EVALUATION OF INVERTEBRATES AS PROTEIN SOURCE IN NILE TILAPIA (*Oreochromis niloticus*) DIETS

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ABSTRACT

A study was conducted to evaluate housefly maggots (HFM) and earthworms (EW) as protein sources in Nile tilapia, *Oreochromis niloticus* diets. HFM and EWM were raised in three different culturing substrates. Cow manure, fermented maize bran and chicken manure were used to culture HFM while cow manure, rabbit manure and chicken manure were used for culturing EW. Yields of HFM and EW from chicken and cow manure respectively were significantly higher ($P<0.05$). Protein content of HFM and EW from cow manure was significantly higher ($P<0.05$) than others. HFM and EWM from cow manure substrates (with high protein contents) were then used to formulate nine isonitrogenous diets (30% crude protein) with fixed five percent of fishmeal. These diets were named as HFM0/EWM0 (0%EWM, 0%HFM), HFM25 (25%HFM), HFM30 (30%HFM), HFM35 (35%HFM), HFM40 (40%HFM), EWM25 (25%EWM), EWM30 (30%EWM), EWM35 (35%EWM) and EWM40 (40%EWM). Feeding trial was done for a period of eight weeks. Every treatment was assigned in triplicates in which 14 fingerlings with a mean weight of 2.52 ± 0.12 grams were allocated using complete randomized design. The fish were fed up to 5% of their body weights twice daily throughout the experimental period. Effect of inclusion levels of either HFM or EWM on growth, feed utilization and cost of production of the fish were determined. There was significant difference ($P<0.05$) in growth and feed utilization of fish. Fish fed diets at35% inclusion level of HFM and EWM had highest performance. Profits were higher in diets with inclusion level of 25 to 40% of HFM and EW. It is included that, inclusion of 35% of either HFM or EWM is the most optimum to achieve high performance of *O. niloticus*.

DECLARATION

I, Tausi Ally, do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my original work and that it has neither been submitted nor being concurrently submitted for degree award in any other institution.

……. ……

Tausi Ally Date

(MSc. MNRSA student)

The above declaration is confirmed by:

…… ……

Dr. Madala, N. A. Date

(Supervisor)

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DEDICATIONS

This dissertation is dedicated to my mother Khadija MussaKibwana and uncles Kondo Mussa and Abdallah Kibwana. It is also dedicated to my fiancee Ismail H. Mokolah and my beloved daughter Asanath I. Mokolah.

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LIST OF ABBREVIATION AND ACRONYMS

ADWG	Average Daily Weight Gain
AOAC	Association of Official Analytical Chemists
AWG	Average Weight Gain
BWG	Body Weight Gain
CM	Cow manure
CF	Crude Fibre
CL	Crude Lipids
CP	Crude Protein
DAARS	Department of Animal, Aquaculture and Range Science
DFI	Daily Feed Intake
EW	Earthworm
EWM	Earthworm Meal
FAO	Food and Agriculture Organization of the United Nations
FCR	Feed Conversion Ratio
FW	Final Weight
FM	Fish Meal
g	Gram
gKg^{-1}	Gram per kilogram
HFM	Housefly Maggots
IW	Initial Weight
Kcal	Kilo calorie
Kg	Kilogram
kJ	Kilo joule
ln	Natural logarithm

mg	Milligram
MGR	Metabolic Growth Rate
MNRSA	Management of Natural Resources for \sustainable Agriculture
MT	Metric tons
PER	Protein Efficient Ration
SEM	Standard Error of the Mean
SUA	Sokoine University of Agriculture
PASW	Predictive Analytics SoftWare
SPSS	Statistical Package for Social Science
TZS	Tanzanian Shillings
USD	United States of America Dollar

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

The world is faced with increased demand of protein for human and animal consumption. Fisheries and aquaculture make a vital contribution towards meeting that demand and contribute up to 17% of global population's intake (FAO, 2014). In 2011 about 164 million metric tons (MT) of total fish production was made by capture fisheries and aquaculture. Approximately 85% (130.8 million MT) of total fish production in 2011was used for human consumption while the remaining (23.2 million MT) was used for non-human uses (Barbaroux *et al.*, 2012). Fish production from capture fisheries and aquaculture is threatened by over-exploitation, climate change, bad fishing practices and environment destruction (FAO, 2014). According to FAO (2014) from 2006 to 2012 global capture fisheries production has approximately remained stagnant at around 90 million MT and it is presented that the trend will remain the same until 2030. Stagnating capture fisheries have given way to the rise of aquaculture seen as an alternative to sustaining demand for fish (FAO, 2014). Global aquaculture production is expected to increase to approximately 120 million MT by 2030 from 66.6 million MT in 2012 (FAO, 2014).

Scarcity and high cost of key ingredients used in making fish feeds is a potential limiting factor in growth of aquaculture industry (Aniebo *et al.,* 2009; Bureau *et al.*, 2009; Huntington and Hasan, 2009a, b;Dedeke *et al.*, 2013). In order for aquaculture to meet the future demand of fish protein, quality ingredients must be available in required quantities. Adequate quantities of nutritional feed enable fish to realize their growth potential and eventually bring profit to the industry. Conventionally, fishmeal (FM)

and/or legumes and cereals have been used as protein and energy sources respectively (Craig and Helfrich, 2002;Huntington and Hasan, 2009;Chapman, 2015). In formulating nutritionally balanced fish diets, FM is the most preferred as a dietary protein source because of its nutritional quality and palatability properties (Tacon and Metian, 2008; Huntington and Hasan, 2009; Mohanta, 2013).

1.2 Problem Statement and Justification

Fishmeal prices are on the rise and increased demands lead to sharply higher prices (Tacon and Metian, 2008). For small fish farmers, this means that fishmeal is less accessible though aquaculture will need to expand sustainably to keep up with increasing demand of fish (FAO, 2012). Recent high demand and consequent high prices for fishmeal, together with increasing production pressure on aquaculture, has led to research into the development of local available proteins for aquaculture and livestock (which could eventually supplement fishmeal) (George and Otubusin, 2007; Gatlin, 2010; van Huis *et al.,* 2013). There has been a keen interest in identifying protein sources with adequate contents of the essential amino acids to support optimum fish performance (Sogbesan *et al.*, 2003: Huntington and Hasan, 2009; Moreau, 2009; Negroni, 2013). Several plant and animal protein sources have been identified as alternatives to fishmeal but have limitations (FAO, 2002). Most of plant sources are deficient in certain essential amino acids such as lysine and methionine. They also contain antinutrients such as mimosine, trypsin, phytic acids, saponins, proteases and lectin inhibitors (Soltan and Hanafy, 2008;Nguyen and Davis, 2009; Ayoola, 2010; Ogbe and George, 2012). On the other hand, the availability of most of these ingredients is seasonal (FAO, 2012).Commonly used plant protein is soybean meal due to its high protein and fairly good amino acid profile though deficient in sulphur containing amino acids (El-Sayed and Tacon, 1997; El-Sayed, 1999; Soltan and Hanafy, 2008; Nguyen

and Davis, 2009). However, in many developing countries such as Tanzania soya bean is scarce and expensive due to low production and competition with human food industry (Beatus, 2005). This necessitates search for other affordable and locally available protein sources such as invertebrates.

Invertebrates such as maggots and earthworms have high nutritional value and they can be easily cultured (van Huis, 2003). These organisms have good amino acids profile, polyunsaturated fatty acids and minerals (Sherman, 2003; Jabir*et al.,*2012, Lourdumary and Uma, 2013). Palatability of these organisms is likely to influence optimal feed intake (Ayoola, 2010; Jabir*et al.,*2012; Miles and Chapman, 2015). Culture of such invertebrates may help to minimize environmental issues associated with accumulation of large amounts of wastes (Teotia and Miller, 1974). Animal wastes like animal manure and household wastes can be used as substrates to produce house fly maggots and earthworms (Newton *et al.,* 2005; Devic, 2014). Therefore, the current study was conducted to evaluate the use of housefly maggots and earthworms as alternative sources of protein for farmed Nile tilapia.

1.3 Objectives of the Study

The main objective of the study was to utilize invertebrates as protein source for aquaculture diets. In order to address the main objective the following specific objectives were formulated:

i. To test different substrates for culturing maggots and earthworms.
ii. To determine effect of different inclusion levels of maggots and earthworms meal on performance of *O. niloticus.*
iii. To determine cost effectiveness of diets containing earthworm and maggot meals.

1.4 Hypotheses

In order to address the above specific objectives the following hypotheses were tested:

i. Substrate type has no influence on yield and nutrient content of housefly maggots and earthworms.

ii. Different inclusion levels containing earthworm and maggot meal have no effect on performance of *O. niloticus.*

iii. There is no difference in cost effectiveness between earthworm and maggot contained diets and FM control diet.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Overview of Invertebrates use in Animal Feeds

Invertebrates are natural food for many animals. For instance, chickens have been found picking worms and larvae from the top soil while worms are used as fishing baits (Sherman, 2003; van Hall *et al.,* 2011). Over the years, use of invertebrates as protein source in animal feeds has been limited. Recent efforts to search for alternative and sustainable protein sources have considered explored several invertebrates. Worms and fly maggots have been used as animal source of protein in animal diets like chickens, guinea fowl, cats, dogs, cattle and rabbits (Sogbesan*et al.,* 2006; Omoyinmi and Olaoye, 2012;Dedeke *et al.,* 2013; Mohanta, 2013). Earthworms and housefly maggot has been shown to be ideal for different fish species like catfish and tilapia (Oyelese, 2007).

2.2 Housefly

The housefly, *Musca domestica*, belongs to Kingdom-Animalia, Phylum-Arthropoda, Class-Insecta, Order-Diptera, Family-Muscidae, Genus-*Musca,* and Species-*M. domestica*. It is commonly found in human dwellings where it thrives on organic wastes (Keiding, 1975). Housefly has a complete metamorphosis with distinct stages of egg, larvae/ maggot, pupa and adult (Keiding, 1986). Eggs of *M. domestica*are whitish in color, and always feed on the materials they were laid to develop into pupae stage. A pupa is the third stage on the life cycle of housefly with a round shaped at both ends. It varies from yellow, red, brown to black in color with ages. At temperature of 32 to 37^{o} C maggots spend two to six days to complete its development (Keiding, 1986). The life cycle is complete in seven to ten days and adult housefly lives for 15 to 30 days (Keiding, 1975). Generations per annum range from more than 20 in tropics and sub-

tropics regions to less than 12 in temperate regions (Keiding, 1986; van Huis *et al*., 2013).

2.2.1 Housefly Maggots

Maggot 'larvae' is intermediate stage between egg and pupae on the life cycle of the housefly. Maggots are creamy whitish in color, cylindrical tapering towards the head with one pair of dark hooks (Keiding, 1986; Hogsette, 1996; Salas, 2007). The legless maggots appear from eggs within eight to twenty hours under optimum temperatures of 25 to 30^{o}C and moist conditions.

