COMPARATIVE GROWTH PERFORMANCE OF AFRICAN CATFISH (*Clarias gariepinus*) FINGERLINGS OFFERED DIFFERENT TARGET DIETS

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DECLARATION

This thesis is my original work and has not been presented for a degree or any other award in any University.

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This thesis is dedicated to my husband Tony Matanda and my children Faith Njeri and Fortune Masinde who have been a great encouragement to me. God bless you all.

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LIST OF ABBREVIATIONS AND ACRONYMS

AA	Amino Acids
ANOVA	Analysis Of Variance
AOAC	Association of Official Analytical Chemists
ATP	Adenosine Triphosphate
BSF	Black Soldier Fly
CP	Crude Protein
DE	Digestible Energy
DE/p	Digestible Energy to protein
DO	Dissolved Oxygen
ESP	Economic Stimulus Program
FAO	Food and Agriculture Organization of the United Nations
FCE	Food Conversion Efficiency
FCR	Food Conversion Ratio
FBW	Final Body Weight
H_2SO_4	Sulphuric Acid
HCL	Hydrochloric Acid
IBW	Initial Body Weight
ICIPE	International Centre of Insect Physiology and Ecology
NaOH	Sodium Hydroxide
$(NH_4)_2SO_4$	Ammoniun Sulphate
pH	Potential of Hydrogen
PUFA	Polyunsaturated Fatty Acids
SR	Survival Rate
SGR	Specific Growth Rate
t	time
TAN	Total Ammonium Nitrate
WG	Weight Gained

ABSTRACT

Aquaculture production is considered the future global solution to declining wild fish capture. However, insufficient production of fish due to lack of affordable quality feeds is a major challenge in aquaculture development. This study investigated the growth performance of fingerlings of African catfish (*Clarias gariepinus*) fed on experimental diets from sun-dried termites (*Macrotermes subhylanus*), cockroaches (*Periplenata americana*), larvae of black soldier fly (*Hermetia illucens*), desert locusts (*Schistocerca gregaria*) and polychaetes (*Marphysa mossambica*) and silver cyprinid (*Rastrineobola argentea*) as Control diet. Six experimental diets were formulated with three replicates per treatment to contain 40% wheat bran and 60% of sun-dried insects and worms. A total of 720 fingerlings, mean weight 0.362 ± 0.089g were obtained. The experiment was conducted for a period of two months (8 weeks). Water quality parameters were recorded twice a week. The nutritive value of the test diets was determined using Association of Official Analytical Chemists (AOAC) methods. The effect of different diets on specific growth rate (SGR%), survival rate (SR%) and water quality parameters was determined. Correlation analysis was done to determine the relationship between diet and tissue amino acids.

Results indicated that water quality parameters such as temperature, dissolved oxygen and PH were within the acceptable range. The nutritional composition of experimental diets ranged as follows; crude protein: 29.3% - 39.1%, ash: 8.9% - 21.3%, fat: 6.7% - 26.3%, moisture: 6.6% - 11.3%, fibre: 4.7 - 14.2% and carbohydrate: 12.9% - 26.4%. The energy content ranged from 281.1Kcal/100g - 409.5Kcal/100g. The amino acid analyses showed that the experimental diets and fish tissue contained six essential amino acids (histidine, isoleucine, leucine, methionine, phenylalanine, and valine) and six non-essential amino acids (arginine, glutamic acid, hydroxyproline, proline, serine and tyrosine). A highly significant positive correlation was observed between the dietary and the tissue amino acids for both essential and non essential amino acids.

Catfish fingerlings body weight increased progressively throughout the experimental period and was significantly higher at the end of the experiment (Mean= 1.1g) compared to the start of the experiment (Mean= 0.4g). Fingerlings fed on black soldier fly larvae recorded the highest weight gain (401.2%) while fingerlings fed on Polychaetes recorded the lowest (54.2%). No significant

variation ($P > 0.05$) in weight gain was however observed among the fingerlings fed on black soldier fly larvae, cockroach and silver cyprinid (Control diet).

The highest specific growth rate was observed on catfish fingerlings fed on black soldier fly larvae (2.64%) and the lowest on fingerlings fed on polychaetes (0.66%). There was no significance difference ($P > 0.05$) in specific growth rate of fingerlings fed on black soldier fly larvae and silver cyprinid. The survival rates of catfish fingerlings significantly varied among the different diets with the highest survival observed in fingerlings fed on cockroach diet (70.0%) and the lowest in fingerlings fed on termites (9.2%). No significant variation ($P > 0.05$) in survival rate was observed in fingerlings fed on termites and polychaetes.

From this study the diets derived from black soldier fly larvae, cockroaches, desert locusts and silver cyprinid contained the recommended range of nutrients necessary for growth of catfish fingerlings. The findings of this study suggest that black soldier fly larvae, cockroach and desert locust can potentially replace silver cyprinid as protein sources in the culture of African catfish. The aforementioned target organisms are cheaply available throughout the year and can be easily harvested by fish farmers for catfish rearing in ponds.

CHAPTER ONE: INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction

Aquaculture is an enterprise that is growing rapidly and improving the economy worldwide. Fish are cultivated for food and source of revenue for the growing human population, restocking of streams, lakes, and rivers to curb the shortage due to the decline in the wild capture and for sport fishing (FAO, 2000). Aquaculture aims at production of fish to provide protein in the diet (Sugunan, 2002). Fish is easily digestible, has ability to prevent and manage heart disorders and neurological diseases (Tan *et al*., 2007).

The global aquaculture fish production rose from 7 percent in 1973 to more than 30 percent in 1997 (Delgado *et al.,* 2003). In 2010, global aquaculture production reached 79 million tons, growing at an annual rate of 9.7 percent since 1998 (FAO, 2012). However, aquaculture global fish production stood at 66.6 million tons in 2012 (FAO, 2014). In 2014, world aquaculture production of fish accounted for 44.1% of total production from capture fisheries and aquaculture up from 42.1% in 2012 (FAO, 2016) and amounted to 73.8 million tons (OECD, 2016). In 2015, global aquaculture production reached 106 million tons, of live weight growing at an average annual rate of 6.6% since 1995 (FAO, 2017a). In sub Saharan Africa, aquaculture growth during 2001-2015 was averaged at 10.4 % (FAO, 2017a).

In Kenya, over 70% of fish and fish products consumed locally are from wild capture fisheries, principally Lake Victoria. “Fish production has increased over the last decade from 1,012 metric tons produced in 2003 to 21,487 metric tons in 2012” (Munguti *et al*., 2014). By 2013, Kenya’s total fish production was 152,711, tons of which 23,501 tons came from aquaculture (KNBS, 2014). However, aquaculture production registered a depressed performance with total fish output dropping by 19.8% to 14,952 tons in 2016 from 18,656 tons in 2015 in the country (World Bank, 2017). In Kenya, increasing fish farming is important since it provides a source of income and revenue to farmers and government, improves food security, creates job opportunities to the growing population, and optimizes use of water resource and conservation of biological diversity (Okechi, 2004).

Fish products play an essential role in food security and meeting the nutritional needs of the human population in developing and developed countries (FAO, 2014; Suvitha *et al*., 2015).

Fish is also an important source of micronutrients such as vitamins, minerals and polyunsaturated omega-3 fatty acids (FAO, 2012). Fish diets with adequate amount of protein are needed for growth, development, survival, reproduction and good health (Suvitha *et al*., 2015). "Amino acids play an important function as building blocks of proteins and are mainly obtained from proteins in diet and the quality of dietary protein is assessed from essential to nonessential amino acid ratio" (Mohanty *et al*., 2014; Suvitha *et al*., 2015). The demand for alternative protein source in fish feed has become more prominent (Sales and Janssens, 2003). "Fish feed is considered good if well accepted by fish, has high digestibility, easily available and is cost effective" (Sogbesan and Ugwumba, 2008). Good quality fish feed should contain all nutritional content supplied in their correct proportions (Wang *et al*., 2006).

Growth and Feed Conversion Ratio (FCR) are excellent means to figure the suitability of feed in fish feeding experiments (Sahu *et al*., 2007). "The food conversion ratio (FCR) in fish is 1.5 times better than chicken, lamb or beef" (Menon, 1991). The proceeds from aquaculture are 2 to 5 fold higher than conventional agriculture. However, "Aquaculture in many developing countries has been faced by many challenges" (Osure, 2011). "These include, lack of certified seeds, poor commercially produced feeds, inadequate training programs for farmers and extension workers, inefficiency in dissemination of technology among others" (Mwangi, 2008; Osure, 2011).

The main cost-effective source of protein for fish is plants and animals. Animal proteins are advanced compared to proteins from plants since they have all essential amino acids and are readily digestible but costly. Traditional farmed ingredients have increased the processing of the feeds, hence has influencing success of aquaculture (Kumar, 2000). The aim of formulating good fish feeds is to attain utmost protein deposition and growth within least inputs of feed at a lowest cost. Experimental fish feeds can either be whole or supplemental. Whole diets provide all the nutrients essential for optimum growth and health of fish when reared in bulk in tanks and ponds. Supplemental diets are intended to sustain the ordinary food usually accessible to fish in ponds or outdoor cages. "In fish farming nutrition is critical because feeds represent 50-60% of production cost" (Steven and Helfrich, 2002).

The development of natural and traditional aqua feeds like, termites, black soldier fly larvae, desert locusts, cockroaches and polychaetes have been used either as fish bait, wild fish food or supplementary diets in animal feeds (Karuri, 2010) but without much research on their nutritional content. Fish farming management is aims at attaining utmost fish biomass within a specific time frame (Schuchardt *et al*., 2008).

1.2 Rationale

Competition for protein for fish feeds in aquaculture with human and other livestock needs is a major challenge in the aquaculture industry. The high demand for protein based feeds points to the need for innovative ways of developing quality fish feeds for aquaculture to become tenable. Studies have shown that insects such as termites, desert locusts, cockroaches, black soldier fly larvae, and worms such as polychaetes have high nutritional value in terms of proteins, fatty acids and mineral salts. This study aims at identifying suitable alternative protein source for catfish fingerlings diet from among known insects and worms.

1.3 Objectives and research questions

1.3.1General objective

To compare the growth performance of catfish fingerlings fed on termites, cockroaches, black soldier fly larvae, desert locusts, polychaetes and silver cyprinid (Control diet) as source of protein in their diets.

1.3.2 Specific Objectives

i. To analyze nutritive composition of target insects (black soldier fly larvae, cockroaches, desert locust, and termites), worms (polychaetes) and silver cyprinid (Control Diet).
ii. To determine growth rate of catfish fingerlings *(Clarias gariepinus)* fed on target experimental diets as source of amino acids.
iii. To assess the content of the amino acids in the catfish fingerlings tissue fed on the target experimental diets.

1.3.3Research questions

i. What is the nutritive value of the target insects (black soldier fly larvae, cockroaches, desert locust, and termites), worms (polychaetes) and silver cyprinid (Control Diet)?
ii. What is the growth rate of catfish fingerlings (*Clarias gariepinus*) fed on target experimental diets as source of amino acids?
iii. What types of amino acids are present in catfish fingerlings tissue fed on target experimental diets?

CHAPTER TWO: LITERATURE REVIEW

2.1 Fish species cultured in Kenya

In Kenya farmers need to identify the types of fish they intend to culture, considering their environments and waters available, whether fresh or salt, warm or cold. Adequate climatic diversity of warm fresh waters has enabled the cultivation of Nile Tilapia (*Oreochromis niloticus*) which represents about 90% of cultivated fish. "The largemouth bass (*Micropterus salmoides*) and common carp (*Cyprinus carpio*) are also cultivated in the country". The rainbow trout (*Oncorhynchus mykiss*) is reared in cool fresh waters (Obiero *et al*., 2014).

The African catfish (*Clarias gariepinus*) species has a ubiquitous distribution in rivers, streams, dams and lakes in Kenya (Okechi, 2004). The *Clarias* species inhabit wetlands and wetland open interface (Greenwood 1966). Successful culture breeding of this species has been done in the country (Campell 1995 and Macharia *et al*., 2002). African catfish is usually stocked in Nile tilapia ponds to control their population and attracts good price in the market due to their good taste and flavor (Obiero *et al*., 2014). Research shows that fish farmers cannot meet the market demand as a result of lack of sufficient protein source for fish feeds. Hence there is a market niche that needs to be filled for aquaculture sustainability and also to meet the food demand in developing countries (Ayieko *et al*., 2010).

2.2 Systematics, classification and distribution of African catfish *Clarias gariepinus*

The African catfish *Clarias gariepinus* (Burchell, 1815) is one of the distinctive groups of fish belonging to Subclass Actinopterygii, Division Teleostei, Order Siluriformes and Family Clariidae. The African catfish species has a wide range and ubiquitous habitat demands. It is a tropical fish occurring in fresh water habitats and has a pan-African distribution but is absent from the Maghreb, the upper and lower Guinea and the Cape Province. It also occurs naturally in Jordan, Israel, Lebanon, Syria and southern Turkey (FAO 2008). These groups of fish (siluriformes) are widely consumed in East Africa. This species is found in most water bodies including swampy vegetated areas of lakes, streams, rivers, and floodplains which are prone to drying up during periods of droughts (Akinsanya and Otubanjo, 2006).

2.3 Performance of African catfish *Clarias gariepinus* in aquaculture

African catfish *Clarias gariepinus* species has shown a considerable potential as fish suitable for intense aquaculture. The farming of *Clarias gariepinus* extended outside natural habitats and was introduced to Argentina; Bangladesh; Brazil; Cambodia; China; Côte d'Ivoire; Czech Republic; Gabon; Greece; India; Indonesia; Iraq; Lao People's Democratic Republic; Lesotho; Mali; Myanmar; Netherlands; Philippines; Singapore; Thailand and Viet Nam (Vitule *et al*., 2006). This fish grows rapidly, is resistant to diseases and stress, sturdy and highly productive even in polyculture (Balogun and Dabrowski, 1992). The predatory, cannibalistic and voracious feeding habitats of African catfish catch the impression of farmers to culture them in inland water bodies (Sambhu, 2004).

2.4 Biology of African catfish *Clarias gariepinus*

2.4.1Reproduction

The African catfish *Clarias gariepinus* migrate laterally from the larger water bodies to temporarily flooded marginal areas in order to breed (Greenwood 1966). Its reproduction is seasonal with gonadal maturation associated to periods of flooding. The maturation process is influenced by changes in water temperature and photoperiod, but the increase of water level is the principal factor for their reproduction (Yalçın *et al*., 2001).

2.4.2 Feeding and Nutrition

The African catfish is omnivorous or can be predatory (Lowe- McConnell 1987) but has a relatively high dietary protein requirement, in the order of 40–50 percent of crude protein on a dry weight basis. Its larvae feed exclusively on zooplankton and phytoplankton in the first week. Juveniles feed mainly on ostracods, aquatic insects and insect larvae (Corbet 1961). Fingerlings feed on micro-crustacean and insect larvae and about 70% of feeding occurs3 at night (Potongkam and Miller, 2016). Adults feed on small fish, aquatic insects, crustacea, mollusks and aquatic plants (Mwebaza-Ndawula 1984). The larvae and early juveniles have a high protein demand of 55 %, a lipid requirement of 9 % and 21% carbohydrates. The basic nutritional requirements during the grow-out phase range from 40 - 43 % for protein, 10 - 12 % for dietary lipid and 15 - 32 % for carbohydrate. The optimum digestible energy is between 14 - 16 kJ/g (FAO, 2017b).

