COMBINED CHICKEN ENTRAILS AND BLOOD MEAL AS REPLACEMENT TO FISH MEAL IN SOYBEAN BASE DIET OF *Clarias gariepinus.*

BY

KAMALU CHIGBOROGU BEDE GABRIEL

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SUPERVISOR: DR. P.C. EGWUI

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CERTIFICATION

This is to certify that I am responsible for the work submitted in this Thesis. I also certify that the original work is mine, except as specified in acknowledgements and references and that this Thesis has not been submitted to this University or any other institution for the award of degree.

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Kamalu, Chigborogu Bede Gabriel Date

NAU/PG/M.Sc/2013596008P

DEDICATION

This project work is dedicated to God almighty for all his guidance and protection throughout the project work.

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ABSTRACT

The effect of replacement to fishmeal with mixed chicken entrails and blood meal (CEBM) in the feed of *Clarias gariepinus* fingerlings on growth, production performance and food utilization were evaluated for the period of 56 days. The experimental diets contained mixed chicken entrails and blood meal at level of 0%, 5%, 10% and 15% of the total dietary protein respectively. A total of 120 fingerlings of homogenous sizes were introduced into 12 tanks of 100 litres volume, each randomly assigned to the four diets. Each treatment had 3 replicates with 10 fingerlings in each tank. The following indices of growth performance monitored showed that CEBM(o%)(112.60±4.85g), which is control diet had the highest mean weight gain, CEBM(5%) (109.00±2.80g), CEBM(10%)(76.13±6.19g) while the least was recorded with group fed with CEBM(15%)(60.44±1.65g). Percentage weight gain was highest with CEBM(0%)(383.19±18.41 g) followed by the group fed with CEBM(5%)(373.71±9.24g), CEBM(10%) (260.27±21.98g) while the least value was recorded by the group fed with CEBM(15%)(257.32±18.15g). The specific growth rate of CEBM(0%)(1.22±0.03g), CEBM(5%)(1.21±0.01g) while the least values were recorded by the groups fed with CEBM10%(0.99±0.05g) and CEBM(15%)(0.90±0.04g). Best food conversion ratio was recorded CEBM(0%)(1.39±0.03g) followed by CEBM(5%)(1.42 ±0.02g) while the poorest was recorded with CEBM(15%)(1.75±0.12g). The best protein efficien cy ratio was observed in highest growth dietary treatments CEBM(0%)(1.64±0.04g) while the poorest was recorded with CEBM(15%)(2.73±0.05g) and their values being significant differenc e ($p \leq 0.05$) between fish fed with control diet and other groups, but there was growth in all treatments with mixed chicken entrails and blood meal. This was an indicator that the diets met the nutritional requirement of the fish to promote growth and tissue development. The separate proximate Analysis of the chicken entrails blood meal revealed that Chicken entrails has moistur e (18.00%), Fats(37.00%), crude protein(28.00%), crude fibre(13.70%), carbohydrate(11.00%), calorific value(489.00kcal/g) and Blood has moisture(20.00%), Ash(8.00%), Fats(12.00%), crud e protein(30.00%), crude fiber(8.60%), carbohydrate(30.00%), Calorific value (348.00kcal/g). T he result proximate composition of the formulated diets used for the growth trial had Moisture(1 0.00%), the diet with CEBM(0%) has the highest crude protein(43.80%), followed by CEBM(5 %)(33.30%), the least crude protein was in the CEBM(15%)(21.00%), Crude Fiber CEBM(0%)(38.50), CEBM(5%)(29.00%), CEBM(10%)(33.00%), CEBM(15%)(23.70%), Fats CEBM(O%)(19.00%), CEBM(5%)(25.00%), CEBM(10)(31.00), CEBM(15%)(43.00%), Ash CEBM(0%)(4.0 0), CEBM(5%)(8.00%), CEBM(10%)(3.60%), CEBM(15%)(12.00%), Carbohydrate CEBM(0%)(23.20), CEBM(5%)(23.70%), CEBM(10%)(30.90) CEBM(15)(14.00%) and Calorific value C EBM(0%)(439.00Kcal/g) CEBM(5%)(453.00Kcal/g), CEBM(10%)(501.00Kcal/g and CEBM(1 5%)(527.00Kcal/g). Mineral elements in chicken entrails and blood meal analyzed showed that S odium(ppm) Chicken entrails(0.010), Blood(0.016), Zinc(ppm) Chicken entrails(0.031), blood(0. 22), Selenium(ppm) Chicken entrails(0.125), blood(0.128), Iron(ppm) Chicken entrails(0.001), b lood(0.001), Potassium(ppm) Chicken entrails(0.048), blood(0.032), Copper(ppm) Chicken entra ils(0.001), blood(0.001), Chromium(ppm) Chicken entrails(0.007), blood(0.001) and Phosphate(Mg/l) Chicken entrails(0.653), blood(2.339) and when compared with recommended FAO/WHO standard showed that chicken entrails blood meal can be used for fish feed formulation or for animal feed. The water quality parameters had the mean values pH 7.50±0.17, Dissolved oxygen 6.23±0.11Mg/l and Temperature 25.75±0.16°C. Their values are within the acceptable range for fish culture in the tropic. Therefore the present study revealed that combined chicken entrails and chicken blood meal can be used as replacement of the fishmeal at the level of 5% inclusion.

TABLE OF CONTENTS

List of Tables

CHAPTER ONE

INTRODUCTION

1.1 General Background

Nutritive value of fish diet depends on quality of the protein ingredients used in diet formulation (Glencross *et al.,* 2007). Protein is the most expensive component in fish feeds and the fishmeal is the major source of protein in fish diet. At present, fishmeal is not only expensive but also a scarce commodity due to its large demand in animal and fish feed industries (Akegbejo and Fasakin, 2008). The global production of fish meal has remained relatively stable over the last two decade, and supplies are unlikely to improve (Lunger *et al.,* 2006).

Therefore, there is a growing concern to identify other alternate animal protein sources which can minimize/lessen the use of fish meal in fish diet. Several plant and animal protein sources have been used in formulating the practical diets for warm water fish with varying degree of success. However, the main draw back to the use the plant protein in fish diets is the presence of variety of endogenous anti-nutritional factors (Glencross *et al.,* 2007).

Generally, the feed stuffs of animal origins are considered better alternative protein sources to fish meal in formulating fish diets because of their higher protein content and the superior indispensable amino acids than that of plant origins (Tiamiyu *et al*., 2018). Future fish diets will include a wide range of alternative ingredients, including combinations of ingredients from animal origins (Glencross *et al.,* 2007). Several animal protein sources were evaluated to formulate the diets for different fish species such as poultry by-product meal, meat and bone meal, blood meal, feather meal, a mixture of meat meal and blood meal, garden snail meal, poultry viscera meal, turkey meal, tad pole meal, fermented silage made from fish meal, shrimp meal, blood meal, maggot meal and tilapia meal, fermented fish offal, fishery by-catch and processing waste, fish waste meal, tuna by-products, a mixture of feather meal, chicken offal and maggot meal, a blend of

animal protein comprised of meat and bone meal, poultry by-product meal and hydrolysed feather meal, chicken concentrate, poultry by-product blend and chicken egg concentrate and *Surumi* by-product meal (Kedar *et al.*, 2013).

In the aqua feed formulation, protein is the main but expensive ingredient and its quality and quantity in fish feeds formulation plays a vital role in promoting fish growth (Tabinda *et al.,* 2013). However fish meal as a protein source is used worldwide in the preparation of feeds but the major challenge is its availability, quality and cost fish meal is unsustainable both environmentally and financially as a protein source for fish feeds (Tabinda *et al.*, 2013).

A diet should supply all essential nutrients and energy in tune with the animal's needs for the maintenance of vital physiological functions such as growth, reproduction and health. Besides, in aquaculture as in other animal production systems, another major issue is that of ensuring flesh and environmental quality, both of which are related to nutrition. Since the nutrient requirements for all the new species under aquaculture are not known, it is rather a common practice to extend data from more or less closely related species. In the formulation of diets, it is essential that, even when the diets are formulated theoretically to contain all the essential nutrients in adequate quantities, the availability of these nutrients from the raw materials used can vary significantly. The diet should be supplied in a form which is easily accepted by the cultivated animal and should have little adverse environmental impact.

1.2 Statement of the Problem

One of the major problems militating against fisheries and livestock development in the tropics is the availability of the feed at economic price (Agbabiaka *et al.*, 2013).

The widening gap between estimated protein intake and actual protein requirement in developing countries like Nigeria, may be attributed to lack of basic information and improper harnessing of the abundant non conventional protein sources that has restricted their being incorporated in commercial feed (Ajah, 2007). Feed producti on account for over 70% of the cost of raising commercial and household livestock as well as cultural fish in Nigeria and this high cost and scarcity of commercial feed are important factor militating against increased livestock and fish production, emphasis should be shifted towards alternative feed ingredients (Ejidike, 2002).

Fishmeal is incorporated in nearly all fish feed due to its high biological value. Nutritionists see it as being the highest quality protein source that is commonly available to the fish manufacturers (Lovell, 1998).

Therefore, there is a need for the research of low cost, locally available protein sources to replace wholly or partially proportion of the fishmeal.

1.3 Justification of the Study

The justification of this study lies in an attempt to alleviate the teething problems militating against fish farming and livestock development due to expensive cost of fish meal. It is imperative to research into the locally available protein sources such as chicken entrails and blood meal if they can be used as alternative to fishmeal

1.4 Aim of the study

The aim of this study is to assess the growth performance of fish fed with mixed chicken entrails and blood meal as replacement to fish meal in soybean diet of *Clarias gariepinus* fingerlings **.**

1.5 Objectives of study

The objectives of the study were to:

- Determine the effect of mixed chicken entrails and blood meal as replaceme nt

  to fishmeal in soybean meal diets, on the growth performance, food utilizatio n and survival of African catfish *Clarias gariepinus*.
- Determine separate proximate analysis of the chicken entrails and blood meal.
- Determine the proximate composition of the formulated diets
- Determine separate mineral elements of chicken entrails and blood meal
- Determine the water quality parameters in experimental tanks containing *Clarias gariepinus.*

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Use of Fish in Feeds

In 2008, global aquaculture production reached 52.5 million tonnes (excluding aquatic plants), growing at an annual rate of 8.4 percent. Its proportional contributi on to total food fisheries output increased from 3.9 percent in 1970 to 42.9 percent in 2008 (FAO, 2010a), indicating the important role it plays in supplying fish for human consumption. There is an increasing trend for aquaculture to be dependent on feeds. In 2008, about 31.5 million tones or 46.1 percent of total global aquacult ure production were dependent upon the direct use of feed, either as a single ingredient, as farm-made *aquafeeds* or by the use of industrially manufactured compound *aquafeeds* (FAO, 2010a).