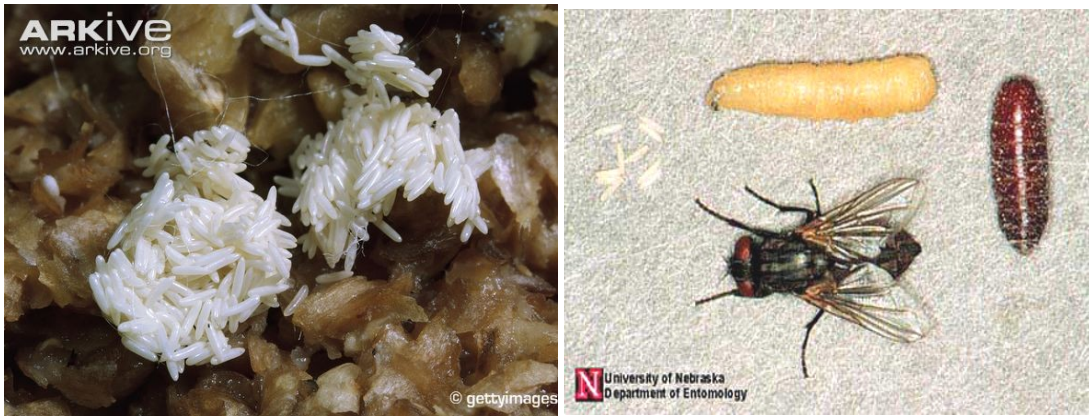

Plate 1: Housefly eggs on culturing substrate and housefly life cycle. (Keiding, 1986)

2.2.2 Effect of Culture Substrates on Yields and Nutritional contents of the Maggots

Mass production and quality housefly maggots are determined by a number of factors including culture conditions, culture substrate and attractants as well as harvesting methods (Nzamujo, 1999; Aniebo and Owen, 2010; Odesanya *et al.,* 2011; Kareem and Ogunremi, 2012; Ajonina*et al.,* 2013; Agbeko*et al.,* 2014). Above or below required limit of culture conditions such as direct sunlight, temperature and humidity may affect negatively the quantity and quality of the maggots (Nzamujo, 1999; Devic, 2014).

Temperature and humidity at a limit of 25 to 33^0C and 50 to 88% respectively a housefly may produce up to 3g eggs that could yield 511g larvae for a day (Devic, 2014).

Also, in the same given culture condition, quantity and quantity of the maggots vary with substrates. Among the three (animal digester, pito waste and poultry manure) substrates, animal digester produces large amount of maggots than others due to its ability to attract the houseflies (Agbeko*et al.,* 2014). For instance, a composed chicken and pig manure have ability of attracting the flies longer due to its odor. Therefore, large amount of the maggots can be produced in the pig and chicken manure (Patricia and Salas, 2007). Additionally, culture substrate can be mixed with fly attractants in order to produce enough maggots. Substrate with fly attractant produces high quantity of the maggots than culture substrate with no attractants (Nzamujo, 2001). Rotten food materials and animal offal added to poultry manure produced (50%) more than the yield (20%) from manure with no attractant (Odesanya*et al.,* 2011).

Likely, nutritional composition of the housefly maggots can be affected by the type of culture substrates (Nzamujo, 1999). The animal digester, poultry manure and brewery spent produced maggots with 59.77, 59.97 and 52.13% CP respectively (Obeng *et al.,* 2014). On the other hand, there is the variation on the crude protein content of the maggots collected from the mixture of wheat bran and cattle blood (44.2% CP) and mixture of saw dust and cattle blood (47% CP) (Aniebo*et al.*, 2009). Therefore, nutritional composition of the maggot meal is the function of amount and quality of nutrients available in the substrates (Patricia and Salas, 2007).

Harvesting method can also affect the quantity and quality of the maggots. Use of screening method during harvesting gives the maggots in large quantity (g/kg of poultry

manure) than floating method (Sogbesan*et al.,* 2006). In floating method it's difficult to sort the maggots if mixed with floating particles of culture substrate, thus more wastage of the maggots. Thus, at present study chicken manure, cow manure and fermented maize bran was used as culture substrates to produce the maggots in large quantity and quality.

2.2.3 Inclusion Levels of Maggot Meal in Fish Diets

There have been several studies which investigated use of maggot meal in fish diets without compromising performance. Yaqub (1999), Ogunji*et al.* (2006), Ahmed *et al.* (2015) and Ezewudo*et al.* (2015) reported the maggot meal can either partially or completely be included in the diet of fish. Yaqub (1999) and Ogunji *et al.* (2006) recommended a complete inclusion of maggot meal in the diets of *O. niloticus* juvenile. Basing on the nutrient utilization and better growth performance of the fish Agbeko *et al.* (2014), Ahmed *et al.* (2015) and Ezewudo*et al.* (2015) suggested the inclusion of maggot meal in the diet of the fish should range from 25 to 75%. This is due to the reason that the meal need to be combined with any other sources of protein for better performance of the fish. A complete replacement of fishmeal with maggot meal may lead to insufficient utilization of nutrient by fish and poor digestibility of the diet (Ahmed *et al.,* 2015).Insufficient utilization of the housefly maggot meal may also increase when the maggot harvested at either late larval stage or beyond (pupae) stage (Mason *et al.,* 1990; Ogunji*et al.,* 2008; Aniebo and Owen, 2010). Thus, present of chitin in the maggot meal limits up 75% inclusion in the diet of Nile tilapia (Gatlin, 2010; Ezewudo*et al.*, 2015).

2.3 Earthworms

Earthworms, *Lumbricusterrestris*are hermaphrodites (each individual carries both male and female reproductive organs). It belongs to kingdom-Animalia, phylum-Annelid, class-Clitellata, family-Lumbriculidae, genus-*Lumbriculus*and species *L. terrestris.* It lacks skeleton, but is able to maintain its' structure using a fluid-filled coelom chamber that function as a hydrostatic skeleton (Dominguez *et al.,* 2001). Different species of earthworms have different life histories (Quillin, 1999; Dominguez and Edwards, 2011)though most of them have typical the same life cycle. However, they are different from other invertebrates in living longer (Quillin, 1999).

Plate 2: Physical appearance of earthworm, *Lumbricusterrestris*.

2.3.1 Effect of Culturing Substrate on the Yields and Nutritional contents of Earthworms

Temperature, moisture, pH and chemical composition of organic matters are some of the factors affecting distribution of the earthworms,*L. terrestris*(Sherman, 2003; Chapman, 2006; Dominguez and Edwards, 2011). At a farm level, a culturing container such as lumber containers, concrete or cinder blocks and bricks, concrete or hollow tile

and plastic containers may affect reproduction and fecundity of earthworms (Sherman, 2003). Insulated containers help to reduce adverse effect of temperature to earthworms. In order to improve culture condition and maintain quality of culturing substrate, bedding materials with optimum pH and nutrient composition are required (Sogbesan*et al.,* 2006; Sherman, 2003; Ibrahim *et al.,* 2010; Monebi and Ugwumba, 2013; Jesikha and Lekeshmanaswamy, 2013).Chemical substances such as polyphenols present in some of the plant bedding materials retards growth of the earthworms (Suthar, 2007a). Therefore, degradable bedding materials are required to maintain moist condition of the substrate, and it may be used as food for worms.

It is reported that, cow manure and pongamia leaves used as culture substrate of the worms supported high productivity of earthworm *Eudrilus Eugenia* (Jesikha and Lekeshmanswamy, 2013). Also, high yields of earthworms can be obtained by placing culture container directly under cages of animals such as rabbit, horse, swine, dairy or steer. This is because the worms receive sufficient nutrients directly from manure (Sherman, 2003). Siddiqui *et al.* (2005) and Sogbesan*et al.* (2006) reported the effect of cow manure on survival, growth and fecundity of *Eiseniafetida* as it resulted into more yields. Also, Dominguez *et al.* (2001) and Ibrahim *et al.* (2010) reported cow manure to have positive influence on productivity of *Eudriluseugeniae* worms. Monebi and Ugwumba (2013) added that cellulose substrate is suitable for weight gain and growth rate of the earthworms*Eudriliseugeniae*.

Likewise, high nutrient content on culturing substrates produce worms with sufficient nutritional composition. Humus substrate is suitable to produce *Libyodrilus violaceus* worms with high adequate nutrients. It produces the worm with 60.46% crude protein content and 4.68% crude fat content (Dedeke *et al.,* 2013). If it is used to produce

different worm species, variation in nutritional content of earthworm may be due to the differences in worm species and ages of the individual (Guerrero, 1983; Dominguez *et al*., 2001;Sogbesan and Ugwumba, 2006;Ibrahim *et al.,* 2010). Sherman (2003) added that most of the earthworms have large amount of protein at sexual maturity stage due to the availability of eggs.

2.3.2 Inclusion Level of Earthworm Meal in Fish Diets

Until now the use of different species of earthworms as alternative to replace fishmeal in the diets of different fish species has been conducted (Yaqub 1999; Olele, 2011; Jabir *et al.* 2012; Olele and Okonkwo, 2012; Dedeke*et al.,* 2013; Monebi and Ugwumba 2013; Pucher *et al.,* 2014). Earthworm meals produced from species such as *Engraulisencrosicolus,Zophobasmorio,Libyodrilus violaceus, Eudriluseuginea*and*Perionyx excavate* can completely replace FM in the diets of fish like *O. niloticus*. This is due to the reason that earthworms contain high nutritional content. However, low feed intake was reported to some fish species fed at high inclusion of earthworm (Jabir*et al,.*2012;Monebi and Ugwumba, 2012). Hence, poor feed intake results into poor feed utilization and reduction in growth performance of the fish. Better feed intake enhanced by feed palatability (Ogunji*et al.,* 2006; Ayoola, 2010; Mile and Chapman, 2010; Devic*et al.,* 2013).

Medina *et al.* (2003), Kostecka and Paczka (2006) reported poor palatability of earthworm meal impart by coelomic fluid. Furthermore, coelomic fluid of earthworm *Eiseniafoetida* has been reported to be toxic to vertebrates (Kobayashi *et al.,* 2001; Kauschke*et al.,* 2007). Thus presence of coelomic fluid is a main limitation to a complete substitution of earthworm meal in the fish diet (Pucher*et al.,* 2014).Therefore, replacement at 25 to 50% is suitable for optimum growth and nutrient utilization (Jabir

et al., 2012; Monebi and Ugwumba, 2012). Inclusion of earthworm meal up to 50% produces fish with similar growth performance to those fed fishmeal based diets.Partial replacement of FM with earthworm meal was also reported to affect body composition of fish in terms of protein, fat and moisture (Jabir *et al.,* 2012; Olele, 2011; Olele and Okonkwo, 2012; Dedeke*et al.,* 2013; Puche*et al.,* 2014).

2.4 Tilapia

2.4.1 General Biology of Tilapia

Tilapia is the generic name of a group of cichlids endemic to Africa. The group consists of three culturally important genera-*Oreochromis, Sarotherodon*and Tilapia (Atz, 1957; Crag and Helfrich, 2002; Negroni, 2013). All tilapia species are nest builders and fertilized eggs are guarded in the nest by brood parents. According to Atz (1957) and Babiker and Ibrahim (1979) the species within this genus were later classified according to differences in their mode of reproduction. Nile tilapia, *Oreochromis niloticus* is the tropical species that prefers to live in shallow water with lower and upper temperature limit of 11^0C and 42^0C (Allanson and Noble, 1964). The species is omnivorous and filter feeder by trapping phytoplankton (Atz, 1957). It reaches sexual maturity at age of 5 to 6 months, with ability to produce number of eggs proportional to its body weight (Fryer and Iles, 1972; Galman, 1980; Alvendia*et al.,* 1998; Fitzsimmons, 2009).

2.4.2 Nutrient Requirements of Nile Tilapia

The nutritional requirements of Nile tilapia are slightly different and vary with life stage i.e. fish size and age (Jauncey and Ross, 1982; Siddiqui *et al*., 1988; El Sayed and Teshima, 1991; Al Hafedh, 1999). Fry and fingerling fish require a diet higher in protein, lipids, vitamins and minerals and lower in carbohydrates as they are developing muscle, internal organs and bone with rapid growth. Sub-adult fish need more calories

from fat and carbohydrates for basal metabolism and a smaller percentage of protein for growth. Adult fish need even less dietary protein for optimum performance, while; broodstock tilapia requires more dietary protein for optimum reproduction, spawning efficiency, larval growth and survival (Table 1).