2.4.3 Survival

The African catfish possesses physiological adaptations such as accessory air breathing organs which enable it to cope with extreme environmental conditions (Claridge *et al*., 1986). The fish can survive in low oxygen concentrations and in water of extreme temperature of 8 - 35°C (Britz and Hecht, 1987), salinity levels between 0 and 10% and a wide range of pH (Safriel and Bruton, 1984). During dry conditions it burrows into the mud and envelopes itself into a cocoon which enables it to survive temporarily and sometimes extended period of desiccation (Greenwood, 1986).

2.5 Factors affecting Aquaculture

The lack of suitable fish feeds at reasonable cost has significantly resulted to a low pace for aquaculture growth in East African region (Munguti *et al*., 2014). “Research has shown that the demand for fish is increasing at a rate of 2.5% per annum, a rate that is expected to increase” (Shamshak and Anderson, 2010). A number of constraints such as inefficient and inexpensive fish feeds for different stages of development, poor feed management skills, limited varieties of the cultured fish species and low quality fish seed have led to low growth rate and poor productivity in aquaculture (Munguti *et al*., 2012). Information gap in African society has led to the inability to select and judge the available information about the best technology for the fish farming.

2.6 Availability of seed

Insufficient supply of certified quality fish larvae has been a great challenge to the advancement of aquaculture causing high mortalities after stocking of ponds with inferior fingerlings and poor quality seed (Munguti *et al*., 2014). “The government of Kenya under the economic stimulus program (ESP) aimed at increasing production of farmed fish by construction of 200 fish ponds in each of 140 constituencies, totaling to more than 28,000 fish ponds nationally in the initial phase”. “This caused an instant short-term demand for about 28 million certified tilapia and catfish fingerlings” (Charo-Karisa and Gichuri, 2010). The economic stimulus program also encouraged more smallholder farmers to come up with their own ponds hence increased demand for seed fish (Musa *et al*., 2012). Besides the impact of the ESP, smallholder farmers embraced fish farming, with new farmers who own bigger ponds practicing commercial aquaculture resulting in higher fish production (Otieno, 2011). “There are 56 operating hatcheries across the country producing tilapia, catfish and common carp, with a total production capacity of over 50 million tilapia fry and 75 million catfish fry per year” (Charo-Karisa and Gichuri, 2010).

Catfish fingerlings seed are normally used as bait for Nile perch fishing in Lake Victoria. “Many farmers in Kenya, Uganda and other countries target fishermen as their customers for the live bait because they get higher prices as compared when sold in the local market”. At the moment hatcheries cannot meet the high demand in the market therefore the need to establish more hatcheries to produce sufficient fish seed to bridge the gap (Merican and Shim, 1996; Bhujel, 2011).

2.7 Available Fish feeds

In fish cultivation, utilization of feed is fundamental for high production and profitability. Nutrition in fish is a key component for aquaculture which accounts for 40-50% of the total productivity on the fish farm (De-Silva and Hasan, 2007). Lack of efficient and cheap locally made feeds for various phases of fish growth is a key challenge in Kenya for aquaculture development (Munguti *et al*., 2012).

Available natural feeds include phytoplankton in earthen ponds that are encouraged by introduction of manures and fertilizers. These require some period of time to be established and have low protein content. Commercial feeds such as a blend of maize, rice, wheat bran and sometimes in addition with Silver cyprinid/Omena (*Rastrineobola argentea)* meals are too expensive for most farmers (Ngugi *et al*., 2007). “Due to the ESP project inception, fish feed shortage stands at14, 000 metric tons per year and has increased to about 50,000 metric tons per year in Kenya “(Charo-Karisa and Gichuri, 2010).

2.8 Non- conventional alternative feeds

The use of unsatisfied feeds has been reported to promote growth and have cost effective value. Focus is on the use of insects, worms, garden snails and tadpoles among others as alternative protein source for cultured fish (Devendra, 1988). The assumption so far is that alternative fish feeds can supply adequate essential nutrients requirements for fish growth and health (McCartney, 1996).

Non-conventional fish feeds are prospective feed components, which are rarely used in fish feed processing, due to insufficient knowledge on protein content during manufacture with a view to commercialize them. Limited findings indicate that non-conventional fish feeds have high quality nutrients that are suitable to conventional types. The expectation is that they are readily available and cheaper since they are from plants or animals not regularly used as food by humans (Roberts, 1989). "The plant protein source of fish diets comprise, leaf protein, leaf meal, aquatic macrophytes, cultivable pulses such as mucuna bean, yam beans, bread beans, winged beans and any legume that can yield pods with seeds. *Azolla piñata* (fresh water fern) is a potential protein fish feed component for the diet of *Oreochromis niloticus"* (CAN, 1993).

Examples of animal protein source for feeds consist of meals from tadpole, earthworm, crab; insects, shrimp and animal wastes from pigs, poultry and blood meal (Devendra, 1988; Abowei and Ekubo, 2011). Animal waste, mainly droppings from pig and poultry can be used to enrich pond for growth of plankton or when uncontaminated, as a direct source of fish feed. "Some fish farmers use poultry droppings without further use of any artificial feed with good results". "Animal wastes are particularly useful in the mixed culture of the local catfishes and tilapias. Earthworm (*Lumbricus terrestris* and *Allobophora long*) have been used as fish bait in artisan fisheries" (Helfrich and Smith, 2001) and therefore may have potential as protein source.

Depending on availability, unprocessed tadpoles' meal is often fed to adult fish or processed with other feed ingredients at 40-50% (Pharmacol and Toxicol, 2011). Scientists in many parts of the world have hinted the use of insect biomass as an excellence feedstuff for animal feed such as poultry, swine and fish. Insects, particularly fly larvae, can translate low value organic material into protein and fat. Insects contain very high in crude protein, ranging above 60% in several species (Finke *et al*., 1989) and vary widely in fat content (Sogbesan, and Ugwumba, 2008). Housefly larvae can be fed whole to fish or processed with a substrate in a mixed form (Houlihan *et al*., 2001). Current research in black soldier fly culture makes this insect favourable for wide scale production. In developing countries many of these insects are found mostly in dried form in the markets and live in research institutes or in the wild.

2.8.1Termites (*Macrotermes subhylanus*)

Climate change favor increased emergence of insect's population like termites and lake flies by influencing soil fauna and its biodiversity that supports them (Saunders, 2008). Notably moisture and temperature are key factors in the insect ecology. Elate termites (*Macrotermes subhylanus*) and lake flies (*Chironomus plumosus)* are available along the Kenya lake region and other parts of Western and Rift valley (Sogbesan and Ugwumba, 2008). Termites are available and collected in most parts of Africa especially semi-arid savannah zones, they are used as diet among non-livestock keeping tribes (Odhiambo, 1978.)

In Kenya termites are collected after rains begin by making a trap using light and fire near their emergent holes. The emerging insects are stupefied by the smoke and drop into the collecting vessel, where they are collected, sun dried and the de-winged (Massam, 1927). Other areas where termites are harvested include Nandi District, in the months of April to May and Wundanyi in Taita Taveta District (Mankowski, 1994), Busia District, Western Kenya, and Siaya District near Yala and Nyangweso and parts of Eastern Uganda. Alternatively, swarming of the sexually mature termites can be made to come out early by simulating rain drops or introducing water to the emergent holes to reinforce the impression of rain (Bryk, 1927).The local communities from these areas use this as food and sell surplus in the local market. Organizations such as KumbeKumbe farm in Western Kenya, process and store the termites for animal feeds (Karp and Karp, 1977).

2.8.2 Cockroaches (*Periplenata americana*)

Cockroaches live in close association with people and are omnivorous scavengers. These insects have been around for over 35 million years and are found living in parts of houses and other buildings in nearly every part of the world. They consume any organic food sources available to them such as sweets, meats, starches, sewage, hair, books, fresh vegetable leaves and decaying matter (Cornwell, 1968).

Cockroaches survive in very harsh conditions for long period of time in form of egg case**.** They are most active at night or in the dark, preferring small spaces and flourish well in warm damp places, congregate in sewers, dumps, outbuildings and wood pile during warm weather. Certain species of roaches have adapted to human lifestyles and food sources. The German cockroach *(Blattaria germanica*) and American cockroach (*Periplaneta americana)* found in Kenya are common in food stores, kitchens, restaurants and grocery stores. Cockroaches are a source of food for small reptiles and bird in the wild as well as domestic chicken. Given favorable condition they can be reared to be used to supply protein in fish feeds (Rust *et al*., 1991).

2.8.3 Desert Locusts (*Schistocerca gregaria*)

Locusts are allied to grasshoppers, however, locusts have another behavioral phase called the gregarious phase. “Common locusts in Africa include the desert locust (*Schistocerca gregaria*) and migratory locust (*Locusta migratoria*)”. These species are notorious and widely distributed due their ability to migrate over long distances (Dingle, 1996). “They are common in Africa, the Middle East, and Asia”. “Locust plague may threaten the economic livelihood of the world's human’s population since each locust can eat amount of food equivalent to its body weight from plants each day” (Morgan, 2014). Breeding of locust is favored by environmental conditions such as heavy rains which produce many green plants that promote breeding, congregation into thick, mobile, ravenous swarms (Watch, 2007).

The locusts are easy to breed, rear and are consumed by several cultures throughout the world and are a delicacy to many communities in Africa, Middle East and Asian countries (Fromme and Alison, 2015). They can be cooked in many ways but are often fried, smoked or dried. Previous studies indicate that locusts contain high nutrient contents such as proteins, fat and mineral salts (Dubois and Sirah, 2015).

2.8.4 Black Soldier Fly (BSF) Larvae *(Hermetia illucens)*

The black soldier flies (*Hermetia illucens)* is a common fly whose adults have no mouthpart or digestive organs and therefore do not feed as adults but survive on fat stored from their larval stages. Healthy adults are approximately 7/8 inches long. The adult has a short life cycle of 5-8 days (Hawkinson, 2005).

A few days after becoming an adult and emerging from a pupal case, female black soldier fly finds a mate, copulates and lays more than 500 eggs in a dry environment near edges or crevices of decaying organic matter. After the eggs are laid and with optimum condition of around 20° to 30°C these remain in this stage for approximately 4-5 days to incubate and hatch (ESR International, 2008).

BSF larvae are photophobic and will bury themselves in the nearest organic material and start feeding on it. The larvae have the ability to digest waste and convert feedstuff and manure into valuable biomass, maximizing efficiency and benefits while minimizing cost. They feed on varied animal matter, including feaces and the softer less cellulosic plant matter whether decaying and high cellulose plant matter once it has begun to decay. The larvae are voracious feeders which can easily consume their own body weight in a day. Given optimum environmental condition, these reach full size of around 20 to 25mm in about 4 weeks at fifth instar (Newton, 2005).

Pre-pupae stages begin at the sixth instar with a final molt phase when the pre-pupae stop feeding and are self-collected as they leave the larval substrate to pupate and are collected by construction of harvesting vessels in such a way that pre-pupae will crawl into collection vessels, from which they can be removed periodically (Newton, 2005).

2.8.5 Polychaete worms (*Marphysa mossambica*)

Polychaetes are commonly used as a natural diet of marine finfish and larger crustaceans, thus forming a larger percentage of interlinkages for those food species. "The commercial worth of polychaetes stems from their high palatability, good characteristics in digestibility and their use as fishing baits as proof of their attractiveness to fish" (Kihia *et al*., 2015)

Furthermore, polychaetes are used as a source of fish oils which contain essential polyunsaturated fatty acids (PUFA) (Luis and Passos, 1995). They provide amino acids and vitamins, which form a vital component for the development of high quality juveniles for both finfish and crustaceans (Cahu *et al*., 1994).

2.9 Protein and amino acids requirements of the African catfish

2.9.1 Protein requirements

"Proteins are made of carbon (50%), nitrogen (16%), oxygen (21.5%), hydrogen (6.5%) and few possess sulfur in low amounts (2.12%)" (Halver and Hardy, 2002). Most proteins are made of 22-26 amino acids but only 18 amino acids are found plant and animal protein. According to function and solubility, various types of proteins are found in fish body and being major organic material in fish tissue, "proteins are estimated to make 65-75% of the total based on dry weight" (Halver and Hardy, 2002). Consumed protein is broken down to release amino acids which are its building blocks, later absorbed and distributed to organs and tissues via blood. Various tissues synthesize new protein from the absorbed amino acids thus their regular intake is required by the fish. New proteins are built during reproduction and growth and for maintenance of body functions. 'The optimal dietary protein fish level is mainly determined by the dietary protein ratio to energy balance, amino acid constituent, protein breakdown and quantities of non-protein energy sources in the diets" (Halver and Hardy, 2002; Davis *et al.,* 2009).

Fish mainly have higher protein requirement during early growth as compared to the later phases of growth. Low protein levels in fish diets leads to cessation of growth and reduced weight as a result of extraction of proteins from less vital organs to sustain the more vital tissues. However, too much supply of protein in diets makes the fish to utilize a proportion to synthesize new proteins and excess converted to energy (Halver, 2013). Studies have reported that the optimal dietary crude protein requirement for catfish is 30-35% (Viveen *et al.,* 1985), 40% to 50% (Hecht *et al.,* 1988) and 28-32% (Halver, 2013). However, protein demands are lower for herbivores, omnivorous fish species and less populated ponds as compared to carnivorous species and highly populated ponds (Craig and Helfrich, 2016). "The optimum dietary protein levels depend on the fish growth rate, feed intake, amount of non-protein energy in the diet, protein quality, presence of natural food and management practices" (Davis *et al.,* 2009) rearing environment, water temperature, water quality, feeding rate and genetic composition (Craig and Helfrich, 2016). Protein is mainly used for growth so long as energy giving nutrients are in adequate levels (Craig and Helfrich, 2016).

2.9.2 Amino acids requirements of the fish

"Amino acids are protein structural components consisting of a carboxyl (-COOH) and amine group (-NH2) on the alpha carbon atom differing in R-group or side chain; usually linked together by a peptide bond to form proteins". There are more than two hundred amino acids but only 20 out of these are focused on. "Amino acids are classified as either essential amino acids or non essential amino acids" (Halver, 2013). Fish requires a proportionate mixture of both essential and non essential amino acids thus is imperative to balance the protein and amino acids required by each fish species (Halver and Hardy, 2002). A fish diet that lacks some of the essential amino acids, results in slow growth and decrease in feeding efficiency (Guillaume, 1999). "Fish being monogastric demands all the ten essential amino acids which include arginine, isoleucine, histidine, leucine, methionine, phenylalanine, lysine, valine, tryptophan and Threonine". Fish are capable of partially substituting non essential amino acids with essential amino acids. "Channel catfish can grow when methionine is the only sulfur containing amino acids as compared to when cystine is the sulfur containing amino acids however cystine replaces about 60% of Methionine". Non essential amino acid, tyrosine replaces one half of channels catfish requirements for phenylalanine (Halver and Hardy, 2002).

