk test and the homogeneity of variance using Levene's test for Equality of Variances.

The stages of maturity of fish was analysed using the multinomial logistic regression models where size (weight), sex and methionine entered as factors and stages of gonads as dependent variable. In order to eliminate cells with zero frequencies, the gonadal stages where grouped into three categories. These were immature, inactive (inactive and inactive – active) and ripe (active, ripe and ripe running) only. The ripe gonads were used as a reference group. The size (weight) was collapsed into 10g intervals. Interactions between and among the factors were tested after the main effects were maintained in the model if significant ($P < 0.05$), and similarly factors not having significant ($P > 0.05$) effect were taken out from the model.

The model used was follows:

$$Y = a + b_1X_1 + b_2X_2 + b_3X_3 + b4X4 + \mathcal{E}$$

Where:

Y = Dependent variable (stages of gonads)
X_1 to X_4 = Independent variables
X_1 = Sex of fish
X_2 = Weight of fish (g)
X_3 = Methionine level (%)
X_4 = interaction effect
b_1 to b_4 = regression coefficients
a = Constant
$\mathcal{E}$ = Residual

Cross tabulations and Chi-square (χ^2) were used to examine the associations between the reproductive status of fish and methionine levels.

Statistical Package for Social Scientist (SPSS) 15.0 (SPSS Inc.) software were used in analysing the data. Microsoft excel was used in the production of figures and graphs. Untransformed data are presented to facilitate interpretation.

Results

The final mean weight, condition factor (K) and standard length (mm) did not differ significantly (P> 0.05) among the treatments. However, the highest final mean weight was recorded at 4% methionine level. Specific growth rate (SGR % day^{-1}) differed significantly (P< 0.05) among methionine levels. However, the control did not differ (P > 0.05) from that of 2% while the 4% methionine level was similar (P> 0.05) to that of 6%. The mean body weight gain differed significantly (P< 0.05) with the higher levels of methionine recording the highest body weight gain. The 4% and 6% methionine did not differ significantly (P> 0.05). Similarly the control and the lowest methionine level (2%) used did not differ significantly (P > 0.05). The same trend was observed in the feed conversion efficiency (%) (Table 1).

There were no significant differences (P> 0.05) observed in the ash and dry matter of the fish among the methionine levels used in the experiment. However, the 4% methionine level recorded the lowest ash and highest dry matter. Dry matter was lowest in the control group (Table 1).

Table 1: Growth parameters of *O. andersonii* subjected to different levels of methionine

Parameter	Treatment (methionine level) (%)			
	0	**2**	**4**	**6**
Initial mean weight (g)	19.833±0.218	19.733±0.203	19.500±0.201	19.267±0.199
Final mean weight (g)	25.259±0.807	24.898±0.678	26.319±0.735	26.136±0.737
Standard length (mm)	92.298±1.034	91.286±0.840	93.604±0.886	92.870±0.904
SGR (%day^{-1})	0.336±0.015^a	0.323±0.014^a	0.414±0.108^b	0.422±0.015^b
Mean body weight gain (g)	5.426±0.218^a	5.165±0.203^a	6.819±0.201^b	6.869±0.199^b
Condition factor (K)	3.099±0.046	3.092±0.033	3.034±0.038	3.051±0.035
GSI	0.126±0.027^a	0.162±0.035^a	0.270±0.092^a	0.356±0.065^b
FCE (%)	12.905±0.638^a	12.304±0.583^a	16.342±0.616^b	16.648±0.605^b
Ash (%)	15.857±0.996	13.485±5.248	10.217±0.743	19.274±7.761
Dry matter (%)	37.861±0.650	41.750±1.983	63.026±13.685	44.536±4.606

Means ± SE with different superscripts differ significantly (P< 0.05).

There was a relationship (P< 0.05) between the independent variables (sex, fish size and methionine levels) and the stages of maturity of the gonads. However, only size and sex had a significant relationship to the maturity stage of the fish and therefore, it was excluded from the final model. The females were less likely to be immature (Odds Ratio = 0.069) or inactive Odds Ratio = 0.271) compared to males. Additionally small fish were more likely to be immature (Odds Ratio = 42.99) or inactive Odds Ratio = 4.959) than the bigger fish. However, the medium sized fish did not differ (P > 0.05) from the reference group which is large fish. This shows that the females are more likely to be in advanced stage of maturity than males indicating that females matured earlier than males. Consequently, large fish seemed to be advanced in maturity status (Table 2).