Table 1: Protein Requirements (dry basis) of Nile tilapia in weight

Life stage/weight (g)	Requirement (% of diet)
First feeding fry	45 - 50
0.02 - 2.0	30 - 40
2.0 - 35	35
35 to harvest	30 - 32
Broodstock	35 - 45

2.4.3 Merits as Cultured Fish

Apart from commercial culturing of several tilapia species, Nile tilapia is the predominant cultured species worldwide (FAO, 2014). The Nile tilapia is much involved in aquaculture due to its desirable characteristics like fast growth, short food chain efficiency in food conversion and high fecundity that ensures high distribution of seeds from farmer to farmer (Fitzsimmons, 2000; Negroni, 2013; FAO, 2014). Also its tolerance to a wide range of environmental parameters, and good tilapia product quality are mostly preferred than other species in the same genus (Atz, 1957; Allanson and Noble,1964; Behrends*et al.,* 1990). Avtalion and Shlarobersky(1994) and Fitzsimmons *et al.* (2014) added that, resistance of Nile tilapia to parasites, pathogens and their suitability in a wide range of farming systems make their production increase rapidly worldwide.

2.4.4 Global, Regional and Local Production Trend of Tilapia

Previously, fisheries industry was contributed most by Carps and Salmons with an average global production of 3.7 million MT in 2010 (FAO, 2011). Looking across species, the fastest supply growth is expected for tilapia, carp, and catfish. Tilapia is one

among the freshwater fish species that is the most of increasing in aquaculture. Its' industry has expanded since it has been an important source of food for human and being the third most important cultured fish group in the world (FAO, 2014).Global tilapia production is expected to almost double from 4.3 million tons to 7.3 million tons between 2010 and 2030 (FAO, 2012). The species is expected to contribute to a 60% predicted total freshwater aquaculture production in 2025 (FAO, 2016).

In 2010, Nile tilapia contributed up to more than 84 percent of total global tilapia production (FAO, 2011). It is anticipated that, in 2020 global Nile tilapia production will value up tomore than $5 billion in a global fish market (Figure 1) (FAO, 2012).

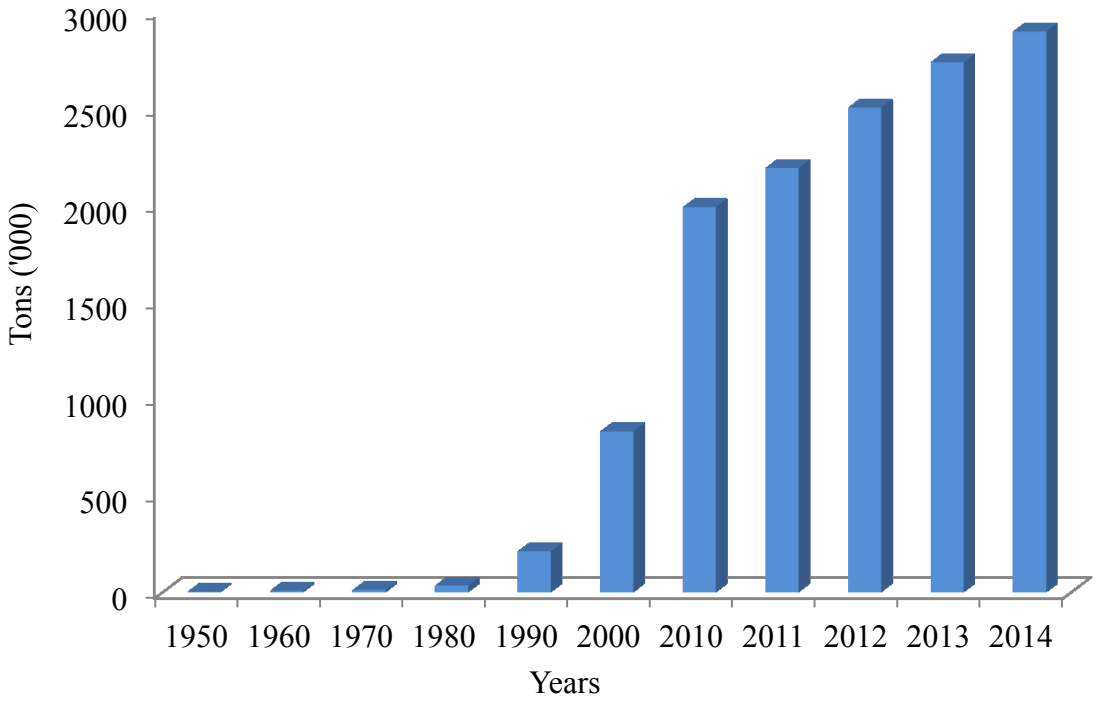

Figure 1: Global tilapia production (FAO Fishstat, 2016)

Fitzsimmons *et al.* (2014) reported tilapias production to continue increasing in popularity globally. In the year 2014 China remained the largest world tilapia producer with total production of nearly 1,650,000 million MT. Egypt reported a production of nearly 790,000 tons in that year, while Indonesia, Philippines and Thailand produced

700,000, 300,000 and 280,000 tons respectively. The other five top ten tilapia producers were the Brazil, Bangladesh, Mexico and Vietnam. Ecuador, Taiwan, Colombia, Malaysia and Myanmar are also major producers of tilapia (FAO, 2014). Global tilapia production is expected to be up to more than 5,200,000 million MT in 2020.

In the United Republic of Tanzania aquaculture production accounts only for a small proportion of total fish produced at the national level. At local level aquaculture is dominated by freshwater fish farming in which small scale farmers practice both extensive and semi-intensive fish farming (Chenyambuga*et al.,* 2014). The dominant species cultured is Nile tilapia (*O. niloticus*), followed by African catfish (*Clariasgariepinus*)(Kaliba*et al.,* 2006). Lack of enough feeds was among the major limitation to Nile tilapia farming (Chenyambuga*et al.,* 2014). Thus, tilapia production is still at a subsistence activity practiced by small scale fish farmers who have low social, cultural, economic status, limited access to technology, markets and feed constraints (Senkondo*et al.,* 2006; FAO, 2012). Therefore, use of housefly maggot and earthworm meals appropriate due to their local availability.

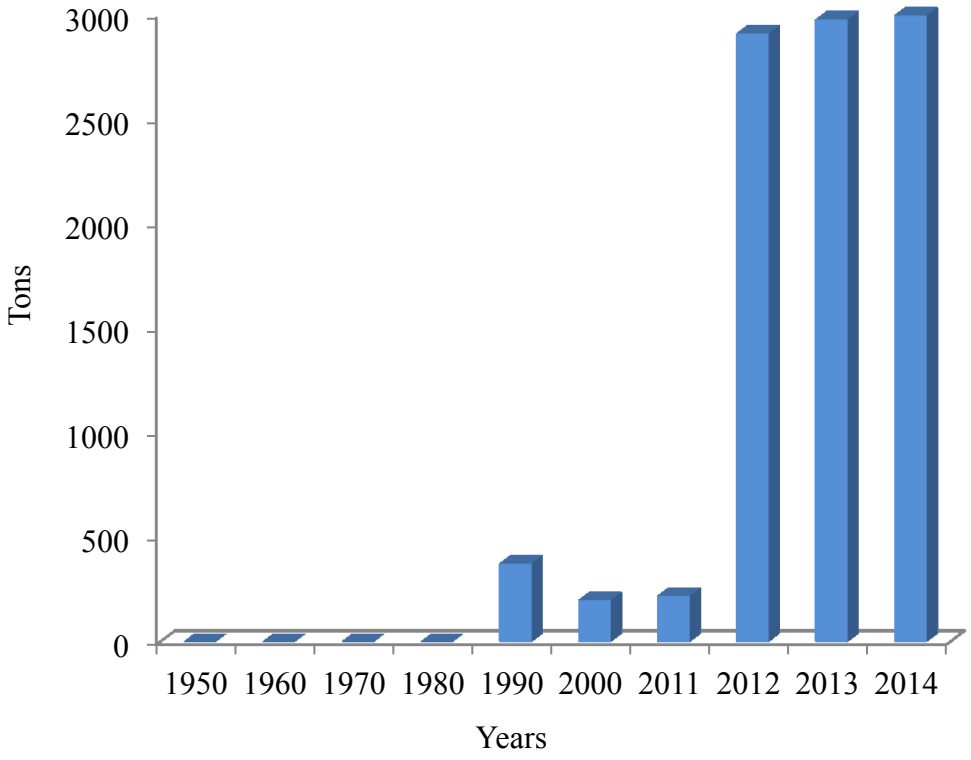

Figure 2: Tilapia aquaculture production trends in Tanzania (FAO Fishstat, 2016)

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Location of the Study

The study was conducted at Aquaculture Research Facility belonging to Department of Animal, Aquaculture and Range science of Sokoine University of Agriculture (SUA), Morogoro, Tanzania. The Facility is situated on the Western part of the University along Town-Mzinga road. SUA is located about 2.5 km South of Morogoro Municipality at an altitude of 550m above sea level. Morogoro receives approximately 880mm of bimodal rainfall annually during October to December and March to May or June. Monthly mean minimum and maximum temperatures are 14.2^0C to 35.5^0C respectively. Relative humidity ranges from 29% to 96%.

3.2 Preparation of Culturing Substrates

3.2.1 Housefly Maggots

Three substrates (fermented maize grain, cow manure and poultry manure) with attractants (cattle offal and fish remains) were used. Every substrate was well mixed with fish and animal wastes as attractants for the houseflies. The mixture was wrapped with a net with a mesh size of 1.2 mm to ease harvesting of maggots. One kg of substrate was placed in a 10 litre plastic buckets. Culturing was done indoors in triplicates for each of the substrates as suggested byNzamujo (2001), Hwangbo *et al.* (2009) and Devic (2014). Observation on the development of the maggots was done once a day. The eggs hatched within two days and were left for further two days to develop into mature maggots. The mature maggots (Appendix 3) were harvested as according to Sogbesan *et al.*(2006) and blanched in hot water (Plate 4). Thereafter, they were weighed to determine total wet weight per harvest and then oven dried at 65^0C for

48 hours. The dried maggots were re-weighed to determine dry weight before ground into a powder.

Plate 3: Set-up of the Housefly Maggots culturing media

Plate 4: Harvested Housefly Maggots blanched in hot water

3.2.2 Earthworms

Plasticbuckets with of 12 cubic liters capacity (Plate 5) were used as culturing containers for earthworms. Different manure (cow manure, rabbit manure and poultry manure) was separately used as sources of nutrients for earthworms with dried rice straws as bedding materials. Two and a half kg of soil from earthworms' naturalhabitat was collected and mixed with mixing 5Kg manure. The culture was set with bedding materials at the bottom of each basin, followed with equal number of adult earthworms in the substrates. Thereafter bedding materials placed on the top of mixtures to improve their housing as advised by Copper *et al.* (1970) and Sherman (2003). The worm cultures were placed under shade and covered with the mosquito nets to prevent escapes.Each culture substrate was moisturized by one liter of water once a week.During harvesting of the adult earthworms, table harvesting method as adopted from Sherman (2003) was used, and then the earthworms were sorted, washed, weighed, sun dried and stored at -5^0C until required amount was obtained. Weighing of the collected adult earthworms (Plate 6) was done with respect to the culture replicates. The earthworms were finally oven dried at 65^0C for 48 hours, and reweighed before grounded into powder form.

Plate 5: Set-up of Earthworms' culturing media

Plate 6: The harvested earthworms from cow manure substrates

3.2.3 Effects of Culturing Substrate on Yields and Nutritional Content of the meals

Mean yields and percentage of nutritional content in terms of crude protein of the HFM and EW meals were used to determine appropriateness of the substrates types. Total yields of the meals were determined by calculating the mean yields (mean±SE) of HFM and EW meals from each substrate. Thus, the higher the yield in dry conditions, the more suitable the substrate was. Nutritional contents of the meals were determined through proximate analysis.