Total industrial compound *aquafeed* production increased almost four fold from 7.6 million tones in 1995 to 29.3 million tones in 2008, with production growing at an average rate of 10.9 percent per year (Tacon *et al.*, 2010). Commonly used key ingredients in *aquafeeds* are Protein sources: fishmeal, soybean meal, various oilse ed cakes and meals; Energy/carbohydrate sources are various cereals and cereal by-products;and lipids/oils: fish oil and vegetable oils (De Silva ; Hasan, 2007). Compound feeds are used both for the production of lower-value (in marketing terms) food-fish species such as non-filter feeding carps, tilapia, catfish and milkfis h *(Chanos chanos)*, as well as higher-value species such as marine finfish, *salmoni ds*, marine shrimp, and freshwater eels and crustaceans. Within the animal husbandry subsectors, aquaculture is now the largest user of fishmeal and fish oil. In 2007, aquaculture is estimated to have used 68.4 percent (3.84 million tonnes) of world fishmeal production and 81.3 percent (0.82 million tonnes) of fish oil production (Tacon *et al.*, 2010). Globally, about five million tonnes of trash

fish/low-value fish are used directly as feed in aquaculture (Edwards *et al.*, 2004). In 20.4 million tonnes (22.4 percent of the global fish and shellfish landings) was reduced into fishmeal and fish oil (FAO, 2O10a). Increased use of fishmeal and fish oil and trash fish/low-value fish in aquaculture can primarily be attributed to the increase in production of carnivorous species, particularly marine crustaceans, marine finfish, *salmonids* and other *diadromous* fishes (Rana *et al.*, 2009). Howeve r, it is projected that over the next ten years or so, the total use of fishmeal by the aquaculture sector will decrease while the use of fish oil will probably remain around the 2007 level (Tacon *et al.*, 2010). Fishmeal is produced through a reducti on process where the fish are cooked, press-dried and milled into meal, Fish oil is a by-product of the process. On average, 4—5 kg of wet fish will yield 1 kg of fishmeal and I00g of fish oil (De Silva and Anderson, 1995). The raw material used in industrial reduction processes consists mainly of low-value fish, often referred to as forage fish or feed fish, obtained from reduction fisheries and as bycatch' resulting mainly from food-fish trawling and artisanal fisheries.

The biggest reduction fisheries are those in the southeast Pacific (example. Peruvian *anchoveta* fishery) and northwest Europe. Some of these fisheries also produce fish for human consumption. While bycatch is a worldwide phenomenon, it is mainly in East Asia where it provides significant quantities of fish for aquacult ure. The main artisanal feed-fish fisheries occur in the Asia-Pacific region(Wijkstr om, 2009).

Globally, the main species used for the manufacture of fishmeal and fish oil are small pelagic species such as anchoveta *(Engraulis ringens),* sand eels *(Animodytes* Species.), Atlantic menhaden *(Brevoortia tyrannus),* capelin (Family Osmeridae, example *Mallotus* species.), Atlantic herring *(Clupea harengus horengus),* Norway pout *(Trisopterus esmat/cil),* European sprat *(Spraltus sprauus),* Chilean jack mackerel *(Trachurus murphyi)* and chub mackerel *(Scomber japonicus)* (De Silva

and Turechini, 2009). In Asia, fishmeal production is based on a mix of species typically derived from trawl fisheries and increasingly from seafood industry processing wastes. Although various feed ingredients of plant and animal origin are often used, whole and chopped trash fish/low-value fish remains the most widely used feed ingredient for feeding high- value, marine carnivorous fish throughout the Asia-Pacific region (De Silva and Turechini, 2009).

However, there is a marked difference among the global regions regarding the sourcing of fish-based protein for compound commercial and farm- made *aquafeeds*. The Asia-Pacific region is the largest consumer of feed fish, reduced or otherwise, as feed in aquaculture. Approximately 25 percent (9.8 million tonnes) of the total capture fishery production of 40 million tonnes in the Asia-Pacific region is currently used other than directly for human consumption for example fishmeal production or as animal/pet food. This contributes towards the production of 2 million tonnes of food fish for human consumption in the region (Funge-Smith *et al.*, 2005; FAO, 2007). In 2003, over 9.9 million tonnes or 47.2 percent of the total fishery catch within the Americas region was destined for reduction and non-food uses (Tacon, 2009), while the farming of mainly carnivorous species in Europe currently uses around 1.9 million tonnes of feed fish to meet fishmeal and fish oil requirements (Huntington, 2009). In Africa and the Near East, around 0.86 million tonnes of pelagic fish were reduced to fishmeal and fish oil in 2004-05 (Hecht and Jones, 2009). Although the majority of fishmeal/fish oil is derived from marine species, there is an emerging trend to use freshwater *pelagics* in *aquafeeds*. In Kenya, between 50 and 65 percent of the silver cyprinid *(Rastrineobola argentea,* local name: "*dagaa*", also known as "*otnena*" in Uganda) catch from Lake Victoria is reduced to fishmeal (Abila, 2003). In 2004, the total recorded "*dagaa*" catch was 31 659 tonnes (FAO, 2006b), suggesting that 15 800 to 20 500 tonnes of fish was reduced to fishmeal. With growing popularity of aquaculture in Africa, it can be

expected that more fish will be used to supply the industry.

2.2 The Limitations for the Used of Fish as Feed in Aquaculture

There is a growing concern that the use of fish as feed in aquaculture has more negative than positive implications for the poor, and that it is not ethically correct to use fish as feed if it can be used for human consumption. There are five main concerns regarding the use of fish as feed; these relate primarily to the supply of low-priced fish as food, income earning possibilities (Wijkstrom, 2009) and direct impacts on ecosystems and biodiversity are when fish is obtained from a reduction fishery and converted into fishmeal that is incorporated into feeds used to grow fish and/or shrimp, then less fish is available as human food-and particularly for the poor; When fish is obtained from the bycatch of commercial fisheries or from surplus landings of small pelagic fisheries and then fed to cultured fish either directly or as fishmeal, the quantities of low-priced fish normally accessible by the poor in port markets are reduced; The growing use of fishmeal in fish and other animal feed contributes to an increase in fishing pressure on reduction fisheries or direct targeting in non-selective trawl fisheries (Kristofersson and Anderson, 2006; Skewgar *et al.,* 2007). This may affect the sustainable use of some wild fish resources, and therefore eventually lead to less fish being available for human consumption, which will affect the poor in particular; if fish is obtained from a reduction fishery and converted into fishmeal, the on-shore job opportunities are lower than if the fish were destined for processing and direct human consumption. This affects the poor in particular, as much of the processing only requires low-skilled labour. Removal of large quantities of forage fish species from marine ecosystems affects other dependent *piscivorous* animal species, including other fish species, birds and mammals (Huntington *et al.,* 2004; Worm *et al.,* 2006; Skewgar *et al*, 2007); The use of trash fish/low-value fish as feed in aquaculture raises the possibility of transmitting diseases/pathogens from non-endemic feed

fish to local wild fish populations, as has been experienced in Australia (WWF, 2005).

Countering these concerns, the global fishmeal industry claims that there is no current demand for direct human consumption for up to 90 percent of the wild-caught fish that is reduced to fishmeal (FIN, 2004). From a global perspective, this is probably correct. However, on a regional or individual country basis, there is evidence to suggest that a proportion of the reduction fishery catch is simply not available for human consumption (Abila, 2003), a though if it had been available it would certainly have been consumed (Kurien, 1998). In Europe and North America, the reduction of fish has no direct consequences because of the low number of poor and undernourished people (Wijkstrom, 2009); and in Africa, reduction fisheries are an exception and aquaculture is nascent and not much dependent on fish as feed (Hecht and Jones, 2009). In the Americas, an increasing proportion of the marine fish catch is expected to be processed for direct human consumption, primarily in the form of easy-to-use and affordable processed fish products, including canned fish and stabilized *surimi*-based products (Tacon, 2009). In Asia, the situation is different. Unlike other aquaculture-producing regions, Asia is largely dependent on imported fishmeal and fish oil (mainly from South America and northwest Europe). The few industrial feed-fish fisheries that exist *in* Asia (mainly in China and Japan) have been declining (Huntington and Hasan, 2009). Manufacturers of fishmeal and fish oil have therefore had to make greater use of trawler by-catch and occasional surplus catches as raw material. The demand for trash fish/low-value fish is now also fuelled by the growth of small-scale rural aquaculture in Viet Nam, which has led to the development of a trash fish/low-value fish fishery that supplies the aquaculture sector. It is clear therefore that the use of trash fish/low-value fish has become a serious issue in certain regions, while in others it is a non-issue.

2.3 Sustainability of Fish Stocks

Irrespective of the region, fisheries that generate excessive by-catch and discards ar e ultimately not sustainable, especially where there are no management strategies f or non target species. Moreover, the removal of large numbers of forage fish from an ecosystem may directly affect their prey and predators and the viability of target and bycatch populations (Huntington and Hasan, 2009).

Although most commercially exploited feed-fish stocks are capable of withstandin g relatively large reductions in biomass (Daan *et al.*, 1990; Jennings *et al;* 2001), the removal of extremely high numbers of spawning stock may lead to recruitment overfishing. Pelagic species are particularly vulnerable to recruitment overfishing, as they are short-lived (Santos *et al.,* 2001).

The incidental catch of non-target species and, in particular, the capture of juveniles of commercial species, is one of the most controversial aspects of feed-fish fisheries, as most undersized fish are landed and processed, resulting in growth overfishing. For example, in North Atlantic waters, juvenile herring are known to shoal with sprat (Santos *et al*., 2001).