Table 2: Logistic regression on likelihood of maturity stages, sex and size of fish

Stage of maturity	**Sex of fish**	**B (Coefficient)**	**S. E**	**Wald**	**Exp (B)**
Immature	Female	2.672	0.459	33.843*	0.069
	Small fish	3.761	1.151	10.681*	42.990
	Medium fish	2.205	1.140	3.737	9.066
Inactive	Female	-1.305	0.404	10.413*	0.271
	Small fish	1.601	0.744	4.637*	4.959
	Medium size	0.821	0.720	1.300	2.272

*Significant different (P< 0.05)

Significant differences ($P< 0.05$) were observed in the stages of maturity with ripe stage recording the highest reproductive stage although this was not significant different ($P> 0.05$) from the immature group. Inactive group was the lowest but was not significant different ($P> 0.05$) from the immature group (Figure 1).

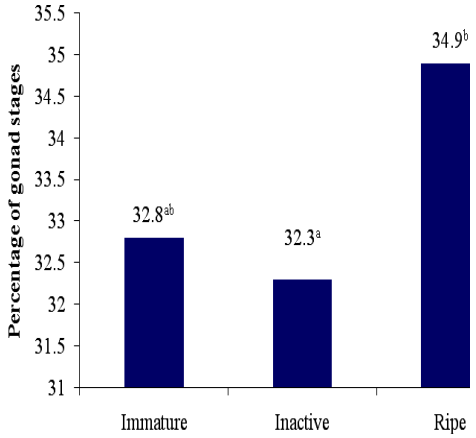

Figure 1: Percentage of gonad stages observed in *O. andersonii*

Water Quality

There were no significant differences ($P > 0.05$) observed in the selected water quality parameters determined (Table 3).

Table 3: Water quality parameters determined in hapas with *O. andersonii*

Parameter	Treatment (methionine level) (%)			
	0	2	4	6
Water Temperature (^oC)	17.292±0.101	17.353±0.104	17.344±0.103	17.4±0.1
Electrical conductivity (µS/cm)	91.341±1.416	91.111±1.435	91.211±1.422	91.791±1.462
Dissolved Oxygen (mg/L)	5.048±0.257	5.407±0.224	4.935±0.267	5.012±0.275
Chlorophyll *a* (mg/L)	0.02±0.007	0.048±0.056	0.051±0.038	0.098±0.111

Discussion

Protein requirements are defined as the minimum amount needed to meet requirements for amino acids and to achieve maximum growth (NRC, 1993).There have several varying results reported by authors probably due to variances in experimental conditions. Wilson (2002) estimated 1.8 to 4.0% dietary methionine of the dietary protein as the requirement for commonly cultured species of fish. Jackson and Capper (1982) reported that *O. mossambicus* appears to have a lower than 0.53% dietary methionine that may provide a satisfactory growth. In the current experiment, a practical diet fortified with methionine did not produce fish with different final mean weight. However, SGR (%) day $^{-1}$, weight gain and FCE (%) differed significantly. However, the 4% and 6% methionine produced similar but superior to the control and 2% methionine level. The rate at which fish grow is dependent on a number of factors including species, age, genetic potential, water temperature, health, and quantity and quality of food. Specific growth rate has been used as a suitable index for determining fish growth. In the current experiment SGR (%) day $^{-1}$ was found to be highest at the methionine level (6%) used although this was not significant different ($P > 0.05$) from that of 4%. This was similar to the weight gain (g) and FCE (%). However, the final mean weight was not significant ($P > 0.05$) although the 4% methionine level produced the fish with the highest final mean weight. Similar results were found by Guyeni and Davis (2009) who did not find any positive effect of methionine addition to soya bean based feed on the final weight on Red tilapia and *O. niloticus*. However, they did not report on SGR (%) day $^{-1}$. Osman *et al* (2008) found that the dietary requirements of methionine for *O. niloticus* fingerlings to be 3.05% of the dietary crude protein. Our results are, therefore, consistent with the finding of Wilson (2002) and Osman *et al* (2008).

To the best of our knowledge there have been no studies conducted on the effect of methionine on the reproduction of *O. andersonii*. Therefore, ours results cannot be referred to any study but can be a benchmark for future studies. Methionine did not affect reproductive index determined in the experiment. Additionally the level of methionine did not affect the rate of maturity of *O. andersonii.* However, sex and size of fish appears to affect the maturity of the *O. andersonii*. The females were found to mature earlier than males and larger fish were more at an advanced maturity stage than small fish. Similar results were observed by Kefi (unpublished).

The study reveals that 4% and 6% methionine level give similar but superior results on the growth rate of *O. andersonii*. However, methionine seems to have no any effect on the reproduction of *O. andersonii.* Therefore, 4% methionine level inclusion in soybean based fish feed would be recommended in the growth enhancement of *O. andersonii*. However, there is need to conduct an economic analysis of utilization of methionine in the soya bean based fish feeds. Furthermore, there is need to study the fortification of the soya bean based fish feeds with methionine for a longer period in aquaculture facility being used in the production of fish for commercial purposes.

