EFFECT OF ORGANIC AND INORGANIC FERTILIZERS ON NATURAL FOOD COMPOSITION AND PERFOMANCE OF AFRICAN CATFISH (*CLARIAS GARIEPINUS*) FRY PRODUCED UNDER ARTIFICIAL PROPAGATION

MSc. (AQUACULTURE) THESIS

SEBASTIAN S. MOSHA

LILONGWE UNIVERSITY OF AGRICULTURE AND NATURAL RESOURCES

BUNDA CAMPUS

DECEMBER, 2015

EFFECT OF ORGANIC AND INORGANIC FERTILIZERS ON NATURAL FOOD COMPOSITION AND PERFOMANCE OF AFRICAN CATFISH (*CLARIAS GARIEPINUS*) FRY PRODUCED UNDER ARTIFICIAL PROPAGATION

SEBASTIAN S. MOSHA

BSc. (Aquaculture), Tanzania

A THESIS SUBMITTED TO THE FACULTY OF NATURAL RESOURCES IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR AWARD OF THE DEGREE OF MASTER OF SCIENCE IN AQUACULTURE

LILONGWE UNIVERSITY OF AGRICULTURE AND NATURAL RESOURCES

BUNDA CAMPUS

DECEMBER, 2015

DECLARATION

I, Sebastian Severin Mosha, declare that the work presented in this thesis is an outcome of my own research effort and assessment and that the results have never been previously presented to the Lilongwe University of Agriculture and Natural Resources or other academic institution for the award of any academic qualification. Any sources of information utilized have been acknowledged by means of references.

Sebastian Severin Mosha

Signature: _____

Date: _____

CERTIFICATE OF APPROVAL

We, the undersigned, certify that this thesis is a result of the author's own work, and that to the best of our knowledge, it has not been submitted for any other academic qualification within the Lilongwe University of Agriculture and Natural Resource or elsewhere. The thesis is acceptable in form and content, and that satisfactory knowledge of the field covered by the thesis was demonstrated by the candidate through an oral examination held on 22nd December 2015.

Major Supervisor: Professor Jeremiah Kang'ombe

Signature: _____

Date: _____

Supervisor: Dr. Nazael Madalla

Signature: _____

Date: _____

Supervisor: Associate Professor Wilson Jere

Signature: _____

Date: _____

DEDICATION

I dedicate this thesis to my wife Rhoda Komanya, all my beloved twins; Moreen and Maureen. To my father Raphael Mosha and mother Monica Mosha, my uncle Severin Mosha and my aunt Sarah Mamboya. To all my brothers Deocrasias and Wolfram; my Sisters Beatrice and Segolina, and all my relatives. Also to all my friends who encouraged me to work hard to make this thesis possible.

ACKNOWLEDGEMENTS

First and foremost, I am very grateful to Professor. J. Kang'ombe (Major Supervisor), Dr. N. Madalla (Co-Supervisor in Tanzania) and Associate Professor. W. Jere (Co-Supervisor) for their advice and assistance they provided to make this thesis possible.

Secondly, I am sincerely thanking iAGRI/USAID and RUFORUM for the financial support in my studies. Also my appreciation goes to all lectures and staffs members in the Department of Aquaculture and Fisheries Science for their support which have contributed to my research work.

Thirdly, my thanks go to Officer In-Charge of Kingorwila National Fish Farming Center for his support in the provision of water quality parameters and other facilities. Also my special thanks go to staff members in Department of Animal Science and Production Laboratory, and Department of Veterinary Medicine Laboratory at Sokoine University of Agriculture (SUA) for their assistance.

In addition, I am extending my thanks to all fellow masters' students; Richard Makanda (Zambia), Mohammed Elshekhe Mohammed (Sudan), Elysee Nzohabonayo (Burundi), Tizita Moges (Ethiopia), Mexford Mulupwa (Malawi) and Benson Lusangasi (Malawi) for their support and advices.

Finally, I am obligated so much to my employer, Ministry of Agriculture Food Security and Cooperative, for giving a study leave permit. Also am grateful to Dr. H. E. Nonga and Dr. H. A. Lamtane (SUA) for their support and timely advices.

ABSTRACT

Demand for fish in both rural and urban areas is very high in Tanzania. In recent times, the African catfish, *Clarias gariepinus,* has gained popularity in the aquaculture sector of Tanzania. However, many aquaculture farmers face the challenge of unreliable supply of seed due to difficulties of reproducing in captivity and high larvae mortality. The study was conducted to determine the effect of organic and inorganic fertilizers on natural food composition and performance of African catfish (*C. gariepinus*) fry produced under artificial propagation. The results indicated that abundance of natural food (phytoplankton) were significantly higher ($P<0.05$) in di-ammonium phosphate (DAP) fertilizer applied tanks compared to other treatments. While zooplankton diversity were higher in chicken manure applied tanks compared to other treatments. Individual final mean weight, weight gain and specific growth rate were not significantly different ($P>0.05$) between DAP fertilizer and chicken manure treatments across stocking densities (5fry/m^2 and 10fry/m^2), but were significantly higher than the control. Survival rates were not significantly different ($P>0.05$) between chicken manure and DAP fertilized treatments across stocking densities but differed significantly ($P<0.05$) from the control. Water quality parameters were found to be within the optimum range for both experiments. In conclusion, the study indicated that higher phytoplankton abundance are attained with DAP fertilized tanks and zooplankton diversity were higher in chicken manure applied tanks. Fish growth performance was higher in chicken manure treatment at low stocking density (5fry/m^2). Therefore, it is recommended that for better growth

and survival in aquaculture practices, catfish fry should be raised in DAP or chicken manure fertilized tanks at low stocking density (5fry/m^2).

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

LIST OF ABBREVIATIONS AND ACRONYMS

DAP	Di-ammonium phosphate
DO	Dissolved oxygen
N:P	Nitrogen Phosphorus ratio
Ppm	Parts per million
SE	Standard error
SGR	Specific Growth Rate
Temp	Temperature
U-DAP	Urea and di-ammonia phosphate

CHAPTER ONE

INTRODUCTION

1.1 Background information

The increase in human population and reports of large numbers of people over 62 percent of the world's population, undernourished or starving (especially in the developing countries such as Tanzania) has made the need for food production a major worldwide issue of concern (Tacon, 2001; Okechi, 2004). There are three major groups of activities that contribute to food production namely agriculture, aquaculture and fisheries. Recent knowledge shows that the world's natural stocks of fish and shell fish, though renewable, have finite production limits, which cannot be exceeded even under best management regimes (Food and Agriculture Organization of the United Nations [FAO], 2000; Okechi, 2004). Most of our lakes, rivers and oceans have exceeded maximum sustainable fishing limit, which calls for an increase in fish supply from aquaculture (FAO, 2012).

In Tanzania, demand for fish in both rural and urban areas is very high (FAO, 2007). Aquaculture which is largely a subsistence activity practiced by poor households in the coastal and inland areas. It has several benefits such as contributing to peoples' requirements for animal protein, particularly in the rural areas where there are no fish caught from the wild. It also providing employment opportunities and is a source of income. However, as capture fisheries are stagnating, the future for Tanzania lies in aquaculture (Tanzania Fisheries Division, 2006).

African catfish (*Clarias gariepinus*) is one of the commonly cultured species in Tanzania (Musiba *et al.,* 2014). The merits of farming African catfish (*Clarias gariepinus*) over other species such as tilapia (*Oreochromis niloticus*) are influenced by both biological and rearing characteristics of the species (Ministry of Livestock and Fisheries Development, 2011). Its demand increases as food, control of over-population in tilapia ponds and as bait for the Nile perch fishery (Musiba *et al.,* 2014). Although catfish farming in Tanzania started during the colonial *era* (i.e. since 1940s), it has remained undeveloped ever since. One of the major obstacles in this respect is the unavailability of quality feeds (Shoko *et al.*, 2011). This problem has driven most fish famers to find alternatives such as plankton that is boosted by fertilizer application. Pond/tanks fertilization can be used to increase natural food production (Grag and Bhatnagar, 2000; Hossain *et al.*, 2008) by enhancing the growth of primary producers which are consumed by fish (Hossain *et al.*, 2006).

1.2 Problem statement and justification

The rearing of *Clarias gariepinus* larvae to juveniles has proved to be challenging due to their small size and lack of functional digestive system (Govoni *et al.,* 1986; Olurin *et al.,* 2012). Great losses are incurred in the hatchery, as fry weans over from yolk absorption to exogenous feeding (Adewumi, 2015). This is due to inability to accept large sized feeds and assimilate protein from dry formulated diets (Govoni *et al.,* 1986; Cahu and Zambonino-Infante, 2001). According to Agadjihouede *et al.* (2012), *Artemia* constitutes an excellent starting food in larviculture of *Clarias gariepinus*. However, its cost is high and not available in developing countries especially in the rural fish farming communities (Agadjihouede *et al.,* 2012). Due to this fact, it has been found important to firstly provide

the larvae with live foods such as zooplankton or algae before they are sequentially acclimatized to accepting formulated diets (Olurin and Oluwo, 2010; Olurin *et al.,* 2012).

Provision of live foods to fish larvae is appreciated as an important aspect because they supply nutrients to the larvae and as well as exogenous enzymes important for the digestion of other feeds, and enhance the development of larvae's pancreas (Lubzen *et al.,* 2001). According to Govoni *et al*. (1986), it is suggested that initial digestion in the fish larvae is carried out by enzymes present in the live prey. The growth of natural feed in the fish ponds/tanks was encouraged by fertilizers application (Chenyambuga *et al.,* 2011). According to Lin *et al*. (1997), fertilization in fish ponds/tanks is known worldwide to improve pond productivity by promoting the growth of phytoplankton thereby increasing natural food available to fish.

Among fertilizers, chicken manure and di-ammonium phosphate fertilizers were chosen because they are cheaper and locally available to fish farmers (Diana, 2012). Also these fertilizers contain a good combination of nitrogen and phosphorus in different proportions which increase the quantity of primary producers (Akand, 1986; Kumar *et al.,* 2014). Although a lot of progress has been made so far in aquaculture, larvae rearing remains the bottleneck in *C. gariepinus* production. The use of natural live food at earlier stage of fry development seems to be one of the solutions to improve growth performance and survival.

1.3 Objectives of the study

1.3.1 General objective

The overall objective of this study was to determine abundance and diversity of live feed composition in tanks applied with chicken manure and di-ammonium phosphate, and performance of catfish fry produced under artificial propagation at different stocking densities.

1.3.2 Specific objectives

The specific objectives were to:

a) Determine the abundance and diversity of different species of live feed produced in tanks applied with chicken manure and di-ammonium phosphate.

b) Assess the growth performance of the catfish fry reared in tanks applied with chicken manure and di-ammonium phosphate at different stocking densities.

c) Evaluate the survival rate of the catfish fry reared in tanks applied with chicken manure and di-ammonium phosphate at different stocking densities.

d) Examine the water quality parameters in tanks applied with chicken manure and di-ammonium phosphate at different fry stocking densities.

1.4 Hypotheses

Study hypotheses were:

a) H_0: There is no significant difference on abundance and diversity of different species of live food produced in tanks applied with chicken manure and di-ammonium phosphate.

b) H_0: There is no significant difference in growth performance of the catfish fry reared in tanks applied with chicken manure and di-ammonium phosphate at different stocking densities.

c) H_0: There is no significant difference in survival rate of the catfish fry reared in tanks applied with chicken manure and di-ammonium phosphate at different stocking densities.

d) H_0: There is no significant difference on water quality parameters in tanks applied with chicken manure and di-ammonium phosphate at different fry stocking densities.

CHAPTER TWO

LITERATURE REVIEW

2.1 Importance of aquaculture and catfish production

Fish has been an important source of food for centuries and contributes around 50 percent of total animal protein in the diets of many Africans (FAO, 2003). At the global level, aquaculture fish production constituted 42.2 % of global fish production (total 158 million tonnes in 2012) from capture fisheries and aquaculture (FAO, 2014). In 2010, Asia accounted for 89 percent of world aquaculture production, and this was dominated by the contribution of China, which accounted for more than 60 percent of global aquaculture production (FAO, 2012). Aquaculture also plays an important role in providing a livelihood to millions of people around the world. In 2004, FAO (2007) estimated that 4.5 million people were engaged directly in aquaculture with most of these located in the developing countries.

In Tanzania, the aquaculture sub-sector has a great potential for expansion, especially due to the fact that demand for fish is increasing as a result of population growth and stagnant production from capture fisheries, both at domestic and global levels (Ministry of Livestock and Fisheries Development, 2010). Many people are employed in a wide range of activities linked to aquaculture such as provision of inputs as well as processing of output (Madalla, 2008).