2.4 Ornamental Fish Farming

Ornamental fish farming is one of the fastest growing fishery sectors throughout the world with an annual trade of 15 million dollar and growth rate of over 10%. Freshwater ornamental fish contribute 85% of the total global ornamental fish trade (Kedar *et al.*, 2013). One of the major problems for the growth of ornamental fish farming is the non-availability of species specific nutritionally balanced diets. So far many of the ornamental fish traders have been using shrimp feeds or other fish feeds meant for rearing the food fishes. Therefore, development of species specific ornamental fish diet as per the nutrient requirement of fish is one of the priority areas in fish nutrition research (Kedar *et al.*, 2013). Among freshwater ornamental fish, blue *gourami*, *Trichogaster trichopterus* is an important sought after

ornamental fish. Using casein-gelatin-dextrin based semi-purified diets, it is reported that the blue *gourami* fingerlings require 350g protein and 80g lipid kg-1 diet with a digestible energy level of 16.7MJ kg-1 for its optimum growth and nutrient utilization (Kedar *et al.,* 2013). In the present study, based on the nutrient requirement of blue *gourami*, nine experimental practical diets were formulated using different animal protein sources such as snail meal, freshwater fish processing waste meal, surimi by-product meal, chicken offal meal, earthworm meal, squid meal, mussel meal, chicken liver meal and lean prawn meal in addition to fish meal used at 10% in all diets. Some of the animal protein sources (freshwater fish processing waste meal, *surimi* by-product meal, chicken offal meal and earthworm meal) used in the present experiment was the agro-industries wastes/ by-products. Although these by-products/waste materials have fairly good amount of protein contents, they are not being utilized so far for any productive purposes and are thrown away by the agro-processors.

2.5 Choice and Quality of Ingredients

Despite much research, both intensive and semi-intensive aquaculture relies upon a relatively small number of feed ingredients. Under semi-intensive culture condition s, cereal bran-oilcake mixture remains the major aqua feed. In intensive aquacultur e, the diets are formulated to be nutrient and energy dense, based mainly on ingredients of marine origin. Since most teleosts are known to utilize dietary carbohydrates rather poorly, the chosen ingredients are necessarily protein and energy-rich (FAO,2010c).

When it comes to finding alternatives to fish meals as a protein and amino acid source, several other agricultural by-products such as animal by-products, cereals (wheat, corn), pulses (*Lupin*, peas, *faba* beans), oil seeds (soybean, rapeseed) hold potential interest, depending upon local availability and cost. (Ajah, 2007; FAO, 2010b).

2.6 Types of Feed

Commercial fish diets are manufactured as either extruded (floating or buoyant) or presure-pelleted (sinking) feeds. Both floating and sinking feed can produce satisfa ctory growth, but some fish species prefer floating, others sinking. Shrimp for exampl e, will not accept a floating feed, but most fish species can be trained to accept a floati ng pellet (Lovell, 1998; Ajah, 2007). Extruded feeds are more due to the higher\ manu facturing costs. Usually, it is advantageous to feed a floating (extruded) feed, because the farmer can directly observe the feeding rates. It is important in maximizing fish growth and feed efficiency (Ajah, 2007).

Feed is available in a variety of sizes ranging from fine crucibles for small fish to large (circumference or lager) pellets. The pellet size should be approximately 20-30% of the size of the fish species mouth gape (Lovell, 1998; Ajah, 2007).

2.7 Feeding Rates, Frequency and Timing

Feeding rates and frequencies are in part a function of fish size. Small Larval fish and fry need to be fed with high protein diet frequently and usually in excess. Small fish h ave a high energy demand and must eat nearly continuously and be fed almost hourly. Feeding small fish in excess is not as much of a problem as over feeding larger fish because small amount of feed relatively to the volume of water in the culture system (Ajah, 2007).

As fish grow, feeding rates and frequencies should be lowered and protein content reduced. However rather than switching to a lower protein diet, feeding less allows the grower to use the same feed (protein level) throughout the grow-out period,

thereby simplifying feed inventory and storage. Feeding fish is labour- intensive and expensive. Feeding frequency dependent on labour availability, farm size, and the fish species and size grown (Lovell,1998; Ajah 2007).

Many factors affect the feeding rates of fish such as time of the day, season, water, temperature, dissolved oxygen levels and other water quality parameters (Ajah, 2007). Feed acceptability, palatability and digestibility vary with the ingredient and feed quality. Fish farmers pay careful attention to feeding activity in order to help determine feed acceptance. Calculate feed conversion ratios and feed efficiencies, monitor feed costs and track feed demand throughout the year (Ajah, 2007).

Published feeding rate tables are available for most commonly cultured fish species. Farmers can calculate optimum feeding rates based on the number of fishes in the tank, raceway, or pond. Farmed fish typically are fed 1- 5% of their body weight per day (FAO, 2007; Ajah, 2007).

2.8 Metal

The commonest causes of metal poisoning are heavy metals: copper, lead, mercury, zinc, chromium, cadmium, manganese and iron. Industrial discharges and seepage from industrial and mining wastes are the commonest sources, although sometimes they occur naturally (Ajah, 2007; Omokheyeke *et al*., 2018). Defining maximum safe levels of any particular metal is difficult, as much ancillary information is required, example pH, acidity or carbonate alkalinity, temperature, dissolved oxygen content, presence of other metals they often act synergistically, example cadmium in the presence of zinc or copper (Ajah, 2007). The pathology of metallic poisoning varies

according to the concentration and length of exposure and is not a reliable diagnostic feature unless historical and analytical evidence is also available (Omokheyeke *et al*., 2018).

2.9 Non-Metals

Many non metals are toxic if present in sufficient quantity. Some of those encountered commonly are Ammonia, Fluorides, Cyanides, Phosphorous, *Sulphides*, Aluminum and beryllium salts, arsenates and halogens, particularly chlorine and the chloramines. Many organic compounds used in agriculture and industry are toxic for fish (Ajah, 2007).

Pesticides are chemicals designed to destroy plants or animal life. The major sources are run-off from treated farmlands, industrial and domestic sewage and spillage. A great variety of such compounds is currently in use and they may be contaminants of the food of both wild and cultured fish or animals (Omokheyeke *et al*., 2018).Aquatic organisms do absorb some inorganic elements not only from their diets but also from their surroundings either in freshwater or saltwater (Ajah, 2007). Many of the trace elements are required in small amounts too difficult to formulate. The Purified diets without mineral supplements results in loss of appetite, growth depression, *hypochromic* anemia, high mortality and cranial deformities. Additional mineral improves growth and survival (Ajah, 2007).

2.10 Nutrient Classes

Carbohydrates, lipids and proteins make up the major part of the dry weight of the ingested food (Ajah, 2007; FAO, 2007). These macronutrients can be used directly as fuels (respiratory substrates) or can be stored within the body for utilization at a later date (Ajah, 2007). In addition to the macronutrients, the food will also contain a rrange of micronutrients. These micronutrients are the vitamins and minerals, which are required to be consumed in small doses (Ajah, 2007; FAO, 2007).

2.11 Carbohydrates

Carbohydrates (starches and sugars) are most economical and inexpensive sources of energy for fish diets. Although not essential, carbohydrates are included in aquacultur e diets to reduce costs and for their binding activity during feed manufacturing (Melci on *et al;* 1993; Ajah, 2007). In fish, carbohydrates are stored as glycogen that can be mobilized to satisfy energy demands. They are major energy source for mammals, but are not used efficiently by fish (FAO, 2007; Ajah, 2007).

2.12 Lipids

The lipids (fats) are heterogeneous class of water- insoluble organic compound. Bioch emical separation techniques yield fractions of the lipids that are distinguishable both on the basis of their abilities to dissolve in different solvents and by the possession of differences in physical and chemical composition (Ajah,2007). Lipids are highly-energy nutrients that can be utilized to partially spare (substitute for) protein in aquaculture feeds. Lipids supply about twice the energy as proteins and carbohydrat es. Lipids typically comprise about 15% of fish diets supply essential fatty acids (EF)

and serve as transporters for fat-soluble vitamins (Ajah, 2007; FAO, 2007).

A recent trend in fish feeds is to use higher levels of lipids in the diet. Although increasing dietary lipids can help reduce the high costs of diets by partially sparing protein in the feed, the problems such as excessive fats deposition in the liver can decrease the health and market quality of fish (FAO, 2007; Ajah, 2007).

2.13 Proteins and Amino Acids

Proteins are large organic molecules that contain carbon, hydrogen, oxygen, nitrogen and often *sulphur* . From a nutritional point of view the amino acids can be divided into non essential and essential groups. Fish and other animals are able to synthesize and interconvert some of the amino acids, but are incapable of the synthesis of others. The essential amino acids are therefore, those that the fish cannot synthesis *de novo*, whereas the non-essential amino acids are those that can be synthesized from precursor molecules. Studied carried out on a range of fish species and other animals, have revealed that ten of the amino acids are essential-a*rginine*, *histidine*, *isolencine*, *leucine* lysine, *methuionine*, phenylalanine, *threonine*, tryptophan and *valine* (Fuglie, 2000; Ajah, 2007).

2.14 Vitamins

Vitamins are organic compounds necessary in the diet for normal fish growth and health. They often are not synthesized by fish and must be supplied in the diet (Ajah, 2007). The two groups of vitamins are water soluble and fat- soluble. Water – soluble vitamins include the B vitamins, *choline*, *inositol*, folic acid, *pantothenic* acid, *biotin* and ascorbic acid (vitamin C) of these, vitamin C probably is the most important

because it is powerful antioxidant and helps the immune system in fish (Ajah, 2007). The fat- soluble vitamins include A vitamins, retinols (responsible for vision); the D vitamins *cholecaciferols* (bone integrity); E vitamins, the tocopherols (antioxidants); and K vitamins such as menadione [blood clotting, (skin integrity) of these, vitamin E receives the most attention for its important role as an antioxidant (Ajah, 2007).

2.15 Aquaculture Production

The aquaculture industry is primarily based on tilapia and catfish, cultivated in intensive (commercial) and semi intensive (artisanal) production system. Both the syst ems involved input of the supplementary formulated feeds, which account for up to 40% and 60% of production costs, respectively (FOA, 2007; Ajah, 2007). African catfish is recognized by its long dorsal and anal fins which give it a rather eel-like appearance. The catfish has a slender body, a flat bony head, and a broad terminal mouth with four pairs of barbells. Its prominent barbells give it image of cat-like whiskers. The catfish is mostly cultured in earthen ponds. However, it can be cultured in other systems such as tanks and hapas. In the wild and riverine systems, the fish reproduces naturally but considerable effort is required to induce spawning under culture conditions (Ajah, 2007). The African catfish is an excellent species for aquaculture as it is omnivorous, grows fast and tolerates relatively poor water quality (Amisah, *et al.,* 2009). The African catfish (*Clarias gariepinus*) is appreciated by consumers for the quality of its meat and is mostly smoked and used in soups (Ajah, 2007; Amisah *et al.,* 2009).