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Rehabilitated spillway at NARDC

The hybrid catfish called NARDCCAT 2013

REPRODUCTIVE PERFORMANCE AND SEX REVERSAL SUCCESS RATE OF FOUR *OREOCHROMIS* SPECIES (*OREOCHROMIS NILOTICUS, OREOCHROMIS ANDERSONII, OREOCHROMIS TANGANICAE* AND *OREOCHROMIS MACROCHIR*) IN AN INDOOR RECIRCULATION HATCHERY

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Abstract
This study was to compare the reproductive efficiency, survival of egg and fry as well as the sex reversal success rate of the four *Oreochromis* species (*O. niloticus*, *O. andersonii*, *O. tanganicae* and *O. macrochir*) in the indoor recirculation hatchery. The breeding stock was reared in eight hapas (8mx 2m x 1m) set up in two semi – concrete ponds (250m^2) and were stocked randomly according to fish species and sex. After 30 days, the breeders were then placed in breeding hapas at sex ratio of 1 Male (182.75 ± 5.32g): 2 females (123.5 ±5.12g). Each fish species was replicated twice with each hapa containing 10 males and 20 females. There were no significant differences ($P > 0.05$) in the brooding efficiency index of the fish species although *O. niloticus* (4.557 ± 0.483%) had the lowest index followed by *O. andersonii* (5.394 ± 1.497%), *O. tanganicae* (5.718 ± 1.185%) and *O. macrochir* (8.968 ± 1.815%). The brown coloured eggs survived the best at 63.6% while survival of yellow and white eggs was only 22.7% and 13.6% respectively. The egg weight of *O. tanganicae* (11.415± 0.675 mg) and *O. macrochir* (11.104±0.213 mg) were insignificant ($P > 0.05$) while the lowest egg weight was observed in *O. andersonii* (7.207±0.141 mg) followed by *O. niloticus* (8.117± 0.163 mg) and were significantly different ($P < 0.05$). The highest sex reversal success was found to be in *O. andersonii* (98.6%) although not significantly different ($P > 0.05$) from *O. niloticus* (95.8%). The lowest sex reversal was achieved in *O. macrochir* (63.9%). *Oreochromis andersonii* presents the best species in sex reversal practices using Methyl testosterone (MT). There is need to study the growth of sex reversed *O. andersonii* and *O. niloticus* which had similar success rate of males. We recommend that only brown eggs should be collected from the brooding females in order to maximise survival and seed production.

Key Words: Breeding efficiency, Hatchability, Sex ratio, Egg colour

Introduction

The cichlids are the largest fish family in Africa with 900 species described and Southern Africa alone having eight genera and 42 species (Skelton, 2001). They are second most important group of food fishes in the world, next to the carps (El – Zaeem, 2011). This is because they are readily acceptable by consumers. Among the Tilapines are the *Oreochromis* species which exhibit mouth brooding of eggs and larvae. Because *Oreochromis* species are mouth brooders they have low fecundity in order to maximise care of progeny. This entails that large numbers of brood fish must be kept at a huge cost, if reasonable numbers of fish seed for stocking in fish ponds are to be produced (Ambali and Little, 1996; Kefi *et al*., 2011). The natural incubation of eggs and fry has allowed removal of the same for indoor hatchery incubation in order to maximize survival and sex manipulation. This is because Tilapia fish exhibits sexually related dimorphic growth in which males grow and reach a larger ultimate size faster than the females (Guerrero, 1975; Mair and Little, 1991; Manosroi *et al*., 2004; Kefi *et al*., 2012). Therefore, male fish provides a major advantage in tilapia culture and in avoidance of early maturation thereby, controlling reproduction.

In Zambia, Tilapias (including *Oreochromis* species) are the major cultured fish species because they are readily acceptable by consumers throughout the country (Mudenda, 2004; Kefi *et al*., 2011). However, scarcity of fingerlings in the country remains one of the key constraints to required aquaculture production. To this effect, the government has been promoting the establishment of indoor hatcheries for the purpose of seed production involving collection of spawn for onward incubation in the hatchery. However, the point at which the eggs have to be collected has not been determined. Furthermore, the most resilient fish species in the hatchery has not been established.

There have been some studies conducted on the sex reversal of *O. andersonii* (Kefi *et al*., 2012) and reproductive performance of *O. andersonii*, *O. macrochir*, and *O. niloticus* (Kefi *et al*., 2012). Sex reversal of *O. macrochir*, *O. niloticus* and *O. tanganicae* using androgen 17 α – methyl testosterone has not been explored and it is possible that the sex reversal rate may be species specific. In the latter experiment, they did not include *O. tanganicae* as a species of interest and survival of

fish in the indoor hatchery. Furthermore, there study concentrated on fish reared in the pond and did not report how the eggs performs in an indoor hatchery, a technology that the government was promoting in the production of fish seed.