2.10 Protein and amino acid characteristics of target organisms

"Black soldier fly larvae are a significant feed source, with high protein and amino acids". "They have about 40-44% crude protein (CP)" (Makkar *et al*., 2014; Tran *et al*., 2015). Black soldier fly larvae have a high profile of amino acids including Alanine , Arginine, Aspartic acid, Methionine, Cystine, Lysine, Isoleucine, Leucine, Phenylalanine, Threonine, Tryptophan, Glutamic acid, Histidine, Proline, Serine, Tyrosine and Valine (Makkar *et al*., 2014). Since BSF larvae are a suitable protein and amino acids source, they are known to maintain good growth in animals (Spranghers *et al*., 2016).

Locust are considered edible and regarded for their high level of protein, minerals and vitamins (Finke, 2002). "Locusts contain CP of (50-65%), though some lower values (<30%) have also been reported" (Makkar *et al*., 2014). The amino acid profiles of desert locust include Alanine, Arginine, Aspartic acid, Methionine, Cystine, Lysine, Isoleucine, Leucine, Phenylalanine, Threonine, Tryptophan, Glutamic acid, Histidine, Proline, Serine, Tyrosine Valine and glutamic acid (Tran *et al*., 2015). "Desert locust meal (*Schistocerca gregaria*) could replace up to 25% dietary protein in *C. gariepinus* juveniles without significant reduction in growth" (Makkar *et al*., 2014).

Silver cyprinid (*Rastrioneobola argentea*) is mostly used for manufacture of fishmeal in the feed production industries (Bille and Shemkai, 2006). *Rastrioneobola argentea* protein values range from 19.1 - 21.8% (Ogonda *et al*., 2010); 53.0- 58.8% in dried *R. argentea* (Owaga *et al*., 2010; Kabahenda *et al.,* 2011). "The high protein value for *Rastrioneobola argentea* is attributed to the edible portion in 100% (whole fish) despite its average length of only 4.6 cm; that is relatively small compared to the deep sea fishes whose lengths range from 14 to 49 cm" (Suseno *et al*., 2010).

Polychaetes contain high nutrients with about 54.72% of their body being protein (Pamungkas, 2015). “The high proportion of protein is due to the occurrence of gametes (either sperm or ovum) filling their bodies when the worms swarm” (Pamungkas, 2015). “The amino acids found in polychaetes include alanine, serine, methionine, aspartic acid, glutamic acid, tyrosine, lysine, Phenylalanine, valine, leucine, isoleucine, threonine and proline and glycine” (Woulds *et al*., 2012).

Termites have high-quality nutrients such as highly digestible proteins (Kinyuru *et al*., 2010). “The crude protein values of termites ranges from 37% to 44.12% (Fadiyimu *et al*., 2003); *Nasutitermes* spp. 43.65 to 44.12 %, (Oyarzun *et al.*, 1996); 46.3 % in *Macrotermes subhyalinus (*Sogbesan and Ugwumba, 2008); 20.4% and 22.1% in *Macrotermes bellicosus* and *M. notalensis*, respectively” (Banjo *et al*., 2006). Termites also contain high profile amino acid content. The essential amino acids comprise Isoleucine, Histidine, Leucine, Lysine, Methionine, Phenylalanine, Threonine, Valine, Tryptophan (Sogbesan and Ugwumba, 2008).

CHAPTER THREE: MATERIALS AND METHODS

3.1 Description of study area

The study was conducted in the University of Nairobi Aquaculture Laboratory between January and November, 2016. The Aquaculture Laboratory is located in the College of Biological and Physical Sciences, School of Biological Sciences, Chiromo Campus.

3.2 Acquisition of experimental fish

Seven hundred and twenty catfish fingerlings (mean weight 0.362 ± 0.089 g) used in this experiment were procured from Green Algae Fish farm in Sagana, Kenya in June, 2016. Forty fingerlings per aquaria were acclimatized to experimental conditions for one week before the experiment started. After acclimation period, ten fingerlings were randomly sampled and weighed using an Electronic balance (Scout Pro Balance, Ohaus) (in order to establish the mean individual weight at the start of the experiment) and mean body weights recorded.

Figure 1: Catfish fingerling (10 cm x 1.8cm) in a fish culture tank

3.3 Collection of target organisms

The desert locusts and cockroaches were reared in the University of Nairobi Insectary. After development to adult stage, the insects were harvested and then frozen in a deep freezer for 10 minutes to immobilize them. Once dead they were oven dried at 70º C for 24 hours and packed in air tight containers. Termites were harvested from Western Kenya (Kakamega County) after the short rains in the months of October and November, 2015. They were sundried and winnowed to remove the dry wings and packed in brown khaki paper bags (No. 10) for transportation to the University of Nairobi aquaculture laboratory. The polychaete worms were dug out from the muddy shores of Western Indian Ocean at Mida Creek. After harvesting they were washed to remove mud and sand and then salted to preserve them. The worms were sundried and packed in airtight containers for transportation to the University of Nairobi aquaculture laboratory.

Black soldier fly larvae were obtained from the International Centre of Insect Physiology and Ecology (ICIPE) laboratory. They were harvested at the pre-pupae stage, sun dried and packed in clear ziplock bags (9 inch) for transportation to the University of Nairobi. Silver cyprinid was bought from the local market (Kangemi, Nairobi) and packed in brown khaki paper bags (No. 10) for transportation to the University of Nairobi aquaculture laboratory where they were unpacked to sort out and remove unwanted materials such as snail shells and stones and then packed in brown khaki paper bags (No. 10).

Black soldier fly larvae, polychaetes, silver cyprinid and termites were further dried in the oven at 70º C for 24 hours to get rid of all the moisture and packed in airtight containers. Silicon pellets wrapped in muslin cloth were put in each package to absorb any moisture during storage.

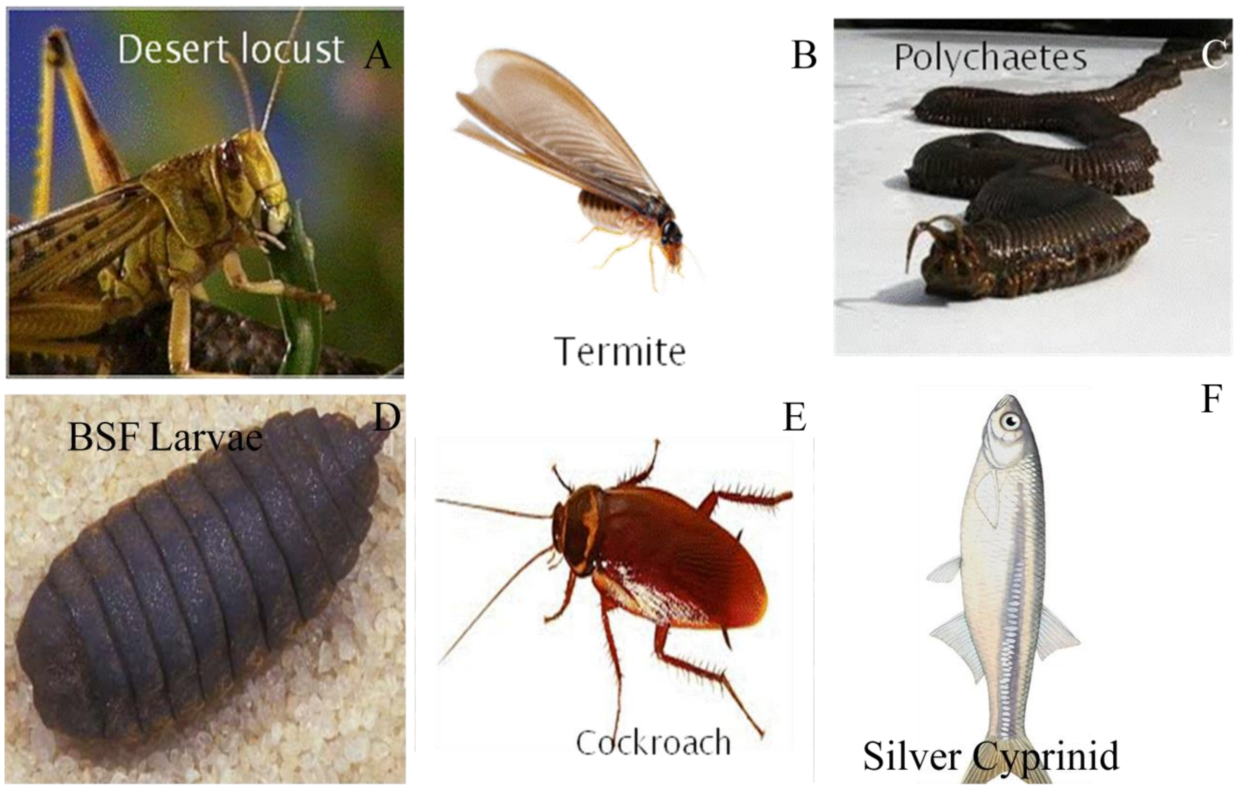

Figure 2: Target organisms; Desert locust (A), Termites (B), Polychaetes (C), Black solder fly larvae (D), Cockroach (E) and Silver cyprinid/Omena (F).

3.4 Proximate analysis of target insects and worms

Proximate analysis was done at the Department of Food Science and Technology, University of Nairobi. The target insects and worms were analyzed for moisture content, total ash, crude fat, crude fiber and crude protein using Control laboratory methods (AOAC, 2005). "The moisture content was determined by drying the samples overnight in a conventional oven (oven-drying method) to a constant weight at 105°C. "Crude fat was determined by extracting ground samples continuously for 6 hours in petroleum ether using Soxtec HT2 1045 extraction system. Crude protein (N x 6.25) was determined by micro kjeldahl method" (Jones, 1991). "The samples were digested in concentrated sulphuric acid using a Digestor 2040 (FOSS, Denmark) followed by distillation using a Kjeltec 2300 auto–analyser (FOSS, Denmark) to determine nitrogen content which was converted to crude protein using a conversion factor of 6.25". "Total Ash was analyzed by ignition at 650°C for 12 hours in electric furnace (Eyela-TMF 3100) to constant weight". Crude fiber was determined by digesting dried lipid-free residue with 1.25% H_2SO_4, 1.25% NaOH and calcined. The carbohydrates were determined as 100 − (moisture content + crude protein + crude fat + ash + fiber). Gross energy was calculated using conversion factors for protein, lipids and carbohydrates.

3.5 Feed formulation

Six experimental diets were formulated to contain 40% wheat bran and 60% crushed dried insects or polychaetes or the silver cyprinid (Control diet). Wheat bran used for this study was obtained from Baraka animal feeds and cereals store, Nairobi Kenya. Each diet mixture was dried in an oven at 50° C for 12 hours. The experimental diets were homogenized into powder using a dry mill kitchen blender (BL335, Kenwood, UK). The dried homogenized diets were sieved using No. 20, Control testing sieve, to remove irregular/and large size pieces before weighing out the quantities required. Samples of all homogenized experimental diets were analyzed for proximate nutrient composition.

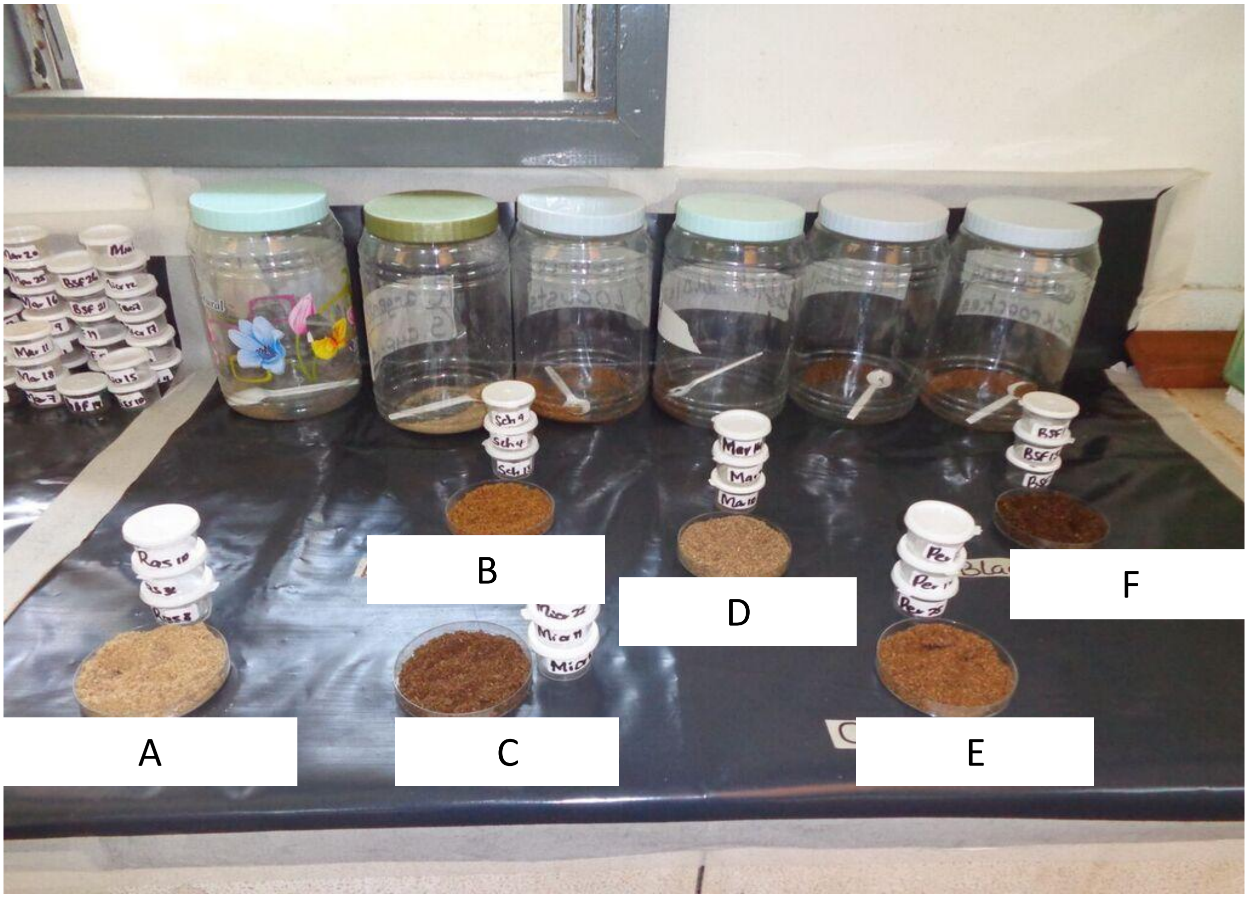

Figure 3: Experimental diets; Silver cyprinid/Omena (Control diet) (A), Termites (B), Desert locust (C), Polychaetes (D), Cockroach (E) and Black solder fly larvae (F).

3.6 Proximate analysis of experimental diets

Proximate analysis of experimental diets was done at the Department of Food Science and Technology, University of Nairobi as described in section 2.3.