2.16 Fish Feeds Production

Two sources of fish feed has been identified. These are farm- made aqua feeds and commercially produced pelleted feeds (Ajah, 2007; FOA, 2010a). There are only a few commercially available pelleted fish feeds only specialized animal feed millers engage in fish feed production and this is only on demand; and as such , the majority of fish feeds produced (69.75 percent) are farm made (Ajah, 2007). Nevertheless, the two main types of feeds produced are for the *tilapia*, which contain 30-35 percent crude protein and catfish, which contain 45- 50 percent crude protein. In 2000, the Nigerian aquaculture industry used an estimated 35570 tonnes of feed representing a n egligible proportion (< 1 percent) of the national feed production (Ejidike, 2002).

2.17 Proximate Principles

This is a type of chemical analysis that gives an estimated and comparative value of moisture content total ash a food materials, crude protein, crude fibre, crude fat, carbohydrate and lipids. These factors are analyzed using the standard method of analysis by AOACS (2005).

2.18 Moisture Content

Moisture content determination is essential in feed storage. The safe limit for storage is 15% moisture. Feedstuffs containing more than 15% moisture should not be preserv ed, this is because it may develop the undesirable moulds and fungus (AOACS, 2005). In the determination of moisture content, temperature variation plays an important role. The even temperature and duration of drying is in relation to the nature of the sa mple. It is not only the water that expelled, it involves volatile matters, therefore

a suitable dry operation that ensures the nearest elimination of moisture without burni ng the test sample is adopted using temperature of 105^0C (AOCS, 2005).

2.19Ash

This is the organic residue remaining on the incineration of any food stuff or organic matter such as fat, protein, sugar, etc in the open under atmospheric condition on a muffle furnace. It contains useful mineral elements (AOACS, 2005).

2.20 Crude Fibre

Crude fibre is the organic residue, which remains after the material has been treated u nder standardized condition that is boiling with *telraoxosulphate (vi)* acid, boiling wit h dilute sodium hydroxide solution and neutralizing with alcohol and water. The amou nt of crude fibre in any material in terms of starch, the higher the crude fibre value, the lower the quality of the starch (AOACS, 2005).

2.21 Crude Protein

Protein may be divided into true protein and non-protein nitrogen. True protein nitrog en estimated by s*tutzer* reagent method, where protein is propitiated with cupric hydro xide in alkali conditions while non- protein portions example urea nitrogen is estimate accurately by convey diffusion techniques. In general pure protein yield 5.25-5.75 cal ories of gross energy per grams. The *Kjedah* method of determining the crude protein involves first, the digestion of the sample of heating with concentrated *tetraoxosulplh ate (vi)* by acid (H_2SO_4) in a long neck digestion flask until a clear solution is obtained . Secondly, the ammonia is distilled into basic acid contained in a conical flash and then, the titration of the boric acid with a catalyst on a sample brings about oxidation

in which the nitrogenous materials in the sample is converted into ammonia s*ulphate* (40%). When the aliquot starts boiling, the ammonia that eventually evolves is distilled into 5.0ml boric acid that is directly estimated by titration. For most routine purposes, the crude protein in the sample is then concentrated by multiplying nitrogen by an empirical factor = N x 6.25 (as the general factor) where N is the nitrogen % factor, 6. 25 is based on the fact that the protein contains 16% nitrogen protein helps in the rebuilding and repair of tissues. Deficiency of protein in children causes Kwashiorkor (AOACS, 2005).

2.22 Nitrogen Free Extract (Carbohydrate)

This is also known as soluble carbohydrate, which consists of water- soluble vitamins. Deficiency of nitrogen free extract in animals causes coronary- heat attack (AOCS, 2005)

2.23 Materials Used In Fish Feeds

There are ten major groups of materials, which can be used in fish and shrimp/ prawn feeds/ they include: grasses, legumes, miscellaneous fodder plants, fruits and vegetabl es, root crops, cereals, oil-bearing see and cakes, feeds of animal origin, miscellaneous feed stuffs and additives (Ajah, 2007).

For artificial fed to be deemed successful, it must meet the requirements for survival and growth of the cultured animal. In nature, animals consume a variety of food types but under culture condition, only one mixture of ingredients is consumed consequentl y, aquatic diet must contain approximate combinations of nutrients, which are effectiv ely and efficiently utilized (Ajah, 2007).

2.24 Options in Commercially Manufactured Feeds

There are three options available currently in commercially manufactured feeds: dry, semi, moist (Ajah, 2007).

2.25 Dry Feed

The major advantage of dry feed is that it can be stored at room temperature for several months without significant deterioration in product quality. It usually contains about 10% (percents) moisture and is the cheapest of the three options. The advantages of dry feed are that it is less palatable to the fish, and less in the nutritional value of the components such as proteins. Another characteristics of dry feed is that the fish use the water from their surrounding environment to build their tissue, which results in better conversion rates (Lovell, 1998; Ajah, 2007) .

2.26 Semi-Moist Feed

Semi–moist feed incorporates some of the benefits of the dry and moist feed. The major advantage being that it does not have to be refrigerated unless the bag is opened. I f feed is utilized without delay, refrigeration may not be needed at all. The main demerit is that, it is the most expensive being twice as expensive as dry feed. Though it is more palatable to the fish, however, based on previous trials using Atlantic salmon, frozen food gave better yield, followed by moist feed and then semi-moist (Ajah, 2007).

2.27 Dry Versus Moist

Dry feed are easier to manufacture on large scale and easier to store, transport and feed. There is evidence that moist feed may be more palatable and attractive to the animals and can give better results than dry feed (Ajah, 2007).

Moist feed (example, Trash fish) can be utilized without employing energy wasting and sometimes quality damaging techniques of fish meal production. Dry and moist feeds are usually formed into a definite physical shape- pellet, crumbles, granules, balls cakes , etc. moist feeds are normally extruded through some form of mixer to form pellets of regular diameter depending again on the diameter of the orifice through which they are extruded (Ajah, 2007).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experimental Site

The experiment was conducted in the Research Centre of the Department of Fisheries Technology, Imo State Polytechnic Umuagwo. It falls within the geographical co-ordinates of latitude 5^{O} -30^{1} and 70^{O} 3^{1} North and longitude 5^{O} 39^{1} and 5^{O} 42^{1}East. It is within the humid area of south eastern Nigeria with almost equal duration of rainfall and dry season. It has an average rainfall of about 2169.8mm and average ambient temperature of 29^{O}C and 34^{O}C as maximum. The vegetation is guinea savannah.

3.2 Collection and Preparation of Samples

The chicken entrails and chicken blood were collected raw in separate 4 liters plastic bucket at Eke-Awka market Anambra State Nigeria. The chicken entrails were washed with tap water. The chicken entrails were cut into chunks and pressure cocked at 100^{O}C for 30 minutes and the blood was also pressure cooked to curtail microbial load/contamination and after which they were oven dried at temperature 105^{O}C. The chicken entrails and blood were milled using a mini harmer mill machine to produce chicken entrails meal and blood meal and were combined in ratio 1:1.The grounded samples were stored in air tight polythene bags (Agbabiaka *et al.*, 2013).

Feed ingredients namely fishmeal, soybean meal, corn starch, cod olive oil, whole wheat, corn gluten meal, lecithin (soy refined), mineral premix, vitamin premix, cholin chloride, bony meal, lysine, methionine were bought from Fidelity Agro Services (NIG) Owerri Imo State. The plastic ponds were bought from Onitsha Ogbo Efere market Anambra State.

3.3 The Study Fish

Total number 120 *Clarias gariepinus* fingerlings were procured from fisheries department of Imo State Polytechnic Umuagwo Nigeria. The fish were conveyed to the site of the study in a 20 liter plastic bucket filed to two third volume of water from the rearing place of procurement to reduce stress. The fish were acclimatized in 12 experimental plastic ponds of 100 liters capacity for one week prior to the start of the experiment.

3.4 Duration of the Study

The research was carried out for 8 weeks (2months) between 14, September to 9th November, 2015.

3.5 Feed Formulation and Processing

The sample of the chicken entrails meal and chicken blood meal produced were analyzed for proximate composition. In order to formulate four Iso-energetic, Iso-nitrogenous and Iso-lipidic diets, different feed ingredients were mixed (Table 1) and fish meal (FM) were replaced with combined chicken entrails and chicken blood meal. Dry ingredients in different ratios were mixed and homogenized for experimental feed formulation. The formulated practical diets were pelleted using locally fabricated pelleting machine with die 1.5mm. Pelleted diets were oven-dried using electric micro oven until crispy(Binatone O-60minutes), packed into air-tight polythene bags and were labeled accordingly.

TABLE 3.1Percentage Composition of Experimental Diets for *Clarias gariepinus* Fingerlings

S/N	INGREDIENTS	FM 15 (g)	FM 10 (g)	FM 5 (g)	FM 0 (g)
1.	Fish meal(FM)	15.0	10.0	5.0	0.0
2.	Chicken entrails and chicken blood meal	0.0	5.0	10.0	15.0
3.	Soyabean meal (SBM)	50.0	50.0	50.0	50.0
4.	Cod olive oil	5.3	5.3	5.3	5.3
5.	Corn starch	5.4	5.4	5.4	5.4
6.	Whole wheat	16.0	16.0	16.0	16.0
7.	Corn gluten meal	5.0	5.0	5.0	5.0
8.	Lecithin (Soy refined)	1.0	1.0	1.0	1.0
9.	Mineral premix	0.3	0.3	0.3	0.3
10.	Vitamin premix	0.4	0.4	0.4	0.4
11.	Cholin chloride	0.2	0.2	0.2	0.2
12.	Bony meal	1.0	1.0	1.0	1.0
13.	Lysine	0.2	0.2	0.2	0.2
14.	Methionine	0.2	0.2	0.2	0.2
	TOTAL	**100g**	**100g**	**100g**	**100g**

3.6 Experimental Design

The experiment comprises of 4 dietary treatments 0%, 5%, 10% 15% combined chicken entrails and chicken blood meal respectively.

Each of the treatments had three replicates .The fishes are of homogenous sizes were randomly arranged and allotted to the respective tanks.

3.7 Feeding and Ration

The fish were fed twice daily between 8.00-9.00am and 4.00-5.00pm at 5% body weight throughout the period of experiment (Agbabiaka *et al*., 2013). The ratio was adjusted, every one week when new mean weights of fish for the various experimental units had been determined.