Production of marketable fish begins with the stocking of fry or juveniles into a rearing environment that assures optimum and rapid growth to allow harvest in the shortest possible time. These fish/seeds can come from wild capture and hatcheries (FAO, 2003). Among the cultured fish species is *Clarias gariepinus* which do not normally reproduce spontaneously in captivity (Pouomogne, 2008). The maturation processes of *C. gariepinus* are influenced by annual changes in water temperature and photoperiod. The final triggering of spawning is caused by a raise in water level due to rainfall (de Graaf *et al.*, 1995). Catfish does not have parental care for ensuring the survival of its offspring except by the careful choice of a suitable site (Bruton, 1979; Ayele, 2015).

Induced breeding has been developed, but production and hatchery management techniques that make catfish seed of good quality readily available to all farmers are yet to be established in most African countries (Brummett *et al.,* 2007). In hatcheries, catfish larvae are fed on a mixture of cow brain plus egg yolk just after vitelline resorption for 4-6 days before being stocked at 50-80/m^2 in nursery ponds fertilized beforehand (usually with chicken manure) to enhance zooplankton development (FAO, 2010). The post larvae are stocked in ponds protected from predators and fed with single ingredients or compounded feeds. Harvesting is carried out after 24-28 days and fingerlings are graded. The average weight of fingerlings at this stage has been 5-7g (FAO, 2010). Since the recommended size at transfer to production ponds should be larger than 10g, additional pre-fattening may be carried out except when the volume of immediate demand drives hatchery operators to sell the fingerlings at 6g. Normally, survival in properly managed nursing systems averages 25-35% (FAO, 2010).

2.2 Enhancing natural food using fertilizers and its impact on catfish fry performance

Primary production in fish pond/tank is limited by phosphate, in particular, and nitrogen (de Graaf and Janssen, 1996). Fertilizer application stimulates the growth of decomposers such as bacteria and fungi which breaks down toxic waste products that can accumulate with the use of prepared feeds (Wurts, 2004). It is an inexpensive method of feeding and plankton is necessary for smaller fish which are small to eat supplemental feeds (Chenyambuga *et al.,* 2011). Ponds with fertilized water will turn into a rich green or reddish color when the plankton becomes abundant. This means that food will be available for the fish. Ponds/tanks with clear water are not fertile and lack plankton (Murnyak, 2010). Due to small size of fish larvae, fertilization is well-suited to the production of plankton and benthic invertebrates which are preferred prey for young and provide high quality protein as well as other essential nutrients (Wurts, 2004). An example of a study conducted in Morogoro, Tanzania showed that, management strategies of fish ponds under small-scale systems involve the use of fertilizer to encourage growth of natural food and to improve the level of dissolved oxygen. Most farmers reported that they fertilize their ponds using manure from domestic animals before stocking the fingerlings. This is supported by the observation that 70 percent of the fish farmers had ponds with greenish water, indicating plankton abundance (Chenyambuga *et al.,* 2011).

The commonly used organic fertilizer is chicken manure (Amisah *et al.,* 2008), which stimulates abundant growth of zooplanktons and insect larvae and other forms of fish food organisms (Akand, 1986; Kumar *et al.,* 2014). Inorganic fertilizers are also commonly used

to fertilize ponds for the rearing of fry and fingerlings under supplementary feeding conditions. Inorganic fertilizers such as di-ammonium phosphate (DAP) mainly increase the quantity of primary producers (Akand, 1986; Kumar *et al.,* 2014). These fertilizers contain a combination of nitrogen and phosphorus in different proportions (Diana, 2012). According to Lin *et al.* (1997), nitrogen is applied as either urea whereas phosphorus is added as DAP. An abundant supply of zooplankton is necessary to successfully rear catfish fry to fingerlings in ponds. Zooplankton abundance is dependent on phytoplankton blooms. Phytoplankton and zooplankton are a rich source of protein often containing 40-60 percent protein on a dry matter basis and are sufficient to support excellent fish growth (De Silva and Anderson, 1995; Sutar *et al*., 2012). Diatoms are important primary producers in aquatic ecosystems and represent an important source of food for aquatic grazing organisms (Round *et al.,* 1990; Debenest *et al.,* 2009). Rotifers are the most important live food organisms for use as starter food for rearing small fish larvae (Watanabe *et al.,* 1983; Shiri *et al.,* 2003). Both the qualitative and quantitative abundance of plankton in a fish pond are of great importance in managing the successful aquaculture operations (Kumar *et al.,* 2014).

Nutritional quality of the feed distributed to the fish larvae determines the success of the delicate larval rearing stage to a large extent (Awaiss and Kestemont, 1998). Moreover, nutritional quality of feed is certainly one of the major parameters, which must be determined in species exhibiting a high growth rate. This is necessary in order to avoid a restricted feeding level, which will frequently exacerbate growth heterogeneity and cannibalism (Awaiss and Kestemont, 1998). An important aspect of larval culture is the

ability to provide feed that is able to sustain growth and development (Mohseni *et al.,* 2012). A variety of artificial dry feeds such as commercial trout starters and microencapsulated egg diet have been tested for primary nursing of *C. gariepinus*. The food intake was considerably reduced especially a few days after commencement of feeding compared to live food items, where growth was poor and mortality high (Verreth *et al.,* 2007). Therefore, feeding live zooplankton from nearby fresh water fish ponds seem to be the most reliable technique for African countries since importation of *Artemia* eggs is either difficult (no foreign exchange or too high importation tax) or prohibited (Brummett *et al.,* 2007).

2.3 Effect of stocking density and water quality on performance of *C. gariepinus* fry

Survival and growth of catfish larvae and fry are influenced by several factors such as stocking density (Schram *et al.,* 2006; Nwipie *et al.,* 2015) and water quality (Brazil and Wolters, 2002; Pangni *et al.,* 2008). Stocking density is one of the main factors determining growth (Rahman *et al.,* 2005; Dasuki *et al.,* 2013). Growth of juvenile African catfish is directly density dependent (Hengsawat *et al.,* 1997; Toko *et al.,* 2007). Juvenile African catfish are normally highly aggressive when confined in small numbers in a large volume of water. Aggressiveness, territorial defense and development of hierarchies and individual dominance are often reduced at high stocking density and fish may start to be stressed once certain threshold densities are attained (Hecht and Uys, 1997; Obirikorang *et al.,* 2014). Aggression can result in stock losses, reduced food conversion efficiency and slower growth. So, there must be a density from which both growth and production decreased with increasing stocking density (Toko *et al.,* 2007). The effects of stocking density on growth

and survival have been studied on some African catfishes such as *Clarias gariepinus* (Haylor, 1992) and *Heterobranchus longifilis* (Ewa-Oboho and Enyenihi, 1999; Coulibaly *et al.*, 2007; Dasuki *et al.,* 2013). Hecht and Appelbaum, (1987) and Haylor, (1992) observed that lower stocking densities always gave the higher growth rate in an experiment with 25 day old *C. gariepinus* fingerlings density range 5/l to 20/l. Suziki *et al.* (2001) observed that increase in stocking density results in increasing stress, which leads to higher energy requirements, causing a reduction in growth rate and food utilization (Olubunmi *et al.,* 2009). However, lower stocking density is also known to increase the rate of cannibalism, for instance, Haylor, (1991) and Hossain *et al*. (1998) found that increasing stocking density from 50 fry/l to 150 fry/l helped to decrease the incidence of cannibalism provided the fish were well-fed.

In addition, the survival rate of *Clarias batrachus* larvae reared in tanks decrease as stocking density increase (Sahoo *et al.,* 2008). Studies of African catfish larvae have shown negative effects of increasing density, reflected by decreased growth performance and increased cannibalism (Hecht and Appelbaum, 1988; Hossain *et al.,* 1998). In contrast, studies of juvenile African catfish showed a positive effect of increasing density, reflected by increased growth performance and decreased aggression (Hecht and Appelbaum, 1988; Hecht and Uys, 1997; Kaiser *et al.,* 1995a, b; van de Nieuwegiessen *et al.,* 2009). Water quality, mainly dissolved oxygen and pH levels are considered as the limiting factors in intensive fish culture. High levels of dissolved oxygen increased growth in channel catfish *Ictalarus punctatus* larvae reared in tanks (Brazil and Wolters, 2002; Pangni *et al.,* 2008). The decreasing trend of dissolved oxygen in tanks with high stocking densities

would be attributed to the gradual increase in biomass, resulting in higher oxygen consumption at varied stocking densities (Pangni *et al.,* 2008).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study area

The study was conducted from March 2015 to June 2015 at aquaculture research facility in Magadu farm at Sokoine University of Agriculture (SUA). SUA is located about 2.5 km South of Morogoro Municipality at an altitude of 550m above sea level. The area receives approximately, 880mm of rain annually with temperature varying from 27^0C to 31^0C. All experiments were conducted in concrete tanks with an area of seven square meters each and supplied with fresh water from Uluguru Mountains. Chicken manure, di-ammonium phosphate fertilizer and no fertilizer (control) were used as treatments. Experiment 1, involved determination of abundance and diversity of natural food in tanks applied with different type of fertilizer. Experiment 2, involved evaluation of growth performance and survival of African catfish fry stocked under two stocking densities (5fry/m^2 and 10fry/m^2), in tanks applied with different type of fertilizer. Two experiments were conducted for 1 week and 2 months, respectively.

3.2 Experiment 1: Determination of abundance and diversity of natural food

Two types of fertilizers (chicken manure and di-ammonium phosphate fertilizer) were used in this experiment. Fresh manure from layers kept in the cages were collected from poultry unit belonging to Department of Animal Science and Production. N (2.55%) and P (0.95%) in chicken manure was determined using proximate analysis at Animal Science laboratory according to Association of Official Analytical Chemists (AOAC, 2002). Nitrogen was

determined using Kjeldahl method where 20g of manure was transferred to Kjeldahl digestion tube. In the fume hood, 3mL of concentrated H_2SO_4 was added, followed by one Kjeldahl tab and a few boiling chips to each digestion tube. Then, digestion tube was placed in 160^0C digestion block for 30 minutes. The temperature was raised from 5^0C min^{-1} to 380^0C, and was hold for 60 minutes until digestion is completed. The tube was removed from block and cooled in acid resistant fume hood for 30 minutes. Then, diluted to graduation mark with deionized water, caped and mixed using vortex mixer for analysis on the Technicon Auto-Analyzer. The following formula was used to calculate N content as percent (%):

$$N = C/(TS/100\%) \quad (1)$$

Where C is the N concentration result from the instrument (%) and TS is total solids of the test portion (%).

Phosphate was determined, in duplicate, by microwave digestion (17 min) with concentrated HNO_3 followed by inductively coupled plasma-optical emission spectroscopy (ICP-OES). Di-ammonium phosphate fertilizer was purchased from a local agro-input shop in Morogoro containing 18% N and 46% P as indicated on the package. Nine concrete tanks having an area of $7m^2$ each were laid out with 10cm of soil (Figure 3.1) and then filled with water to a depth of 0.8m. Three treatments: 2kg of chicken manure, 42g of di-ammonium phosphate fertilizer and no fertilizer (control) were randomly assigned to the tanks. Each treatment was replicated thrice.

Figure 3.1 Tanks laid out with soil during preparation

One week after fertilization, water samples were collected with a 10L bucket from 4 locations within each concrete tank for all 9 tanks at 08:00am. Water sample was placed into a plankton net (KC Denmark A/S, Denmark) with 20μm mesh size and left for about 20min (Figure 3.2). The concentrated sample from plankton net was transferred into 200 ml plastic bottles. Five drops of 70 percent alcohol were added to each sample to fix the organisms and taken to Faculty of Veterinary Medicine Laboratory at Sokoine University of Agriculture for analysis.

Figure 3.2 Concentrating the water sample using plankton net

3.2.1 Plankton identification and quantification

Plankton identification was done on a light microscope (BRESSER, Gutenbergstr .2. DE-46414 Rhede/Westf, Germany) 100x magnification using identification keys (United Nations Educational Science and Cultural Organization [UNESCO], 2006). One milliliter (1ml) of the water sample was taken from the collecting bottles (200ml bottles) using micropipette and transferred to the Sedgewick Rafter cell (Made in England) then covered with slide at the top and placed under microscope (Figure 3.3). From 10 randomly selected squares of cell, planktonic organisms were enumerated and numerical abundance was calculated. Phytoplankton and zooplankton abundance were calculated using the following formulas as described by Greenberg *et al.* (1992) and Wetzel and Likens (1991), respectively:

$$Phytoplankton\ Abundance = \frac{C}{F \times V} \quad (2)$$

Where C is the number of organisms counted, F is the number of fields counted and V is the volume of sample settled.

$$Zooplankton/L = \frac{(C \times Va)}{(Vb \times Vc)} \quad (3)$$

Where C is the number of zooplankton counted, V_a is the volume of the concentrated sample (L), V_b is the volume of counted sample (L) and V_c is the volume of water filtered (L).