3.7 Determination of amino acids in the experimental diets and catfish tissue

Amino acid determination was carried out at International Centre of Insect Physiology and Ecology (ICIPE). A sample of experimental diets and harvested preserved catfish fingerlings were dried in the oven at 70^{o}C for 24 hours to get rid of all the moisture and packed in clear zip lock bags (9 inch) for amino acid analysis. Protein extraction and amino acid quantification was done using a method adopted from (Hamilton *et al.*, 2012). The experimental diet samples were "snap-frozen in liquid nitrogen and crushed into fine powder". The samples (2 g each) were extracted for 1hr in ice cold 5 v/w 100 mm 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid

(HEPES) pH 7.2, 2 mm dithiothreitol (DTT), 2.5% Polyvinylpyrrolidone (PVP), 0.5 mm Ethylenediaminetetraacetic acid (EDTA), 1 mm benzamidine 0.1 mm phenylmethanesulfonylfluoride (PMSF) in a magnetic stirrer. “The samples were filtered through KERLIX™ Gauze Bandage Rolls Sterile Soft Pouch 5.7 cm × 2.7 m centrifuged at 8000 rpm for 30 min at 4°C to remove solid debris”. “Protein was precipitated between 45% and 80% $(NH_4)_2SO_4$ and the pellet recovered by centrifugation at 21,000 rpm for 30 min at 4°C”. “The protein pellets were desalted in 20 mm HEPES–NaOH pH 8 containing 2 mm DTT using Sephadex G-25 gel filtration chromatography (PD-10 columns, GE Healthcare) to give 10g in each sample” (Hamilton *et al.*, 2012).

Ten miligrams of each sample was “separately transferred into a 5 ml micro- reaction vial into which 2ml of 6N HCl was added and closed after careful introduction of nitrogen gas”. “The samples were hydrolyzed for 24 h at 110°C. For tryptophan analysis, 10 mg from each of the samples were separately transferred into a 5 ml micro-reaction vial into which 2 ml of 6N NaOH were added and then capped after careful introduction of nitrogen gas”. “The samples were hydrolyzed for 24 h at 110°C”. “After the hydrolysis, the mixtures were evaporated to dryness under vacuum”. “The hydrolysates were reconstituted in 1 ml 90:10 water: acetonitrile, vortexed for 30 s, sonicated for 30 min, and then centrifuged at 14,000 rpm and the supernatant analysed using LC-Qtof-MS”. The analysis was done in triplicates.

3.8 Experimental set up

The feeding experiment was conducted at the University of Nairobi Aquaculture Laboratory from June to August, 2016. A total of 18 aquaria, each measuring 80x35x40cm, were used in this study. Forty catfish fingerlings were randomly sampled and each batch put into an aquarium of 112 Liters (80x35x40cm) by volume at a stoking density of 0.14g/litre. Eighteen aquaria in total were used for the six treatments (black soldier fly larvae, cockroaches, desert locusts, marine polychaetes, silver cyprinid and termites) each replicated three times. The aquaria were connected to a temperature (24 ± 2^oC) controlled system (heaters) (JAD microprocessor digital heater, DR 9200) and air pumps (Resun, AC 9908) for aeration. All aquaria were placed on laboratory benches for ease of accessibility and observation. Clean water was supplied from a reservoir tank into each aquarium through inlet pipes and the effluent water was drained by siphoning mechanism. A constant photoperiod of 12 hours of light and 12 hours of darkness was maintained through the experimental period (8 weeks).

Figure 4: Laboratory set up at the University of Nairobi Aquaculture laboratory where the feeding trials were done from January to November, 2016.

3.9 Feeding protocol

The six experimental diets were randomly assigned to each aquarium tanks in triplicate. The fingerlings were fed daily to satiation at10% body weight two times a day at 10.00am in the morning and 4.00pm in the evening for two months (8 weeks). Aquaria were cleaned daily prior to feeding by siphoning out the water with fecal matter and left over feed at the bottom of the tank using siphoning pipes. Experimental tanks were refilled with warm (25±2°C) dechlorinated water from a reservoir tank. The water quality parameters which included temperature, dissolved oxygen, total Ammonium Nitrate and pH were measured twice a week.

3.10 Determination of Specific Growth Rate of catfish fingerlings

The growth performance of catfish fingerlings was assessed in terms of weight gain and specific growth rate for each treatment. A sample of ten fingerlings were scooped out using a hand scoop net from each aquarium. To achieve accurate fingerlings weight, the excess water on the hand scoop net was carefully wiped using a kitchen towel. The fingerlings were then introduced into a zeroed half-filled water bowl on a weighing balance (Scout Pro Balance, Ohaus) and the weight recorded. After weighing, the fingerlings were returned to their respective aquarium immediately. Growth of the fingerlings was expressed as specific growth rate (SGR) using the formula:

$$\text{SGR\%} = \frac{\text{In FBW} - \text{In IBW}}{\text{t}} \text{ x } 100$$

Where FBW is final body weight; IBW is initial body weight; In = natural logarithmic; t = time in days.

3.11 Determination of Survival Rate of catfish fingerlings

Dead catfish fingerlings, if any, were removed daily and the mortality used to determine the number of live fingerlings left in each aquarium. Survival rate was determined at the end of the experiment by counting the number of live catfish fingerlings remaining in each aquarium and expressed as a percentage of the stocked fish as follows;

$$\text{SR\%} = \frac{Nt}{N0} \text{ x } 100$$

SR is the survival (in %); *Nt* is the number of fish collected at sampling time *t; N0* is the number of fish initially stocked.

The live fingerlings were thereafter put in a freezer to immobilize to death. Once dead, they were oven dried at 70^{o}C for 24 hours and ground into powder for amino acid analysis.

3.12 Data analysis

The data obtained was analyzed by using the analysis of variance (ANOVA) PROC ANOVA procedure of GENSTAT version 15. The effect of different diets on specific growth rate (SGR%), survival rate (SR%) and amino acids composition were compared by analysis of variance (one way ANOVA) followed by Tukey's multiple range test which was used to compare statistical difference among treatment means. Differences between the treatment means were considered significant at 95% confidence level. Correlation analysis was done to compute the relationship between the diet and tissue amino acids.

CHAPTER FOUR: RESULTS

4.1 Water quality parameters

The physicochemical water quality parameters of fish culture environment in aquaria were observed over the two months period of the experiment (Table 1). Water temperature in the aquaria ranged from 23.8°C to 27.7°C with a mean of 24.6 ±3°C but the variation among the different treatments was not significant ($F_{5,12}$=0.76, p > 0.05). There was no significant variation in the mean pH values among the six treatments ($F_{5,12}$=6.05, p >0 .05). The dissolved oxygen (DO) concentration ranged from 3.1 to 4.7 ml/L with a mean of4.0 ml/L but did not differ significantly among the different treatments ($F_{5, 12}$=1.68, p > 0.05). The mean Total Ammonium Nitrogen (TAN) significantly varied among the treatment within the two months of the study ($F_{5,12}$=26.17, p < 0.05) being highest (3.4mg/L) in the aquaria in which polychaetes were tested and lowest (2.5 mg/L) in the aquaria where black soldier fly larvae were tested. There was however no significant variation (p > 0.05) in concentration of TAN among the aquaria where cockroach, desert locust and silver cyprinid diets were tested.

Table 1: Physicochemical water parameters in the aquaria where different experimental diets were fed on catfish

	Dietary treatments					
Water Parameters	Silver cyprinid	Termites	Desert locust	Polychaetes	Cockroach	BSF larvae
Temperature	$24.2±0.21^{a}$	$24.6±0.24^{a}$	$25.5±1.08^{a}$	$24.6±0.21^{a}$	$24.5±0.13^{a}$	$24.4±0.05^{a}$
pH	$7.1±0.02^{a}$	$7.1±0.01^{a}$	$7.1±0.01^{a}$	$7.1±0.01^{a}$	$7.1±0.01^{a}$	$7.1±0.01^{a}$
DO	$4.2±0.16^{a}$	$4.1±0.03^{a}$	$3.9±0.24^{a}$	$4.4±0.16^{a}$	$3.9±0.34^{a}$	$3.9±0.15^{a}$
TAN	$2.8±0.02^{b}$	$2.7±0.03^{bc}$	$2.8±0.04^{b}$	$3.4±0.45^{a}$	$3.0±0.05^{b}$	$2.5±0.14^{c}$

Mean (± SE) values followed by the same superscript letter(s) in the same row are not significantly different at (p ≤ 0.05)

4.2 Nutrient Composition of the target organisms

The crude protein content was significantly higher in silver cyprinid (67.4%) compared to the rest while it was significantly lower in termite (32.9%) (P < 0.05) compared to the rest except BSF larvae. No significant variation in protein content was however recorded in cockroach (65.5%) and desert locust (63.4%) (P > 0.05) (Table 2). The ash content was significantly higher in polychaete worms (26.2%) compared to BSF larvae (11.7%), cockroach (5.1%), termites (4.1%) and desert locust (3.2%) ($F_{5,12}$=1, $p < 0.05$). The fibre content varied significantly among the six target organisms ($F_{5,12}$=1.89, $p < 0.05$). The desert locust recorded the highest fibre content (13.4%) and BSF larvae the lowest (0.5%) (Table 2). Carbohydrate content ranged between 0.7% and 5.5% and varied significantly among the target organisms ($p < 0.05$). Similarly, the fat content varied significantly ($F_{5,12}$=0.51, $p < 0.05$) with termites having the highest fat content of 48.4% while polychaete worms had the lowest (6.6%) (Table 2). A significant variation was also recorded in the moisture content which ranged from 2.47-6.82% ($F_{5,12}$=1.05, $p < 0.05$).

Table 2: Nutrient composition (%) of target organisms

	Target Organisms (%)					
	Silver cyprinid	Termites	Desert Locust	Polychaetes	Cockroach	BSF Larvae
Moisture	3.5 ± 0.00^{c}	2.9 ± 0.00^{d}	3.5 ± 0.00^{c}	6.8 ± 0.00^{a}	5.7 ± 0.00^{b}	2.6 ± 0.04^{e}
Fat	12.9 ± 0.04^{c}	48.4 ± 0.46^{a}	15.6 ± 0.04^{c}	6.6 ± 0.61^{e}	9.7 ± 0.00^{d}	26.2 ± 1.00^{b}
Protein	67.4 ± 0.00^{a}	32.9 ± 0.00^{d}	63.4 ± 0.23^{ab}	53.7 ± 0.00^{bc}	65.5 ± 0.00^{ab}	45.3 ± 6.20^{cd}
Ash	13.1 ± 0.00^{b}	4.1 ± 0.00^{cd}	3.2 ± 0.00^{cd}	26.2 ± 0.00^{a}	5.1 ± 0.00^{c}	11.7 ± 0.76^{b}
Fibre	0.3 ± 0.00^{f}	8.3 ± 0.01^{c}	13.4 ± 0.09^{a}	1.4 ± 0.00^{d}	10.6 ± 0.00^{b}	0.5 ± 0.03^{e}
CHO	2.9 ± 0.04^{ab}	3.6 ± 0.37^{a}	1.4 ± 0.38^{b}	4.9 ± 0.30^{a}	3.4 ± 0.00^{ab}	2.9 ± 0.81^{ab}

Mean (± SE) values followed by the same superscript letter(s) in the same row are not significantly different at ($p \leq 0.05$)

The value of energy in the six target organisms were in the range of 321.0-341.5Kcal/100g in BSF larvae, 358.9-366.0Kcal/100g in cockroach, 393.0-400.0Kcal/100g in desert locust, 393.3-400.4Kcal/1009 in silver cyprinid, 289.8-296.8Kcal/100g in polychaetes and 576.3-585.3Kcal/100g in termites respectively (Figure 5).

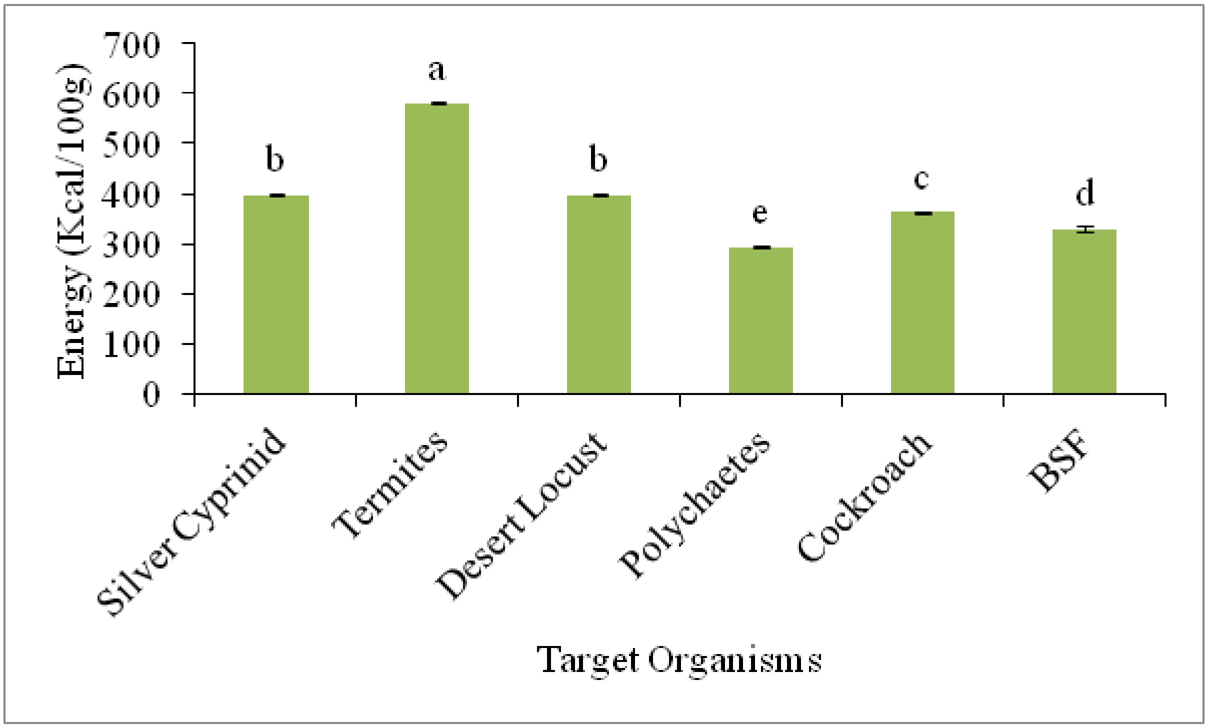

Figure 5: Energy content (Kcal/100g) of target organisms

Bar graphs accompanied by similar letters are not significantly different Tukeys test at ($p \leq 0.05$); Error bars represent Control error of means.