3.2.4 Sources and Preparation of the Ingredients

Four ingredients namely, fishmeal (FM), maize meal (MM), wheat meal (WM) and sunflower oil (SFO) were bought from Main Morogoro Food Market. Cotton seed meal (CSM) and minerals and vitamin (Min/Vit) premix were purchased from Morogoro agricultural input suppliers. Fishmeal, Maize and wheat were well sorted and careful washed with hotwater to remove mites, sand and dusts that might reduce the quality of diets. Thereafter, eachingredient wassun dried for two days,thoroughly groundedand sieved through 1 mm diameter sieve.

3.2.5 Biochemical Analysis

Determinations of nutrient contents of all meals were done using procedures described by AOAC (2005). Dry matter and ash were determined by weighing 1 gram of the samples by using a 160g capacityanalytical weighing balance (Precisa 180A, Oerlikon, Switzerland), oven-drying (E 115, WTB binder 7200, Germany) at 70^0C to constant weight, re-weighing and ashing the samples in Muffle furnace (N31R, Nabertherm, West Germany) at 500^0C for three hours.

The crude protein was determined by weighing the samples (Presica 180A, Oerlikon, Switzerland) followed by three stages of Kjeldahl system namely digestion (Digestion System 12 1009, Digester, Tecator, Sweden), distillation (2200 Kjeltec Auto Distillation, Foss Tecator, Sweden) and titration (Digitrate, Tecator, Sweden). This was multiplied by the factor of 6.25 to get the amount of crude protein.

Crude fat of the samples was determined by weighing the samples (Presica 180A, Oerlicon, Switzerland), and fat extracted by Soxhlet extraction method (Soxtec system HT 1043 Extraction unit, Tecator, Sweden). Thereafter, the extraction cups containing fat material were dried at 105^0C for 30 minutes to remove traces of moisture. Then cups containing fat material were cooled in a desiccator for about 10 minutes and weighed to calculate amount of crude fat of the feeds by using the following formula:

$$\%\ EE = \frac{(Wt.ofcupwithextractedfat - Wt.ofcupwithboilingchips)}{Wt.ofSample} x100$$

Where by,

EE = Ether extract, Wt = Weight.

Crude fibre was obtained by weighing samples (Presica 180A, Oerlicon, Switzerland) into filter bags, digesting the sample in weak sulphuric acid and rinsed in weak NaOH solution in at 100^0C for 30 minutes in Ankom machine (ANKOM^{220}, ANKOM Technology, USA) and then washing and rinsing using distilled water. Then weak sodium hydroxide (alkaline) solution was added to remove acids in the samples by heating the samples in the solution at 100^0C for another 30 minutes. Rinsed with distilled water and then acetone was added to remove fat remaining in the residues. Then the samples were dried and ashed. The difference between the residues and the weight of ash gave the amount of crude fibre. The Nitrogen-free extracts percentages (NFEs) were obtained by subtracting the sum of percentages of Crude protein, Crude fibre, ash and Crude lipid from 100%. Gross Energy (GE) of the feed ingredients were estimated according to Jobling (1983) as follows:

$GE = (\%\ CPx\ 5.65) + (\%\ EEx\ 9.45) + (\%\ NFEx\ 4.2)\ Kcal/g$

Whereby,

CP = Crude protein, NFE = Nitrogen free extract.

3.2.6 Formulation of the Experimental Diets

Proximate analysis (Table 4 and 5) indicated that, in both cases, cow manure when used as substrate resulted into meals with highest crude protein contents. As such the meals were used to formulate the experimental diets. Separately,the HFM and EW meals were included in the formulated diets at 0%, 25%, 30%, 35% and 40% respectively. All diets were isonitrogenous (30% crude protein) (Tables 2 and 3).

Appropriate quantities of the ingredients in each diet were weighed and mixed thoroughly in a container. Thereafter, water was added and continuously mixed, and then the feeds were pressed into pellets using pelleting machine (NMG-745, Nikai

Japan Ltd, Japan) of 5 mm diameter. Pellets were then sun dried for two days throughout and stored in a refrigerator at temperature of 5^0C for the whole feeding trial period.

Table 2: Five formulated HFM Based Diets (g/100g diet)

	Diets				
Ingredients	**HFM0**	**HFM25**	**HFM30**	**HFM35**	**HFM40**
Fish meal	5.0	5.0	5.0	5.0	5.0
Housefly maggot meal	0.0	12.0	24.5	40.0	48.8
Cottonseed meal	50.0	39.0	25.0	9.0	0.0
Maize meal	40.0	38.5	40.5	42.0	42.2
Wheat meal	2.0	2.0	2.0	2.0	2.0
Sunflower oil	1.0	1.5	1.0	0.0	0.0
Vitamin/mineral premix*	2.0	2.0	2.0	2.0	2.0

HFM = Housefly maggot meal, HFM0 = 0% HFM, HFM25 = 20% HFM, HFM30 = 30% HFM, HFM35 = 35% HFM, HFM40 = 40% HFM.

*Vitamin A 25,500,000 IU, Vitamin D3 5, 000, 000 IU, Vitamin E 5,050 IU, Vitamin B2 mg 4,750, Vitamin B6 mg 2,750, Vitamin B12 mcg 11, 750, Vitamin K3 mg 4,850, CAL PAN mg 5,750, Niacinamide mg 16, 500, Vitamin C 10, 000 mg, Iron 5,250 mg, Manganese 12, 760 mg, Copper 13, 250 mg, Zinc 13, 250 mg, Sodium chloride 48, 750 mg, Magnesium 12, 750 mg, Potassium acetate 73, 750 mg, Lysine 15,000 mg, Methionine 12, 000 mg, antioxidant and anticaking qsf 1 kg.

Table 3: Five formulated EW Based Diets (g/100g diet)

	Diets				
Ingredients	**EWM0**	**EWM25**	**EWM30**	**EWM35**	**EWM40**
Fish Meal	5.0	5.0	5.00	5.0	5.0
Earthworm meal	0.0	12.0	24.0	39.8	45.0
Cottonseed meal	50.0	38.0	24.5	5.0	0.0
Maize meal	40.0	38.0	39.5	42.7	42.0
Wheat meal	2.0	2.0	2.0	2.0	2.0
Sunflower oil	1.0	3.0	3.0	3.5	4.0
Vitamin/Mineral premix*	2.0	2.0	2.0	2.0	2.0

EWM = Earthworm meal, EWM0 = 0% EWM, EWM25 = 25% EWM, EWM30 = 30% EWM, EWM35 = 35% EWM, EWM40 = 40% EWM

*Vitamin A 25,500,000 IU, Vitamin D3 5, 000, 000 IU, Vitamin E 5,050 IU, Vitamin B2 mg 4,750, Vitamin B6 mg 2,750, Vitamin B12 mcg 11, 750, Vitamin K3 mg 4,850, CAL PAN mg 5,750, Niacinamide mg 16, 500, Vitamin C 10, 000 mg, IRON 5,250 mg, Manganese 12, 760 mg, Copper 13, 250 mg, ZINC 13, 250 mg, Sodium chloride 48, 750 mg, Magnesium 12, 750 mg, Potassium acetate 73, 750 mg, Lysine 15,000 mg, Methionine 12, 000 mg, antioxidant and anticaking qsf 1 kg.

Plate 7: Nine formulated isonitrogenous experimental diets

3.3 Experiment Setup

The experiment was conducted at Aquaculture Research Facility on a recirculation system that comprises two large tanks and five medium water tanks (Plate 8). The upper tank receives clean water from major water pipe to the inner system. Used water from inner system (Plate 9) passed through a series of pipes to the filtering tanks sequentially. From lower large tank a pumping machine pumps treated water to the upper tank and repeatedly to the inner system for reuse.

Plate 8: The water treatment system of the fish hatchery building at Magadu farm

Plate 9: The indoor rearing system of fish hatchery at Magadu Farm

Nine diets were randomly allocated in triplicates of 27 rearing tanks in a completely randomized block design. Each rearing tank was stocked with fourteen fingerlings withinitial mean weight of 2.5±0.84gramsto make a total of 42 fingerlings per treatment. Experimental fish were fed twice a day at 0900Hrs and 1100Hrs according to

their feeding response, while limited to 5% of their body weight. The amount of feed was adjusted in response to changes in fish's body weights. Rearing tanks were flushed twice a week and the water holding tanks cleaned once a week to enhance aeration and avoid the risks of infection and diseases. The experiment lasted for eight weeks.

3.4 Growth Performance and Feed Utilization

For determination of growth performance and feed utilization; average daily weight gain (ADWG), specific growth rates (SGR), protein efficiency ratio (PER), feed conversion ratio (FCR) and feed intake (FI) were calculated. Before stocking initial body weight of each experimental fish was weighed and recorded. Weighing of fish was done using a 1kg digital weighing balance (Mettler PM11, Mettler Instrument LTD, Switzeland). Subsequent body weights were weighed and recorded after every seven days. Before weighing fish were starved for a day. The body weights of fish from each replicate were recorded in bulky and finally mean weights was calculated. Performance characteristics were calculated using the following formulae as adopted from Olvera-Novoa*et al.* (1990).

(i) Average daily weight gain (ADWG)

$$ADG = \frac{Finalbodyweight(g) - Initialbodyweight(g)}{Time\ (days)}$$

(ii) Specific Growth Rates (SGR%)

$$\%\ SGR\ = \frac{LN(Finalbodyweight) - LN(Initialbodyweight)}{Experimentalperiod\ (days)} x\ 100$$

(iii) Percentage Survival (PS)

$$PS = \frac{Finalnumberoffishatharvest}{Initialnumberoffishatstocking} x\ 100$$

(iv) Protein Efficiency Ratio (PER)

$$PER = \frac{Bodyweightgain\ (g)}{Crudeproteinintake\ (g)}$$

(v) Feed Conversion Ratio (FCR)

$$FCR = \frac{Feedsupplied\ (g)}{Bodyweightgain\ (g)}$$

(vi) Daily Feed Intake (DFI)

$$DFI = \frac{FeedSupplied\ (g)}{Time\ (days)}$$

3.5 Cost Effectiveness Analysis

The cost of feed required to produce 1kilogram fishfrom feeding various experimental diets was estimated by using the cost of ingredients used to formulate experimental diets. Major assumptions were that all other operating costs i.e. labor, rearing facilities, water and electricity remained the same for all diets. The costs of other feed ingredients were based on the current prices at time of purchase. The costs of producing HFM and EW meals were put as the cost of time used during collection and processing of culturing substrates. The value used in the calculation of the cost to produce 1Kg market sized fish was in TZS.Incidence cost, profit index and cost to produce 1 kg fish was determined as follows:

(i). Incidence cost

$$Incidencecost = \frac{Valueoffish\ (Kg)}{Costoffeed\ (Kg)}$$

(ii). Profit index ratio

$$Profitindex = \frac{Costoffeed\ (Kg)}{Weightoffishproduced\ (Kg)}$$

(iii). Cost to produce 1Kg fish (TZS)

$Costtoproduce\ 1Kgfish = (FeedcostXFCR)$

3.6 Statistical Analysis

The collected data were analyzed using Statistical Package for Social Sciences program version 10 (SPSS Richmond, VA, USA) as described by Dytham (1999). Data were tested for normality and homogeneity of variance before being analyzedby using One-way analysis of variance (ANOVA).Treatment means were considered significant at $P<0.05$. Post–hoc analysis was also done where significant differences existed between treatments means using Tukey's Honest Significant Difference Test (Steele and Torrie, 1980).