Shannon-Wiener diversity index (H') and evenness (J') were also estimated. Diversity index of plankton was calculated by using the formula as described by Krebs (1989):

$$H' = \frac{n \ln(n) - \sum_{i=1}^{k} f_i \ln(f_i)}{n} \quad (4)$$

Where *k* is the number of categories, f_i is the number of observations in category *i*, *n* is the sample size.

Species evenness or homogeneity or relative diversity (J`) was calculated from the observed species diversity and from the equation of H_{max} as described by Sundar *et al.* (1995). Index of species evenness was measured by using the following formula:

$$J' = \frac{H'}{H'_{max}} \quad (5)$$

Where, $H'_{max} = \ln(k)$

After plankton quantitative analysis, phytoplankton and zooplankton were further identified up to genus level following the guide of Bellinger (1992).

Figure 3.3 Plankton identification and quantification under microscope

3.3 Experiment 2: Determination of growth and survival of African catfish *Clarias gariepinus* fry

The objectives of this experiment were to assess growth performance and evaluation on survival of catfish fry reared in concrete tanks fertilized using chicken manure and di-ammonium phosphate treated tanks at different stocking densities.

3.3.1 Artificial propagation and rearing of catfish larvae in the hatchery

The brood stock was collected from Mindu dam at Morogoro Municipal. A well distended, swollen abdomen female weighing 355g and a male weighing 350g were selected according to de Graaf and Janssen (1996). Both female and male were kept separately in a tank for 24hrs without feeding. A fresh catfish pituitary gland extract was used to induce breeding. According to Parker (1984), this hormone involved in the control of maturation and spawning of fishes by stimulating hypothalamus to produce the small peptide hormone (Releasing hormone) and Gonadotropine hormone which pass into the blood to reach the gonads. The pituitary gland extract (0.09g) was obtained by killing a male (344g) and was ground in a porcelain mortar then mixed with salt solution (9 g of common salt/litre of water). The reason behind was that, the mixture (saline solution and pituitary gland) should be isotonic with the blood of the recipient fish. A syringe was filled with the solution and female was injected intramuscularly to induce final maturation and ovulation of eggs. The fish were left in the hatchery tanks for 18 hours to allow ovulation of eggs. Stripping of the female was carried out after 18 hours of pituitary hormonal injection by gently pressing their abdomen with a thumb from the pectoral fin towards the genital papilla. Ovulated eggs were flowing out easily in a thick jet from the genital vent and collected into a dry plastic container (Figure 3.4).

Figure 3.4 Brood stock selection, hormonal injection and stripping (From left to right)

The sperm was obtained by killing a male, testes (1.6g) were then dissected and placed in a mortar or a tea cup. The testes were rapidly cut into small pieces using a scissor and finally the milt was pressed out with a pestle according to procedures reported by de Graaf and Janssen (1996). The eggs were mixed with milt by stirring with a feather and then spread out onto mosquito netting (mesh size 0.5 mm) placed in an incubator. Continuing water flow was maintained to ensure supply of oxygen (Figure 3.5).

Figure 3.5 Eggs fertilization and incubation (From left to right)

After 1 hour, the dead eggs and unfertilized eggs were removed by using forceps. Eggs hatched after 27 hours of incubation at 28°C and the hatchlings passed through the screen while shells remained on the screen. The hatchlings were collected and kept in an aquarium ($0.6m^2$) placed in a dark place for three days without being fed as they relied on the food resource within their yolk sac. Aquarium was connected to three aerators which continued supplying air for 24 hours. The temperature was maintained at 30^oC with automated glass heater. Larvae were fed 3g of formulated feed (55% crude protein) at an interval of two hours to satiation for a period of 2 weeks. The remaining feeds were removed by siphoning every morning using a pipe. After the larvae attained 0.2g, they were transferred to concrete tanks ($3.6m^2$) and kept for another 2 weeks. The tanks were supplied with flowing water for aeration and larvae were fed 25% body weight (0.05g/individual) in a day at the interval of every 6 hours. The tanks were covered with black nylon to prevent sun light. After 2 weeks the larvae attained 0.5g (Figure 3.6).

Figure 3.6 Catfish fry attained in two weeks

3.3.2 Experimental procedure

Random fertilization of tanks used in this experiment was done one week before fry stocking. Total of 18 concrete tanks (7m^2 each) were used. Six tanks were fertilized with chicken manure and the other 6 tanks fertilized with di-ammonium phosphate at the rate of 286g/m^2 and 6g/m^2, respectively. While the remaining 6 tanks were not fertilized (control). The experiment was laid out in a Factorial Design where catfish fry assigned randomly in 18 concrete tanks. *Clarias gariepinus* fry (1 month old) with average weight of 0.5g ± 0.01g from hatchery tank were randomly assigned to treatment tanks. Two stocking densities (5fry/m^2 and 10fry/m^2) were used. Each stocking density was assigned at once and replicated three times. All 18 concrete tanks were covered with plastic nets to prevent

predators (Figure 3.7). Fertilization was repeated every week for a period of 2 months. During this period, all treatments were supplemented with the same diet formulated at 25% body weight per day.

Figure 3.7 Concrete tanks covered with plastic nets

3.3.3 Data collection

Once every week in the rearing period of 2 months, the catfish fry from each concrete tank were harvested by using seine net, placed in 10L buckets and taken to hatchery building where they were weighed using analytical balance (BOECO, Germany) and recorded. The following formulae were used to determine fry performance as outlined by Kang'ombe *et al.* (2006).

a) Weight gain (g):

Weight gain (g) = Final mean weight (g) - Initial mean weight (g) (6)

b) Increase in mean weight (%):

$$Increase\ mean\ weight(\%) = \frac{Final\ mean\ weight\ (g) - Initial\ mean\ weight\ (g)}{Initial\ mean\ weight\ (g)} x\ 100 \quad (7)$$

c) Specific growth rate (SGR):

$$Specific\ growth\ rate(\%) = \frac{\ln(Final\ mean\ weight\ (g) - Initial\ mean\ weight\ (g))}{Time\ (days)} x\ 100 \quad (8)$$

d) Survival rate:

$$Survival\ rate\ (\%) = \frac{Initial\ number\ of\ fry - Number\ of\ dead\ fry}{Initial\ number\ of\ fry} x\ 100 \quad (9)$$

3.4 Analysis of water quality parameters

In both experiments, water quality parameters were monitored. Temperature (^{o}C) and dissolved oxygen (mg L^{-1}) were monitored on daily basis at 08:00hrs and 16:00hrs and measurements were taken by using DO meter (OAKTON, 300 Series. Singapore). The pH was monitored every week (08:00hrs-16:00hrs) and measurements were taken by using pH meter (EXSTICK, USA). Ammonia and nitrite (ppm) were determined once a week by using the following methods.

3.4.1 Ammonia-nitrogen by salicylate method

A clean calorimetric tube was rinsed with water sample and then filled up to 10ml mark. The tube was inserted into the calorimeter chamber, the lid closed and scan blank selected. The tube was removed and 2.0ml of salicylate ammonia was added, closed and well mixed.

Then, 0.3g of salicylate reagent # 2 was added, mixed and waited for 1 min. At the end of 1 min period, 0.2g of Salicylate reagent #3 powder was added, closed and shaken vigorously for at least 30 sec until all solid were dissolved and then waited for another 12 min for maximum colour developed. The tube was then inserted into the calorimeter (SMART3, USA) for reading and to convert into unionized ammonia. The result obtained was multiplied with 1.2 factor.

3.4.2 Nitrite-nitrogen by diazotization method

The same procedures were repeated as above up to calorimetric tube scanning. 5ml of mixed acid reagent was then added into the 5ml tube, closed and mixed. 0.2g of colour developing reagent was later added, mixed by gently inverting for 1 min. The mixture was kept for 5 min for maximum colour development. At the end of 5 min period, the tube was inserted into the calorimeter for reading. To convert nitrite-nitrogen into nitrite, the value obtained was multiplied by 3.3 factor.

3.5 Data analysis

One way and two way analysis of variance (ANOVA) (Experiments 1 and 2, respectively) were used to compare differences between treatment means at 5% level of significance. Post-hoc analysis was done where significant differences existed between treatments means using Tukey's Test. Analyses were performed using SPSS software version 20 (SPSS Inc.).

3.6 Model

The following one way and two way linear models were used respectively:

$Y_{ij} = \mu + t_i + e_{ij}$

Y_{ij} is the observed plankton number of the j^{th} treatment within i^{th} fertilizer

μ is an overall mean

t_i is the fixed effect of the i^{th} fertilizer (i=1,3,3)

e_{ij} is the random deviation of the j^{th} treatment from the average of the fertilizer

$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + e_{ijk}$

Y_{ijk} is the observed growth parameters of the k^{th} treatment within the i^{th} fertilizer and j^{th} stocking density

μ is an overall mean

α_i is the fixed effect of i^{th} fertilizer (i=1,2,3)

β_j is the fixed effect of the j^{th} stocking density (j=1,2)

$(\alpha\beta)_{ij}$ is the fixed effect of interaction between fertilizer and stocking density

e_{ijk} is the random deviation of the k^{th} treatment from the average of fertilizer x stocking density

CHAPTER FOUR

RESULTS

4.1 Effects of fertilizers type on abundance and diversity of natural food in concrete tanks

Generally, fertilized tanks had higher abundance and diversity of phytoplankton and zooplankton compared to unfertilized tanks (Tables 4.1, 4.2, 4.3 and 4.4). Phytoplankton abundance was represented by four diverse groups, namely *chlorophyta*, *cynophyta*, *euglenophyta* and *diatomae.* Among the phytoplankton, *chlorophyta*, *cryophyte* and *diatomae* were highly significant (P<0.05) in tanks fertilized with chicken manure and di-ammonium phosphate fertilizer compared to control. *Euglenophyta* showed to be highly significant (P<0.05) when di-ammonium phosphate fertilizer was used in comparison to other treatments used in this study as shown in Table 4.1 and Appendix 1.

Table 4.1 Phytoplankton abundance (individuals/L) observed in tanks fertilized with different type of fertilizers (Mean ± SE)

Parameters (Individual/L)	**Treatment**			
	Chicken manure	**DAP fertilizer**	**No fertilizer**	**P-Value**
Chlorophyta	52750 ± 10528^{a}	71833 ± 11383^{a}	12542 ± 2648^{b}	***
Cynophyta	18833 ± 3346^{a}	25458 ± 4616^{a}	3333 ± 417^{b}	***
Euglenophyta	9917 ± 1109^{a}	14917 ± 1500^{b}	2500 ± 224^{c}	***
Diatomae	10375 ± 2376^{ab}	19833 ± 3840^{a}	1542 ± 384^{b}	***

Means with different superscript within rows indicate significant differences (Tukey's multiple range test at P<0.05); ***-P<0.001

Phytoplankton diversity was higher in tanks applied with DAP fertilizer, followed by tanks applied with chicken manure and least in tanks with no fertilizer. The diversity was indicated with the high values of Shannon-Wiener indices (H') and evenness (J') as shown in Table 4.2.

Table 4.2 Shannon-Wiener indices of phytoplankton diversity (individuals/L) in tanks applied with chicken manure DAP fertilizer and no manure application.

Phytoplankton/L	H'	H'_{max}	J'
Chicken manure	1.1300	1.3863	0.8151
DAP fertilizer	1.1797	1.3863	0.8509
No fertilizer	1.0489	1.3863	0.7566

Zooplankton was represented by three major groups, namely rotifers, copepods and cladocerans. All zooplankton groups had significantly ($p<0.05$) higher in abundance in tanks fertilized with di-ammonium phosphate fertilizer (DAP) compared to chicken manure and control as shown in Table 4.3 and Appendix 2.

Table 4.3 Zooplankton abundance (individuals/L) observed in treatment tanks (Mean ± SE).