The study is, therefore, aimed at comparing the breeding efficiency, survival of fry as well as the sex reversal success rate of the four *Oreochromis* spp (*O. niloticus, O. andersonii, O. tanganicae* and *O. macrochir*) in the indoor hatchery. The information will be necessary in adapting good hatchery management practices that enhance quality and increased *Oreochromis* fish species seed production. In addition, the information is indispensable in developing egg development schemes that are necessary in the management of aquaculture production schedules.

Materials and Methods

Eight (8) hapas each measuring 8m x 2m x 1m were set up in two semi-concrete ponds measuring $250m^2$ each and were stocked with fish according to fish species and sex randomly. After 30 days, the brooders were then placed in breeding hapas at sex ratio of 1:2 (Male: Females). Each fish species was replicated twice with each hapa containing 10 males and 20 females. During this period the brooders were fed with isonitrogenous (30% crude protein) and isocaloric (10% crude lipid) feed at a feeding rate of 3 % of their total live body weight twice a day split in equal amounts.

Table 1: Initial mean weight (g), Standard length (mm) and Total length (mm) of *Oreochromis* species brood stock (Mean ±S.E)

Parameter	*O. andersonii*	*O. macrochir*	*O. niloticus*	*O. tanganicae*
Initial weight (g)	182.75 ±13.021	191.00 ± 16.127	173.75 ± 5.825	123.5 ± 7.137
Standard length (mm)	163.95 ± 4.660	160.45± 4.501	155.20 ± 3.816	141.55 ± 3.273
Total length (mm)	207.6 ± 5.557	204.80 ± 5.609	199.25 ± 2.714	177.30 ± 3.802

Fourteen days after mixing the males and females, the latter were checked for egg incubation and all those found with eggs or fry were recovered and taken to the laboratory for enumeration according to fish species. The colour of the eggs at the time of recovery from the fish was also noted coded as white = 1, yellow = 2 and brown = 3. In the laboratory, a random sample of fifty (50) eggs from each fish was taken and randomly incubated in jars mounted on a recirculatory in door hatchery. The remainder of the eggs and fry were counted and weighed on a citizen analytical balance. The eggs were assigned to the jars in a completely randomised design. The number of days taken by each species to complete yolk absorption was recorded. After hatching, the survival was then determined by counting the hatched fry. The procedure was repeated thrice.

Two hundred (200) fry of each species that had just exhausted their yolk were then collected and stocked in hapas in a complete random design for sex reversal using 17 α – methyl testosterone (MT). Each fish species was replicated twice. To prepare the feed (50% CP) for masculinisation, 60 mgMT was dissolved in 15 ml/kg feed of ethanol (Teichert – Coddington *et al*., 2000). The mixture of feed ingredients, ethanol and MT were then allowed to dry at room temperature with no direct sunlight. The feed was then stored at 8^oC till used. The fry were then fed with the MT incorporated feed for 21 days at 10% live body weight fed four times a day in equal portions. After wards, the fry were fed with same feed but without MT incorporation for 90 days. The fingerlings were then caught, killed and preserved in 10% formalin for 10 days before sex reversal examination by aceto – carmine gonadal squash method as described by Guerrero and Shelton (1974).

Water temperature, pH, electrical conductivity and dissolved oxygen were measured and recorded twice daily both in the hapas and jars using the YSI professional water checker.

Data analysis

Analysis of variance (ANOVA) was performed to determine if any significant differences ($P < 0.05$) existed in the weight of eggs and fry, brooding efficiency and time taken for egg absorption among the fish species. If significant Duncan's multiple Range Tests (DMRT) (Duncan, 1955) was applied to separate the means. Cross tabulations and Chi – square test were performed to determine if any association existed between colour and survival and between species and survival. If significant, Mann Whitney test was performed to identify where the differences existed between the fish species. Correlation analysis was performed to determine the relationship between survival and colour of eggs, time of yolk absorption and egg weight. It was further used to determine the differences in the sex reversal success among the fish species. Multinomial regression was employed to determine the relationship between stage (colour) and egg weight. All statistical tests were performed in the Statistical Package for Social Scientists (SPSS)

15.0 (SPSS Inc) while excel was used in the production of graphs.

Results

There were no significant differences ($P > 0.05$) in the brooding efficiency index of the fish species although *O. niloticus* (4.557 ± 0.483%) had the lowest index followed by *O. andersonii* (5.394 ± 1.497%). *Oreochromis macrochir* had the highest brooding efficiency index (8.968 ± 1.815%) (Table 2).