The model was:

$Y_{ij} = \mu + T_i + L_{ij} + \varepsilon_{ij}$

Where,

Y_{ij} = Observed value (yields, nutritional composition of experimental meals, fish growth performance and feed utilization)

μ = General means

T_i = the effects of treatment (I = 1, 2)

ε_{ij} = Residual Error

L_{ij} = Levels within treatments (j = 1, 2, 3, 4, 5)

CHAPTER FOUR

4.0 RESULTS

4.1Substrate for Culturing the Invertebrates

4.1.1 Housefly Maggot Meal (HFM)

Yield of housefly maggots was significantly different between thedifferent substrates. Yield of 457.0 ± 2.0 g/kg from chicken manure was significantly higher (P<0.05) than yields of 307 ± 17.4 and 364.8 ±40.3 obtained from cow manure and maize bran respectively.

There was significant difference in the protein content of HFM harvested from three culturing substrates (P<0.05) isshown in Table 4. HFM from cow manure had significantly higher (P<0.05) crude protein content than those from chicken manure and maize bran substrate. Crude fibre and ether extracts of the maggots produced from all culturing substrates had no significant differences (P>0.05). The maggots from chicken manure had significantly higher (P<0.05) ash content than those from fermented maize bran and cow manure. The maggots harvested from cow manure and fermented maize bran was creamy in color with exception of those from chicken manure observed to be black in color.

Table 4: Chemical Composition of HFM meals (means±SE)

	Substrate		
Item (%)	**Cow Manure[1]**	**Fermented Maize bran**	**Chicken Manure**
Dry Matter	97.52 ±0.47[a]	96.42±1.47 [a]	96.71±0.08 [a]
Crude Protein	48.55±0.81[a]	42.63±0.23[b]	40.43±0.21[c]
Crude Fibre	5.71±0.25[a]	5.02±0.25[a]	6.00±0.25[a]
Ether Extract	19.07±0.46[c]	20.40±0.42[c]	20.00±0.06[c]
Ash	11.13±0.23[a]	10.70±0.48[a]	18.47±0.19[b]

[1]Maggot meal used in the formulation of fish diets
Means with different superscript letters within a row are significantly different at P<0.05.

4.1.2 Earthworm Meal (EWM)

Earthworm yields of 442.3±60.4from cow manure and of 421.0±25.7from rabbit manure were not significantly different (P>0.05). Earthworm yield 286.7±13.6 of chicken manure was lower than that from cow and rabbit manures. Earthworms cultured in cow manure had significantly higher (P<0.05) crude protein content than those from rabbit and chicken manure substrates as shown in Table 5. Percentage ether extract and ash contents of earthworms meals from all culturing substrates was not significantly different (P>0.05).

Table 5: Chemical Composition of EW meals (means±SE)

	Substrate		
Item (%)	**Cow manure[1]**	**Chicken manure**	**Rabbit manure**
Dry matter	95.02±0.96[a]	97.20±0.47[a]	95. 26±0.67[a]
Crude protein	48.61±0.18[a]	40.83±0.43[b]	39.80±0.41[b]
Ether extract	6.80±0.49[a]	5.60±0.22[a]	5.23±0.12[a]
Ash	28.60±0.11[a]	28.77±0.48[a]	29.77±0.10[a]

[1]Earthworm meal used in the formulation of fish diet
Means with different superscript letters within a row are significantly different at P<0.05.

4.1.3 Chemical Composition of Feed Ingredients and Experimental Diets

Percentage crude protein of experimental fishmeal was higher than EWM and HFM meal (Table 6). Cottonseed meal had higher percentage crude fibrecontent than any other ingredient.Crude protein contents of formulatedexperimental diets were within the recommended(not more than 30% CP) protein required for the Nile tilapia fingerlings to grow. Percentage crude fibre (4.22 to 8.17% CF) and ash (5.03 to 12.96%Ash) contents of experimental diets werebetween a range accepted in Nile tilapia diet while; ash contents of all diets increased with an increase in inclusion levels of the HFM and EWM (Tables 6 and 7). Gross energy of study diets ranged from 18.07 to 18.99 (Kcal/g).

Table 6: Chemical composition and gross energy of the feed ingredients used in the formulation of *O. niloticus* diets

	Ingredients			
Item (%)	**Fish Meal**	**Maize Meal**	**Wheat Meal**	**Cotton Seed Meal**
Dry matter	98.96	88.01	96.9	97.50
Crude Protein	69.20	10.5	11.74	41.60
Ether Extract	10.28	3.60	1.80	8.5
Crude Fibre	1.0	2.3	2.31	14.37
Ash	22.76	1.30	1.91	6.70
Nitrogen free extract[1]	2.38	84.30	79.15	23.34
Gross energy(Kcal/g)	18.99	18.03	18.10	18.88

[1] Nitrogen free extract + fibre (NFE) = 100 - (% protein + % fat + % ash).

Table 7: Chemical composition of diets contained different inclusion levels of HFM meal fed to *O. niloticus*

	Diets				
Item (%)	**HFM0**	**HFM25**	**HFM30**	**HFM35**	**HFM40**
Moisture	6.45	7.29	8.52	9.98	6.93
Crude Protein	30.39	30.19	30.18	30.20	30.25
Ether Extract	8.49	10.42	10.50	10.80	11.90
Crude Fibre	8.17	5.20	4.89	3.45	2.43
Ash	5.03	6.52	7.95	9.80	11.12
Nitrogen Free Extract	46.86	43.99	43.41	42.19	38.84
Gross Energy	18.35	18.07	18.34	18.77	18.85

Table 8: Chemical composition of the diets contained different inclusion levels of EW meal fed to *O. niloticus*

	Diets				
Item (%)	**EWM0**	**EWM25**	**EWM30**	**EWM35**	**EWM40**
Moisture	6.45	4.21	4.21	7.50	8.10
Crude Protein	29.59	30.31	30.31	29.58	29.80
Ether Extract	8.49	10.34	9.34	9.53	9.84
Crude Fibre	8.17	4.76	3.76	3.00	2.22
Ash	5.03	7.90	7.90	11.80	12.96
Nitrogen Free Extract	46.86	41.61	41.61	40.32	39.10
Gross Energy	18.35	17.21	17.21	16.30	16.07

4.2 Effect of Diets on Performance and Nutrient utilization of *O. niloticus*

4.2.1 Housefly Maggot Meal (HFM) Diets

Average body weights of fish fed diets containing different inclusion levels of HFM meal during the experiment is shown in Figure 3. Difference in body weightsbetween control diet and other diets with different inclusion levels of the HFM meal was noticeable after one week. Weekly body weightsof fish fed HFM0 diet was lowcompared toother diets throughout the experimental period. On the other hand, fish fed diet HFM0 and HFM40 had more less the same averagebody weights.

Growth performance i.e. body weight gain, average daily weight gain, specific growth rate; feed intake and feed utilization i.e. feed conversion ratio, protein efficiency ratio of experimental *O. niloticus* fed HFM based diets are shown in Table 9. There was no significant difference ($P>0.05$) in the initial weight, final weight and body weight gains of the fish fed housefly maggot based diets (Appendix 1). The diets had a significant effect ($P<0.05$) on average daily weight gain (ADWG), feed intake (FI) and feed conversion ratio (FCR). Fish fed diet HFM30 had significantly higher ($P<0.05$) ADWG, while highest PER value was observed from fish fed HFM35. The lowest FI was observed in HFM0 and HFM40. The means for specific growth rate (SGR) was significantly different ($P<0.05$) among diets. Fish fed diets HFM30 and HFM35 had significantly ($P<0.05$) higher SGR and PER. The FCR also differed significantly ($P<0.05$) among dietary treatments, with lowest FCR observed on the fish fed HFM35 and HFM30. On the other hand, survival did not show any significant difference among the different dietary treatments.

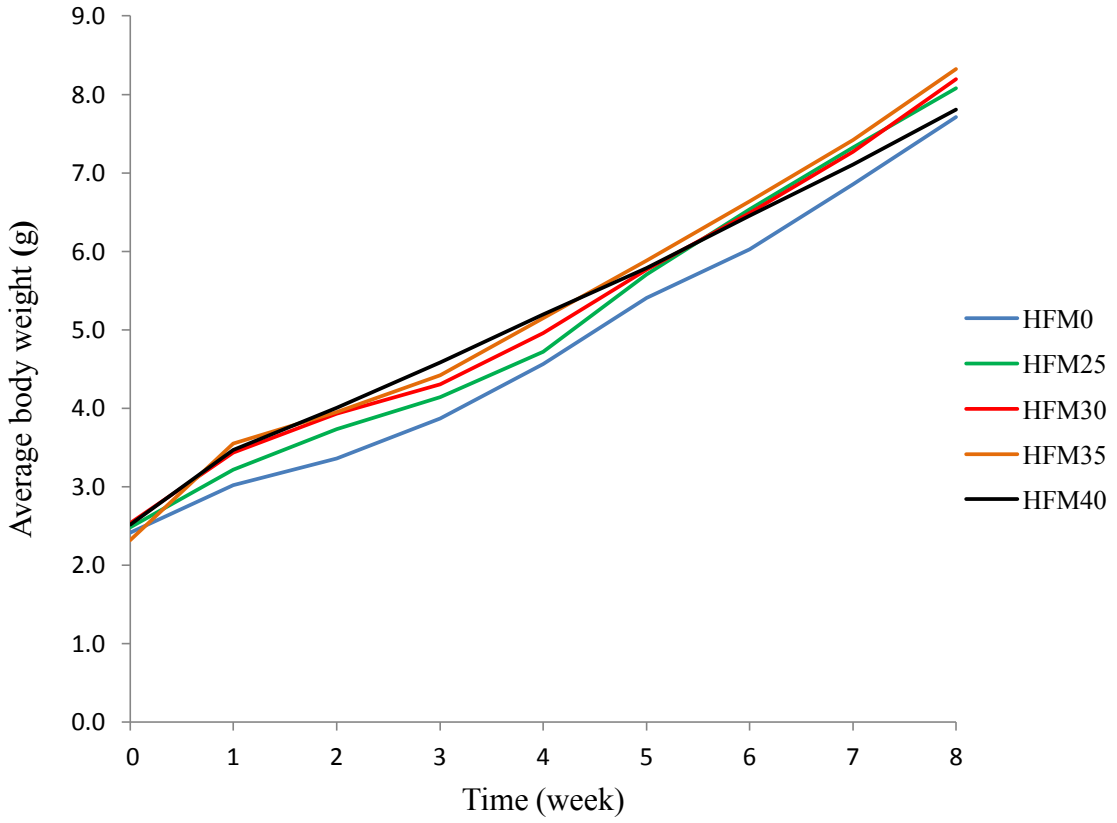

Figure 3: Average Weight Gain of *O. niloticus* fed different Inclusion levels of HFM based Diets

Table 9: Effects of the diets contained Housefly maggot meal on growth performance and nutrient utilization of *O. niloticus* fed different diets at different inclusion levels (mean±SE)

	Diets				
Parameter	**HFM0**	**HFM25**	**HFM30**	**HFM35**	**HFM40**
Initial body weight (g)	2.41±0.17^{a}	2.48±0.11^{a}	2.54±0.12^{a}	2.32±0.08^{a}	2.52±0.04^{a}
Final body weight (g)	7.71±0.07^{a}	8.07±0.19^{a}	8.19±0.13^{a}	8.33±0.20^{a}	7.81±0.37^{a}
Body weight gain(g)	5.30±0.25^{a}	5.59±0.08^{a}	5.65±0.20^{a}	5.01±0.27^{a}	5.29±0.34^{a}
Average daily weight gain(gday^{-1})	0.096±0.004^{b}	0.100±0.0041^{a}	0.101±0.001^{a}	0.100±0.005^{a}	0.091±0.006^{b}
Feed intake (gfish^{-1}day^{-1})	0.22±0.01^{b}	0.26±0.01^{ab}	0.30±0.01^{ab}	0.28±0.01^{a}	0.24±0.01^{b}
Feed conversion ratio	2.47±0.01^{c}	2.24±0.07^{abc}	2.05±0.11^{b}	1.85±0.04^{a}	2.66±0.16^{dc}
Specific growth rate	2.00±0.15^{a}	2.11±0.04^{a}	2.18±0.06^{b}	2.16±0.09^{a}	1.88±0.06^{b}
Protein efficiency ratio	1.35±0.01^{b}	1.49±0.05^{b}	1.64±0.09^{ad}	1.81±0.03^{a}	1.26±0.08^{c}
Survival (%)	88.1±8.6^{b}	97.6±2.4^{a}	95.2±4.8^{a}	97.6±2.4^{a}	95.2±2.1^{a}

Means with different superscript letters within a row are significantly different at P<0.05.