Parameters (Individual/L)	**Treatment**			
	Chicken manure	**DAP fertilizer**	**No fertilizer**	**P-Value**
Rotifers	157± 23^{a}	1207± 388^{b}	33± 4^{a}	***
Copepods	257± 8^{a}	667± 133^{b}	40± 10^{a}	***
Cladocerans	167±14^{a}	640± 69^{b}	17± 7^{a}	***

Means with different superscript within rows indicates significances (Tukey's multiple range test at $P<0.05$); ***-$P<0.001$

Chicken manure had high values of Shannon-Wiener indices (H') and evenness (J') compared to other treatments. This suggests that zooplankton diversity was higher in tanks applied with chicken manure, followed by tanks applied with DAP fertilizer and least in tanks with no fertilizer as shown in Table 4.4.

Table 4.4 Shannon-Wiener indices of zooplankton diversity (individual/L) in tanks applied with chicken manure, DAP fertilizer and no manure application.

Zooplanktons/L	H'	H'_{max}	J'
Chicken manure	1.0728	1.0986	0.9765
DAP fertilizer	1.0526	1.0986	0.9581
No fertilizer	1.0431	1.0986	0.9494

The highest number of phytoplankton and zooplankton genera (14 and 8, respectively) were recorded in DAP fertilizer and the lowest number (10 and 4, respectively) were recorded in control as shown in Table 4.5 and 4.6.

Table 4.5 Phytoplankton diversity at genus level observed in different fertilizers type during the study.

Parameters (Individual/L)	**Treatment**		
	Chicken manure	**DAP fertilizer**	**No fertilizer**
Chlorophyta	*Actinastrum, Scenedesmus, Kirchneriella, Ankistrodesmus, Franceia, Closterium, Oscillatoria and Anabaena*	*Scenedesmus, Actinastrum, Franceia, Treubaria, Stichoccocus, Ankistrodesmus, Straurastrum, Pediastrum and Gloeotila*	*Franceia, Closterium, Oschillatoria, Anabaena, Scenedesmus and Kirchnerilla*
Cynophyta	*Oscillaria*	*Oscillatoria and Anabaena*	*Oscillatoria*
Euglenophyta	*Trachelomonas and Phacus*	*Trachelomonas*	*Phacus and Trachelomonas*
Diatomae	*Tabelaria*	*Stauroneis and Tabelaria*	*Surirella,*

Table 4.6 Zooplankton diversity at genus level observed in different treatments during the study.

Parameters (Individual/L)	**Treatment**		
	Chicken manure	**DAP fertilizer**	**No fertilizer**
Rotifers	*Lecane, Branchioni and Trochocera*	*Lecane, Branchioni and Trochocera*	*Lecane and Branchioni*
Copepods	*Claidae*	*Cyclops and Claidae*	*Claidae*
Cladocerans	*Daphnia and Moina*	*Daphnia, Alona and Moina*	*Daphnia*

Measured water quality parameters indicated that there was no significant difference (P>0.05) on temperature and dissolved oxygen in all treatments (Appendix 4), while NO_2 and NH_3 were significantly different (P<0.05) among the treatments as shown in Table 4.7 and Appendix 6. Ranges in pH values were closely related in tanks fertilized with chicken manure and DAP fertilizer compared to control tanks (Table 4.10).

Table 4.7 Water quality parameters observed in different types of fertilizer during experiment 1 (Mean ± SE).

Parameters	**Treatments**			**P-value**
	Chicken manure	**DAP fertilizer**	**No fertilizer**	
Temp (^{0}C) at am	26.5±0.1	26.4±0.1	26.4±0.1	NS
Temp (^{0}C) at pm	29.1±0.1	29.1±0.1	29.3±0.1	NS
DO (mg L $^{-1}$) at am	4.5±0.1	4.2±0.1	4.4±0.1	NS
DO (mg L $^{-1}$) at pm	6.2±0.1	6.2±0.1	6.0±0.1	NS
NO_2 (ppm)	0.1±0.1^a	0.3±0.1^b	0.03±0.1^a	***
NH_3 (ppm)	0.2±0.1^a	0.6±0.1^b	0.1±0.1^a	*

Means with different superscript within rows indicates significances (Tukey's multiple range test at P<0.05); NS-Non significant, *-P<0.05 and ***-P<0.001

4.2 Effect of fertilizer type and stocking density on growth performance of *C. gariepinus* fry

Fry performance was higher in fertilized tanks compared to unfertilized tanks. At low stocking density (5fry/m^2) fry had better growth performance compared to high stocking density (10fry/m^2) across all fertilizer types. Individual final mean weights (g), weight gain (g) and specific growth rate (%) were significantly different (P<0.05) between stocking

densities. While were not significantly different (P>0.05) between chicken manure and di-ammonium phosphate fertilizers but significant higher than the control. Initial mean weight (g) and increase mean weight (g) were not significantly different (P>0.05) across stocking densities and fertilizers type as shown in Table 4.8 and Appendix 3. In addition, average weight (g) of fry was higher in low stocking density than high stocking density across all fertilizer types as shown in Figure 4.1.

4.3 Effect of fertilizer type and stocking density on survival of *C. gariepinus* fry

Survival rate was significantly (P<0.05) higher under stocking density and the interaction but were not significantly different among fertilizer types (Appendix 3). Higher survival rate was observed in tanks fertilized with di-ammonium phosphate fertilizer and control tanks under low stocking density (Table 4.8).

Table 4.8 Growth parameters and survival observed in chicken manure, di-ammonium fertilizer and no fertilizer at different stocking densities (Mean ± SE).

Parameters	**Initial mean weight (g)**	**Individual final mean weight (g)**	**Weight gain (g)**	**Increase mean weight (g)**	**Specific growth rate (%)**	**Survival rate (%)**
Stocking densities	NS	*	**	NS	*	***
5fry/m^2	0.5±0.01	24.1±2.1[a]	6.3±0.4[a]	90.7±10.9	21.6±1.7[a]	97.8±0.9[a]
10fry/m^2	0.5±0.01	18.3±2.1[b]	4.6±0.4[b]	80.6±10.9	16.5±1.7[b]	87.2±0.9[b]
Fertilizers	NS	*	*	NS	*	NS
Chicken manure	0.5±0.01	25.6±2.5[a]	6.5±0.5[a]	90.8±13.4	22.5±2.1[a]	92.1±1.2
DAP fertilizer	0.5±0.01	20.5±2.5[a]	5.2±0.5[a]	84.3±13.4	18.7±2.1[a]	93.9±1.2
No fertilizer	0.5±0.01	17.5±2.5[b]	4.5±0.5[b]	81.7±13.4	15.8±2.1[b]	91.5±1.2
Stocking density* Fertilizer	NS	NS	NS	NS	NS	****
5fry/m^2*chicken manure	0.5±0.01	29.2±3.6	7.6±0.8	92.6±19.0	25.6±2.9	93.3±1.7[a]
5fry/m^2*DAP fertilizer	0.5±0.01	24.3±3.6	5.9±0.8	90.9±19.0	20.6±2.9	98.1±1.7[b]
5fry/m^2*no fertilizer	0.5±0.01	18.8±3.6	5.2±0.8	88.5±19.0	15.6±2.9	97.3±1.7[b]
10fry/m^2* chicken manure	0.5±0.01	21.9±3.6	5.4±0.8	89.0±19.0	19.6±2.9	90.8±1.7[a]
10fry/m^2*DAP fertilizer	0.5±0.01	16.7±3.55	4.4±0.8	77.7±19.0	16.7±2.9	87.9±1.7[a]
10fry/m^2* no fertilizer	0.5±0.01	16.3±3.6	3.9±0.8	74.9±19.0	13.1±2.9	82.9±1.7[c]

Means with different letters in the same columns are significantly different (Tukey's multiple range test at P<0.05); NS-Non significant, *-P<0.05, **-P<0.01 and ***-P<0.001.

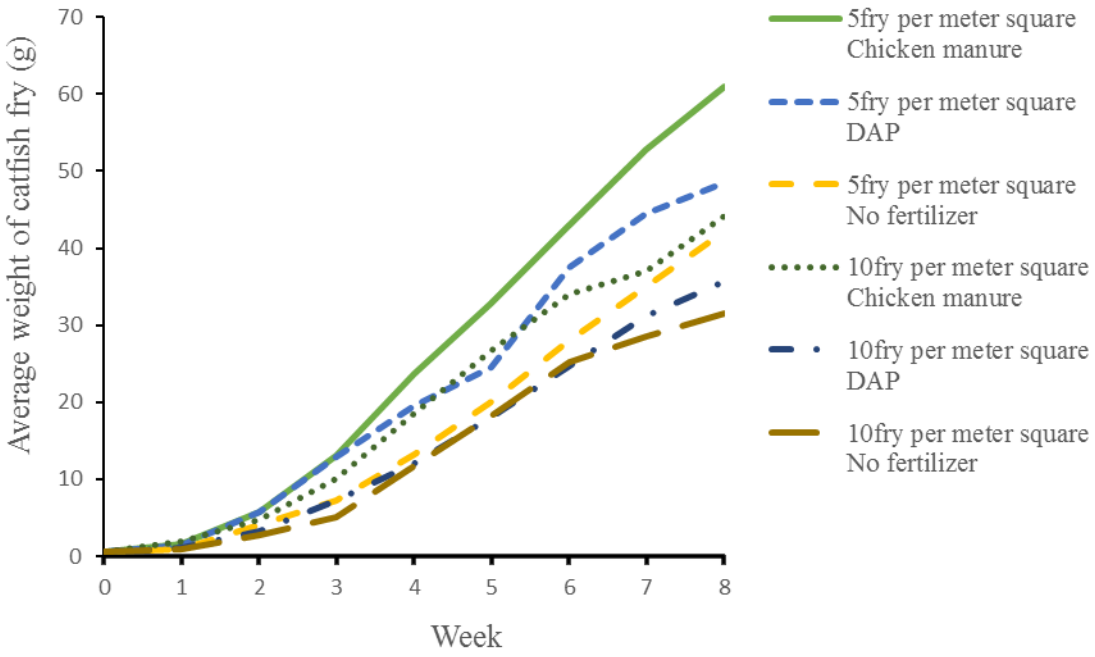

Figure 4.1 Average weight of catfish fry reared in tanks fertilized with different types of fertilizer at different stocking densities for 8 weeks.

4.4 Effect of fertilizer type and stocking density on water quality

There were no significant differences (P>0.05) in morning and afternoon temperature, dissolved oxygen (morning), and ammonia between stocking density and fertilizer type (Appendix 5 and 7). Afternoon dissolved oxygen and nitrite were significantly (P<0.05) higher among stocking densities and fertilizer types (Table 4.9). The range in pH values were similar to all stocking densities under chicken manure and DAP fertilizer but different ranges were observed in the control experiments (Table 4.10).

Table 4.9 Water quality parameters observed in fertilizer type and different stocking densities (Mean ± SE).

Parameters	**Temp (^{0}C) at am**	**Temp (^{0}C) at pm**	**DO (mgL^{-1}) at am**	**DO (mgL^{-1}) at pm**	**NO_2 (ppm)**	**NH_3 (ppm)**
Stocking densities	NS	NS	NS	NS	NS	NS
5fry/m^2	26.8±0.1	28.6±0.1	4.2±0.1	6.2±0.1	0.2±0.1	0.5±0.2
10fry/m^2	26.8±0.1	28.5±0.1	4.3±0.1	6.3±0.1	0.3±0.1	0.5±0.2
Fertilizers	NS	NS	NS	*	*	NS
Chicken manure	26.7±0.1	28.6±0.1	4.3±0.8	6.1±0.1^a	0.2±0.1^{ab}	0.3±0.2
DAP fertilizer	26.8±0.1	28.5±0.1	4.3±0.8	6.5±0.1^b	0.4±0.1^a	0.8±0.2
No fertilizer	26.8±0.1	28.6±0.1	4.3±0.8	6.0±0.1^a	0.04±0.1^b	0.3±0.2
Stocking density*Fertilizer	NS	NS	NS	*	*	NS
5fry/m^2*chicken manure	26.7±0.2	28.7±0.2	4.2±0.1	6.1±0.2^a	0.1±0.1^a	0.4±0.3
10fry/m^2*chicken manure	26.8±0.2	28.5±0.2	4.3±0.1	6.1±0.2^a	0.3±0.1^a	0.3±0.3
5fry/m^2*DAP fertilizer	26.8±0.2	28.5±0.2	4.2±0.1	6.3±0.2^b	0.3±0.1^b	0.9±0.3
10fry/m^2*DAP fertilizer	26.8±0.2	28.5±0.2	4.3±0.1	6.7±0.2^b	0.5±0.1^b	0.9±0.3
5fry/m^2*no fertilizer	26.8±0.2	28.7±0.2	4.2±0.1	6.0±0.2^a	0.02±0.1^c	0.3±0.3
10fry/m^2*no fertilizer	26.9±0.2	28.6±0.2	4.3±0.1	6.0±0.2^a	0.07±0.1^c	0.3±0.3

Means with different letters in the same columns are significantly different (Tukey's multiple range test at P<0.05); NS-Non significant, *-P<0.05and ***-P<0.001

Table 4.10 The pH values observed during the experiments.