Significant differences ($P < 0.05$) were observed in the egg weight obtained from the female brooding fish. *Oreochromis tanganicae* and *O. macrochir* eggs had similar ($P > 0.05$) weights. The lowest egg weight was observed in *O. andersonii* followed by *O. niloticus* (Table 2).

Egg survival was maximised in *O. macrochir* at 74.3% and this was significantly higher ($P < 0.05$) than in other fish species under study. *Oreochromis tanganicae* had the lowest survival rate (Table 2). Aggregating the data, the highest survival was obtained in brown eggs (63.6%) and the lowest in whitish looking eggs (13.6%) (Figure 3).

Table 2: Reproduction parameters of fish species in the study

Reproduction parameter		*O. macrochir*	*O. andersonii*	*O. niloticus*	*O. tanganicae*
[1]Brooding efficiency (%)	Mean ± SE	8.968 ± 1.815	5.394 ± 1.497	4.557 ± 0.483	5.718 ± 1.185
	Minimum	3.94	0.74	2.97	2.61
	Maximum	14.13	11.32	5.77	10.36
Egg weight (mg)	Mean ± SE	11.104 ± 0.213[c]	7.207 ± 0.141[a]	8.117 ± 0.163[b]	11.415 ± 0.675[c]
	Minimum	5.10	2.60	4.10	1.80
	Maximum	15.60	11.70	11.20	22.80
Fry weight (mg)	Mean ± SE	47.160 ± 5.100	41.300± 5.262	42.783 ± 4.222	40.493 ± 4.222
	Minimum	11.60	3.3	2.10	2.40
	Maximum	99.0	92.8	99.90	91.90
[2]Survival of eggs (%)	Mean ± SE	74.3[c]	54.8[b]	57.3[b]	42.3[a]
Time taken for yolk absorption (hours)	Mean ± SE	212.77 ± 2.67[b]	190.08 ± 3.08[a]	224.58 ± 3.75[c]	215 ± 2.38[b]
	Minimum	168	120	156	120
	Maximum	264	252	264	240

[1](Weight of eggs (g)/weight of fish (g) – weight of eggs (g))*100 (Kefi *et al*., 2011). [2]Minimum and maximum not determined as the percentages taken are whole

Table 3: Percentage of eggs survived according to the colour of eggs

Colour of eggs	*O. macrochir*	*O. andersonii*	*O. niloticus*	*O. tanganicae*
White (just fertilized)	0	50	0	13.6
Yellow	0	0	0	22.7
Brown (advanced)	100	50	100	63.6

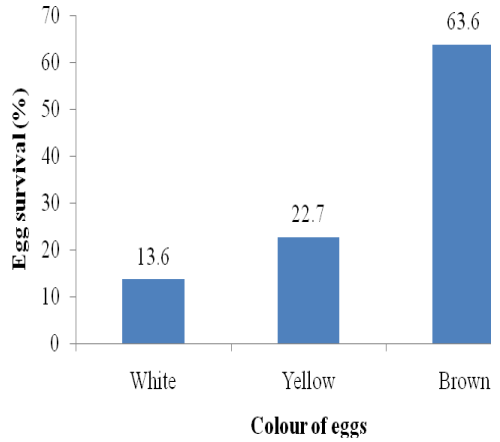

Figure 3: Survival of eggs according to egg colour

Table 4: Mean egg weight (g) according to egg colour

Egg colour	Mean egg weight (mg)
White	7.873±2.012
Yellow	6.617±1.497
Brown	7.746±1.358

Sex reversal success

The study shows that sex was skewed towards males in all the fish species. However, the highest sex reversal success was observed in *O. andersonii* (98.6%) although the percentage was not significantly different (P> 0.05) from *O. niloticus*. The lowest sex reversal was achieved in *O. macrochir* (63.9) (Table 5).

Table 5: Sex reversal success according to fish species

Species	Males (%)	Females (%)
O. andersonii	98.6^{c2}	1.4^{a1}
O. macrochir	63.9^{a2}	36.1^{c1}
O. niloticus	95.8^{c2}	4.2^{a1}
O. tanganicae	79.4^{b2}	20.6^{b1}

Different superscripts in a row and column indicate significant difference (P < 0.05)

Water quality parameters are presented in Table 6. Significant differences (P < 0.05) were only observed in the electrical conductivity observed in the water quality parameters determined with *O. andersonii* and *O. tanganicae* having water of similar (P > 0.05) conductivity (Table 6).