3.8 Cleaning of Tanks

Fecal materials and left over feeds were siphoned out daily before feeding and water replaced. Complete cleaning and changing of water was done once every week on the sampling day.

3.9 Weight Measurement

The weights of the fish were recorded in the group by group on weekly basis using *Kero* electronic scale (KlS 10001) weighing balance with sensitivity of 0.001

3.10 Determination of Indices of Growth and Feed Utilization

The following indices of growth performance were monitored such as weight gain, percentage weight gain, specific growth rate, feed conversion ratio, and protein efficiency ratio and were calculated using the formular of Agbabiaka *et al* (2013).

3.11 Weight Gain (WG)

Weight gain was expressed as the weight increase of individual in the organism life time ($t_2 - t_1$) and was expressed or determined as weekly final mean weight minus initial mean weight divided by duration of the study.

$$WG = \frac{W_2 - W_1}{T_2 - T_1} \times \frac{100}{1}$$

Where W_1 = Initial weight of fish

W_2 = final weight of fish

T_1 = initial weight

T_2 = final weight

3.12 Percentage Weight Gain (%WG)

This is percentage increase in weight. This is the percent difference between the final weight and the initial weight.

Mathematically it is stated thus:

$$PWG = \frac{W_2 - w_1}{W_1} \times \frac{100}{1}$$

3.13 Specific Growth Rate (SGR)

Specific growth rate (SGR) was determined from the relationship of the difference in the weight of fish within an experimental period in days.

$$SGR = \frac{\text{Log}e w_2 - \log e\ w_1}{T_2 - T_1} \times \frac{100}{1}$$

Were W_2 = final weight of fish at time (T_2)

W_1 = initial weight of the fish at time (T_1)

Loge = logarithmic exponent.

3.14 Feed Conversion Ratio (FCR)

This was determined as the ratio of food consumed by the fish to the weight gain of the fish expressed as

$$FCR = \frac{\text{Weight of food consumed}}{\text{Weight gain of the fish}}$$

3.15 Protein Efficiency Ratio (PER)

Protein in feed makes about 5% of the body dry weight, making them the most abundant macronutrients in the body. However, not all proteins are created equal. To better assess the quality of proteins in various feeds, analyst often use the protein' efficiency ratio, or PER, a measure of a specific proteins ability to promote fish to the crude protein consumed, measured in grams. It is based on the weight gain of a test subject divided by its intake which was evaluated from the quality of protein in feed. Commercial feed efficiency ratio as the standard for evaluating protein quality of food or feed. It is stated as:

$$PER = \frac{\text{Mean weight gain of fish}}{\text{Protein intake}}$$

$$\text{protein intake} = \frac{\text{total feed consumed x\% crude protein in feed}}{100}$$

3.16 Statistical Analysis

Data obtained were subjected to analysis of variance (ANOVA) in a completely randomize design (CRD) and the mean were differentiated using least significant difference (LSD).

3.17 The Proximate Analysis

The samples of feeds, chicken entrails and chicken blood meal were analyzed for proximate composition at the fisheries laboratory of the Imo State Polytechnic Umuagwo using the methods of the Association of Official Analytical Chemists society (AOACS, 2005), For crude fat content, Ash content, moistures contents, crude protein, crude fibre, carbohydrate contents and calorific value.

3.18 Crude fat content

The *soxhlet* extractor was set up and exactly 20g of the dried powder specimen sample was weighed out using the sensitive weighing balance (KIS10001). The weight of a dried extraction thimble (W_1) with the sample was measured out as W_2. The thimble containing the sample was placed inside the *soxhlet* extractor which was fitted into the neck of the 500m1 capacity round bottom flask containing chips which prevented bumping or trotting due to pressure. Enough quantities of the solvent that petroleum ether was poured into the round bottom flask and into the extractor to wet the sample wrapped in the thimble. The *soxhlet* extractor containing the thimble fixed to the condenser was fixed into the round bottom flask fitted a little bit above or on top of a heating mantle. The heating was maintained at a temperature of $60^{O}C$ while it lasted for 18 hours after which the extractor was disconnected and the thimble removed and then the ether was reclaimed by fixing back the extractor without the thimble with continuous heating which separated a high percentage of the petroleum ether leaving the mixture of oil and a little quality of ether in the flask. (AOACS, 2005).

Oil Recovery

The remaining petroleum ether in the oil was poured into a beaker and the ether was removed by distillation over a water bath after which the ether was left to dry off at 105°C for 30minutes. Over 60% of the petroleum ether was recovered. Therefore, the percentage by mass of the oil in the sample was calculated using the formular below:

FORMULAR

$$\% \text{ Fat} = \frac{\text{Weight of Oil}}{\text{Weight of sample}} \times \frac{100}{1}$$

3.19 Ash Content Determination

A clean crucible was dried in an oven at 105^{o}C for 30 minutes and allowed to cool. After cooling, the sample was weighed into the crucible and the weight was noted. The crucible containing the ground sample was placed in a muffle furnace and was heated at 600^{o}C. The heating continued until total decarbonization was achieved. The furnace was switched off and allowed to cool. (AOACS, 2005)

Therefore the weight of the ash content was calculated from the formular:

$$\text{Ash \%} = \frac{\text{Weight of ash}}{\text{Weight of sample}} \times \frac{100}{1}$$

3.20 Moistures Content Determination

The flat silica dish was washed, cleaned, dried and weighed empty; 1.0g of the sample was weighed into the silica dish using the weighing balance. The silica dish containing the sample was transferred into an oven at 105^{o}C and allowed to dry for 24 hours in order to achieve possible minimal moisture content. This was later removed, allowed to cool in desiccators and then weighed. (AOACS, 2005).

Calculation was done using the formular:

$$\text{Moisture content \%} = \frac{\text{Weight of sample before drying- weight of sample after drying}}{\text{Weight of sample before drying}} \times \frac{100}{1}$$

3.21 Crude Protein Determination Principle

The total nitrogen was determined by a modified *Kjeldahl* method and the result was multiplied by 6.25 to give the crude protein content. This method does not include nitrogen from protein alkaloids, nucleic acid, etc. The organic matter was oxidized by concentrated *sulphuric* acid in the presence of catalyst and the nitrogen was converted to animonium *sulphate.* This was made alkaline and the librated ammonia was distilled and estimated as percentage nitrogen since large part of the nitrogen present in food was derived from protein. The crude protein (cp) was estimated by multiplying the percentage of nitrogen by an appropriate factor; which in this case is 6.25.

Digestion

1g of the ground sample was weighed into the digestion flask then 0.5g of mercuric oxide was weighed into the flask, followed by the addition of 5g of potassium *tetraoxosulphate* (iv) acid and then 20cm^3 of concentrated *tetraoxosulhate* (iv) acid was also added. The mixture in the flask was digested using a heating mantle. The temperature of the heating mantle was regulated and digestion was continued until a clear coloured solution was acquired. The digested sample was made up to 100cm^3 in a 100cm^3 volumetric flask to form a standard solution.

Distillation

With the distillation apparatus in working order and having been steamed out for several minutes, 5ml of saturated solution of boric acid was poured into a 50cm^3 flask with the addition of two drops of the mixed indicator. The flask containing the mixture was attached to the receiving end of the condenser. Then 10cm^3 aliquot

of the digest was transferred to the distillation apparatus with the external steam vent on the steam boiler remaining open. Exactly 10cm^3 of 40% alkaline NaOH, 10% sodium *thiosuiphate* mixture slowly added from the funnel controlled by a punch cock down inside of the distillation so that the alkali tended to form a layer at the bottom. The outside vent of the boiler was continued until about 25cm^3 solut ions was received (which gave a light orange colour).

Titration

All the nitrogen in the sample held as ammonia in the boric solution was titrated with 0.lm hydrochloric acid to a light blue colour.

Calculation

The protein content was calculated using the formular:

$$\text{Nitrogen content \%} = \frac{\text{Cm}^3\text{ (acid) x (m acid) x 0.0 14 x 20}}{\text{g(sample)}} \times \frac{100}{1}$$

cm^3 = volume of acid used

m = molarity of acid used = 0.1m

Crude protein % = Nitrogen x 6.25

3.22 Crude Fiber Determination

The crude fibre was determined from the fraction remaining after extraction of oil; 2.0g of the fat free sample was weighed into a round bottom flask. 2000cm^3 of hot, 0.1275M suphuric acid solution was added to the flask containing the 2.0g of the defatted sample. The flask was later placed quickly under reflux condenser and was made to boil gently for 45minutes using distilled water to maintain the volume and to wash down particles adhering to the side of the flask Excessive foaming was checked with antifoam where necessary. The content of the flask was then filtered through whatman No 54 filter paper. The residue was washed with boiling water and then transferred back to the round bottom flask and 200cm^3 of hot 0.313

sodium hydroxide (NaOH) solution was added. The flask was replaced under reflux condenser and was again made to boil for 1 minute. After 30minutes, it was filtered through ashless filter paper. Finally, it was washed first with boiling water, then 1% hydrochloric acid and then washed with ethanol, dried overnight at 100^0C in an oven, cooled and weighed. The weighed sample and the filter paper were placed in a crucible and were ashed in a muffle furnace at 500^0C for 7 hours or till it de carbonized. After switching off the furnace, the crucible with ash was allowed to cool and then weighed. The crude fibre (%) was then, calculated. (AOACS, 2005).

Formular:-

$$\text{Crude fibre} = \frac{\text{weight of crucible + dried residue - weight crucible +Ash}}{\text{Weight of sample}} \times \frac{100}{1}$$

3.23 Carbohydrate Content Determination

The carbohydrate content of samples was calculated from the percentage of the moisture, protein, ash and fat as shown below.

Carbohydrate = 100 - *(%* protein + % ash + % moisture + % fat) (AOACS, 2005)

3.24 Calorific Value Determination

The calorific value was determined from the relation below.

Calorific value = (% protein x 4) + *(%* fat x 9) + *(%* carbohydrate x 4)

3.25 Determination of Mineral Analysis

Mineral elements in chicken entrails and blood meal were analyzed using Varian AA240 Atomic Absorption *Spectrophometer* according to the methods (APHA, 1995).

3.26 Water Quality Parameters

The physico-chemical parameters of water monitored throughout the study period include temperature, pH, dissolved oxygen using APHA method (1998) and were weekly determined before sampling of the fishes of the various unit.

1. Temperature: The temperature of water in the pond was measured with mercury in glass thermometer. The thermometer which was caliberated from O^0C to 360^0C was lowered into the water for 5 minutes and the reading was taken immediately the thermometer was raised.