4.2.2 Earthworm Meal (EWM) Diets

Average body weights of *O. niloticus* fish in all dietary treatments is shows in figure 4. Body weightsof experimental fish were more less the same throughout the study. Up to the end of experiment there was no significant difference in the body weightsamong the fish fed diets EWM25, EWM30 and EWM35. The average body weights of the fish fed diets EWM0 and EWM40 was significantly lower than the average body weights of the fish fed diets EWM25, EWM30 and EWM35

Growth performance i.e. body weight gain, average daily weight gain and specific growth rate; feed intake and feed utilization parameters i.e. feed conversion ratio and protein efficiency ratio of *O. niloticus* fed diets with different inclusion levels of EWM are shown in Table 10. There was significant difference ($P<0.05$) in feed intake (FI), final body weight, average daily weight gain (ADWG) and specific growth rate (SGR) among dietary treatments. There was no significant difference in growth performance of the fish fed EWM35 and EWM30. Feed conversion ratio (FCR) and protein efficiency ratio (PER) differed significantly ($P<0.05$) among dietary treatments. Fish fed diet EWM35 and EWM30 had significantly lower ($P<0.05$) FCR and higher PER compared to others. Percentage survival of experimental *O.niloticus* in all dietary treatments was not significantly different ($P>0.05$). Both feed EWM35 and 30 showed higher performance since there was no significant difference in growth variables between fish fed EWM35 and EWM30.

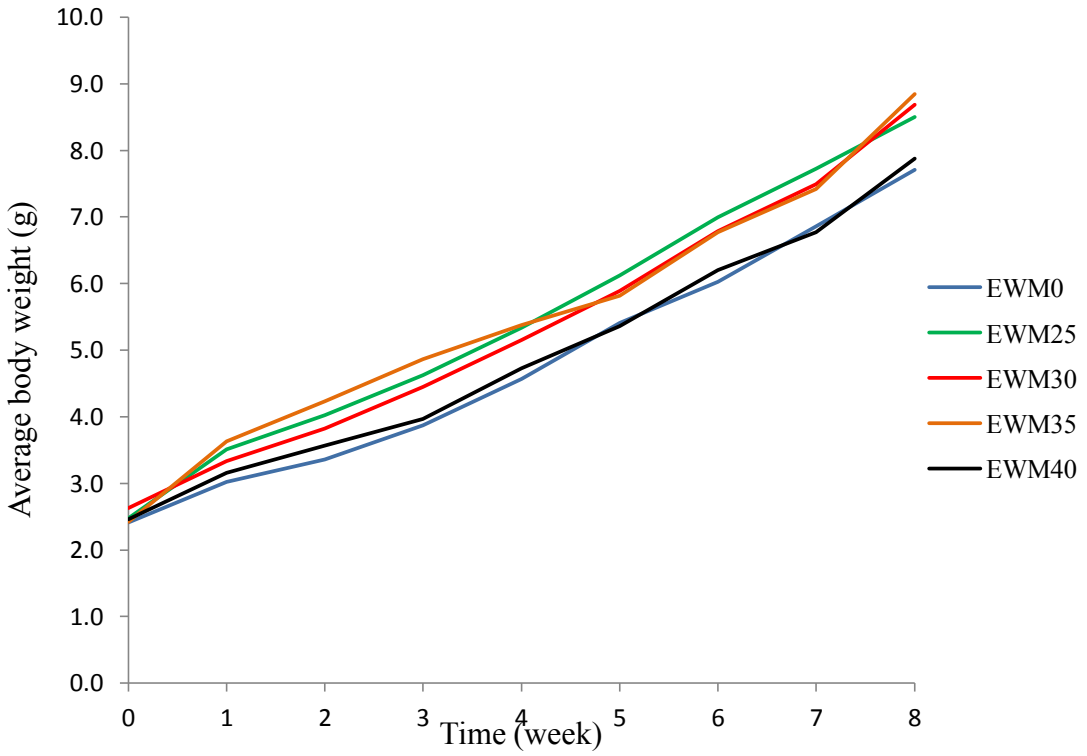

Figure 4: Average weights gain of *O. niloticus* fed different Inclusion levels of EWM based diets.

Table 10: Effects of diets contained earthworm meal at different inclusion levels on growth and nutrient utilization of *O. niloticus* (mean ± SE)

	Diets				
Parameter	**EWM0**	**EWM25**	**EWM30**	**EWM35**	**EWM40**
Initial body weight (g)	2.41±0.18^{a}	2.48±0.05^{a}	2.42±0.08^{a}	2.43±0.05^{a}	2.46±0.40^{a}
Final body weight (g)	7.71±0.071^{c}	8.50±0.28^{b}	8.84±0.48^{a}	8.92±0.06^{a}	7.71±0.02^{c}
Body weight gain	5.30±0.25^{c}	6.02±0.25^{b}	6.42±0.44^{a}	6.49±0.10^{a}	5.25±0.38^{c}
Average daily weight gain(gday^{-1})	0.096±0.004^{d}	0.104±0.005^{c}	0.115±0.008^{b}	0.118±0.002^{a}	0.096±0.007^{d}
Feed intake (gfish^{-1}day^{-1})	0.22±0.01^{c}	0.24±0.01^{ab}	0.26±0.01^{a}	0.28±0.01^{a}	0.23±0.01^{c}
Feed conversion ratio	2.47±0.14^{c}	2.22±0.07^{b}	2.10±0.09^{a}	1.85±0.31^{a}	2.43±0.04^{c}
Specific growth rate	2.11±0.15^{c}	2.26±0.05^{b}	2.31±0.08^{a}	2.38±0.05^{a}	2.18±.31^{c}
Protein efficiency ratio	1.35±0.01^{c}	1.57±0.05^{c}	1.80±0.21^{b}	1.81±0.07^{a}	1.68±0.19^{b}
Survival (%)	88.1±8.6^{a}	97.6±2.4^{a}	97.6±2.4^{a}	97.6±2.4^{a}	92.9±0.0^{a}

Means with different superscript letters within a row are significantly different at P<0.05.

4.3 Cost Effectiveness

Cost effectiveness in Tanzanian shillings (TZS) in terms incidence cost and profit indexof HFM and EWM diets is shown in Table 11 and 12. The cost of producing a Kg fish fed on HFM25, HFM30, HFM35 and HFM40 was lower than the cost of producing a Kg of fish fed on control diet. Cost effectiveness of experimental diets formulated with HFM based meal was significantly different ($P<0.05$) among dietary treatments. Among all dietary treatments, cost to produce a 1Kg fish was significantly low with diet HFM35 compared to other diets ($P<0.05$). Diets HFM35 and HFM40 demonstrated slightly low incidence cost and high profit index compared to control diet. There was a slightly low increase in the cost of feeds as inclusion level of EWM increased in the diets up to 40%. Diet EWM0 had significantly ($P<0.05$) high feed costs. Cost effectiveness increased slightly with an increase in the level of EWM. The differences in the cost effectiveness varried among dietary treament, and diet EWM35 had significantly ($P<0.05$) low cost to produce a Kg of *O. niloticus*than other diets. The lowest incidence cost was shown by diets EWM25 and EWM30, while all diets with eathworm meal had important effect on the profit index.

Table 11: Cost effectiveness of practical diets containing HFM meal fed to *O. niloticus* reared in tanks for 8 weeks.

	Diets				
Item	HFM0	HFM25	HFM30	HFM35	HFM40
Costoffeeds (TZS/Kg)	1509	1374	1359	1329	1329
Costeffectiveness (TZS/Kg of fish)	3727.2 ± 215.9^{c}	3077.4 ± 7.6^{b}	2779.2 ± 143.2^{ab}	2453.41 ± 47.2^{a}	3534.8 ± 102.6^{c}
Incidence cost	1.51	1.37	1.36	1.33	1.33
Profit index	0.66	0.73	0.74	0.75	0.75

Table 12: Cost effectiveness of practical diets containing EW meal fed to *O. niloticus* reared in tanks for 8 weeks.

	Diets				
Item	EWM0	EWM25	EWM30	EWM35	EWM40
Cost of feeds (TZS/Kg)	1509	1419	1419	1434	1449
Cost effectiveness (TZS/Kgof fish)	3727.2±215.9^{d}	3150.2±7.6^{b}	2979.8±348.7^{ba}	2659.8±104.7^{a}	3521.1±280.4^{c}
Incidence cost	1.51	1.42	1.42	1.43	1.45
Profit index	0.66	0.70	0.70	0.70	0.70

CHAPTER FIVE

5.0 DISCUSSION

5.1 Suitable Substrate for Culturing Housefly Maggots and Earthworms

5.1.1 Housefly Maggots

The higher yield of maggot from chicken manure substrate was probably due to long lasting odor (Nzamujo, 1999; Agbeko*et al.,* 2014) which strongly attracted the flies. Similar result was reported by Calvert (1979) and Patricia and Salas, (2007) where chicken manure producedlarge quantity of the maggots. Nzamujo (1999)and Agbeko*et al.* (2014) reported that the more the quantity and long lasting odor of substrate, the more number of flies and the greater the number of maggots produced.

Results of proximate composition of maggots produced from all substrates were within the range reported elsewhere (Nzamujo, 1999; Fasakin*et al.,* 2003;Idowu*et al.,* 2003; Aniebo *et al.,* 2009;Hwangbo *et al.,* 2009; Odesanya *et al.,* 2011). The mean crude protein (43.87%CP) of the housefly maggots from all three culture substrates produced in the present study was higher than 37.5% and 28.63%CP reported by Ogunji*et al.* (2006) and 22.97% reported by Omoyinmi *et al.*(2012). Crude protein contents of produced maggots fell within a range of 40 to 61.4% as previously reported by other workers(Nzamujo, 1999; Adeniji, 2007;Odesanya*et al.,* 2011). Crude protein contents of HFM meals produced during the study were differently affected by culturing substrates. Cow manure was very efficient in influencing the crude protein content which reached 48.55%. This agrees Ajonina and Nyambi (2013) and Agbeko*et al.* (2014) that animal manures are appropriatein producing the maggots with high crude protein content.Patricia and Salas (2007)reported that protein content depends mainly on the relative nutrient content present in the substrate and ability of the organisms to feed and utilizing it.Patricia and Salas(2007) and Aniebo *et al.*(2009) also reported that

cow manure has the ability of influencing microbial activity. The highest crude protein obtained in this study was more than crude protein contents reported byAttey and Ologbenla (1993), Fasakin*et al.* (2003) and Anieb*oet al.* (2009). However, it is far from Hwangbo *et al.* (2009)who reported the maggot protein content of 63%.In contrast to Sogbesan*etal.* (2006) and Odesanya *et al.* (2011) chicken manure used in this study produced the HFM with relatively low protein content (40.43%CP). Probably chicken manure was of low quality. Lowered protein content of the maggots produced from chicken manure agrees with Horn (1998) who reported the negative impact of age of manure and storage time on the nutrients content of manure and its compositional effect on the invertebrates fed on that manure. The author reported reduction on the amount of nutrients in the fresh manure with time. This is due to the loss of Nitrogen after itsconversion into ammonia and then lost to the atmosphere. The collected chicken manure in the present study was black in color. According to Merry and Mcallan (1883) and Claudio and Patricia (2007) is a result ofdark color of manure. Low crude protein content housefly maggot cultured on chicken manure was reportedbyOgunji*et al.* (2006).Fasakin*et al*. (2003) reported that crude protein content of maggot may be related to the quality of substrate used to produce the maggot meal.