4.3 Nutrient composition of the experimental diets

The proximate composition of experimental diets was determined and nutrient composition ia as shown in Table 3. Crude protein ranged between 24.6% and 43.5% in the six experimental diets and was significantly higher ($p < 0.05$) in silver cyprinid compared to termites, polychaete worms and BSF larvae. The ash content significantly varied among the six diets ($F_{5,12} = 3.57$, $p < 0.05$) with polychaetes larvae having the highest content (21.3%) and cockroach the lowest (8.9%) (Table 3). The fat content of the diets was significantly higher ($p < 0.05$) in termites (26.3%) while silver cyprinid (11.3%) had the highest moisture content. The fibre content significantly varied among the treatments ($F_{5,12}= 0.14$, $p <0.05$). Polychaete worms had the highest carbohydrate content of 26.4% while termites had the lowest (12.9%) (Table 3).

Table 3: Nutrient composition (%) of the experimental diets

	Experimental diets					
	Silver cyprinid	Termites	Desert Locust	Polychaetes	Cockroach	BSF larvae
Moisture	11.3 ± 0.82^{a}	6.6 ± 0.10^{c}	8.6 ± 0.14^{b}	11.2 ± 0.11^{a}	7.8 ± 0.18^{bc}	7.3 ± 0.06^{bc}
Fat	11.9 ± 0.53^{cd}	26.3 ± 0.61^{a}	18.4 ± 2.59^{bc}	6.7 ± 0.62^{d}	20.6 ± 2.67^{ab}	23.5 ± 0.57^{ab}
protein	39.1 ± 1.67^{a}	30.5 ± 2.31^{b}	34.7 ± 0.10^{ab}	29.5 ± 1.07^{b}	35.3 ± 1.01^{ab}	29.3 ± 2.55^{b}
Ash	15.7 ± 0.49^{b}	12.0 ± 0.59^{cd}	10.7 ± 0.36^{de}	21.3 ± 0.07^{a}	8.9 ± 0.61^{e}	12.6 ± 0.52^{c}
Fibre	8.7 ± 1.67^{bc}	11.8 ± 0.60^{ab}	13.6 ± 0.68^{a}	5.3 ± 0.19^{c}	10.3 ± 1.43^{ab}	12.6 ± 0.49^{ab}
CHO	13.4 ± 2.27^{b}	12.9 ± 2.72^{b}	14.0 ± 2.00^{b}	26.4 ± 1.33^{a}	17.0 ± 2.66^{ab}	14.8 ± 2.67^{b}

Mean (± SE) values followed by the same superscript letter(s) within columns are not significantly different at ($p \leq 0.05$).

The energy content was highest in termites (409.5Kcal/100g), followed by cockroach (395.1Kcal/100g) then black soldier fly larvae (387.5Kcal/100g). However there was no significant variation ($p > 0.05$) in energy content among termites, cockroach and black soldier fly larvae diets. The lowest energy content was recorded in polychaete worms' diet (281.1Kcal/100g) (Figure 6).

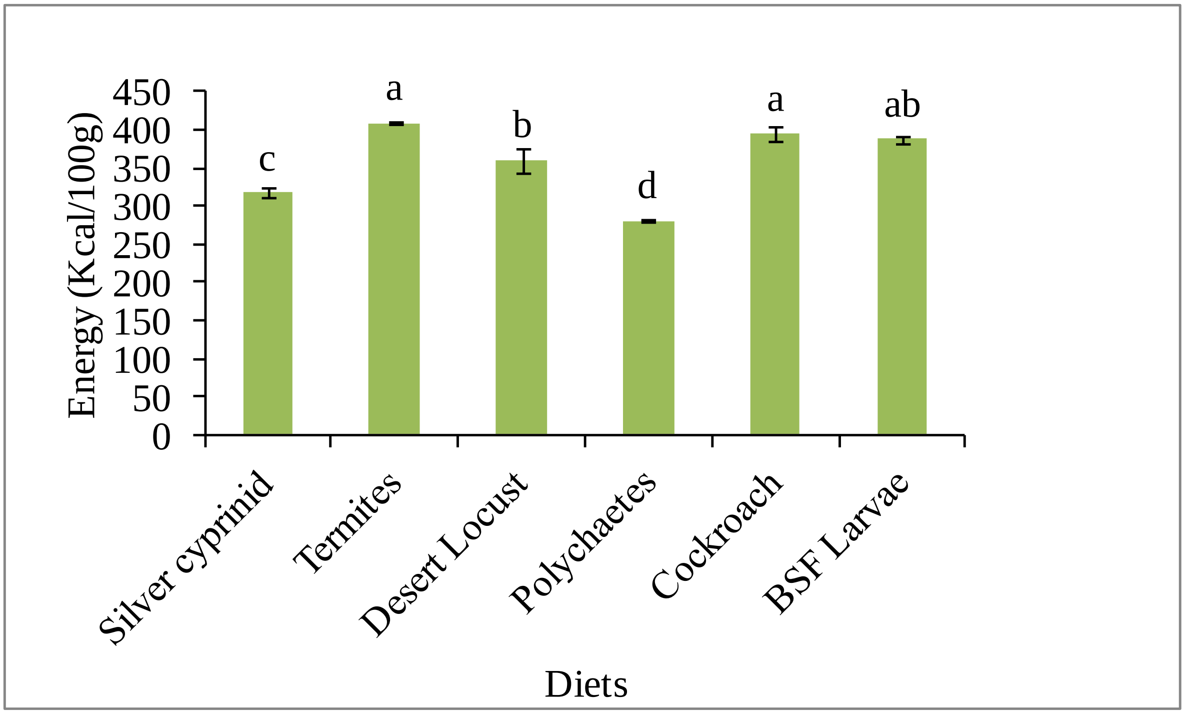

Figure 6: Energy content (Kcal/100g) of experimental diets fed to catfish fingerlings

Bar graphs accompanied by similar letters are not significantly different ($p \leq 0.05$); Error bars represent Control error of means.

4.4 Amino acid composition in the experimental diets

Six essential amino acids (histidine, isoleucine, leucine, methionine, phenylalanine, and valine) were present in the experimental diets (Table 4) except cockroach diet that lacked histidine (Table 4). The amino acid composition of experimental diets was significantly different ($p < 0.05$) among the diets (Table 4).

There was considerable presence of non-essential amino acids (arginine, glutamic acid, hydroxyproline, proline, serine and tyrosine) in experimental diets. The concentration of non-essential amino acids was significantly variable ($P < 0.05$) among the six experimental diets (Table 4).

Table 4: Amino acid composition (ug/mg) of the experimental diets fed on catfish fingerlings

Amino acid composition	Experimental diets					
	Silver Cyprinid	Termites	Desert Locust	Polychaetes	Cockroach	BSF Larvae
Essential amino acids						
Histidine	5.6±0.20[a]	5.6±0.24[a]	6.1±0.46[a]	5.8±0.44[a]	-	6.0±0.25[a]
Valine	19.8±0.7[a]	16.4±0.7[b]	9.2±0.69[d]	16.7±1.26[b]	13.0±0.55[c]	17.7±0.73[b]
Methionine	11.1±0.40[b]	23.6±1.00[a]	6.8±0.51[d]	6.8±0.51[d]	10.4±0.44[b]	8.7±0.36[c]
Isoleucine	7.9±0.28[ab]	7.3±0.31[bc]	8.3±0.62[a]	6.1±0.46[d]	8.3±0.35[a]	7.0±0.29[c]
Leucine	6.5±0.23[bc]	5.6±0.24[d]	6.5±0.49[bc]	6.2±0.47[c]	10.2±0.43[a]	7.0±0.29[b]
Phenylalanine	15.5±0.55[c]	17.5±0.74[bc]	22.0±1.65[a]	12.2±0.92[d]	6.3±0.27[e]	18.8±0.78[b]
Non essential amino acids						
Arginine	58.6±2.08[a]	-	-	46.6±3.50[b]	41.8±1.78[b]	5.7±0.24[c]
Proline	28.8±1.02[a]	20.8±0.88[c]	17.4±1.31[d]	16.6±1.25[d]	21.7±0.92[c]	25.9±1.07[b]
Tyrosine	7.2±0.25[bc]	7.5±0.32[ab]	6.8±0.51[c]	6.6±0.50[c]	7.6±0.32[ab]	7.9±0.33[a]
Serine	35.6±1.26[b]	20.7±0.88[c]	33.5±2.52[b]	47.6±3.58[a]	8.0±0.34[d]	20.1±0.83[c]
Glutamic acid	26.7±0.95[b]	35.1±1.49[a]	27.0±2.03[b]	36.2±2.72[a]	22.4±0.95[c]	34.6±1.44[a]
Hydroxyproline	6.2±0.22[bc]	6.2±0.26[bc]	6.6±0.50[b]	6.0±0.45[c]	7.5±0.32[a]	6.5±0.27[bc]

Mean (± SE) values followed by the same superscript letter(s) within rows are not significantly different at ($p \leq 0.05$)

4.5 Growth performance of catfish fingerlings fed on experimental diets

The catfish fingerlings were sampled once per week during the experimental period to determine the growth performance. The initial weight of the catfish fingerlings ranged between 0.3 and 0.4g (Figure 7). The body weight of fish increased progressively during the two months experimental period and was significantly higher ($p < 0.05$) at the end of the experiment (Mean= 1.1g) as compared to the start of the experiment (Mean= 0.4g). At the end of the experiment, fish fed on black soldier fly larvae had a significant higher ($p < 0.05$) weight gain (Mean = 401.2%) compared to desert locust (Mean = 172.3%) (Table 5). However weight gain on fish fed on black soldier larvae was not significantly different ($p > 0.05$) from those fed on silver cyprinid (Control) and cockroach. The lowest weight gain was recorded on fish fed on termites (78.6%) and polychaete worms (54.2%) (Table 5).

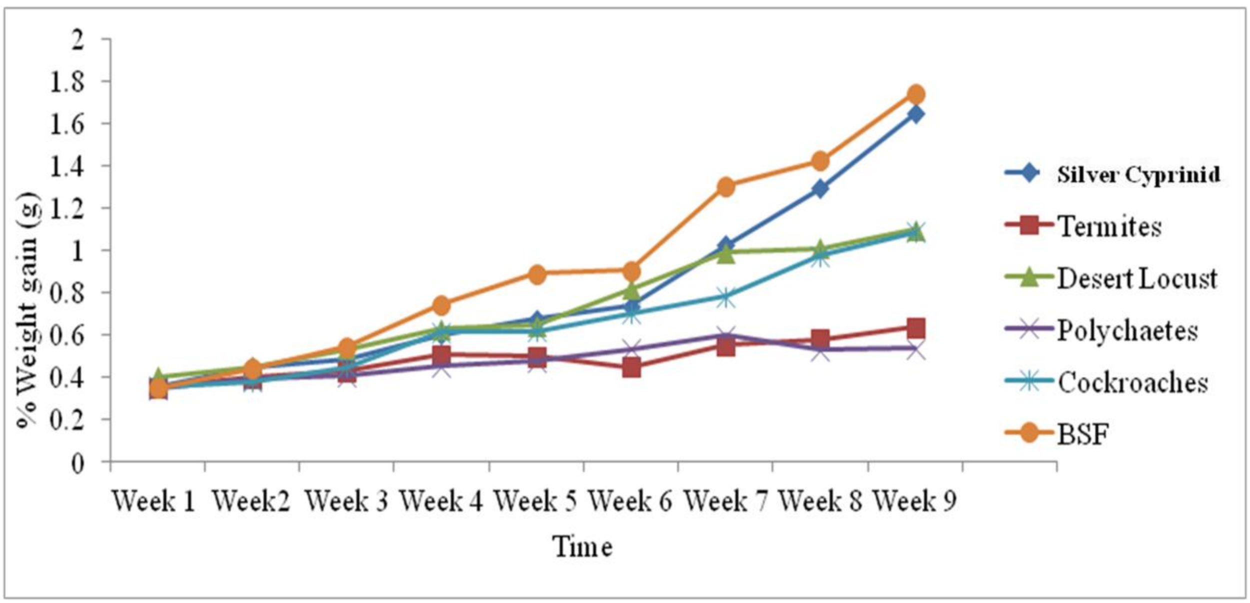

Figure 7: Weight gain throughout the experimental period

Table 5: Growth performance and survival of catfish fingerlings in different experimental diets (means ± SE)

	Initial mean weight (g)	Final mean weight (g)	Mean weight gain (g)	Weight gain (%)	SGR (g day−1)	Survival (%)
Silver Cyprinid	0.4±0.01^{a}	1.6±0.38^{ab}	1.2±0.37$_{ab}$	362.3±92.8^{ab}	2.5±0.38^{a}	45.8±6.67^{b}
Termites	0.4±0.01^{a}	0.6±0.1^{ab}	0.2±0.09^{bc}	78.6±21.85^{c}	1.0±0.26^{b}	9.2±0.83^{c}
Desert Locust	0.4±0.00^{a}	1.1±0.11^{ab}	0.7±0.10^{abc}	172.3±24.28^{bc}	1.7±0.15^{ab}	62.5±9.01^{ab}
Polychaetes	0.3±0.01^{a}	0.5±0.11^{b}	0.2±0.11^{c}	54.2±31.05^{c}	0.7±0.33^{b}	9.2±5.47^{c}
Cockroach	0.4±0.02^{a}	1.1±0.11^{ab}	0.7±0.12^{abc}	209.7±45.65^{abc}	1.9±2.5^{ab}	70.0±2.50^{a}
BSF Larvae	0.3±0.02^{a}	1.7±0.31^{a}	1.4±0.30^{a}	401.2±85.91^{a}	2.6±0.27^{a}	60.8±2.21^{ab}

Mean (± SE) values followed by the same superscript letter(s) within columns are not significantly different at ($p \leq 0.05$). Tukey's test

The specific growth rate of catfish fingerlings significantly varied ($F_{5,12}$=1.8, $p < 0.05$) among the experimental diets (Figure 8). Specific growth rate was highest in fingerlings fed on black soldier fly larvae (2.64%) followed by silver cyprinid (Control diet) (2.47%) but the difference between these two diets was not significant. The lowest specific growth rate was observed on fingerlings fed on polychaete diet (0.66%). Overall, the specific growth rate was significantly higher ($p < 0.05$) in fish fed on black soldier fly larvae compared to other treatments (Figure 8).