Parameters	**Experiment 1**	**Experiment 2**	
		5fry/m^2	**10fry/m^2**
Chicken manure	6.0-9.0	6.0-8.0	6.0-8.0
DAP fertilizer	6.0-8.0	5.0-7.0	5.0-7.0
Control	7.0-9.0	6.0-8.0	7.0-8.0

CHAPTER FIVE

DISCUSSION

5.1 Effects of fertilizer type on abundance and diversity of natural food in concrete tanks

The results indicated that abundance of plankton was higher in tanks fertilized with di-ammonium phosphate (DAP) than in chicken manure and the control (Tables 4.1 and 4.3). Similar results have been reported by Kumar *et al.* (2014) where they reported higher plankton composition in DAP fertilizer treatments. Padmavathi (2009) reported that phytoplankton (*Cyanophyceae, Chlorophyceae, Bacillariophyceae, Eugleninae* and *Dinophyceae*) were numerically abundant when 60 kg/ha of DAP and urea fertilizer applied in ponds. The significantly higher abundance and diversity of phytoplankton in DAP fertilizer could likely be due to easily available of nutrients to the water. Inorganic fertilizers have much higher concentrations of nutrients such as nitrogenous compounds than manures (Boyd and Massaut, 1999). Gamal (2008) reported significantly high chlorophyll *a* (total algal biomass), lutein (green algal biomass), and fucoxanthin (diatom biomass) in ponds fertilized with inorganic fertilizer than organic fertilizers and control ponds. Also, Soderberg (2012) reported that inorganic fertilizers have a drastic and immediate effect on primary production from phytoplankton. Most of the phytoplankton genera were observed in all treatments including the control but differences were identified in DAP fertilized tanks where genera *Treubaria, Stichoccocus, Straurastrum, Pediastrum* and *Gloeotila* were identified (Table 4.5). Similarly, Ahmed *et al.* (1997) found some

common phytoplankton genera in all treatments which were different either in quality or in quantity.

Rotifer, copepods and cladocerans showed a higher abundance as phytoplankton trend. This might be due to feeding effects of zooplankton on phytoplankton earlier observed by Guangjun (2013). Similarly, Ghosh *et al.* (1974) and Kumar *et al.* (2014) reported rotifers to be the common and dominant zooplankton group in fertilized ponds. In addition, Mischke and Zimba (2004) reported that copepod nauplii and cladocerans were significantly higher in ponds fertilized with inorganic fertilizer (Urea +Super phosphate) than in control ponds and organically fertilized ponds. Higher zooplankton diversity were recorded in chicken manure followed by DAP fertilizer and the least was recorded in the control (Table 4.4). This might be due to phytoplankton particle size and cell abundance which influences zooplankton communities (Bell, 2002). The larger size of phytoplankton cells were not consumed by smaller zooplankton hence high abundance of phytoplankton communities in DAP fertilized tanks than zooplankton. Soderberg (2012) reported that organic fertilizers require bacteria and other microbes for decomposition, and thus offer a wider diversity of fish ponds, particularly zooplankton. Also, chicken manure have been reported to provide a substrate for zooplankton production (FAO, 2003) which enhance high diversity.

In addition, zooplankton genus *Cyclops* was identified in DAP fertilized tanks only (Table 4.6). This could be due to higher amount of phosphate also reported by Hossain *et al.* (2006) and Kumar *et al.* (2014). *Cyclops* are phosphate limited organisms which increased

as phosphate increased (Law *et al.*, 2005). Similarly, Thingstad *et al.* (2005) reported increased in number of zooplankton due to increased phosphate. Also, genus *Surirella* (diatom) was identified in control treatment because it does not require high nutrient loading and dissolved oxygen (<5mg/l) (Kumar *et al.*, 2008). Therefore, amount of available phosphate in DAP fertilizer was higher indicating better improvement in zooplankton abundance as reported earlier by Schroeder (1980), Kumar *et al.* (2014) and Hossain *et al.* (2006). Planktons are therefore important on survival of catfish fry (Mischke *et al.*, 2009).

Among water quality parameters, temperature and dissolved oxygen (Table 4.9) were within the optimal range 16^0C to 32^0C (Hecht, 2013) and 3.5mg/l to 9.2mg/l (Abdelhamid *et al.*, 2010) respectively. In the present study, pH ranges (Table 4.10) were similar to those reported by Kumar *et al.* (2014). Concentration of NO_2 and NH_3 were high in the chicken manure and DAP fertilizer treatments as those reported by Boyd (1997). The reason might be due to temperature and pH dependence of ammonia and ammonium ion from the fertilizer. Ammonium fertilizer such as DAP increases total ammonia nitrogen concentrations in water while manures have low nitrogen content and tend to decompose slowly and incompletely (Boyd, 1997).

5.2 Effect of fertilizer type and stocking density on growth performance of *Clarias gariepinus* fry

The overall results showed that catfish fry had higher performance in fertilized tanks than unfertilized. The similar results have been reported by Muendo *et al.* (2006). Individual

mean weight and weight gain showed to be high at low stocking density (5fry/m^2) compared to high stocking density (10fry/m^2) across all fertilizer type (Table 4.8). This indicated that the performance of catfish fry was influenced by stocking density and fertilizer type. Jamabo and Keremah (2009) reported significantly higher growth rate at a stocking density of 5fry compared to 10fry and 15fryper 55m^3 tank. Among fertilizer applied tanks, chicken manure treated tanks had better catfish fry performance than the rest (Table 4.4). This could be due to high zooplankton diversity observed in tanks applied with chicken manure than DAP and control treated tanks. Hossain *et al.* (2006) suggested the capacity of phosphorus released from poultry manure might be more efficient than other organic fertilizers and inorganic fertilizers.

Specific growth rate of African catfish (*Clarias gariepinus)* fry were observed to be better in tanks fertilized with chicken manure and DAP fertilizers compared to unfertilized tanks under both stocking densities (Table 4.8). Hossain *et al.* (1998), Jamabo and Keremah (2009) reported that growth of catfish fry was strongly influenced by stocking density. This might be due to high natural food organisms in fertilized tanks at low stocking densities compared to high stocking densities (Bahnasawy, 2009). These results indicated that the addition of fertilizers to the fish ponds/tanks increase the level of nitrogenous compounds and minerals which are considered as a good source for phytoplankton growth. Lin *et al.* (1998), Dang and Dalsgaard, (2012) and Saad *et al.* (2014) reported that addition of manures and fertilizers to the fish ponds improve the level of phytoplankton that is responsible for growth fish. This was supported by Silva *et al.* (1995) who reported that phytoplankton and zooplankton are a rich source of protein often containing 40-60%

protein on a dry matter basis and is sufficient to support excellent fish growth. Saad *et al.* (2014) found that, nitrogenous and phosphorus compound fertilizers improve the growth of phytoplankton and zooplankton that in turn improve growth performance of the fish.

5.3 Effect of fertilizer type and stocking density on survival of *Clarias gariepinus* fry

Results in the present study showed that survival rate can be influenced by stocking density and its interaction with fertilizer type. Similarly, Haylor (1991) found that survival rates in African catfish fry were directly related to stocking density. The study recorded high survival rate under low stocking density with DAP and control treatments interaction compared to chicken manure interaction. While the lowest survival rate recorded under high stocking density in control treatment interaction (Table 4.8). This might be due to low dissolved oxygen and NO_2 concentration recorded (Table 4.9). Similar results were reported by Sophin and Preston (2001) in tilapia where poorer survival rates were observed in the ponds fertilized with chicken manure as compared to the inorganic fertilizers (urea and DAP) and unfertilized ponds.

In addition, aggressive behavior of catfish in taking in food and cannibalism in control tanks might influence low survival rate. Similar results have been reported by Gamal (2008) who found that high stocking density (5 fingerlings/m^2) using chemical fertilizer (urea + monophosphate) and chicken manure had the highest survival rate (96.48%) of catfish showing increased predatory behavior of catfish in higher stocking densities. In addition, Baskerville-Bridges and Kling (2000) reported greater risk of mortality at high stocking densities as a result of deteriorating water quality.

5.4 Effect of fertilizer type and stocking density on water quality

The values of water quality parameters such as temperature and dissolved oxygen during the experiment (Table 4.9) were within the acceptable ranges for growth of catfish fry 26^0C to 28^0C (Hecht, 2013) and 4mg/L to 6mg/L (Francis-Floyd, 2014), respectively. This might be due to high abundance of planktons (Gamal, 2008). The concentration of NO_2 was high at 10fry/m^2 under DAP fertilizer compared to other treatments. This might be due to ammonia utilization by phytoplankton (Boyd, 1998) or oxidation of ammonia nitrite especially in high dissolved oxygen level conditions (Boyd, 2000).

In addition, pH ranges (Table 4.10) were optimal as recorded by Tucker (1991). However, there were similarly higher pH values under both stocking densities in chicken manure and DAP fertilized tanks compared to control. Boyd (1990) reported that the application of ammonium and urea-based fertilizers can cause acidification of pond water because of nitrification, which produces two hydrogen ions from each ammonium ion. Similarly, Saad *et al.* (2014) reported high pH values in treatments that received inorganic and organic fertilizers compared to group given feed only. Therefore, results in this study may suggest that addition of fertilizers to the fish tank increased some of the physicochemical properties of the water.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

The study demonstrated both DAP and chicken manure fertilized tanks had statistically highest phytoplankton abundance compared to control tanks. Zooplankton abundance were statistically higher in tanks fertilized with DAP fertilizer followed by chicken manure and control tanks. While chicken manure applied tanks had high zooplankton diversity compared to other treatments. There were significant differences on abundance and diversity of different species of live food produced in chicken manure and di-ammonium phosphate treated tanks.

Growth performance parameters (individual final mean weigh, weight gain and specific growth rate) did not differ significantly between chicken manure and DAP fertilizer treatments across stocking densities. There was no significant difference in growth performance of the fry produced in chicken manure and di-ammonium phosphate at different stocking densities.

The study also proved that survival rate of catfish fry did not depend on fertilizer type. However, it was dependent on stocking density where high growth was recorded at low stocking density (5fry/m^2) across all fertilizer types. There were no significant differences in survival rates of the fry produced in chicken manure and di-ammonium phosphate at different stocking density.

In addition, the study also revealed that some water quality parameters such as dissolve oxygen during the afternoon and nitrite depended on fertilizer types. While some parameters such as temperature, dissolved oxygen during the morning and ammonia did not. However, there were no significant differences among the water quality parameters in chicken manure and di-ammonium phosphate at different stocking densities.

6.2 Recommendations

Based on the results of the study, it is recommended that catfish fry should be raised in chicken manure fertilized tanks at low stocking density (5fry/m^2) for better growth and survival in aquaculture practices. However, the following areas could be focused to improve this type of research and further research can be conducted as follows:

Determine the optimal stocking density of catfish fry raised under different size/area of concrete tanks to achieve better growth and survival rate by using DAP fertilizer.

Determine the effects of different fertilization rates of chicken manure and DAP fertilizer on fry/fingerling reared in concrete tanks.

REFERENCES

Abdelhamid, A.M., Radwan, I.A., Mehrim, A.I. and Abdelhamid, A.F.B. (2010). Improving the Survival rate of African Catfish, *Clarias gariepinus. Journal of Animal and Poultry Production*, 1 (9): 409 – 414.

Adewumi, A.A. (2015). Growth performance and survival of *Clarias gariepinus* hatchlings fed different starter diets. *European Journal of Experimental Biology,* 5 (3): 1-5. Available at http://pelagiaresearchlibrary.com/european-journal-of-experimental-biology/vol5-iss3/EJEB-2015-5-3-1-5.pdf [Accessed on 10th April, 2015].

Agadjihouede, H., Chikou, A., Bonou, C.A. and Laleye, P.A. (2012). Survival and Growth of *Clarias gariepinus* and *Heterobranchus longifilis* Larvae fed with fresh water zooplankton. *Journal of Agricultural Science and Technology,* 2: 192-197.

Ahmed, G.U., Islam, M.F., Khan, M.N.A., Haque, M.M. and Kibria, M.G. (1997). Culture feasibility of African catfish (*Clarias gariepinus* Lin.) fry in glass tank and synthetic hapa system using supplemental diets. *Bangladesh Journal of Fish Research,* 1 (2): 59-66. Available at http://aquaticcommons.org/16374/1/BJFR1.2_059.pdf [Accessed on 20th May, 2015].