Percentage ether extracts of the HFM meals for present study were closely to 19.3%EE(Nzamujo, 1999) and 19.8%EE(Ogunji*et al.,*2006) but higher than 10%EE and 9.67%EE reported by Omoyinmi*et al.* (2005) and Okah and Onwujiariri (2012) respectively.Also, percentage ether extract of present study was lower than 23.3%EE (Ogunji, 2008),24.31% EE (Hwangbo*et al.,* 2009), 25.3%EE (Anieb*oet al.,*2010), 31.76% EE (Odesanya*et al.,* 2011) and 28.95%EE (Osse*yet al.,* 2014).

Ash content of the experimental housefly maggot mealfound in the present study was at a range of 10 to 19%.The results agree with those reported by Yaqub (1999),Nzamujo (1999) and Okah and Onwujiariri (2012). Ash content of maggots produced from cow manure and fermented maize bran from the present study was the similar to that reported by Sogbesan (2006) and Odesanya*et al.* (2011). Elevated ash content in the maggot meal from chicken manure might be due to the lower quality of the chicken manure used during the study.

5.1.2 Earthworms

The high yield of earthworm meals observed from cow manure in the present study is supported by the findings ofCooper *et al.* (1970), Sogbesan and Ugwumba (2006) and Jesikha and Lekeshmanaswamy (2013). Similarly, earthworm yield from rabbit manure agrees with findings bySherman (2003). Also Edwards and Niederer (1988) and Ismail (2005)reported that cow and rabbit manure werebest growing substrates for different worm species like *Lumbricusrubellus* and *Dendrobaenarubida*. In the present study culture substrates containing cow manure and rabbit manure produced relatively largequantities of earthworms. High yields from these substrates weredue to the effect of great number of young earthworms observed during culture period. Theseresults suggestthat both cow and rabbit manures are good substrate to influence high yields of earthworms (Copper, 1970; Dominguez *et al.,*2001; Ismail, 2005; Mounroe, 2007;Jesikha and Lekeshmanaswamy,2013). Low yield of earthworm obtained from chicken manure substrate was a result of low production of new young earthworms.Sherman (2003) reportedchicken manure tend to generate heatdue to high concentration of salts and ammonia with negative effects onearthworms. The effect is more pronounced on young earthworms thus only few young earthworms are able to survive and grow in the chicken manure substrate (Sherman, 2003 and Sbryce, 2013).

The present study investigated the possibility of using different culturing substrates to enhanceproduction of earthworms with different protein contents. Results of the study show that among all substrates, cow manure resulted in earthworms with highest crude protein content. Crude protein content of earthworms (48.61%CP) produced from cow manure in the present study was higher than 46.57%CP, 47.43%CP and 44.3%CP reported by Hasanuzzamanand Das (2010), Jabir *et al.* (2012) and Puche*et al.* (2014) respectively. But lower than protein content reported by Nguyen *et al.* (2010). During this study the highest protein content of earthworm produced from cow manure substrate probably due to its relative high nutrient contents generated during microbial protein synthesis (Merry and Mcallan, 1983;Munroe, 2007;Rana, 2007; Nguyen *et al.,* 2010) and ability of the worms to feed on it. Sogbesan and Ugwumba (2006), Rana (2007) and Nguyen *et al.* (2010) reported that the quality of different substrates influence differently composition processes through microbial activities.

Earthworm meal from rabbit manure had low crude protein despite the fact that rabbit manure is richer in Nitrogen (Smith, 2001; Sherman, 2003). This might be due to the lowered quality of rabbit manure. Results of the present study agrees with those byHorn (1998)who reported the effect of manureto organism produced on itand poor manure storage (timetaken to store and management) on the composition of manure. However, chicken manure which is also rich in nitrogen content produced earthworm with least protein content. This is because chicken manure was not a good medium for microbial activity (Nguyen *et al.*, 2010). Sherman (2003), Munroe (2007) andDominguez and Edwards (2011)reported factors such as quality and quantity of fertilizer, species specific, seasons, life stages, specificecology and processing methods used during preparation of earthworm meals to influence the quality of the worm meal.Despite protein contents of earthworm from rabbit and chicken manure being low, these values

were slightly higher than 31.7%CP,12% CP, 10.5% CP and 44.3% CP reported by Lourdumary and Uma (2013), Yaqub(1999), Finke (2002) and Puche*et al.* (2014) respectively. However, the crude protein content of earthworm meals of the present study was lower than reported range of 60 to 70% CP (Tacon*et al.,* 2006).

The mean crude lipid of experimental earthworm meals in the present study (5.88%EE) was similar to the values reported by Sogbesan*et al.* (2006) and Puche*et al.* (2014). Thus, nutritional variations may be because of differences in the species of the earthworm used. Therefore, results of present study agrees with previous reports that nutritional content of earthworm meals varies due to the number of factors as discussed bySherman (2003), Munroe (2007) and Dominguez and Edwards (2011).

The mean ash content of experimental earthworm meals was lower than 45.7% and 39.53% reported by Barker *et al.* (1998) and Dedeke *et al.* (2013) respectively, but, higher than the ash content ranges from 6.0 to 8.9% (Yaqub, 1999;Sogbesan*et al.,* 2006; Olele, 2011; Adesina, 2012). This implied that experimental EWM had optimal ash content that can not affect other nutrientsinthe diets (McClements, 2005, 2007 and 2009). Generally, nutrient contents of experimental earthworm meals were adequate to be used as the major source of protein in tilapia diets.

5.2 Effects of Inclusion of HFM and EWM on performance of *O.niloticus*

5.2.1 Growth Performance

The increase in the body weight gain, average daily weight gain and specific growth rate of *O. niloticus*fed on HFM and EWM inclusion of 25, 30 and 35% implies that maggot andearthworm meal can successfully be included in the diets of Nile tilapia up to 35%. Higher growth perormance of *O. niloticus* fed on HFM and EWM diets reflects nutritional quality and acceptance of the diets as well as good utilization of the meals at

agiven level (Idowu *et al.,* 2003). This may be because of both reduced fibre content and the combining effect of three sources of protein from fishmeal, cottonseed meal and either HFM or EWM in the diet (Ogunji and Manfred, 2001; Olvera-Novoa *et al.,* 2002). These authors reported that, the decrease in fibre content inhances the acceptability of feed to fish that leads to increased feed utilization and growth performances of fish. Also synergisticeffect of three crude protein sources from fishmeals, cottonseed meal and HFM imporoved the quality of these diets.The authors added that, the synergisticeffect of more than two protein sources for formulating a single diet produce a good quality diet with well balanced nutrients. Theresults of present study is corroborated by those from Hilton (1983), Sogbesan*et al*. (2006) and Khan *et al.* (2013)thata combination of different nutritional ingredients of diets produce superior single diet for better growth performance of fish.

It has been reported that the nutritional value of maggot meal is equivalent to that of fishmeal (Ajani *et al*., 2004), while Olele (2011) reported adequate amino acid contents in EWM meals that have the similar effect to fishmeal on the performance of fish.Ogunji*et al.* (2006) andMakkar*et al.* (2014) added that, the nutritional value of maggot meals were equivalent to that of whole fishmeal. Interestingly, the larvae of maggots does not contain any anti-nutritional or toxic factors often found in alternative plant protein sources (Ogunji*et al.,* 2006; Makkar *et al.,* 2014). The nutrients contained in EWM which was limiting in most of ingredients were qualified as growth promoting factors that are highly needed for cultured fish. This fact is supported by the observed increase in growth performance and good feed utilization observed in dietary treatments that contained 35% level of either HFM or EWM. The results are also supported by those reported by Olele (2011), Jabir *et al.* (2012), Monebi and Ugwumba (2013) and

Dedeke*et al.* (2013) who observed positive response of the inclusion levels of these meals on growth performance i.e. ADWG and SGR of experimental *O. niloticus*.

Nevertheless, there was a decline in body weight gain, average daily weight gain and specific growth rate of the fish fed diets with 40% inclusion level of either HFM or EWM. Poor growth performance of fish fed diets HFM40 and EWM40 contrasts with the findings of Yaqub (1999), Ogunji *et al.* (2006) andAnieboet *al.* (2010) who recommended feeding fish up to 100% inclusion level of these invertebrates. These results agree Dedeke *et al.* (2013), Olaniyi and Salau (2013) and Pucher *et al.* (2014) who reported negative impact of HFM and EWM ongrowth performance of fish if high level included in the diet.

Increased use of HFM in experimental diets could have led to the increased chitin content that caused adverse effect on growth performance of fish. The results of present study are in agreement with Olaniyi and Salau (2013) that, the HFM can not be used as a whole protein source in the diet of fish because it may contain chitin from their exoskeleton. On the other hand present results is supported by those of Ogunji *et al.* (2006) who reported the reduced growth performance of *Carassius auratus* when fed at high level of housefly maggot meal. The author also added that chitin was difficult to be digested by fish and considered to be the anti-nutritional factor that affects the animal's digestion and absorption functions.

Moreover, poor growth performance of the fish fed diets EWM40 might be caused by low feed intake of fish fed this diet. Palatability is one among the factors affecting feed intake of fish. Medina *et al.* (2003) and Barroso *et al.* (2014) reported unpalatability of the diet with high inclusion of earthworms due to presence of ceolomic fluid. This fluid

is unpalatable to fish, accordingly, causes poor feed intake and feed utilization hence poor average daily weight gain of fish. This observation is in agreement to the report of Rameshguru and Govindarajan (2011) that, earthworm meal has ceolomic compounds which if not well mixed with different feed ingredients during feedpreparation of fish diets acts as toxins to fish. Poor growth of fish such as trout fed on high levels of earthworm meal was reported by Tacon (1983). This was attributed to the presence of ceolomic fluid and high inclusion levels of earthworm meal which was unfavourable to fish (Tacon, 1983; Medina *et al.,* 2003). Thus, the decrease in growth performance of the fish with increased inclusion level of EWM in the fish diet might be associated by the effect of ceolomic fluid. Additionally, reduced performanceof *O. niloticus* fed on HFM40 and EWM40 diets was perhapsdue to the effect of imbalanced nutrientscontributed by few feed ingredients. Present results support the reports of Hilton (1983), Sogbesan*et al*. (2006), Khan *et al.* (2013) and Makkar *et al.* (2014) that combination of different ingredientsproduce a single superior diet for better fish growth performance.