2**. pH:** The pH of the water was determined by using pH meter (Model KC 101) and universal pH paper was used to double check the meter.

3. **Dissolved oxygen (DO):** This dissolved oxygen in each experimental unit was determined in the laboratory using Winkler titration method

CHAPTER FOUR

4.0 RESULT

4.10 Indices of Feed Utilization

4.11 Weight Gain

The data presented in Table 4.11 shows that the highest weekly weight gain was recorded by the fish fed with CEBM (0%) diet (112.60±4.85g) followed by those fed with CEBM (5%) diet (109.00±2.80g) while the least was recorded by those fed with CEBM (15%) diet (60.44±11.65g) respectively. The data in Table 4.11 was also subjected to analysis of variance (ANOVA) presented in Table 6 in the appendix. The analysis of variance result revealed that there was significant difference between the weight gain of *Clarias gariepinus* fingerlings fed with various percentages of the combined chicken entrails and chicken blood meal ($p< 0.05$) at 5 % level, of significance.

Table 4:1.1: Summary of mean weight gain of *Clarias gariepinus* fingerlings fed with CEBM for 8 weeks S

	Weekly mean weight of fish (g) ± SE									
Treatments										
	(W0)	**W1**	**W2**	**W3**	**W4**	**W5**	**W6**	**W7**	**W8**	**Weight gain± SE**
Control 0%	**29.40±0.12^{a}**	**34.67±0.04^{b}**	**41.73±2.78^{ab}**	**49.93±4.59^{ab}**	**64.20±4.45^{a}**	**80.47±5.26^{a}**	**104.50±6.88^{a}**	**126.97±3.53^{a}**	**142.00±4.70^{a}**	**112.60±4.85^{a}**
CEBM5%	**29.17±0.03^{a}**	**34.63±1.52^{a}**	**41.37±1.45^{ab}**	**51.50±2.21^{a}**	**63.17±1.66$^{a`}$**	**77.90±0.71^{b}**	**96.53±0.79^{b}**	**118.90±2.15^{b}**	**138.17. ±2.83^{b}**	**109.00±2.80^{b}**
CEBM10%	**29.27±0.09^{a}**	**34.77±0.90^{b}**	**38.47±1.24^{a}**	**41.77±1.82^{b}**	**49.80±3.16^{a}**	**59.87±4.95^{a}**	**74.13±3.87^{ab}**	**86.20±3.27^{a}**	**105.40±6.10^{ab}**	**76.13±6.19^{ab}**
CEBM15%	**29.23±0.09^{a}**	**38.37±1.20^{ab}**	**41.30±18.02^{a}**	**45.87±2.42^{c}**	**52.30±3.12^{ab}**	**62.30±6.46^{c}**	**73.23±7.77^{ab}**	**89.67±7.03^{c}**	**104.43±5.11^{ab}**	**60.44±11.65^{c}**

W = week, mean ±SE= standard Error; CEBM 0% = Chicken entrails and blood meal 0 %; CEBM 5% = Chicken entrails and blood meal 5%; CEBM 10% = chicken entrails and blood meal 10%; CEBM 15% = chicken entrails and chicken blood meal 15%.

4.12 Percentage Weight Gain

The data presented in Table 4.12 shows the percentage weight gain of *Clarias gari epinus* fingerlings was highest in those fed with CEBM (0%) (383.19 ± 18.41) followed by those fed with CEBM (5%) diet (373.71±92) while the least was recorded by those fed with CEBM(15%) diet (257.32±18.15) . The data presented in table 4.12 was further studied by analysis of variance (Table 7) in the Appendix.

From the Table 7 in the Appendix indicated that there was significant difference ($p<0.05$) in the percentage weight gain between *Clarias gariepinus* fingerlings fed with various percentage of chicken entrails and chicken blood meal for 8 weeks at 5% level of significance.

Table 4.12 Percentage Weight Gain of *Clarias gariepinus* Fingerlings Fed with Various Percentage of Chicken Entrails and Chicken Blood Meal for 8 Weeks.

Treatments	Replicates			Total	Mean Percentage Weight Gain±SE
	C	B	C		
Control (0%)	403.78	346.46	399.32	1149.56	383.19±18.41[a]
CEBM (5%)	373.63	389.73	357.73	1121.09	373.71±9.24[a]
CEBM (10%)	226.87	301.72	252.22	780.81	260.27±21.98[b]
CEBM (15%)	225.51	258.08	288.36	771.95	257.32±18.15[b]

Columns sharing similar superscripts are not significantly different at P>0.05, ±SE = mean standard error, CEBM= Chicken entrails and chicken blood meal.

4.13 **Specific Growth Rate (SGR)**

The data presented in Table 4.13 showed that the highest mean specific growth rate was recorded by *Clarias gariepinus* fingerlings fed with CEBM(0%) control diet (1.22±0.03) followed by those fed with CEBM (5%) diet(1.21±0.01) while the least specific growth rate was recorded in *Clarias gariepinus* fingerlings fed with the diet CEBM 15% (0.90±0.04). The data in table 4.13 was further studied by analysis of variance (Table 8) in the appendix.

Table 8 in the Appendix, there was a significant difference($p<0.05$) between specific growth rate of the *Clarias gariepinus* fingerlings fed with various percentage of chicken entrails and chicken blood meal at 5% level of significance.

Table 4.13 Specific Growth Rate of *Clarias gariepinus* Fingerling Fed with Various Percentages of Chicken Entrails and Chicken Blood Meal for 8 Weeks.

Treatments	Replicates			Total	Mean SGR±SE
	A	B	C		
Control (0%)	1.25	1.16	1.25	3.66	1.22±0.03[a]
CEBM (5%)	1.21	1.23	1.18	3.63	1.21±0.01[a]
CEBM (10%)	0.92	1.08	0.98	2.98	0.99±0.05[b]
CEBM (15%)	0.92	0.99	1.05	2.96	0.90±0.04[b]

Columns sharing similar superscripts are not significantly different at P>0.05, ±SE=Mean standard error, CEBM =Chicken entrails and chicken blood meal.

4.14 **Food Conversion Ratio (FCR)**

The data on food conversion ratio (FCR) of *Clarias gariepinus* fingerlings fed with various percentage of the chicken entrails and chicken blood meal in Table 4.14 showed that the best food conversion ratio was recorded in *Clarias gariepinus* fingerlings fed with CEBM (0%) diet (1.39±0.03), while the poorest was recorded in *Clarias gariepinus* fingerlings fed with CEBM (15%)(1.75±0.12) for 8 weeks. The data was further studied by analysis of variance (Table 9) in the Appendix.

Table 9 in the Appendix showed the analysis of variance of food conversion ratio of *Clarias gariepinus* fingerlings fed with various percentages of chicken entrails and blood meal. From the table, there was significant difference ($p<0.05$) between the food conversion ratio of the *Clarias gariepinus* fingerlings fed with various percentage of chicken entrails and chicken blood meal at 5% level of significance.

Table 4.14 Food Conversion Ratio (FCR) of *Clarias gariepinus* Fingerlings Fed With Various Percentages of Chicken Entrails and Chicken Blood Meal.

Treatments	Replicates	Initial Weight (g)	Final Weight (g)	Weight Gain	Food Fed(g)	FCR	MeanFCR ±SE
Control(0%)	A	29.10	146.60	117.50	156.60	1.33	1.39 ± 0.03^{c}
	B	29.70	132.6	102.90	144.60	1.41	
	C	29.40	146.80	117.40	169.20	1.44	
CEBM(5%)	A	29.20	138.30	109.10	153.60	1.41	1.42 ± 0.02^{c}
	B	29.20	143.00	113.80	156.60	1.38	
	C	29.10	133.20	104.10	153.60	1.48	
CEBM(10%)	A	29.40	96.10	66.70	134.60	2.02	1.73 ± 0.15^{b}
	B	29.10	116.90	87.80	133.80	1.52	
	C	29.30	103.20	73.90	122.40	1.66	
CEBM(15%)	A	29.40	95.70	66.30	115.80	1.75	1.75 ± 0.12^{b}
	B	29.10	104.20	75.10	135.00	1.80	
	C	29.20	113.30	84.20	142.20	1.69	

Columns sharing similar superscripts are not significantly different $p>0.05$, ±SE =Standard error, CEBM =Chicken entrails and Chicken blood meal.

4.15 Protein Efficiency Ratio (PER)

The data on protein efficiency ratio presented in Table 4.15 showed that the best value for protein efficiency ratio was recorded in the *Clarias gariepinus* fingerlings fed with control diet (1.64± 0.04) which is the lowest value. However, the poorest protein efficiency ratio was recorded in *Clarias gariepinus* fingerlings fed with CE BM(15%)(2.73±0.05). The data was further studied by analysis of variance (Table 10) in Appendix.

Table 10 in the Appendix, indicated that there was significant difference($p<0.05$) in the protein efficiency ratio between *Clarias gariepinus* fingerlings fed with various percentage level of chicken entrails and chicken blood meal for 8 weeks at 5% level of significance.

Table 4.15 Protein Efficiency Ratio (PER) of *Clarias gariepinus* Fingerlings Fed with Various Percentage of Chicken Entrails and Chicken Blood Meal for 8 Weeks.

Treatments	Replicates	Total Feed Consumed	% Protein	Protein Intake	Initial Weight (g)	FinalWeight(g)	Weight Gain	PER	Mean PER±SE
Control(0%)	A	156.60	43.80	68.59	29.10	146.60	117.50	1.71	1.64±0.04 [c]
	B	144.60	43.80	63.33	29.70	132.60	102.90	1.62	
	C	169.20	43.80	74.11	29.40	146.80	117.40	1.58	
CEBM(5%)	A	153.60	33.30	51.15	29.20	138.30	109.10	2.13	2.12±0.04[d]
	B	156.60	33.30	52.15	29.20	143.00	113.80	2.18	
	C	153.60	33.30	51.15	29.10	133.20	104.10	2.04	
CEBM(10%)	A	134.60	24.50	32.98	29.40	96.10	66.70	2.02	2.39±0.19[b]
	B	133.80	24.50	32.78	29.10	116.90	87.80	2.68	
	C	122.40	24.50	29.99	29.30	103.20	73.90	2.46	
CEBM(15%)	A	115.80	21.00	24.32	29.40	95.70	66.30	2.73	2.73±0.05[a]
	B	135.00	21.00	28.35	29.10	104.20	75.10	2.65	
	C	142.20	21.00	29.86	29.20	113.30	84.20	2.82	

Columns sharing similar superscript are not significantly different at P >0.05, ±SE =Mean standard error, CEBM =chicken entrails and chicken blood meal.