Akand, M.A. (1986). Effect of organic fertilizers on water quality and fish production in carp ponds. *Bangladesh Journal of Fisheries Research*, 9: 47-53.

Amisah, S., Adjei-Boateng, D. and Afianu, D.D. (2008). Effects of bamboo substrate and supplementary feed on growth and production of the African catfish, (*Clarias*

gariepinus). *Journal for Applied Science and Environmental Management,* 12 (2): 25-28.

Association of Official Analytical Chemists, (2002). *Official Methods of Analysis,* Association of Official Analytical Chemists (AOAC). JRC-European Commission, Washington D.C, USA. Available at https://law.resource.org/pub/us/cfr/ibr/002/aoac.methods.1.2002.pdf [Accessed on 24th July, 2014].

Awaiss, A. and Kestemont, P. (1998). Feeding sequences (rotifer and dry diet), survival, growth and biochemical composition of African catfish, (*Clarias gariepinus, Burchell Pisces*: *Clariidae*), larvae. *Aquaculture Research,* 29: 371-741.

Ayele, T.A. (2015). *Growth performance and survival rate of African catfish larvae Clarias gariepinus (Burchell 1822) fed on different types of live and formulated feeds*. MSc Thesis, University of Natural Resources and Life Science (BOKU), Vienna, Austria. Available at https://zidapps.boku.ac.at/abstracts/download.php?dataset_id=12898&property_id=107 [Accessed on 22nd August, 2014].

Bahnasawy, M.H. (2009). Effect of dietary protein levels on growth performance and body composition of monosex Nile tilapia, *Oreochromis niloticus* L. reared in fertilized tanks. *Pakistan Journal of Nutrition,* 8 (5): 674-678.

Baskerville-Bridges, B. and Kling, L.J. (2000). Larval culture of Atlantic cod (*Gadus morhua)* at high stocking densities. *Aquaculture,* 181: 61-69.

Bell, T. (2002). The ecological consequences of unpalatable prey: Phytoplankton response to nutrient and predator additions. *OIKOS,* 99 (1): 59-68. Available at https://www.zoology.ubc.ca/~schluter/reprints/bell%202002%20oikos%20ecological%20consequences%20of%20unpalatable%20prey.pdf [Accessed on 10th February, 2015].

Bellinger, E.D. (1992). *A key to common algae*. The Institution of Water and Environmental Management, London, UK. Available at http://onlinelibrary.wiley.com/doi/10.1002/9780470689554.refs/pdf [Accessed on 04th February, 2014].

Boyd, C.E. (1990). *Water quality in ponds for aqua culture.* Alabama Agriculture. Experiment Station Auburn University, Alabama, USA. Available at https://books.google.mw/books?id=oVY5AQAAIAAJ&source=gbs_navlinks_s&redir_esc=y [Accessed on 20th February, 2015].

Boyd, C.E. (1997). Practical aspects of chemistry in pond aquaculture. *The Progressive Fish Culturist,* 59: 85-93.

Boyd, C.E. (2000). *Water Quality: An Introduction.* Kluwer Academic, Bosto, Massachusetts, USA. Available at https://books.google.co.in/books/about/Water_Quality.html?id=aKgK83ANjk8C [Accessed on 20th December, 2014].

Boyd, C.E. and Massaut, L. (1999). Risks associated with the use of chemicals in pond aquaculture. *Aquacultural Engineering,* 20: 113-132.

Boyd, C.E. and Tucker, C.S. (1998). *Pond Aquaculture Water Quality Management.* Kluwer Academic. Boston, USA. Available at http://www.springer.com/cn/book/9780412071812# [Accessed on 10th September, 2014].

Brazil, B.L. and Wolters, W.R. (2002). Hatching success and fingerling growth of channel catfish cultured in ozonated hatchery water. *North American Journal of Aquaculture*, 64: 144-149.

Brummett, R.E., Yong-Sulem, S., Tabi, T.E. and Tchoumboué, J. (2007). Towards the maximum profitability of smallholder catfish nurseries: Predator defense and feeding-adapted stocking of *Clarias gariepinus*. *Aquaculture,* 271: 371-376.

Bruton, M.N. (1979). The food and feeding behaviour of *Clarias gariepinus* (*Pisces: Clariidae*) in Lake Sibaya, South Africa, with emphasis on its role as a predator of cichlids. *The Transactions of the Zoological Society of London*, 35 (1): 47-114.

Cahu, C.L. and Zambonino-Infante, J.L. (2001). Substitution of live food by formulated diets in marine fish larvae. *Aquaculture*, 200 (2): 161-180.

Chenyambuga, S.W., Madalla, N. and Mnembuka, B. (2011). *Management and value chain of Nile tilapia cultured in ponds of small-scale farmers in Morogoro region, Tanzania.* Department of Animal Science, Sokoine University of Agriculture. Morogoro, Tanzania. Available at https://ir.library.oregonstate.edu/xmlui/bitstream/handle/1957/37878/Chenyamb

uga-IFFET%20paper%20July%20201.pdf?sequence=3 [Accessed on 19th July, 2015].

Coulibaly, A., Ouattara, I.N., Koné, T., N'Douba, V., Snoeks, J., Gooré Bi, G. and Kouamelan, E.P. (2007). First results of floating cage culture of the African catfish *Heterobranchus longifilis* Valenciennes, 1840: Effect of stocking density on survival and growth rates. *Aquaculture,* 263: 61-67

Dang, S.T. and Dalsgaard, A. (2012). *Escherichia coli* contamination of fish raised in integrated pig-fish aquaculture systems in Vietnam. *Journal of Food Protection,* 75 (7): 1317-1319.

Dasuki, A., Auta, J. and Oniye, S.J. (2013). Effect of stocking density on production of *Clarias gariepinus* (tuegels) in floating bamboo cages at Kubanni reservoir, Zaria, Nigeria. *Bayero Journal of Pure and Applied Sciences,* 6 (1): 112-117.

De Graaf, G. and Janssen, H. (1996). Artificial reproduction and pond rearing of the African catfish *Clarias gariepinus* in sub-Saharan Africa. Fisheries Technical Paper. No. 362. Rome. Available at http://www.nefisco.org/downloads/Clarias.PDF [Accessed on 20th January, 2014].

De Graaf, G.J., Galemoni, F. and Banzoussi, B. (1995). The artificial reproduction and fingerling production of the African catfish *Clarias gariepinus* (Burchell 1822) in protected and unprotected ponds. *Aquaculture Research,* 26: 233-242.

De Silva, S.S. and Anderson T.A. (1995). *Fish nutrition in aquaculture*. New Delhi, India: Chapman and Hall.

Debenest, T., Silvestre, S., Coste, M., Delmas, F. and Pinelli, E. (2009). A new cell primo-culture method for freshwater benthic diatom communities. *Journal of Applied Psychology*, 21 (1) 65-73.

Diana, J.S. (2012). Some Principles of Pond Fertilization for Nile Tilapia Using Organic and Inorganic Inputs In: Mischke. C. C (Ed.), *Aquaculture Pond Fertilization: Impacts of Nutrient Input on Production*, John Wiley & Sons, USA. (pp. 163-177). Available at http://onlinelibrary.wiley.com/doi/10.1002/9781118329443.ch12/pdf [Accessed on 17th July, 2015].

Ewa-Oboho, I. and Enyenihi, U.K. (1999). Aquaculture implications of growth and variation in the African catfish: *Heterobranchus longifilis* (Valenciennes, 1840) reared under controlled conditions. *Journal of Applied Ichthyology*, 15: 111-115.

Food and Agricultural Organization of the United Nations, (2000). *Fisheries statistics: aquaculture production 1998.* FAO Yearbook. Food and Agriculture Organization of the United Nations, Rome. Available at http://www. fishbase.org/ manual/ fishbasefao_statistics00002679.htm [Accessed on 15th June, 2015].

Food and Agricultural Organization of the United Nations, (2003). *Seed production. An overview*. FAO Fisheries and Aquaculture Department, Rome. Available at http://www.fao.org/docrep/016/i2727e/i2727e.pdf [Accessed on 30th February, 2014].

Food and Agricultural Organization of the United Nations, (2007). *The state of word fisheries and aquaculture 2006*. FAO Fisheries and Aquaculture Department, Rome. Available at http://www.fao.org/docrep/009/a0699e/a0699e00.HTM [Accessed on 05th August, 2014].

Food and Agricultural Organization of the United Nations, (2010). *Cultured Aquatic Species (Clarias gariepinus). Information Programme.* FAO Fisheries and Aquaculture Department, Rome. Available at http://www.fao.org/figis/pdf [Accessed on 07th August, 2014].

Food and Agricultural Organization of the United Nations, (2012). *The state of word fisheries and aquaculture.* FAO Fisheries and Aquaculture Department, Rome. Available at http://www.fao.org/docrep/016/i2727e/i2727e00.htm [Accessed on 10th August, 2014].

Food and Agricultural Organization of the United Nations, (2014). *The State of World Fisheries and Aquaculture 2014.* FAO Fisheries and Aquaculture Department, Rome. Available at http://www.fao.org/3/a-i3720e.pdf [Accessed 16th August, 2014].

Francis-Floyd, R. (2014). Dissolved Oxygen for Fish Production. Fisheries and Aquatic Sciences Department, *UF/IFAS Extension*, University of Florida, Gainesville. (pp. 1-3). Available at https://edis.ifas.ufl.edu/pdffiles/FA/FA00200.pdf [Accessed on 10th April, 2014].

Gamal, O.E., Ibrahim, N.A. and Abou-Zead. M.Y. (2008). Influence of fertilizers' types and stocking density on water quality and growth performance of Nile tilapia-

African catfish in polyculture system. *8th International Symposium on Tilapia in Aquaculture*, World Fish Center, Egypt. (pp.151-171). Available at http://cals.arizona.edu/azaqua/ista/ISTA8/NabilIbrahem.pdf [Accessed on 02nd April, 2014].

Ghosh, A.L., Rao, H. and Banerjee, S.C. (1974). Studies on hydrobiological conditions of seawage fed pond with a note of their role in fish culture. *Journal of Inland Fisheries Society of India,* 6: 51-61.

Govoni, J.J., Boehlert, G.W. and Watanabe, Y. (1986). The physiology of digestion in fish larvae. *Environmental Biology of Fish,* 16: 59-77. Available at https://swfsc.noaa.gov/publications/CR/1986/8628.PDF [Accessed on 15th June, 2015].

Grag, S.K. and Bhatnagar, A. (2000). Effect of fertilization frequency on pond productivity and fish biomass in still water ponds stocked with *Cirrhinus mrigala*. *Aquaculture Research,* 31: 409-414.

Greenberg, A.E., Clesceri, L.S. and Eaton, A.D. (1992). *Standard Methods for the Examination of Water and Wastewater* (18th ed). Baltimore, Maryland: American Public Health Association.

Guangjun, L. (2013). Structure and diversity of zooplankton communities in four reservoirs with varying nutrient compositions in the Yangtze River Basin, China. *International Conference on Remote Sensing, Environment and Transportation Engineering,* Southwest University, Rongchong, China. (pp. 566-572).

Available at http://www.atlantis-press.com/php/download_paper.php?id=8252 [Accessed on 20th June, 2015].

Haylor, G.S. (1991). Controlled hatchery production of *Clarias gariepinus* (Burchell, 1822): growth and survival of fry at high stocking density. *Aquaculture and Fisheries Management*, 22: 405-422.

Haylor, G.S. (1992). Controlled hatchery production of *Clarias gariepinus* (Burchell, 1822): growth and survival of fry at higher stocking density. *Aquaculture and Fisheries Management*, 22: 405-422.

Hecht, T. (2013). A review of on-farm feed management practices for North African catfish (*Clarias gariepinus*) in sub-Saharan Africa. In: Hasan, M.R. and New, M.B. (Eds.). *On-farm feeding and feed management in aquaculture*. FAO Fisheries and Aquaculture Technical Paper. No. 583. Rome. (pp. 463-479). Available at ftp://ftp.fao.org/fi/Cdrom/T583/root/17.pdf [Accessed on 14th June, 2015].

Hecht, T. and Appelbaum, S. (1987). Notes on the growth of Israeli sharp tooth catfish *Clarias gariepinus*. During the primary nursing phase. *Aquaculture*, 63: 195–204.