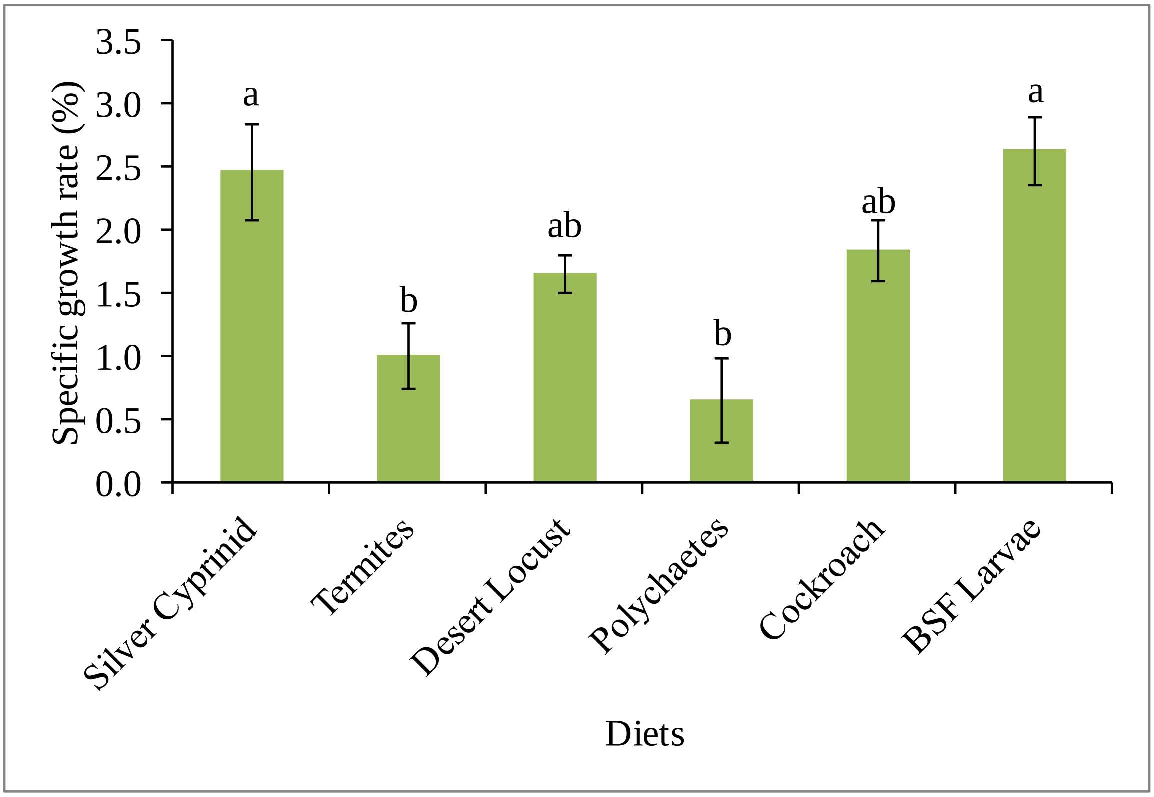

Figure 8: Specific growth rate of catfish fingerlings fed on different experimental diets

Bar graphs accompanied by similar letters are not significantly different ($p \leq 0.05$); Tukey's test; Error bars represent Control error of means.

4.6 Survival rate of the catfish fingerlings fed on different experimental diets

At the end of the experiment, the survival rates of catfish fingerlings ranged from 2.5% to 75% and varied significantly ($F_{5,12}$=0.44, $p < 0.05$) among the different experimental diets (Figure 9). The highest survival was recorded in fingerlings fed on cockroach diet (70.0%) followed by those fed on desert locust (62.5%) then black soldier fly larvae (60.8%) diets but survival rate was not significantly different ($p > 0.05$) for these three diets. The lowest survival rate was recorded in fingerlings fed on termite diet (9.2%) which was not significantly different from those fed on polychaetes (Figure 9).

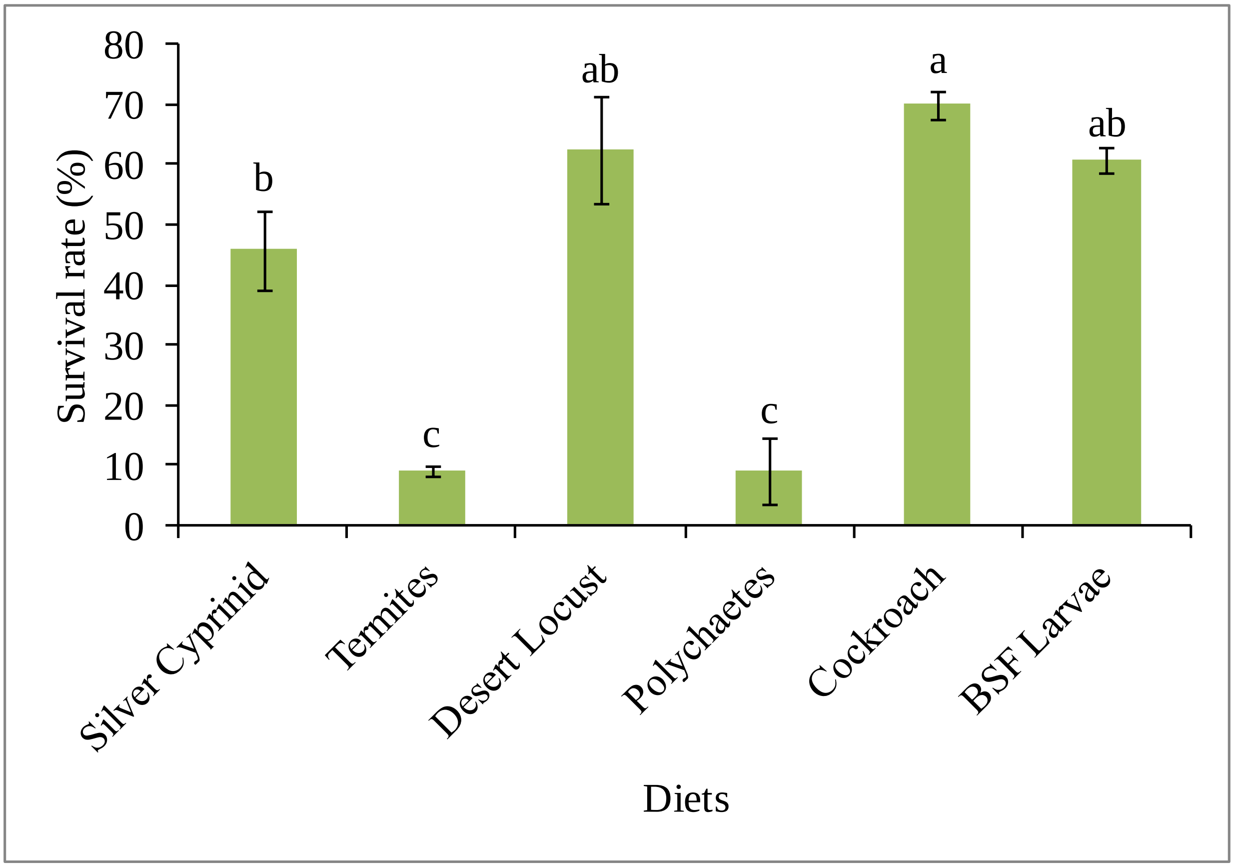

Figure 9: The survival rate of catfish fingerlings fed on different experimental treatment diets Bar graphs accompanied by similar letters are not significantly different ($p \leq 0.05$); Tukey's test; Error bars represent Control error of means.

4.7 Amino acid composition in the fish tissue

All the essential amino acids (histidine, isoleucine, leucine, methionine, phenylalanine, and valine) were present in the fish tissue. The composition of amino acid in the catfish tissue varied significantly ($p < 0.05$) among the experimental diets. In addition, non-essential amino acids such as Arginine, glutamic acid, glutamine, hydroxyproline, proline, serine and tyrosine were also present in the fish tissue. However, no arginine was detected in the tissue of fingerlings fed on termites and polychaetes (Table 6).

Table 6: Amino acid composition (µg/mg) of catfish fingerlings tissue fed on experimental diets

	Catfish Tissue					
Amino acid composition	**Silver Cyprinid**	**Termites**	**Desert Locust**	**Polychaetes**	**Cockroach**	**BSF Larvae**
Essential amino acids						
Histidine	6.8±0.01[b]	7.0±0.23[ab]	6.9±0.01[b]	6.8±0.22[b]	-	7.9±0.04[a]
Valine	18.2±6.00[a]	13.8±0.50[a]	49.4±36.22[a]	26.9±0.97[a]	19.5±2.91[a]	10.7±2.09[a]
Methionine	15.7±5.26[a]	7.1±0.25[b]	7.7±1.56[b]	12.6±0.46[ab]	11.6±2.02[ab]	7.3±1.34[b]
Isoleucine	7.1±0.72[ab]	5.5±0.20[b]	6.8±0.70[ab]	9.5±0.34[a]	7.5±1.02[ab]	6.7±0.86[ab]
Leucine	6.4±5.60[a]	5.9±0.21[a]	8.7±2.48[a]	10.8±0.39[a]	6.4±0.15[a]	8.9±2.78[a]
Phenylalanine	6.1±6.34[a]	5.5±0.20[a]	6.7±0.76[a]	6.3±0.23[a]	5.9±0.14[a]	6.3±0.37[a]
Non essential amino acids						
Arginine	-	-	40.4±20.40[a]	-	32.6±16.42[a]	23.1±23.12[a]
Proline	30.1±5.577[ab]	26.5±0.95[ab]	34.3±6.08[ab]	35.1±1.27[a]	27.3±3.56[ab]	18.3±9.02[b]
Tyrosine	7.3±0.36[a]	6.3±0.23[a]	6.4±0.50[a]	6.6±0.24[a]	11.0±4.59[a]	6.0±0.21[a]
Glutamine	38.8±19.41[ab]	58.8±2.12[a]	16.9±16.86[ab]	57.2±2.06[a]	17.3±17.26[ab]	37.1±18.91[ab]
Serine	25.2±9.27[a]	21.6±0.78[a]	30.2±10.17[a]	29.9±1.08[a]	22.7±7.88[a]	-
Glutamic acid	29.2±1.83[a]	21.2±0.77[b]	28.8±9.52[ab]	22.3±0.80[b]	28.3±2.06[ab]	-
Hydroxyproline	6.6±0.29[a]	6.0±0.22[a]	6.4±0.39[a]	6.3±0.23[a]	10.5±3.89[a]	6.3±0.11[a]

Mean (± SE) values followed by the same superscript letter(s) within rows are not significantly different at ($p \leq 0.05$); Tukey's test

4.8 Amino acid composition in the experimental diets and fish tissue

The experimental diets and tissue showed great similarity in the amino acid composition (Figure 10). Most of the essential amino acids detected in the diets were present in the fish tissue although varying in concentration. Significant variation ($p < 0.05$) in essential amino acid composition was observed between experimental diets and tissue. Histidine was present in some treatment diets and consequently in the corresponding fish tissues, although in some instance there was significant difference ($p < 0.05$) in the composition of essential amino acid between black soldier fly larvae and fish tissue fed on the same diet (Figure 10). Phenylalanine and valine were significantly higher ($p < 0.05$) in black soldier fly larvae while leucine was more abundant in the tissue. However, methionine, phenylalanine and valine were significantly higher ($p < 0.05$) in termites compared to tissue. The amino acids valine was significantly higher in the tissue compared to cockroach diet. There was significant variation ($p < 0.05$) in the concentration of amino acids between the desert locust diet and fish tissue fed on desert locust (Figure 10). Valine was more abundant and significantly higher ($p < 0.05$) in the tissue compared to desert locust diet.

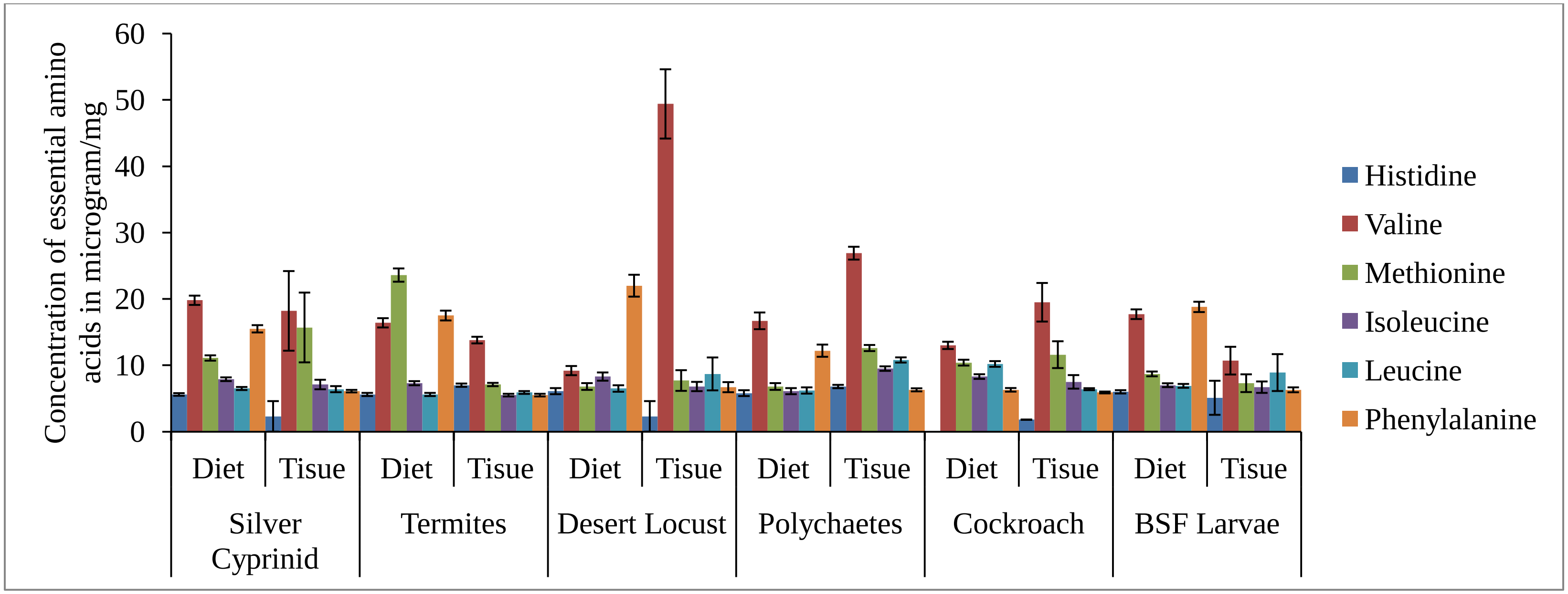

Figure 10: Essential amino acid composition of experimental diets and tissue of catfish fingerlings

Error bars represent Control error of means; Tukey's test

The concentration of non essential amino acids was significantly different ($p < 0.05$) between experimental diets and fish tissue (Figure 11). Arginine was present in the diet but lacked in the fish tissue. For instance in termites and desert locust, Arginine lacked in the diet and the fish tissue fed on termites but present in the fish tissue fed on desert locust. In silver cyprinid, arginine content was the most abundant of all the amino acids. However no arginine content was detected in fish tissue fed on silver cyprinid. No arginine content was detected in both termites and fish fed on the same diet. Analysis of the amino acid content in cockroach diet and tissue indicated that arginine was present in both the tissue and in the diet; however, the concentration was higher in the tissue compared to the diet. Significant higher ($p < 0.05$) concentration of amino acid arginine was detected in polychaetes diet while proline was higher in fish fed on the same diet. Except in black soldier larvae, the amino acids glutamic acids and serine were the most abundant non-essential amino acids in diets and low concentration in the tissue. In cockroach, glutamic acid, hydroxyproline, serine and tyrosine was more abundant in tissue than the diet (Figure 11). Non essential amino acid serine was most abundant in silver cyprinid and desert locust diets while glutamic acid was most abundant in their tissues. Significant higher ($p < 0.05$) concentration of glutamic acid was observed in termite diet compared to tissue. However no significant variation was observed in the hydroxyproline and tyrosine content between tissue and diet. In polychaetes, glutamic acid and serine were significantly higher ($p < 0.05$) in the diet than in the tissue (Figure 11).