4.16 Different Proximate Analysis of the Chicken Entrails and Chicken Blood Meal Results.

The Table 4.16 shows the results of proximate analysis of the chicken entrails and chicken blood meal used in feed formulation. The chicken entrails meal contained moisture (18.00%), Ash (6.00%), Fat (37.00%), crude protein (28.00%), crude fibre (13.70%), Carbohydrate (11.00%) and Calorific value (489.00Kcal). Chicken blood meal contained moisture (20.00%), Ash (8.00%), Fat (12.00%), Crude Protein (30.00%), Crude fibre (8.60%) Carbohydrate (30.00%) and calorific value (348.00Kcal).

Table 4.16 Different Proximate Analysis of the Chicken Entrails and Chicken Blood Meal Results.

Parameters	**Chicken Entrails{%}**	**Blood(%)**
Moisture	18.00	20.00
Ash	6.00	8.00
Crude Fat	37.00	12.00
Crude Protein	28.00	30.00
Crude Fibre	13.70	8.60
Carbohydrate	11.00	30.00
Calorific Value	489.00Kcal/g	348.00Kcal/g

4.17 Proximate Composition of the Formulated Diets with CEBM Used for the Gro wth Trial.

The result of proximate composition of the formulated diets used for the experimen t is presented in Table 4.17. From the table, the diets used for the growth trial had the moisture (10.00%) the diet with CEBM(0%) inclusion had the highest crude protein content (43.80%) while the least crude protein was in the CEBM(15%). The crude fat was highest in the diet with CEBM(15%)while lower in the control diet. Crude fiber was highest in the control diet and least CEBM(15%). Ash content was highest in the diet CEBM(15%) and least in the control diet. The diet with CEBM(15%) had the highest calorific value and least in the control diet. The diet with CEBM(5%) had highest Carbohydrate content and lowest value was in the diet with CEBM(15%). The energy content of some of the diets was probably different and would probably produce different effects.

Table 4.17 Proximate Composition of the Formulated Diets with CEBM Used for the Growth Trial.

Parameters	**CEBM (0%)**	**CEBM(5%)**	**CEBM(10%)**	**CEBM(15%)**
Moisture	10.00	10.00	10.00	10.00
Crude Protein	43.80	33.30	24.50	21.00
Crude Fibre	38.50	29.00	33.00	23.70
Crude Fat	19.00	25.00	31.00	43.00
Ash Content	4.00	8.00	3.60	12.00
Carbohydrate	23.20	23.70	30.90	14.00
Calorific Value	439.00Kcal/g	453.00Kcal/g	501.00Kcal/g	527.00Kcal/g

Chicken entrails and chicken blood meal (0%), CEBM(5%)=Chicken entrails and chicken blood meal(5%), CEBM(10%)=Chicken entrails and chicken blood meal(10%), CEBM(15%)=Chicken entrails and chicken blood meal(15%).

4.18 Mineral Elements in Chicken Entrails and Chicken Blood Meal.

The Table 4.18 shows the separate results of the mineral elements in chicken entrai ls and blood meal used in feed formulation for *Clairas gariepinus* fingerlings and world health organization standard for fish health and recommended safe level.

Table 4.18 Mineral Elements in Chicken Entrails and Chicken Blood Meal.

Parameters	Sample Chicken Entrails	Chicken Blood	WHO Standard
Sodium (ppm)	0.010	0.016	5.000
Zinc (ppm)	0.031	0.022	≤ 5.000
Selenium (ppm)	0.125	0.128	≤ 0.010
Iron (ppm)	0.001	0.001	≤ 1.000
Potassium (ppm)	0.048	0.032	≤ 10.000
Copper (ppm)	0.001	0.001	≤ 1.000
Chromium(ppm)	0.007	0.001	≤ 1.000
Phosphate(mg/l)	0.653	2.339	≤ 2.000

ppm= part per million, ≤= Lessthan, WHO=World Health Organization

4.19 Water Quality Parameters

The table 4.19 shows the result of the following mean value parameters pH 7.50± 0.17, dissolved oxygen 6.23±0.11Mg/l and temperature 25.75±0.16ºC respectively

Table 4.19 Water Quality Parameters Monitored During the Experiment

Water Quality Parameters	Range	Mean value ±SE
pH	7.43-7.54	7.50±0.17
Dissolve oxygen (Mg/l)	6.06-6.53	6.23±0.11
Temperature (^{o}C)	25.75	25.75±0.16

Mg/l=Milligram per liter, ^{O}C=Degree Celsius, ±SE=Mean standard error

CHAPTER FIVE

5.0 DISCUSSION

5.1 Weight gain

The present study showed that the results from the feeding trial indicated that highest weight gain was recorded in the treatment fed with CEBM 0% has mean weight gain (112.60 ±4.85) which is the control diet, followed by CEMB (5%) (109.00±2.80g) had similar growth as compared to control CEBM (0%). The overall significantly higher growth were recorded in CEBM (0%) and CEBM (5%) as compared with (CEBM10% and CEBM(15%) (76.13±6.19g and 60.44±11.65g). This result is contrary to the observation of Tibinda *et al* (2013). However the weight gain increased linearly with increased in fish meal. The reason the control diet which is sole animal protein source performed better than other groups fed with mixed chicken entrails and chicken blood meal will not be totally explained. Nevertheless, all the diets supported growth of the fish. This was an indication that all the diets met the nutrient requirements (Crude Protein 20-40%) in fish to promote growth and tissue development (Ajah, 2007; Agbabiaka *et al.*, 2013). It was also been reported that biological value of any protein source does not only depend on its amino acid profile but also on digestibility (Sotolu, 2009).

5.2 Percentage Weight Gain

The result of this study indicated that when fish were fed with test diets of different percentage levels of chicken entrails and chicken blood meal. The highest percenta ge weight gain was in CEBM(0%)(383.19±18.41g), CEBM(5%)(373.71±9.24g) had almost similar percentage weight gain as compared to control CEBM(0%)

while the least were recorded by those fed with CEBM(10%)(260.27±21.98g) and CEBM(15%)(257.32±18.15g). Similarly to the observation of Agbabiaka *et al* (2013) that percentage weight gain increased linearly with increase in fish with highest weight gain.

5.3 Specific Growth Rate

Specific growth rate (SGR) was recorded highest with the group fed with control diet CEBM(0%)(1.22±0.03g) followed by CEBM (5%) (1.21±0.01g) while CEBM (15%)(0.99±0.04g) had the least specific growth rate. Similarly to the observation of Agbabiaka *et al* (2013) that specific growth rate was highest in treatment with highest weight gain.

5.4 Food Conversion Ratio

The best food conversion ratio in the group of fish fed diets of four different percentage level of chicken entrails and chicken blood meal was recorded with group fed with CEBM (0%)(1.39±0.03g)which was the least value, followed by CEBM(5%)(1.42±0.02g). The poor food conversion ratio were recorded with groups fed with CEBM(10%)(1.73±0.15g) CEBM (15%) (5.24±3.34g). Similarly to the observation of Tibinda *et al* (2013) that food conversion ratio slightly decreased by increased fishmeal level in diets.

5.5 Protein Efficiency Ratio

The best value for protein efficiency ratio was obtained in fish fed test diet of

CEBM (0%) (1.64±0.04g) had the least value. The CEBM(5%)(2.12±0.04g) had similar protein efficiency ratio as compared to CEBM(10%)(2.39±0.19g) and CEBM(15%) (2.73±0.05g). But CEBM (15%) had the highest value as compared to control diet CEBM (0%). Similarly to the observation of Tibinda *et al* (2013) that the lowest protein efficiency ratio was observed in highest growth dietary treatments and vice versa.

5.6 The Proximate Analysis of Chicken Entrails and Blood Meal

The proximate analysis of chicken entrails and chicken blood meal showed that the ash contained some quality of mineral elements such as sodium, zinc, selenium, iron, potassium, copper, chromium, phosphate and calcium which make it useful in fish feed formulation. High levels of ash in fishmeal lead to trace element deficienc y and caused 2-4 million blind salmon in Canada and a while ago 4 million blind or partially sighted fish in United State of America (Ajah, 2007). The low moisture content (18% and 20%) suggested that the chicken entrails and chicken blood meal can retain its storage for a longer time. The crude fat contents of (12% and 37%) showed that they had low fat contents, therefore, their crude fat content may not be used for commercial purposes. There protein contents (28% and 30%), showed that it is proteinous and can be incoperated in fish feed as it is higher than the 20 percent baseline for protein ingredients in fish feed (FAO,2007; Lovell, 1998). The presence of crude fibre, also showed that chicken entrails and chicken blood

meal contain some mineral nutrients. Carbohydrate content of 11% and 30% and high caloric values 489Kcal/g and 348Kcal/g showed that they have high energy contents, therefore, good when utilized in the fish feed formulation.

5.7 Proximate Composition of the Formulated Diet

Result obtained from the proximate composition of the test diet showed that the crude protein and crude fibre were highest in the control diet and lowest in CEBM (15%) diet. Crude protein of the test diet showed a pattern of increase with increasi ng level of fishmeal. The levels of these dietary components have been found to be adequate for normal growth for the size of fish used in this study (Tibinda *et al*., 2013). Quite a point of concern is the fact that the group of fish fed with the control diet performed better than other groups, though, the CEBM 15% diet have the lowest crude protein content. The results obtained from the crude protein source, Alternative to fishmeal with mixed chicken entrails and blood meal at all level showed that cat fish do not require as much dietary protein for maximum growth as has typically been assumed (Agbabiaka *et al*.,2013). Lipids are the most energy rich class of nutrients. In addition to satisfying the essential fatty acid requirements of the fish, dietary lipids may supply energy and spare the more valuable protein for growth . Once protein is used for energy, ammonia becomes metabolic waste products (Ajah, 2007). Herbivorous fish that normally consumed high amount of carbohydrates, could be expected to used protein and lipids as

energy source to a large extend (Ajah, 2007). Using high dietary level of lipids will yield high caloric values increasing weight gains and improve protein retention and lead to increased lipids deposition (Ajah, 2007).