5.2.2 Feed Intake

Comparable feed intakes between the control diet and diets with 35, 30 and 25% HFM and EWM was probably due to the effect of frequent feeding response of fishto feeds shown by the fish on these dietary treatments throughout experimental period. Fish fed diets with 25, 30 and 35% inclusion levels showed high feed intake probably due to palatability effect of diets. Meena (2015) reported palatability of the diet as among the factors that largely affect acceptance of feedto fish. Ingredients based on animal protein rather than plant proteins sources have great effect on improving palatability of the diets. Thus, high feeding intake shown by experimental fish fed diets 35% inclusion either HFM35 or EWM35 was a reflection of the meals palatability and nutritive quality

of these biomaterials. In addition, palatability of these diets might be contributed by the combined effect of fishmeal, cottonseed meal and HFM or EWM as previously reported by Hilton (1983), Sogbesan*et al*. (2006), Khan *et al.* (2013) and Makkar *et al.* (2014). Nevertheless, fish fed diets with 40% inclusion level of either HFM or EWM had more or less the same low feed intake as those fed control diet. These results differ from Yaqub (1999),Ogunji*et al.* (2006) and Monebi and Ugwumba (2013) who reported that FI of Nile tilapia improved when EWM was included in the diet of fish up to 100%. Diet HFM40 would have been expected to have shown the best feed intake since it contains neither compound which was unpalatable for fish but this was not so. However, FI of the fish fed diet EWM40 diet might be a result of unpalatable coelom fluid available in the earthworms(Tacon, 1983; Medina *et al.,* 2003;Coyle *et al.,* 2004; Kostecka and Paczka, 2006; Dedeke*et al.,* 2010).Similar findings was reported byHilton (1983) that the vertebrates such as fish shows poor acceptability of feed when fed diets contained high inclusion level of earthworm meal.

5.2.3 Feed Utilization

The decrease in FCR as the level of either HFM or EWM increased up to 35% indicate good feed utilization. The lower the FCR the more efficient the conversion efficiency i.e. better utilization of the feed by the fish. Thus, FCR observed on fish fed diets HFM30, HFM35 and EWM35 indicated good FCR of *O. niloticus*.During present study FCR of fish fed with experimental diets was in agreement with Ogunji *et al.* (2006), Ogunji *et al.* (2008),Jabir*et al.* (2012),Omoyinmi (2012) and Mekhamar *et al.* (2015).However, FCR of the fish fed diets included with HFM and EWM was than those reported by Mohanta *et al.*(2013)and Olaniyi and Salau (2013) whosevalues ranged from3.13 to 5.07.Contrarily, the FCRof present studywas better than that reported by Yaqub (1999) who fed fish with HFM and EWM.

During the present study, the PERs were increasing with increased HFM and EWM level in the diet up to 35%. Increase in PER observed through out experimental period was because of decreased crude fibre content andadded nutritive quality as well as acceptance of fish tothe experimental feed ingredients used in formulation of the diets. The present results arein agreementwith previous findings which showed that maggot and earthworm meals were well accepted and utilized by the fish (Jonathan, 2012;Monebi and Ugwumba, 2012;Omoyinmi *et al.,* 2012). It has been suggested that the good nutrient utilization capacity of fish fed insect based diets stem from the high nutritional value and digestibility of the ingredient (Sogbesan*et al.,* 2006). According to Coyle *et al.* (2004) inclusion level of HFM in the fish diet produced the highest PER due to protein contents of these meals. This was in accordance with the findings of Odesanya*et al.* (2011),Ossey *et al.* (2014),Pucher *et al.* (2014) and Vodounnou *et al.* (2015) who reported that the FCR and PER of Nile tilapia were slightly improved when fed the EWM diets contained 25 and 50% inclusion levels. But the trends of an increase in PERs obtained under this study were different from Monebi andUgwumba (2012)who reported a decreased PER above 25% EWM inclusion level.Use of HFM and EWM in the diet of *O. niloticus* up to 35% has no adverse effect on feed utilization of the fish. But above that level there was a decrease in PER of the fish. This agrees with the findings ofStafford and Tacon (1988), Dedeke*et al.* (2010), Hasanuzzaman and Das (2010) and Sogbesan (2014)who advised to include earthworm meal in the diets of the fish in order of improving their feed intake and utilization for better growth performance.

5.3 Cost Effectiveness of Experimental Diets

Based on cost effectiveness, maggot and earthworm meals is a good protein source in the diets of the fish. The results of this study have shown that the cost per Kg

production of HFM and EWM meal was low, and the cost of producing diets decreased with an increase in inclusion levels (Okah and Onwujiariri, 2012).The presentresults agree with those of Sogbesan and Ugwumba (2006), Olele and Okonkwo (2012), Ossey *et al.* (2014) and Sogbesan (2014) whominimizedproduction cost of 1Kg HFM and EWM if compared to the production of 1Kg of fish meal.Cost of production of fish could also be reduced due to the diets formulated with housefly maggots and earthworms being sourced at little costs(Barry *et al.,* 2004; Sogbesan, 2014;Okah and Onwujiariri, 2012; Olele and Okonkwo, 2012; Sogbesan, 2014).Based on formulated diets inclusion of HFM and EWM at 35% had more effect on reducing cost of producing a Kg of diet.During present study the diets with 35% inclusion levels of HFM and EWM had optimized cost effectiveness. The optimized cost effectiveness of these diets was due to the lowered feed conversion ratioMgheni and Christensen (1985). The least cost per Kg of fish observed at 35% inclusion levelsof HFM35 and EWM35 was an effect ofminimized cost of HFM and EWM meals (Newton *et al.,*2005; Aniebo*et al.,* 2010; Devic*et al.,* 2013). This finding agrees with Ijaiya and Eko(2009) and Olele (2014) that, cost effectiveness and crude protein contents could be potential in reducing production cost of cultured fish.

All inclusion levels of HFM and EWMin all dietsof present study had the same effect in improving profit index on production of *O. niloticus.* This is an indication of the benefit economic efficiency and implied that feed cost was reduced with increase in the inclusion levels of either HFM or EWM (Omondi*et al.,* 2001; Sogbesan *et al.,* 2003; Ezewudo*et al.,* 2015). Several studies on utilization of insects and others invertebrates as alternative to fishmeal have demonstrated its beneficial effects on fish production. Present study agrees with Atteh and Ologbenla(1993), Adesulu and Mustapha (2000), Ogunji and Wirth (2004), Sogbesan*et al.*(2003), Nguyen and Davis (2009) and Ijaiya

and Eko(2009)that, there is economic benefit of using housefly and earthworm meals in fishdiets such as *O. niloticus.* Based on cost effectiveness, availability and high crude protein content of HFM and EWM,the diets seem to be potential for fish farmers similar to observations byBarbaroux*et al.*(2012) andDevic *et al.*(2014).

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1 Conclusions

Based on the findings obtained from the study the following conclusions are made;

i. Chicken and cow manure are good substrates in supporting high yields of HFM and earthworm meal respectively, while cow manure is good substrate to produce housefly maggots and earthworms with highest crude protein level.

ii. Inclusion of either HFM or EWM up to 35% donot compromise growth and feed utilization of *Oreochromis niloticus.*

iii. Inclusion of 35% of either HFM or EWM is most cost effective.

6.2Recommendations

In view of the present study the following recommendations are pointed out:

i. There is a need of sensitizing production of housefly maggots and earthworms to the small scale fish farmers in efforts to improve nutritive value of fish diets.

ii. Further research on HFM and EWM basing on different protein levels and combination with several other locally available protein sources should be explored.

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APPENDICES

Appendix 1: ANOVA tables for the Housefly Maggots based Diets

Dependent Variable: Yields

Source	Df	Sum of Square	Mean square	F value	Pr>F
Between groups	2	27011.3	13505.6	3.795	0.086
Within groups	6	21352.8	3558.8		
Total	8	48364.0			

Dependent Variable: %CP

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	2	74.736	37.368	49.502	0.000
Within groups	6	4.529	0.755		
Total	8	79.266			

Dependent variable: Initial number of fish

Source	Df	Sum of Squares	Mean Square	F value	Pr>F
Between Groups	4	0.000	0.000	.	.
Within Groups	9	0.000	0.000		
Total	13	0.000			

Dependent variable: Final number of fish

Source	Df	Sum of squares	Mean Square	F value	Pr>F
Between Groups	4	4.190	1.048	0.884	0.511
Within Groups	9	10.667	1.185		
Total	13	14.857			

Dependent variable: Initial weight

Source	Df	Sum of squares	Mean Square	F value	Pr>F
Between Groups	4	0.328	0.082	1.288	0.344
Within Groups	9	0.574	0.064		
Total	13	0.902			

Dependent Variable: FW

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	3.159	0.790	3.684	0.048
Within groups	9	1.929	0.214		
Total	13	5.088			

Dependent Variable: BWG

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	2.124	0. 531	2.119	0.161
Within groups	9	2.255	0.251		
Total	13	4.378			

Dependent Variable: ADG

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	0.001	0.000	2.119	0.161
Within groups	9	0.001	0.000		
Total	13	0.001			

Dependent Variable: FI

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	0.005	0.001	3.780	0.045
Within groups	9	0.003	0.000		
Total	13	0.008			

Dependent Variable: FCR

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	0. 715	0. 179	3.106	0.073
Within groups	9	0. 518	0.058		
Total	13	1.233			

Dependent Variable: SGR

Source	Df	Sum of Square	Mean Square	F value	Pr>F
Between Groups	4	0.096	0.024	0.540	0.711
Within Groups	9	0.400	0.044		
Total	13	0.496			

Dependent Variable: PER

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	213.8	53.45	0.884	0. 511
Within groups	9	544.2	60.469		
Total	13	758.0			

Dependent Variable: % Survival

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	4.000	1.000	0.787	0.559
Within groups	9	12.704	1.270		
Total	13	16.704			

Dependent Variable: Cost effectiveness

Source	Df	Sum of square	Mean Square	F value	Pr>F
Between groups	4	1910729.800	477682.450	9.961	0.002
Within groups	9	479538.484	47953.848		
Total	13	2390268.283			

Appendix 2: ANOVA tables for Earthworm based Diets

Dependent variable: Yields

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	2	42732.7	21366.3	4.758	0.058
Within groups	6	26943.3	4490.6		
Total	8	69676.0			

Dependent Variable: %CP

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	2	127.873	63.936	141.958	0.000
Within groups	6	2.702	0.450		
Total	8	130.575			

Dependent Variable: Initial number of fish

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	0.000	0.000	.	.
Within groups	9	0.000	0.000		
Total	13	0.000			

Dependent Variable: Final number of fish

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	4.190	1.048	0.885	0.511
Within groups	9	10.667	1.185		
Total	13	14.857			

Dependent Variable: IW

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	0.328	0.082	1.288	0.344
Within groups	9	0.574	0.064		
Total	13	0.902			

Dependent Variable: FW

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	3.159	0.790	3.684	0.048
Within groups	9	1.929	0.214		
Total	13	5.088			

Dependent Variable: BWG

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	2.124	0. .531	2.119	0.161
Within Groups	9	2.255	0. .251		
Total	13	4.378			

Dependent Variable: ADG

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	0.000	0.000	2.119	0.161
Within Groups	9	0.000	0.000		
Total	13	0.000			

Dependent Variable: FI

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	0.005	0.001	3.780	0.045
Within Groups	9	0.003	0.000		
Total	13	2.008			

Dependent Variable: FCR

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	0.715	0.179	3.106	0.073
Within groups	9	0.518	0.058		
Total	13	1.233			

Dependent Variable: PER

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	0.429	0.107	2.574	0.110
Within Groups	9	0.375	0.042		
Total	13	0.804			

Dependent Variable: SGR

Source	Df	Sum of square	Mean square	F value	Pr>F
Between Groups	4	0.096	0.024	0.540	0.711
Within Groups	9	0. 400	0.044		
Total	13	0. 496			

Dependent Variable: % Survival

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	213.800	53.450	0.884	0.511
Within groups	9	544.218	60.469		
Total	13	758.017			

Dependent variable: Cost effectiveness

Source	Df	Sum of square	Mean square	F value	Pr>F
Between groups	4	458306.02	114576.50	0.874	0.512
Within groups	9	1310510.47	131051.05		
Total	13	1768816.49			

Appendix 3: The housefly maggots on culturing substrate contained cow manure before harvest