Table 6: Water quality parameters in the hatchery (H) and hapa (h) (mean ± standard error (S.E))

Water quality parameter	Species			
	O. andersonii	*O. macrochir*	*O. niloticus*	*O. tanganicae*
Water temperature (ºC) (H)	23.853 ± 0.124	24.068 ± 0.148	24.063 ± 0.149	23.872 ± 0.152
Water temperature (ºC) (h)	27.421±0.212	27.469±0.207	27.430±0.211	27.473±0.209
Dissolved Oxygen (mg/L) (H)	7.329 ± 0.235	7.037 ± 0.368	7.001 ± 0.350	7.147 ± 0.300
Dissolved Oxygen (mg/L) (h)	6.836±0.463	6.622±0.465	6.674±0.463	6.571±0.468
Conductivity (µmho/cm) (H)	60.183 ± 2.619^{a}	68.565 ± 2.281^{b}	67.223 ± 2.374^{b}	57.710 ± 2.609^{a}
Conductivity (µmho/cm) (h)	207.970±4.188	207.969±4.187	207.944±4.191	207.958±4.175

* n.s, not significant (P > 0.05) difference between groups

Values in the same row not sharing a superscript are significantly different

Discussion

Several fish reproductive characteristics have been proposed to define the quality of the progeny (Carrillo *et al.*, 2000). Jegede (2008) recommended the use of the total egg weight in calculating the gonadosomatic index (GSI) if its main aim is determining the reproductive potential. Results of the brooding efficiency indicate a lower index for *O. niloticus* followed by *O. andersonii* although the differences were not significant (P > 0.05). These findings are consistent with the observation made by Kefi *et al.* (2011) who observed the lowest brooding efficiency in *O. niloticus*. The lower brooding index indicates that *O. niloticus* directs the lowest energy towards reproduction allowing somatic growth even when it attains maturity and when brooding. The energy investment towards reproduction is greater in

O. macrochir since the brooding efficiency was the highest. This is because brooding efficiency is a good indicator of the GSI that indicates an investment into brooding the progeny and lower GSI indicates high reproductive efficiency.

Choosing the stage at which eggs are incubated in the hatchery is crucial if survival is to be maximised. Several authors (Geffen *et al*., 2006; Ahmed *et al*., 2007; Dhaneesh *et al*., 2009) have described the stages of egg development. However, none described the simple and quick visual methods that would make collection of eggs from brooding fish easier. In the current experiment fish eggs were categorised according to the colour of eggs. The brown colour survived the best at 63.6% while the survival of the yellow eggs was only 22.7%. Whitish eggs accounted only for 13.6%. However, in terms of fish species egg survival in the hatchery was maximised in *O. macrochir* followed by *O. niloticus* although both of the species recorded the highest mortality of the eggs that never reached the advanced stage. McGurk (1986) reported that fecundity alone may not be related to the probability of survival of the individual eggs because egg mortality is scaled to egg size. It is, therefore, expected the egg size to affect the survival of eggs in the hatchery system. In the current experiment, advanced eggs survived better than the other two stages of eggs. However, there was no any evidence of a relationship of egg development to egg size. The contradiction shows that the survival potential of individual fish eggs may not in itself be an important factor in controlling the success of fish reproduction (Duarte and Alcaraz, 1989). Therefore, it shows that embryo development in an egg does not increase the size of an egg. This phenomenon, however, requires further investigation. The longest time recorded in *O. tanganicae* for the eggs to hatch would be attributed to the fact that the majority of the eggs were not advanced therefore taking time to pass in all the stages before hatching and consequently yolk absorption. The time of egg collection is cardinal in maximising the survival of eggs in the hatchery. Furthermore, the variances in egg colour among fish species and within species justifies the need to conduct a study on the actual period for egg collection. However, within the fish species variations were observed.

The use of androgen in sex reversal of Tilapia fish species has been found to be effective and simple (Green and Teichert – Coddington, 2000; Mateen, 2007; Kefi *et al*., 2012). This is because culture of monosex *Oreochromis* species, preferably males, has been recognized as the most effective way of avoiding early maturation and uncontrolled reproduction. In the current experiment, 60mgMT/kg feed was utilised as it is the rate that provided a higher male percentage of *O. andersonii* in conditions similar to the current experiment (Kefi *et al*., 2012). The males were significantly ($P < 0.05$) higher than the females within the fish species. Similarly, the percentage of males across the species differed ($P < 0.05$). Several factors have been described to affect sex reversal (Phelps and Popma, 2000). To the best of our knowledge the species among the tilapine group has not been mentioned before. This could be due to the administration of the hormone to single fish species in experiments. Therefore, the differences in the percentage of males in the fish species could be due to variances in fish species and that the rate of sex reversal could be species specific. Therefore, *O. andersonii* provides the best alternative in sex reversal hatchery operations followed by *O. niloticus* since the percentage of males was highest at 98.7% for *O. andersonii*. Kefi *et al*. (2012) reported 94.4% of sex reversal success in *O. andersonii*.