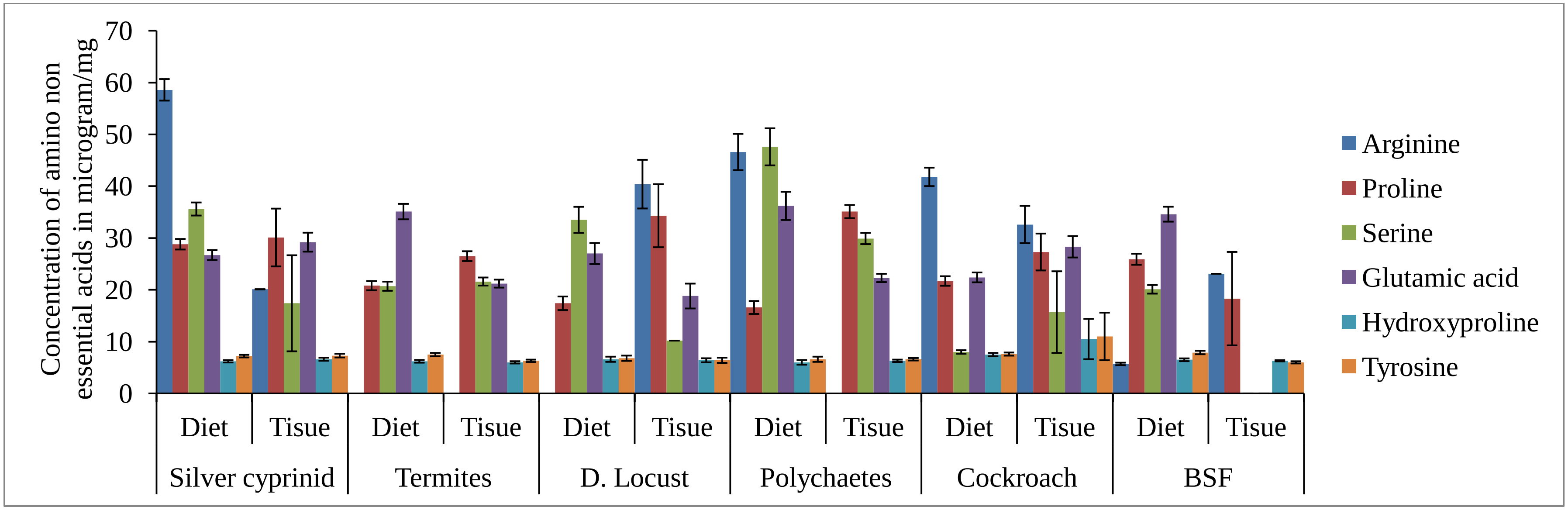

Figure 11: Non-essential amino acid composition of experimental diets and tissue of catfish fingerlings

Error bars represent Control error of means; Tukey's test

The results of correlation analysis of the catfish (*Clarias gariepinus*) fingerlings tissue and their diets show a strong relationship between the dietary and the tissue amino acids (for both essential and non essential amino acids) (Table 7). A positive relationship between dietary and tissue amino acids was observed in all groups. There was a highly significant positive correlation ($P < 0.01$) between the dietary and the fish tissue amino acids in black soldier fly larvae, silver cyprinid, cockroach, Polychaete worms and termites. A significant positive correlation ($P < 0.05$) was observed between dietary and tissue amino acids in desert locust (Table 7).

Table 7: Correlation analysis of amino acids in experimental diets and fish tissue amino acids

	Silver cyprinid Diet	Silver cyprinid Tissue	Termite Diet	Termite Tissue	Desert Locust Diet	Desert Locust tissue	Polychaete Diet	Polychaete Tissue	Cockroach Diet	Cockroach Tissue	BSF Larvae Diet	BSF Larvae Tissue
Silver cyprinid Diet	1											
Silver cyprinid Tissue	0.80**	1										
Termite Diet	0.76**	0.64**	1									
Termite Tissue	0.92**	0.83**	0.59**	1								
D. Locust Diet	0.72**	0.39*	0.68**	0.50*	1							
D. Locust tissue	0.83**	0.70**	0.54**	0.93**	0.50*	1						
Polychaete Diet	0.81**	0.61**	0.76**	0.69**	0.78**	0.64**	1					
Polychaete Tissue	0.87**	0.82**	0.53**	0.94**	0.32^{ns}	0.83**	0.60**	1				
Cockroach Diet	0.86**	0.84**	0.65**	0.95**	0.49*	0.89**	0.75**	0.89**	1			
Cockroach Tissue	0.77**	0.72**	0.58**	0.86**	0.38^{ns}	0.82**	0.68**	0.81**	0.82**	1		
BSF Diet	0.95**	0.68**	0.78**	0.82**	0.88**	0.76**	0.92**	0.71**	0.80**	0.70**	1	
BSF Tissue	0.66**	0.72**	0.50*	0.67**	0.50*	0.50*	0.66**	0.60**	0.76**	0.52**	0.66**	1

**Correlation coefficient significant at $p < 0.01$; *correlation coefficient significant at $p < 0.05$; ns - not significant.

CHAPTER FIVE: DISCUSSION, CONCLUSION AND RECOMMENDATIONS

5.1 Water quality parameters

In this study water quality parameters such as temperature, PH and dissolved oxygen were within the optimum range (for various treatments thus did not have any adverse effect on the growth of catfish fingerlings. However, the amount of TAN content was above the set Control of between 0.3mg/l and1mg/l which might have contributed to the general slow growth of the catfish fingerlings.

5.2 Nutrient composition of target organisms and experimental diets

The proximate composition of protein, fat, carbohydrate, lipid, moisture and ash contents was determined in target organisms and the experimental diets. The knowledge of the proximate composition of fish feeds is important because of the dietary and medical emphasis on the role of these nutrients in human health (Shaji and Hindumathy, 2013). Fish feed constituents play a vital role in acceleration of fish growth, reproduction and determination of survival rate. Protein is an indispensable nutrient that must be incorporated in the diet at correct proportion to guarantee sufficient growth and health of fish (Ogunji *et al*., 2008).

In this study, it was observed that the crude protein (CP) content of target organisms was highest in silver cyprinid, cockroach and desert locust compared to BSF larvae, polychaete worms and termites. Likewise the protein content in experimental diets was highest in silver cyprinid and significantly varied among the different treatments. The high protein content in the diets could have contributed to higher growth of catfish fingerlings in silver cyprinid and cockroach compared to polychaetes and termites. Previous studies have reported high crude protein (CP) content of insects which varies from 42 to 63% (Tran *et al*., 2015). Catfish fingerlings require diets with at least 35 percent protein in the diet (Robinson *et al*., 2006). So except for termites that had 32% protein all the other protein sources had adequate protein for catfish fingerlings. Dietary protein plays significant role in supplying amino acids for the biosynthesis of the body proteins which are essential for growth of fish (Alam *et al*., 2016). A protein deficiency often leads to reduced growth which may partly explain the reason for low growth in termite base treatment. On the other hand, excessive protein tend to increase the feeding cost needlessly (Obeng *et al*., 2015) together with compromising water quality in the system especially in terms of TAN concentration (Sallenave, 2012). The optimum dietary protein levels however depend on "the fish growth rate, feed intake, amount of non-protein energy in the diet, protein quality, presence of natural food and management practices" (Davis *et al.*, 2009).

Fats are highly digestible and an important source of concentrated energy and play several key roles in the growth and development of fish (Robinson and Li, 2005). In this study, the fat content varied among the target organisms but was significantly higher in termites compared to the rest and significantly lower in polychaetes. It seems however like the high fat content in the termites was not advantageous in fish growth since this treatment recorded the lowest weight gain, Control growth rate and survival of the fish. A diet lacking in essential fatty acids decreases the weight gain of fish (Obeng *et al*., 2015). It is likely therefore that the high fat content in termites may be composed of non-essential fatty acids and therefore not able to promote growth and survival of fingerlings. "Insects often accumulate fat, especially during their immature stages" (Manzano-Agugliaro *et al*., 2012). According to Robinson and Li (2012), lipid level in catfish feeds should be between 5 to 6 %. However, "the nature and quantity of lipid used in catfish diets are based on essential fatty acid requirements, economics, constraints of feed manufacture, and quality of fish flesh desired" (Robinson and Li, 2005). Thus it is important to profile the fatty acids from different feed sources to establish presence of the important essential fatty acids especially for termites that have such high levels of fats.

The ash content significantly varied among the target insects and worms and was significantly higher in polychaete worms compared to the BSF larvae, cockroach, desert locust, silver cyprinid and termites. Likewise, polychaete diet had the highest ash content compared to other diets. The high ash content in polychaete diet did not positively effluence specific growth and survival rates of catfish fingerlings. On the other hand, the high ash content in BSF larvae, desert locust and silver cyprinid in this study could have positively influenced the growth of fingerlings as a result of the presence of mineral that promotes growth. "High ash content of >12% in feed has been reported to produce better growth performance in *Clarias* species" (Kiriratnikom and Kiriratnikom, 2012). Ali and Jauncey (2004) observed that growth performance of *Clarias gariepinus* was better on diet containing 9.3% ash content. Ash content is a measure of the total amount of mineral elements such as calcium and phosphorous within a feed (Obeng *et al*., 2015). It has been reported that ash content in the feed of *C. gariepinus* should not be < 8% in order to provide sufficient mineral for the growth, survival of fish and good performance of immune system and healthy skin (Alam *et al.*, 2012).

Fibre is very important in fish feed as it gives it the physical bulkiness (Obeng *et al*., 2015). The presence of fibre in feed improves "binding and moderates the passage of feed through the alimentary canal" (Ayuba, and Iorkohol, 2012). However, high levels of fibre content more than 8-12 % in fish feed are not advisable as it "lowers digestibility of nutrients" and slows the growth (De Silva and Anderson, 1995). The fibre content observed in this study significantly varied among the experimental diets and was highest in BSF larvae, cockroach, desert locust and termites diet but lowest in polychaete worm diet. The high crude fibre content in termites might have contributed to slow growth rate of catfish fingerlings possibly due to the inability of the fish to digest and utilize the high crude fibre in the diet unlike in BSF larvae, cockroach, desert locust and silver cyprinid diets. In the results of Agokei *et al.* (2011), significant high growth performance of *C. gariepinus* juveniles was found in the diet that contained <2% fibre content.

This study showed that polychaetes diet had a higher proportion of carbohydrates while termite diet had the least. The high content of carbohydrates recorded in polychaete diet could be positively attributed to poor growth rate observed in catfish fingerlings. "Atypical catfish feed contains 25 percent soluble (digestible) carbohydrates plus 3 to 6 percent more carbohydrates that are generally present as crude fiber (mainly cellulose)" (Robinson *et al*., 2006). Tan *et al.* (2007) reported that carbohydrate in the diet of *Clarias* species should not exceed 20%, if it thus, food conversion ratio begins to decrease. Other studies showed that *C. gariepinus* does not utilize large amount of carbohydrates for growth (Mustapha *et al*., 2014). Therefore, carbohydrates being essential nutrients in fish diets should be provided in the required amounts since they serve as a source of energy for growth and reproduction (Obeng *et al*., 2015).

The moisture content recorded in this study significantly varied among the target organisms with polychaetes having the highest content while BSF larvae the lowest. Likewise, the highest value of moisture content in experimental diets was also observed in polychaetes. According to Robinson and Li (2012), the moisture content in catfish feeds should not exceed 12% to prevent the feed from molding and maintain palatability. In this study therefore, I suppose the high moisture content in polychaete diet might have contributed to deterioration which might have been responsible for slow growth rates observed in fingerlings. High moisture level predispose fish feeds to decompose if unpreserved for extended period after harvest (Adefemi, 2011) and therefore should be maintained "below 8% to increase shelf-life of fish feed during storage" (Soorensen, 2003).

In this study the value of energy significantly varied among the target insects and worms and was higher in termites compared to the rest. On the other hand, the energy content in experimental diets was significantly higher in BSF larvae, cockroach and termites compared to polychaetes and silver cyprinid. The energy content was below the optimum requirement in all the experimental diets, however, the low energy content might have played a role in slow growth and survival rates of catfish fingerlings fed on termites and polychaetes diets. "Energy requirements reported for catfish, generally been expressed as a ratio of digestible energy (DE) to crude protein (DE/P), range from 7.4 to 12 kilocalorie/ gram (kcal/g)" (Robinson *et al*., 2006). "A DE/P ratio of 8.5–10 kcal/gram is adequate for use in commercial catfish feeds" (Robinson *et al*., 2006). "Increasing the levels of energy in catfish diets above the required range may increase fat deposition, a reduction in food intake and thus reduce nutrient intake and if the energy value is too low, the fish will grow slowly" (Robinson *et al*., 2006).

5.3 Amino acids

"Amino acids are the major constituents of non-protein nitrogen in fish, accounting for 50–85%" (Ozden, 2005). The most vital amino acids required by fish include alanine, arginine, glutamic acid, glycine, histidine, proline, and taurine (Ozden, 2005). In this study, essential amino acids such as histidine, isoleucine, leucine, methionine, phenylalanine, and valine were present in the fish diets. These dietary amino acids are essential for growth, survival and fish metabolism. The same spectrum of essential amino acids observed in the experimental diets was also detected in the fish tissue an observation similar to those of Alam *et al*. (2016). The high concentration of essential amino acids in the tissue could be as a result of correspondingly higher concentration of the same amino acids in the diet. "Amino acids such as alanine, isoleucine, phenylalanine, and serine together with glycine form polypeptides that support re-growth and tissue healing in fish" (Peng *et al.*, 2013). "It is very important to supply all essential amino acids to fish in an appropriate quantity for optimal protein synthesis" (Alam *et al*., 2016). In this study, arginine, glutamic acid, hydroxyproline, proline and serine tyrosine non-essential amino acids were detected in fish diets. Likewise, non-essential amino acid glutamic acid, glutamine, hydroxyproline, serine and tyrosine were present in fish tissues. Osibano *et al*. (2009) reported that serine, glutamic acid, tyrosine and hydroxyproline are the main non-essential amino acids found in catfish.

The results of this study indicate that histidine was highest in desert locust and BSF larvae diets and lowest in silver cyprinid and termite diets. The high content of histidine in desert locust and black soldier fly larvae diets might have influenced the growth of the catfish fingerlings. On the other hand, histidine content was significantly higher in fish tissue fed on black soldier fly larvae and termites compared to desert locust, polychaetes and silver cyprinid. "Histidine is required for growth and repair of tissue, maintenance of the myelin sheaths, and removing heavy metals from the body" (Heimann, 1982). According to Førde-Skjærvik *et al*. (2006), "dietary supplementation of histidine increases intramuscular histidine levels and pH, while reducing muscle gapping in Atlantic cod". Moreover, "dietary supplementation of histidine improves sensory attributes such as flavor of aquacultured seafoods since histidine and its related imidazole derivatives (anserine and carnosine) confer desirable taste and texture" (Ogata, 2002). The high levels of intramuscular histidine which contributes to a higher quality fillet (Førde-Skjærvik *et al*., 2006) could explain the high growth performance observed in fish fed on black soldier fly larvae.