Hecht, T. and Appelbaum, S. (1988). Observation of inter-specific aggression and coeval sibling cannibalism by larval and juvenile *Clarias gariepinus Claridae: Pisces.* under controlled conditions. *Journal of Zoological Society of London*, 214: 21–44.

Hecht, T. and Uys, W. (1997). Effect of density on the feeding and aggressive behaviour in juvenile African catfish, *Clarias gariepinus*. *South African Journal of Science*, 93: 537-541.

Hengsawat, K., Ward, F.J., Jaruratjamorn, P. (1997). The effect of stocking density on yield, growth and mortality of African catfish (*Clarias gariepinus*, Burchell 1822) cultured in cages. *Aquaculture,* 152: 67-76.

Hossain, K.Z., Miah, M.J.U. and Ali, M.H. (2008). Effect of inorganic fertilizer in the growth and production performance of exotic Carps in polyculture systems. *Progressive Agricultural*, 19 (2): 129-137. Available at http://www.banglajol.info/index.php/PA/article/download/16953/11906 [Accessed on 18th June, 2015].

Hossain, M.A.R., Beveridge, M.C.M. (1998). The effects of density, light and shelter on the growth and survival of African catfish (*Clarias gariepinus* Burchell, 1822) fingerlings. *Aquaculture,* 160: 251-258.

Hossain, M.Y., Begum, M., Ahmed, Z.F., Hoque, M.A., Karim, M.A. and Wahab, M.A. (2006). A study on the effects of Iso-Phosphorus Fertilizers on plankton production in fish ponds. *South Pacific Studies,* 26 (2): 101-110.

Jamabo, N.A. and Keremah, R.I. (2009). Effects of stocking density on growth and survival of fingerlings *Clarias gariepinus* (Burchell, 1822). *Journal of Fisheries International*, 4 (4): 55-57. Available at https://www.was.org/documents/MeetingPresentations/WA2009/WA2009_0623.pdf [Accessed on 14th June, 2015].

Kaiser, H., Weyl, O. and Hecht, T. (1995a). Observations on agonistic behavior of *Clarias gariepinus* larvae and juveniles under different densities and feeding frequencies in a controlled environment. *Journal of Applied Ichthyology,* 11: 25-36.

Kaiser, H., Weyl, O. and Hecht, T. (1995b). The effect of stocking density on growth, survival and agonistic behavior of African catfish. *Aquaculture International,* 3: 217-225.

Kang'ombe, J., Brown, J.A. and Halfyard, L.C. (2006). Effect of using different types of organic animal manure on plankton abundance, and on growth and survival of *Tilapia rendalli* (Boulenger) in ponds. *Aquaculture Research,* 37: 1360-1371.

Krebs, C.J. (1989). *Ecological Methods for Field and Laboratory Investigations*. New York, USA: Harper Collins.

Kumar, A., Sharma, L.L. and Aery, N.C. (2008). Physicochemical Characteristics and Diatom as Indicators of Trophic Status of Kishore Sagar, Rajasthan. In: Sengupta, M. and Dalwani, R. (Eds.) *Proceedings of Taal 2007: The 12th World Lake Conference*, India. (pp. 1804-1809). Available at http://www.moef.nic.in/sites/default/files/nlcp/Indian%20Case%20Studies/Q-4.pdf [Accessed on 17th June, 2014].

Kumar, S.D., Kumar, S.P., Kumar, V.U. and Anbuganapathi, G. (2014). Efficacy of Bio-fertilizer Enriched Flower Waste Vermicompost on Productionand Growth of Primary Producers and Freshwater Aquarium Fishes. *Global Veterinaria*, 13

(2): 215-220. Available at http://www.idosi.org/gv/gv13 (2)14/12.pdf [Accessed on 15th July, 2015].

Law, C.S., Abraham, E.R., Woodward, E.M.S., Liddicoat, M.I., Fileman, T.W., Thingstard, T.F., Kitidis, V. and Zohary, T. (2005). The fate of phosphate in an in situ Lagrangian addition experiment in the Eastern Mediterranean. *Deep-Sea Research II: Topical Studies in Oceanography*, 52: 2911-2927. Available at http://www.sciencedirect.com/science/article/pii/S0967064505001980 [Accessed on 11th June, 2015].

Lin, C.K., Teichert-Coddington, D.R., Green, B.W. and Veverica, K.L. (1997). Fertilization regimes. In: Egna, H. and Boyd, C.E. (Eds.) *Dynamics of Pond Aquaculture*. Boca Raton, Florida: CRC Press.

Lin, C.K., Yang, Y., Shivappa, R.B. and Kabir, C.M.A. (1998). Optimization of Nitrogen Fertilization Rate in Freshwater Tilapia Production Ponds. *PD/A CRSP Sixteenth Annual Technical Report*, Oregon State University, USA. (pp. 49-56). Available at http://www .pdacrsp.oregonstate.edu/pubs/technical/16tch/TOC.pdf [Accessed on 11th October, 2014].

Lubzen, E., Zmora, O. and Barr, Y. (2001). Biotechnology and aquaculture of rotifers. *In Rotifera IX Developments in Hydrology*, 153: 337-353.

Madalla, N. (2008). *Novel Feed Ingredients for Nile Tilapia* (*Oreochromis niloticus* L.). PhD Thesis. Institute of Aquaculture University of Stirling, Scotland, UK.

Available at https://dspace.stir.ac.uk/bitstream/1893/795/1/Thesis27012008.pdf [Accessed on 24th October, 2014].

Ministry of Livestock and Fisheries Development, (2010). *Investment opportunities in the fisheries industry. Fishery sector Development Program*, Dar es Salaam. Available at http://www.tanzania.go.tz/egov_uploads/documents/FSDP_sw.pdf [Accessed on 24th December, 2014].

Ministry of Livestock and Fisheries Development, (2011). *Investment opportunities in the fisheries industry. Fishery sector Development Program*, Dar es Salaam. Available at http://www.tanzania.go.tz/egov_uploads/documents/development_strategy-Livestock_sw.pdf [Accessed on 24th February, 2014].

Mischke, C.C. and Zimba, P.V. (2004). Plankton community responses in earthen channel catfish nursery ponds under various fertilization regimes. *Aquaculture*, 233: 219-235.

Mischke, C.C., Wise, D.J. and Byars, T.S. (2009). Evaluation of Zooplankton in Hatchery Diets for Channel Catfish Fry. *North American Journal of Aquaculture*, 71: 312-314.

Mohseni, M., Pourkazemi, M., Hassani, S.H., Okorie, O.E., Min, T.S. and Bai, S.C. (2012). Effects of different three live foods on growth performance and survival rates in Beluga *(Huso huso)* larvae. *Iranian Journal of Fisheries Sciences,* 11 (1): 118-131. Available at http://www.jifro.ir/files/site1/user_files_eb12be/admin-A-10-1-142-cffc0bf.pdf [Accessed on 24th August, 2014].

Muendo, P.N., Milstein, A.A., Dam, N., Gamal E.L., Stoorvogel, J.J. and Verdegem, C.J. (2006). Exploring the trophic structure in organically fertilized and feed driven tilapia culture environments using multivariate analyses. *Aquaculture Research*, 37: 151-163.

Murnyak, D. (2010). *Basics of Raising Tilapia and Implementing Aquaculture Projects:* A Farmer's Guide to Tilapia Culture. Heifer Project International, Arkansas, USA. (pp. 1-25). Available at https://c.ymcdn.com/sites/members.echocommunity.org/resource/collection/E66CDFDB-0A0D-4DDE-8AB1-74D9D8C3EDD4/Fish_Farming.pdf [Accessed on 24th February, 2015].

Musiba, M.J., Ngupula G.W., Kashindye, B.B., Elison, M., Shoko1, A.P., Ndikumana, J., Zziwa, E. and Katunzi, E.F.B. (2014). Performance of locally formulated feeds for rearing of African catfish in Tanzania. *African Crop Science Journal*, 22 (4): 979-986. Available at http://www.bioline.org.br/pdf?cs14050 [Accessed on 14th February, 2015].

Nwipie. G.N., Erondu, E.S. and Zabbey, N. (2015). Influence of Stocking Density on Growth and Survival of Post Fry of the African Mud Catfish, *Clarias gariepinus*. *Fish and Aquaculture,* 6 (1): 2-4.

Obirikorang, K.A., Madkour, H.A. and Adjei-Boateng, D. (2014). A study of Intra-cohort Cannibalism in Juveniles of the African Catfish, (*Clarias gariepinus*) under Controlled Conditions. *International Journal of Science and Technology*, 3 (1): 23-26. Available at

https://www.researchgate.net/profile/Kwasi_Obirikorang/publication/267514992 [Accessed on 14th December, 2014]

Okechi, K.J. (2004). Profitability assessment: *A case study of African catfish (Clarias gariepinus) farming in the Lake Victoria basin.* Kenya Marine and Fisheries Research Institute (KMFRI). Kisumu Research Centre, Kenya. Available at www.unuftp.is/static/fellows/document/okechiprf04.pdf [Accessed on 27th February, 2015].

Olubunmi, A., Olateju, A., Latifat, M. and Oluniyi, A. (2009). Production, Growth and Effect of Varying Stocking Density of *Clario branchus* Fry. *Journal of Fisheries International,* 4 (4): 73-78. Available at http://docsdrive.com/pdfs/medwelljournals/jfish/2009/73-78.pdf [Accessed on 25th March, 2014].

Olurin, K.B., Iwuchukwu, P.O. and Oladapo, O. (2012). Larval rearing of African catfish, *Clarias gariepinus* fed decapsulated *Artemia*, wild copepods or commercial starter diet. *African Journal of Food Science and Technology*, 3 (8): 182-185. Available at http://www.interesjournals.org/full-articles/larval-rearing-of-african-catfish-clarias-gariepinus-fed-decapsulated-artemia-wild-copepods-or-commercial-starter-diet.pdf?view=inline [Accessed on 04th February, 2015].

Olurin, K.B., Oluwo, A.B. (2010). Growth and survival of African catfish (*Clarias gariepinus*) larvae fed decapsulated *Artemia*, live daphnia, or commercial starter diet. *The Israeli Journal of Aquaculture*, 62 (1): 50-55. Available at

http://cmsadmin.atp.co.il/Content_siamb/editor/62_1_7_Olurin.pdf [Accessed on 29th May, 2014].

Padmavathi, P. and Veeraiah, K. (2009). Studies on the influence of *Microcystis aeruginosa* on the ecology and fish production of carp culture ponds. *African Journal of Biotechnology*, 8 (9): 1911-1918.

Pangni, K., Atsé, B.C. and Kouassi, N.J. (2008). Effect of stocking density on growth and survival of the African catfish *Chrysichthys nigrodigitatus*, *Claroteidae* (Lacépède 1803) larvae in circular tanks. *Livestock Research for Rural Development,* 20 (7): 1-9.

Parker, N.C. (1984). Third Report to the Fish Farmer: The Status of Warm water Fish Farming and Progress in Fish Farming Research. In Dupree, H.K. and Huner, J.V. (Eds.). *U.S. Fish and Wildlife*, Washington, D.C. USA. (pp. 270). Available at http://www.aces.edu/dept/fisheries/education/.../HistoryofAquaculture.pdf [Accessed on 07th March, 2015].

Pouomogne, V. **(**2008). *Capture-based aquaculture of Clarias catfish:* Case study of the Santchou fishers in western Cameroon. In: Lovatelli A. and Holthus P.F. (Eds.). *Capture-based aquaculture. Global overview*. FAO Fisheries Technical Paper. No. 508. Rome. (pp. 93-108). Available at http://www.fao.org/3/a-i0254e/i0254e03.pdf [Accessed on 11th March, 2015].

Rahman, M.A., Mazid, M.A., Rahman, M.R., Khan, M.N., Hossain, M.A. and Hussain, M.G. (2005). Effect of stocking density on survival and growth of critically

endangered mashseer, *Tor putitora* (Hamilton), in nursery ponds. *Aquaculture,* 249: 275-284.

Round, F.E., Crawford, R.M. and Mann, D.G. (1990). *The diatoms. Biology and morphology of the genera.* Cambridge, UK: Cambridge University Press.

Saad, T.T., El-Hawary, W.N. and Abel-lateef, M.A. (2014). Growth and Economic Performance of *Oreochromis niloticus* in Earthen Ponds. *Journal of Middle East Applied Science and Technology (JMEAST),* 11: 463-475.

Sahoo, S.K., Giri, S.S. and Chandra, S. (2008). Rearing performance of *Clarias batrachus* larvae: Effect of age at stocking on growth and survival during fingerling production. *Aquaculture,* 280: 158-160.