The study shows *O. niloticus* to have the highest reproductive efficiency while *O. andersonii* presents the best species in sex reversal practices using MT. However, there is need to study the growth of sex reversed *O. andersonii* and *O. niloticus* which had similar success rate of males. The advanced stage of eggs had achieved the survival at 63.6% while the least survival was observed in white looking eggs. Therefore, we recommend that only brown eggs should be collected if survival should is to be maximised. However, the survival rate of even brown eggs will depend on handling.

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Oreochromis andersonii masculisation verification on a profile microscope

Biological and chemical laboratory at NARDC

GROWTH COMPARISON OF *OREOCHROMIS ANDERSONII, OREOCHROMIS MACROCHIR* AND *OREOCHROMIS TANGANICAE* IN SEMI CONCRETE PONDS

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Abstract

The objective was to compare the growth rate of *Oreochromis tanganicae*, *Oreochromis macrochir* and *Oreochromis andersonii* using commercial fish feed from National Milling Company. Fish of mixed sex with an average weight of 6.7g were used. Three treatments; were used in this study; treatment 1 was *Oreochromis tanganicae*, treatment 2 was *Oreochromis macrochir* and treatment 3 was *Oreochromis andersonii* under a complete randomized design (CRD) for a period of 120 days. The study demonstrated that the *O. andersonii* produced higher final mean weight and specific growth rate though there was no significant (P>0.05) difference with *O. macrochir*. *Oreochromis tanganicae* recorded significantly lower survival rate than other treatments.

Key words: *Oreochromis,* growth and survival.

Introduction

Tilapias are the most popular food fish in Zambia. More than 20 species of tilapia are cultured in developing counties where animal protein is lacking. Although native to Africa, tilapias are cultured in many parts of the world (Maclean *et al*., 2002). In Zambia *Oreochromis* species are the most farmed finfish. In captivity, the success of tilapia is dependent on their ability to thrive on readily available natural food organisms and prepared diets (Teichert – Coddington, Popma, and Lovshin, 1997; Popma and Masser, 1999) In tropical pond waters under semi-intensive culture management, the Nile tilapia can grow to 150 – 250 g in 4 to 6 months, 500 to 800 g in 10 to 12 months and 2 – 3 kg in 2 years. In the great lakes of Africa, several scientists (Worthington and Ricardo 1936; Lowe-McConnell 1958) recorded maximum sizes of 61- 64 cm, weights 4 – 7 kg.

The main drawback to the culture of tilapia is their excessive recruitment in earthen ponds resulting in low yield of harvestable size of fish. Though more research has been done on the growth rate of tilapias, much has been done on *Oreochromis niloticus* while less has been done on other *Oreochromis* species in semi concrete ponds. This study therefore tries to compare and contrast the growth performance of O. *tanganicae, O. macrochir* and *O. andersonii* in semi concrete.

Objective

- To compare and contrast the growth performance of Oreochromis *andersonii, O. tanganicae and O. macrochir* in semi concrete ponds.

Materials and Methods

Experimental design

The experiment was carried out at Misamfu fish farm for a period of 120 from July to September, 2012. Nine semi concrete ponds measuring 80m^{2}each were used in the experiment. The experiment had three (3) treatments in triplicate. Treatment 1 was O. *tanganicae*, treatment 2 was *O. macrochir* and treatment 2 was *O. andersonii*. The treatments were randomly assigned to the semi concrete ponds.

Stocking of experimental tanks

Fish of mixed sex with an average weight of 6.9g were used in the experiment. The fingerlings were obtained locally at the fish farm. Each semi concrete pond was stocked with 320 fingerlings.

Determination of fish growth

Mean weight of individual fish was determined at stocking and 10% of fish were randomly sampled from each pond every three weeks using a seine net to determine the growth rate. Weight (g) of each fish was measured using a balance and total length (cm) was measured using measuring board.

Growth performance parameters of the harvested fish were calculated according to Kang'ombe, Brown, Halfyard (2006) and NACA (1985) using the following formulae:

- Average daily gain weight (ADG, g d^{-1}) = weight gain (g) / time (days);
- Specific growth rate (SGR, % d^{-1}) = 100 x (Ln final weight (g) – Ln initial weight (g))/ time (days)
- Survival rate (%) = 100 x (initial number of fish - number of dead fish)/Initial number of fish.

Physiochemical and biological parameters of water

Temperature and pH were measured using a multi probe water checker by dipping it into the pond. Temperature was measured twice a day at 08:00 and 14:00 hours throughout the growing period.