Isoleucine is a branched chain essential amino acid that is required for muscle formation and proper growth (Charlton, 2006). In this study, isoleucine was significantly higher in cockroach and desert locust diets compared to BSF larvae, polychaetes and termites. On the other hand, isoleucine content was significantly higher in fingerlings fed on polychaetes and lowest in termites. It has been reported that "isoleucine is necessary for haemoglobin formation, stabilizing and regulating blood sugar and energy" (Osibano *et al*., 2009). Therefore the high concentration of isoleucine in cockroach and desert locust diet might have influenced high growth performance, survival and high quality of the fingerlings.

Leucine content was highest in cockroach, black soldier fly larvae, silver cyprinid and desert locust diets respectively in this study. On the other hand, concentration of leucine in the tissues was higher in polychaetes, BSF larvae and desert locust but lowest in termites. "As a dietary supplement, leucine has been found to slow the degradation of muscle tissue by increasing the synthesis of muscle proteins" (Mohanty *et al*., 2014). Moreover, leucine encourages the healing of bones, skin and muscle tissue (Osibano *et al*., 2009); and these could have been the reason why the catfish fingerlings performed better in terms of growth and survival in fingerlings fed on black soldier fly larvae, cockroach, desert locust and silver cyprinid.

Methionine is a requisite amino acid which is involved in protein formation and other key physiological functions (Ghomi and Alizadehna, 2012). "Dietary methionine increases growth performance of a number of fish species such as Juvenile Cobia *Rachycentron canadum* (Zhou *et al*., 2006) and carp *Cyprinus carpio* (Schwarz *et al*., 1998) gibel carp *Carassius auratus gibelio"* (Hu *et al*., 2008). In this study, the high methionine content recorded in silver cyprinid, cockroach and BSF larvae diets might have influenced the growth rate of catfish fingerlings. Likewise, high content of methionine in fingerlings tissue fed on silver cyprinid, cockroach and desert locust positively correlated with proper growth and survival rates.

In the present study, phenylalanine was significantly higher in desert locust and BSF larvae diets compared to cockroach diet. Likewise, fish fed on desert locust and black soldier fly larvae respectively had the highest concentration of phenylalanine. "Dietary phenylalanine has a great impact on feed intake, growth performance, immunity and survival of fish in natural environment" (Li *et al*., 2009). Therefore, the high concentration of phenylalanine in desert locust and black soldier fly larvae diets and respective tissues might be accountable for the high growth performance and survival rates observed in the catfish fingerlings.

The "amino acid Valine plays an crucial role in protein synthesis, optimum growth of fish tissue, repair and maintenance of nitrogen (N) balance in the fish body" (Ahmed and Khan, 2006). Valine in the current study was higher in silver cyprinid and black soldier fly larvae compared to desert locust diets. Presence of high dietary valine levels in silver cyprinid and black soldier fly larvae might have contributed to high growth rates. As an indispensable amino acid involved in many metabolic activities (Zehra and Khan, 2014), the addition of an optimum amount of valine is a prerequisite for formulating a diet balanced in amino acids (Ahmed and Khan, 2006). In this study however, the concentration of valine in fingerlings tissue was higher in fish fed on desert locust, polychaetes and cockroach which indicate that the quantities of valine in the given diets were well assimilated.

Amino acid content in experimental diets revealed that control diet had the highest concentration of non essential acid arginine while black soldier fly larvae had the least. On the other hand, arginine content was higher in fish tissues fed on desert locust and least in black soldier fly larvae. No arginine content was however detected in termite diet, desert locust diet and on catfish tissues fed on polychaetes, silver cyprinid and termites which could explain the slow growth rates observed especially in fish fed on polychaetes and termites. “Fish require high dietary levels of arginine, since it is plentiful in protein inform of peptide bound amino acids and tissue fluid- phosphoarginine, a major reservoir of ATP; and its de novo synthesis is limited” (li *et al*., 2009). Hence in this study, arginine as a conditionally essential amino acid was synthesized and detected in fish tissue fed on desert locust whereas it was not present in the diet. Buentello and Gatlin (2001) found that the “survival of channel catfish in response to challenge with Enteric Septicaemia of catfish (*Edwardsiella ictaluri*) critically depended upon dietary arginine levels.” “Growth and health-promoting effect of dietary arginine beyond meeting requirement for protein synthesis has been reported in fish” (Li *et al*., 2009).

Non-essential amino acid tyrosine is a common “precursor for essential hormones and neurotransmitters, including thyroxine (T4), triiodothyronine, epinephrine, norepinephrine, dopamine, and melanin” (Chang *et al*., 2007). In the present study, tyrosine was most abundant in black soldier fly larvae and cockroach diets. It was also observed that fish fed on cockroach, silver cyprinid and desert locust diets, respectively had higher concentration of tyrosine in their tissues. Studies have reported that “dietary levels of tyrosine could profoundly influence pigmentation development, feed intake, growth performance, immunity, and survival of fish in natural environment” (Li *et al*., 2009). For this reason, the high level of tyrosine in black soldier fly larvae and cockroach diets could be the reason fingerlings fed on the respective diets exhibited the highest growth rates.

In this study, proline was significantly higher in silver cyprinid, then BSF larvae and cockroach diets but lowest in desert locust diet. However, the concentration of proline in tissue was significantly higher in fingerling fed on polychaetes, desert locust and silver cyprinid compared to BSF larvae. Proline, being a conditionally essential amino acid for fish in both early life and adult stages, promotes feed intake (Li *et al*., 2009) making it vital in the fish nutrition. The high concentration of proline in silver cyprinid, BSF larvae and cockroach diets and fingerling fed on desert locust diet might have influenced high growth and survival of the fingerlings. Studies have reported that "proline concentration in muscle of rainbow trout was dependent on dietary proline" (Zhang *et al*., 2006). In this study, the high concentration of proline in the tissue fingerlings fed on silver cyprinid could be attributed to the correspondingly high amino acid in the diet.

The highest concentration of glutamic acid in this study was observed in polychaete diet and least in cockroach diet. On the other hand, tissues of catfish fingerlings fed on silver cyprinid, desert locust and cockroach diets had the highest concentrations of glutamic acid. "Glutamic acid is necessary for trans-amination reactions and synthesis of molecules, such as glutathione required for removal of highly toxic peroxides and polyglutamate folate cofactors" (Mohanty *et al*., 2014). It is also fundamental for cell proliferation in fish (Zhao *et al*., 2010; Peng *et al.*, 2013). Therefore the high assimilation of glutamic acid in fish fed on silver cyprinid, desert locust and cockroach diets could explain the high growth rates observed. Although the concentration of glutamic acid was higher on termites and polychaetes, no significant effect on growth was observed on catfish fed on these diets.

The results of this study indicate that hydroproxyline was significantly higher in cockroach diet which could explain the high growth and survival rates observed in fingerlings fed diet. Aksnes *et al*. (2008) reported that "dietary supplementation of hydroxyproline, increased growth rate and modified bone composition of salmon". Hydroproxyline is responsible for collagen synthesis (Liu *et al*., 2014) and in fish it is estimated at 7% of the amino acid residues in collagen (Sato *et al*., 1989). It is also involved in stimulation of tissue protein synthesis through multiple signaling pathways (Phang *et al*., 2008; Wu *et al*., 2011). "Muscle hydroxyproline has been reported as the only free amino acid in tissues that is positively associated to the growth rate of juvenile salmon" (Dabrowski *et al*., 2005). In this study, fingerlings fed on cockroach, silver cyprinid and desert locust, respectively had the highest concentration of hydroproxyline in their tissues. High concentrations of hydroxyproline in muscle of fast growing animals are an indicator of a high rate of collagen turnover (Adams and Frank, 1980). Therefore, the high concentration of hydroproxyline especially in tissues of fingerlings fed on cockroach, silver cyprinid and desert locust could be responsible for high growth rates.

Glutamine which is a crucial amino acid to the immune response in fish (Li *et al*., 2007) was only detected in fish tissue in this study. The concentration was more abundant in fish fed on termites and polychaetes diets and lowest in desert locust diet. Glutamine is an important energy substrate in fish and the most plentiful free a-amino acid in fish plasma and muscle (Li *et al*., 2009). Due to the stress attributed to high level of TAN in polychaetes diet and high fat content in termite diet, this might have led to synthesis of glutamine in high amounts in the aforementioned experimental diets.

"Amino acids, the building blocks of body proteins, are responsible for growth, development, repair and maintenance of cells" (Marques *et al*., 2010). They participate in healing processes (Mohanty *et al*., 2014) and deficiencies in amino acid hinder many recovery processes (Osibona *et al*., 2009; Peng *et al*., 2013; Mohanty *et al*., 2014). Therefore, knowledge of the amino acid requirements in fish is crucial for proper diet formulation, enhanced growth, yield and higher nutritive value (Shaji and Hindumathy, 2013).

In this study, a significant positive correlation was obtained between the dietary and tissue amino acid. The result shows that the amino acid profile of catfish tissue was positively influenced by the dietary amino acids which are in conformity with the study by Zakeri *et al*. (2014). According to Mambrini and Kaushik (1995), the whole body amino acid profile better reflects the amino acids requirements of the fish. In this study, the best correlation results were observed in dietary and tissues amino acids in silver cyprinid and cockroach diets. The amino acid profile in the tissue of catfish replicated the respective dietary amino acids, an observation consistent to that of (Zakeri *et al*., 2014).

5.4 Growth performance of catfish

The weight of catfish fingerlings in this experiment significantly increased during two months experimental period with the highest mean weight gain observed on fish fed on black soldier fly larvae, results consistent with those of Talamuk (2016). The high nutritional composition in the diets could be attributed to the increase in body weight especially in fingerlings fed on black soldier fly larvae, cockroach, desert locust diets and silver cyprinid. Thus, "the high nutritive value of feed promotes better growth and higher yield in fish" (Madu *et al*., 2003).

The specific growth rates of catfish fed on black soldier fly larvae was significantly higher compared to other experimental diets. The good growth performance of fish fed on black soldier fly larvae is an indication that the diet contained well balanced nutrients as well as its high digestibility and nutrient utilization. A previous study in rainbow trout has shown that insects such as the black soldier fly larvae (BSF) can effectively replace fishmeal (St. Hilare *et al*., 2007). De Silva and Anderson (1995) stated that “the quality of a feed is a function of how well that feed meets the nutrient requirement of a fish”. The results of this study indicate that fish fed on polychaetes and termites resulted in the least specific growth during the study period. This could be attributed to the fact that termites contained very high fat content which reduce digestibility of the diet. Therefore, the growth rate and survival of fish depend on the quantity of feed available and its nutritional composition.

In this study, the survival rate of catfish fingerlings was higher on fish fed on cockroach diet followed by those fed on desert locust and then black soldier fly larvae diets. However, lowest survival rates were recorded in fish fed on termites and polychaetes diets. Previous studies have shown that the lowest survival of 80% was recorded in 75% and 100% termite meal diets with the highest survival rate of 93.3% recorded in control diets (Sogbesan and Ugwumba, 2008). Termites have been shown to be “suitable replacement for soybean, fish meal and vitamin premix without any reduction in growth performance” (Men *et al*., 2005). This was not the case in this study as low survival rate of catfish fingerlings fed on termites was observed. This was probably due to the fact that as the fingerlings advanced in age, the high fat content in termite diet hindered assimilation of other nutrients and hence poor sustenance and satisfaction in the growing fingerings.

5.5 Conclusion

From this study, water quality parameters such as temperature, pH, dissolved oxygen and total ammonium nitrate (TAN) in different treatments remained constant throughout the study period. The water parameters; temperature, pH and dissolved oxygen were within the recommended range and did not negatively influence the growth of catfish fingerlings. However, total ammonium nitrate (TAN) was above the recommended range which could have negatively influenced the general growth rate of fingerlings. The assessment of nutrient composition (crude proteins, fat, ash, crude fibre, carbohydrates, moisture and energy) in target insects and worms and in experimental diets was within the recommended range for catfish fingerlings rearing. It was established that BSF larvae, cockroach, desert locust and silver cyprinid can be used as a potential source of the aforementioned nutrients without compromising the nutrient utilization, whole-body composition, nutrient digestibility, growth and survival rate.

The amino acid analysis of experimental diets indicated that, the target insects and worms are particularly good sources of essential and non-essential amino acids- arginine, glutamic acid, glutamine, histidine, hydroxyproline, isoleucine, leucine, metionine, phenylalanine, proline, serine, tyrosine and valine are the most important amino acids for fish growth and survival. In this study we confirmed that the catfish tissues examined had high levels of the aforementioned essential and non-essential amino acids for promoting tissue growth, good health, and high fillet quality.

Weight gain, specific growth rate and survival rates of the catfish were best when fed with black soldier fly larvae, desert locust, cockroach and silver cyprinid (Control diet). The highest weight gain and growth rates observed in catfish fed on black soldier fly larvae, cockroach, desert locust and silver cyprinid, respectively correlated to the balanced nutritional content and the availability of amino acids responsible for growth. The slow growth performance of fingerlings fed on polychaetes and termites diets may be attributed to high anti-nutritional factors which depressed the feed intake, growth and low palatability of these diets. The results of this study indicated that the highest survival rates observed in catfish fingerlings fed on cockroach, desert locust, black soldier fly larvae and silver cyprinid diets, respectively are associated to proper nutrients in these diets. The low survival rate of fingerlings fed on polychaetes and termites diets could have been due to low digestibility and unpalatability of these diets.

Growth, health and reproduction of fish are chiefly dependent upon an adequate provision of well balanced nutrients and amino acids both in quality and quantity. It is imperative to feed fish with suitable feed containing the required nutrients and amino acids, in their required proportions in order to get optimum body weight gain, growth rate and survival rate of the fish. The result of this study showed that there are nutritional benefits from using cheaply available dried insets such as cockroach, desert locust and BSF larvae to substitute fish meal in diets of catfish fingerlings.

5.6 Recommendations

The following recommendations are made:

1. There is need to evaluate the effectiveness, acceptability and utilization of insects and worms in fish farms with different farmed fish species.
2. There is need for further investigation on fats and lipids in the insects and worms and fish tissue which was not possible during the study due to low amounts of samples collected.
3. Experimental diets were fed to the catfish fingerings during their early stages for approximately eight weeks. Further studies are recommended to determine the growth trend of the catfish fed on the same diets to maturity.
4. The insects used as target materials for feeds had a positive response towards fish growth, therefore, they can be potential protein source for fish feeds at a lower cost. Ways should be established on how to rear the insects for mass production for fish feeds to be used by Kenyan fish farmers.
5. There is need for further studies to compare growth rate of catfish fed on conventional feeds in the market and the experimental fish diets.
6. Further analysis of the experimental fish diets is required to determine other micronutrients like vitamins, minerals and their effects on the growth rate of the fish.
7. Further work should be done on better storage techniques of target insects and worms to avoid deterioration during storage.
8. Future experiments should focus on the shelf life of experimental feeds to determine their rate of degradation.

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