Schram, E., Van der Heul, J.W., Kamstra, A. and Verdegem, M.C.J. (2006). Stocking density dependent growth of dover (*Solea solea*). *Aquaculture,* 252: 239-247.

Schroeder, G.L. (1980). Fish farming in manure loaded ponds. In: Pullin, R.S.V. and Shehaded, Z.H. (Eds.). *Integrated agriculture-aquaculture farming systems: ICLARM Conference Proceedings* 4, 258. Manila, Philippines: International Centre for Living Aquatic Resources Management.

Shiri, H.A., De Charleroy, D., Auwerx, J., Vught, I., Van Slycken, J., Dhert, P. and Sorgeloos, P. (2003). Larval rearing of burbot (*Lota lota* L.) using *Brachionus calyciflorus* rotifer as starter food. *Journal of Applied Ichthyology,* 19 (2): 84-87.

Shoko, A.P., Lamtane, H.A., Wetengere, K., Kajitanus, O., Msuya, F.E., Mmochi, A.J. and Mgaya, Y.D. (2011). The status and development of aquaculture in Tanzania,

East Africa. In: Natarajan, P., Wondimu, L., Boyossa, T., Zuberi, M.I., Nair, A.S., Beyeh, A. and Aga, E. (Eds.). *Technical Proceedings of the International Conference on Ecosystem Conservation and Sustainable Development (ECOCASD),* Ambo University, Ambo, Ethiopia. (pp. 85-97). Available at http://tafiri.weebly.com/uploads/4/6/1/1/46116859/shoko_s_cv__latest_16-9-2015.pdf [Accessed on 26th September, 2015].

Soderberg, R.W. (2012). Organic and Inorganic Fertilization. In: Mischke C. C. (Ed.), *Aquaculture Pond Fertilization: Impacts of Nutrient Input on Production*, USA: Wiley-Blackwell.

Sophin, P. and Preston, T.R. (2001). Effect of processing pig manure in a biodigester as fertilizer input for ponds growing fish in polyculture. *Livestock Research for Rural Development.* Available at http://www.lrrd.org/lrrd13/6/pich136.htm [Accessed on 28th September, 2015].

Sundar, S., Raina, H.S., Mohon, M. and Singh, B. (1995). Ecology and fisheries potentials of the Guala River with special reference to proposed impoundment (Jamrani dam) on the system. *Journal of Inland Fisheries Society of India,* 27 (2): 33-45.

Sutar, V.B., Chavan, B.R., Bonde, V.R. and Balamani, C.H. (2012). Efficacy of farm yard manure and vermicompost on growth and survival of *Catla catla, Labeo rohita* and *Macrobrachium rosenbergii* in rearing pond. *DAV International Journal of Science,* 1 (2): 128-131.

Suziki, N., Kondo, M., Gunes, E., Ozongun, M. and Ohno, A. (2001). Age and growth of turbot *Psetta maxima* in the Black Sea. *Turkey Journal of Fisheries and*

Aquatic Science, 1: 43-53. Available at http://dergipark.ulakbim.gov.tr/trjfas/article/viewFile/5000045202/5000042671 [Accessed on 15th March, 2015].

Tacon, A.G.J. (2001). Increasing the contribution of aquaculture for food security and poverty alleviation. In: Subasinghe, R.P., Bueno, P., Phillips, M.J., Hough, C., McGladdery S.E. and Arthur, J.R. (Eds.). *Aquaculture in the Third Millennium. Technical Proceedings of the Conference on Aquaculture in the Third Millennium*, Bangkok. (pp. 63-72). Available at http://library.enaca.org/NACA-Publications/AquaMillenium.pdf [Accessed on 19th March, 2014].

Tanzania Fisheries Division, (2006). *Status of Aquaculture in Tanzania.* Ministry of Natural Resources and Tourism Fisheries Division, Dar es Salaam. Available at http:// ftp://ftp.fao.org/fi/document/fcp/en/FI_CP_TZ.pdf [Accessed on 09th March, 2015].

Thingstad, T.F., Krom, M.D., Mantoura, R.F.C., Flaten, G.A.F., Groom, S., Herut, B., Kress, N., Law, C., Pasternak, A., Pitta, P., Psarra, S., Rassoulzadegan, F., Tanaka, T., Tselepides, A., Wassmann, P., Woodward, E.M.S., Wexels Riser, C., Zodiatis, G. and Zohary, T. (2005). Nature of P-limitation in the ultra-oligotrophic Eastern Mediterranean. *Science*, 309: 1068-1071.

Toko, I., Fiogbe, E.D., Koukpode, B. and Kestemont, P. (2007). Rearing of African catfish (*Clarias gariepinus*) and vundu catfish (*Heterobranchus longifilis*) in

traditional fish ponds (whedos): Effect of stocking density on growth, production and body composition. *Aquaculture*, 262: 65-72.

Tucker, C.S. (1991). *Water Quantity and Quality Requirements for Channel Catfish Hatcheries.* Southern Regional Aquaculture Center, Delta Research and Extension Center, Mississippi State University, USA. Available at http://fisheries.tamu.edu/files/2013/09/SRAC-Publication-No.-461-Water-Quantity-and-Quality-Requirements-for-Channel-Catfish-Hatcheries.pdf [Accessed on 29th March, 2014].

United Nations Educational Science and Cultural Organization, (2006). *Manual on aquatic cyanobacteria: A photo guide and a synopsis of their toxicology*, Lund University, Sweden: Institute of Ecology/Limnology.

Van de Nieuwegiessen, P.G., Olwo, J., Khong, S., Verreth, J.A.J. and Schrama, J.W. (2009). Effects of age and stocking density on the welfare of African catfish, *Clarias gariepinus* (Burchell, 1822). *Aquaculture*, 288: 69-75.

Verreth, J., Eding, E.H., Rao, G.R.M., Huskens, F. and Segner, H. (2007). A Review of Feeding Practices, Growth and Nutritional Physiology in Larvae of the Catfishes *Clarias gariepinus* and *Clarias batrachus. Journal of the World Aquaculture Society,* (24) 2: 135-144.

Watanabe, T., Kitajima, C. and Fujita, S. (1983). Nutritional values of live organisms used in Japan for mass propagation of fish: a review. *Aquaculture*, 34: 115-143.

Wetzel, R.G. and Likens, G.E. (1991). *Limnological Analyses*. (2nd ed), New York, USA: Springer-Verlag.

Wurts, W.A. (2004). Organic fertilization in culture ponds. *Aquaculture,* 35(2): 64-65.

APPENDICES

Appendix 1: ANOVA for phytoplankton analyzed in experiment 1

Dependent variable: Chlorophyte

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	10992770833.333	5496385416.667	11.107	0.001
Residual	15	7423010416.667	494867361.111		
Total (Corrected)	17	18415781250.000			

Dependent variable: Cynophyte

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1547312500.000	773656250.000	11.838	0.001
Residual	15	980343750.000	65356250.000		
Total (Corrected)	17	2527656250.000			

Dependent variable: Euglenophyte

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	468361111.111	234180555.556	33.204	0.0001
Residual	15	105791666.667	7052777.778		
Total (Corrected)	17	574152777.778			

Dependent variable: Diatomae

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1004145833.333	502072916.667	12.221	0.001
Residual	15	616229166.667	41081944.444		
Total (Corrected)	17	1620375000.000			

Appendix 2: ANOVA for zooplankton analyzed in experiment 1

Dependent variable: Rotifers

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	4988844.444	2494422.222	8.275	0.004
Residual	15	4521400.000	301426.667		
Total (Corrected)	17	9510244.444			

Dependent variable: Copepods

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1215511.111	607755.556	17.108	0.0001
Residual	15	532866.667	35524.444		
Total (Corrected)	17	1748377.778			

Dependent variable: Cladocerans

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1270177.778	635088.889	63.312	0.0001
Residual	15	150466.667	10031.111		
Total (Corrected)	17	1420644.444			

Appendix 3: ANOVA for growth performance analyzed in experiment 2.

Dependent variable: Initial mean weight

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1579.704	789.852	2.606	0.077
Stocking density	1	1230.126	1230.126	4.058	0.056
Fertilizer * Stocking density	2	194.912	97.456	0.322	0.726
Residual	138	41829.247	303.110		
Total (Corrected)	143	44833.989			

Dependent variable: Weight gain

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	95.977	47.988	3.473	0.034
Stocking density	1	101.627	101.627	7.355	0.008
Fertilizer * Stocking density	2	3.629	1.814	0.131	0.877
Residual	138	1906.786	13.817		
Total (Corrected)	143	2108.018			

Dependent variable: Increase mean weight

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	2098.577	1049.289	0.121	0.886
Stocking density	1	3677.533	3677.533	0.424	0.516
Fertilizer * Stocking density	2	785.128	392.564	0.045	0.956
Residual	138	1196468.645	8670.063		
Total (Corrected)	143	1203029.884			

Dependent variable: Specific growth rate

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1089.777	544.889	2.649	0.074
Stocking density	1	943.560	943.560	4.586	0.034
Fertilizer * Stocking density	2	26.270	13.135	0.064	0.938
Residual	138	28391.012	205.732		
Total (Corrected)	143	30450.619			

Dependent variable: Survival rate

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	162.500	81.250	1.143	0.322
Stocking density	1	4011.111	4011.111	56.435	0.0001
Fertilizer * Stocking density	2	1318.056	659.028	9.272	0.0001
Residual	138	9808.333	71.075		
Total (Corrected)	143	15300.000			

Appendix 4: ANOVA for dissolved oxygen and temperature analyzed in experiment 1.

Dependent variable: Dissolved oxygen during the morning (am)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1.736	0.868	1.700	0.187
Residual	123	62.822	0.511		
Total (Corrected)	125	64.558			

Dependent variable: Dissolved oxygen during the afternoon (pm)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.903	0.452	0.517	0.598
Residual	123	107.534	0.874		
Total (Corrected)	125	108.437			

Dependent variable: Temperature during the morning (am)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.674	0.337	0.926	0.399
Residual	123	44.772	0.364		
Total (Corrected)	125	45.446			

Dependent variable: Temperature during the afternoon (pm)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1.050	0.525	0.876	0.419
Residual	123	73.680	0.599		
Total (Corrected)	125	74.730			

Appendix 5: ANOVA for dissolved oxygen and temperature analyzed in experiment 2.

Dependent variable: Dissolved oxygen during the morning (am)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.034	0.017	0.033	0.967
Stocking density	1	0.568	0.568	1.126	0.290
Fertilizer * Stocking density	2	0.082	0.041	0.081	0.922
Residual	228	115.012	0.504		

Total (Corrected)	233	115.696

Dependent variable: Dissolved oxygen during the afternoon (pm)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	8.510	4.255	4.228	0.016
Stocking density	1	0.629	0.629	0.625	0.430
Fertilizer * Stocking density	2	2.277	1.139	1.131	0.324
Residual	228	229.487	1.007		
Total (Corrected)	233	240.903			

Dependent variable: Temperature during the morning (am)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.477	0.239	0.237	0.789
Stocking density	1	0.267	0.267	0.265	0.607
Fertilizer * Stocking density	2	1.100	0.050	0.050	0.951
Residual	228	229.270	1.006		
Total (Corrected)	233	230.114			

Dependent variable: Temperature during the afternoon (pm)

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.563	0.281	0.162	0.850
Stocking density	1	0.246	0.246	0.142	0.707
Fertilizer * Stocking density	2	0.420	0.210	0.121	0.886
Residual	228	394.819	1.732		
Total (Corrected)	233	396.047			

Appendix 6: ANOVA for nitrite and ammonia analyzed in experiment 1

Dependent variable: Nitrite

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.282	0.141	5.833	0.013
Residual	15	0.363	0.024		
Total (Corrected)	17	0.645			

Dependent variable: Ammonia

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.571	0.286	2.762	0.095
Residual	15	1.551	0.103		
Total (Corrected)	17	2.122			

Appendix 7: ANOVA for nitrite and ammonia analyzed in experiment 2.

Dependent variable: Nitrite

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	0.441	0.220	4.725	0.031
Stocking density	1	0.083	0.083	1.776	0.207
Fertilizer * Stocking density	2	0.019	0.009	0.204	0.819
Residual	12	0.560	0.047		
Total (Corrected)	17	1.102			

Dependent variable: Ammonia

Source	DF	Sum of square	Mean square	F Value	Pr>F
Fertilizer	2	1.290	0.645	2.623	0.113
Stocking density	1	0.008	0.008	0.031	0.863
Fertilizer * Stocking density	2	0.003	0.002	0.007	0.993
Residual	12	2.952	0.246		
Total (Corrected)	17	4.254			