Statistical analysis

One way ANOVA was used to analyze the data. Significant differences (P <0.05) among means were tested by Duncan's multiple range test (Duncan, 1955). All the tests were performed using SPSS 17.0 statistical software.

Results and discussion

The mean values of water quality parameters measured in the semi concrete ponds are shown in Table 1. There were no significant (P>0.05) differences in the water quality parameters in all the treatments. Water temperatures ranged from 18.6 to 18.9 ^{o} C in the morning and ranged from 24.8^{o} C to 24.9 ^{o} C in the afternoon and were within the recommended range for tilapia growth and high yield (Caulton, 1977; Meske, 1985; Buttner *et al.,* 1993; Popma and Masser, 1999).

The pH was within optimum ranges for tilapia growth. Tilapia can survive in pH ranging from 5 to 10, but they do best in a pH range from 6 to 9 (Popma and Masser 1999). Lawson (1994) and Popma and Masser (1999) recommended that pH value between 6.5 and 9 was the best for fish production. The differences in pH in fish ponds are usually associated with differences in phytoplankton standing crops and subsequent differences in photosynthetic activity (Boyd, 1990).

Table 1. Water quality parameters measured in semi concrete ponds

Parameters	Treatments		
	Treatment 1 *O. tanganicae*	Treatment 2 *O. macrochir*	Treatment 3 *O. andersonii*
Morning temperature (^{o}C)	18.9 ± 0.10	18.8 ± 0.12	18.6 ± 0.10
Afternoon temperature (^{o}C)	24.8 ± 0.13	24.9 ± 0.23	24.8 ± 0.12
pH	6.7 ± 0.09	6.6 ± 0.03	6.7 ± 0.09

Values without superscripts in rows are not significantly different *(P>0.05)*

Growth performance data are presented in Table 2. The average initial weight of the stocked fish ranged from 6.9g to 7.1g. The final mean weight ranged from 91.7g to 102.69g and differed significantly (P < 0.05) with *O. tanganicae* recording the lowest growth rate.

There were no significant (P > 0.05) differences in the growth performance between *O. macrochir* and *O. andersonii*. Specific growth rate varied from 2.16 % day^{-1} to 2.24 % day^{-1} and were significantly (P < 0.05) different with treatment 1 recording the lowest. However, there was no significant (P > 0.05) difference between treatment 2 and 3. Specific growth rates in this study compare very well with the results reported by El-Zaeem SY (2011) for *O. niloticus* (ranging from 1.68% to 1.96 % day^{-1}) and for *O. aureus* (ranging from 1.7% to 2.0 % day^{-1}) raised in the laboratory.

Table 2: Initial weight, final weight, weight gain, specific growth rate (SGR) of *O. macrochir*, *O. tanganicae* and *O. andersonii*

Parameters	Treatments		
	Treatment 1 *O. tanganicae*	Treatment 2 *O. macrochir*	Treatment 3 *O. andersonii*
Initial mean weight (g)	6.9 ± 0.05	7.1 ± 0.05	7.0 ± 0.09
Final weight (g)	91.7 ± 1.07^{a}	102.4 ± 1.45^{b}	102.69 ± 1.32^{b}
Weight gain (g)	84.8 ± 0.01^{a}	95.3 ± 0.02^{b}	$95.89 \pm 0{,}01^{b}$
SGR (% day^{-1})	2.16 ± 0.03^{a}	2.22 ± 0.01^{b}	2.24 ± 0.02^{b}
Survival rate (%)	62.0 ± 0.01^{a}	70.8 ± 0.02^{b}	80.6 ± 0.03^{c}

Values without superscripts in rows are not significantly different (P>0.05)
Values with different superscripts in a row are significantly different (P<0.05)

In this study, reproduction occurred in *O. macrochir* and *O. andersonii* treatments at 100g. Eggs were found in the mouth of *O. macrochir* and *O. andersonii* and some fry were observed in the ponds signaling the onset of breeding.

Survival rates in this study were lower than 90% as reported by Kang'ombe *et al*. (2006) and Kang'ombe *et al.* (2007) for *T. rendalli* in concrete ponds fertilized with animal manure and chicken manure supplemented with a diet. However, the results in this study were higher than was reported by Muendo *et al* (2006) who reported survival rates of *O. niloticus* ranging from 41% to 54 % in fish ponds treated with chicken manure, field grass and floating pellets. The lower survival rates in this study were because of predation by the birds as well.

The study demonstrated that the *O. andersonii* produced higher final mean body weight and specific growth rate though there were no significant differences with *O. macrochir*. *Oreochromis tanganicae* recorded significantly lower survival rate than other treatments.

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Research concrete ponds at Misamfu Aquaculture Research Station