5.8 Mineral elements

The results of the selected mineral elements such as sodium, zinc, selenium, iron, potassium copper chromium and phosphate that are important for fish health (Ajah, 2007) which were analyzed separately for the chicken entrails and chicken blood meal when compared with recommended safe levels of FAO/WHO standard showed that chicken entrails and blood meal can be use for fish feed formulation or used for animal feed. However, it has been reported that mobility of mineral elements depends not only on the total concentration in the soil and sediment but also on the soil or sediment properties; metal properties and environmental factors (Hatje *et al*., 1998). Omokheyeke *et al* (2018) reported that sediments are the major depository of mineral elements; in some cases holding over 99% of the total amounts of mineral elements present in amounts several times higher than their natural background levels and pollute sediments in regions near large industrial and urban areas. Consequently, sediments contaminated by mineral elements constitute a threat to the health of aquatic organisms (Omokheyeke *et al*., 2018).

5.9 Water Quality parameters

The mean Value of the water quality parameter Monitored during the experimental period showed that pH (7.50±0.17) was within the range recommended 5-9 pH levels for fish culture (Ajah, 2007., Tiamiyu *et al.*, 2018). The dissolved oxygen (DO)(6.23±0.11Mg/l) was similar to recommended dissolved oxygen content level for tropical water fish culture (Durojaiye *et al*., 2018). Ajah (2007) recommended that for the best growth of fish that the dissolved oxygen level should be above 5Mg/l to avoid super-saturation. The mean temperature value (25±0.16ºC) is within the acceptable range for fish culture in the tropics as reported by Adeparrussi (1990). Ajah (2007) recommended that warm water fish culture grows best at 25ºC-35ºC for tilapia species, common carp and catfish species.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The present study revealed that there was significant difference in the growth of fish fed with control diet and other group, but was growth in other groups fed with combined chicken entails and chicken blood meal. This was an indication that combined chicken entrails and chicken blood meal met the nutritional requirement of the fish to promote growth and tissue development. Therefore combined chicken entrails and chicken blood meal can be used as replacement of the fishmeal at the level of 5% inclusion based on food conversion ratio result.

6.2 Recommendation

- It will be recommended that, for further research, works should be done on the use of mixed chicken entrails and blood meal as alternative to fish meal in a great or proportion as to ascertain its efficiency.
- It is further recommended that experiments with the same feed formulation under natural earthen pond condition, concrete pond and recirculating system are recommended for future study to elucidate the growth and survival under other condition.

- It is therefore inferred from this study that, chicken entrails and blood meal may be used in fish to reduce the fish feed cost.

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APPENDIX

TABLE 1. TITRATION OF DISSOLVED OXYGEN

WEEK I

FM0% (CEBM15%)

	R1	R2	R3
Final	21.00	28.00	27.06
Initial	19.75	26.74	25.80
Vol. used	1.25	1.26	1.26

The mean volume = 1.25

FM5% (CEBM10%)

	R1	R2	R3
Final	6.57	16.78	21.37
Initial	5.40	15.60	20.20
Vol. used	1.17	1.18	1.17

The mean volume = 1.17

FM10% (CEBM5%)

	R1	R2	R3
Final	11.72	17.03	9.35
Initial	10.60	15.90	8.20
Vol. used	1.12	1.13	1.15

The mean volume = 1.13

FM15% (CEBM0%)

	R1	**R2**	**R3**
Final	19.96	25.44	6.76
Initial	18.70	24.20	5.50
Vol. used	1.26	1.24	1.26

The mean volume = 1.25

WEEK II

FM0% (CEBM15%)

	R1	**R2**	**R3**
Final	16.61	21.30	21.10
Initial	15.50	20.20	20.00
Vol. used	1.11	1.10	1.10

The mean volume = 1.10

FM5% (CEBM10%)

	R1	**R2**	**R3**
Final	11.56	16.84	11.04
Initial	10.50	15.80	10.00
Vol. used	1.06	1.06	1.04

The mean volume = 1.05

FM10% (CEBM5%)

	R1	R2	R3
Final	11.54	17.07	6.37
Initial	10.30	15.80	5.10
Vol. used	1.24	1.27	1.27

The mean volume = 1.26

FM15% (CEBM0%)

	R1	R2	R3
Final	16.29	20.87	11.77
Initial	15.00	20.20	10.50
Vol. used	1.29	1.27	1.27

The mean volume = 1.28

WEEK III

FM0% (CEBM15%)

	R1	R2	R3
Final	6.34	11.13	15.85
Initial	5.00	9.80	14.50
Vol. used	1.34	1.33	1.35

The mean volume = 1.34

FM5% (CEBM10%)

	R1	R2	R3
Final	1.23	5.73	10.03
Initial	0.00	4.50	8.80
Vol. used	1.23	1.23	1.23
The mean volume =		1.23	

FM10% (CEBM5%)

	R1	R2	R3
Final	16.80	22.02	16.40
Initial	15.50	20.70	15.10
Vol. used	1.30	1.32	1.30
The mean volume =		1.31	

FM15% (CEBM0%)

	R1	R2	R3
Final	13.71	17.01	23.01
Initial	12.50	15.80	21.80
Vol. used	1.21	1.21	1.21
The mean volume =		1.21	

WEEK IV

FM0% (CEBM15%)

	R1	R2	R3
Final	11.16	15.99	9.12
Initial	10.00	14.80	8.00

Vol. used	1.16	1.19	1.11
The mean volume =	1.18		

FM5% (CEBM10%)

	R1	**R2**	**R3**
Final	16.30	20.90	13.78
Initial	15.00	19.60	12.50
Vol. used	1.30	1.30	1.28
The mean volume =	1.29		

FM10% (CEBM5%)

	R1	**R2**	**R3**
Final	15.78	21.08	11.77
Initial	14.50	19.80	10.50
Vol. used	1.28	1.28	1.27
The mean volume =	1.28		

FM15% (CEBM0%)

	R1	**R2**	**R3**
Final	11.81	17.44	22.94
Initial	10.50	16.10	21.60
Vol. used	1.31	1.34	1.34
The mean volume =	1.33		

WEEK V

FM0% (CEBM15%)

	R1	**R2**	**R3**

Final	6.24	10.92	15.24
Initial	5.00	9.70	14.00
Vol. used	1.24	1.22	1.24
The mean volume =		1.23	

FM5% (CEBM10%)

	R1	**R2**	**R3**
Final	11.25	15.55	19.74
Initial	10.00	14.30	18.50
Vol. used	1.25	1.25	1.24
The mean volume =		1.25	

FM10% (CEBM5%)

	R1	**R2**	**R3**
Final	11.23	16.63	11.74
Initial	10.00	15.40	10.50
Vol. used	1.23	1.23	1.24
The mean volume =		1.23	

FM15% (CEBM0%)

	R1	**R2**	**R3**
Final	11.73	17.33	23.01
Initial	10.50	16.10	21.80
Vol. used	1.23	1.23	1.21
The mean volume =		1.22	

WEEK VI

FM0% (CEBM15%)

	R1	**R2**	**R3**
Final	6.25	10.95	16.25
Initial	5.00	9.70	14.00
Vol. used	1.25	1.25	1.25

The mean volume = 1.25

FM5% (CEBM10%)

	R1	**R2**	**R3**
Final	8.65	12.75	16.97
Initial	7.50	11.60	15.80
Vol. used	1.15	1.15	1.17

The mean volume = 1.16

FM10% (CEBM5%)

	R1	**R2**	**R3**
Final	11.24	16.64	22.06
Initial	10.00	15.40	20.80
Vol. used	1.24	1.24	1.26

The mean volume = 1.25

FM15% (CEBM0%)

	R1	**R2**	**R3**
Final	11.31	16.91	22.13
Initial	10.00	15.60	20.80
Vol. used	1.31	1.31	1.33

The mean volume = 1.32

WEEK VII

FM0% (CEBM15%)

	R1	**R2**	**R3**
Final	6.22	10.53	14.93
Initial	5.00	9.30	13.70
Vol. used	1.22	1.23	1.23

The mean volume = 1.23

FM5% (CEBM10%)

	R1	**R2**	**R3**
Final	6.20	10.90	17.01
Initial	5.00	9.70	15.80
Vol. used	1.20	1.20	1.21

The mean volume = 1.20

FM10% (CEBM5%)

	R1	**R2**	**R3**
Final	11.28	16.68	16.29
Initial	10.00	15.40	14.00
Vol. used	1.28	1.28	1.29

The mean volume = 1.28

FM15% (CEBM0%)

	R1	**R2**	**R3**
Final	16.78	21.99	16.39
Initial	15.50	20.70	15.10
Vol. used	1.28	1.29	1.29

The mean volume = 1.29

WEEK VIII

FM0% (CEBM15%)

	R1	**R2**	**R3**
Final	21.00	19 .96	28.00
Initial	19.75	18.70	26.74
Vol. used	1.25	1.26	1.26

The mean volume = 1.25

FM5% (CEBM10%)

	R1	R2	R3
Final	21.00	6.57	16.61
Initial	19.75	5.40	15.50
Vol. used	1.25	1.17	1.11

The mean volume = 1.18

FM10% (CEBM5%)

	R1	R2	R3
Final	11.23	16.64	17.33
Initial	10.00	15.40	16.10
Vol. used	1.23	1.24	1.23

The mean volume = 1.23

FM15% (CEBM0%)

	R1	R2	R3
Final	22.02	11.81	20.90
Initial	20.70	10.50	19.60
Vol. used	1.32	1.31	1.30

The mean volume = 1.31

Calculation of dissolved oxygen (DO) Mg/l

$$DO = \frac{\text{Volume of titrat x 0.025N x 8 x 1000mg/l}}{\text{Volume of sample x volume of sample} - 2 \text{ Volume of Winkler bottle used}}$$

Where m/ titrant = volume of $Na_2S_2O_3$ used in titrant
N = Normality of $Na_2S_2O_3$
8 = Oxygen concentration equivalent to 1ml of $Na_2S_2O_3$
1000 = Conversion factor to 1 litre

WEEK I
0% FM

$$DO = \frac{1.25 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.38mg/l$$

5% FM

$$DO = \frac{1.17 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.01mg/l$$

10% FM (CEBM 5%)

$$DO = \frac{1.13 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 5.80mg/l$$

15% FM (CEBM 0%)

$$DO = \frac{1.25 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.40mg/l$$

WEEK II
0% FM (CEBM 15%)

$$DO = \frac{1.10 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 5.50mg/l$$

5% FM (CEBM 10%)

$$DO = \frac{1.05 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 5.34 mg/l$$

FM10% (CEBM 5%)

$$DO = \frac{1.26 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.45 mg/l$$

FM15% (CEBM 0%)

$$DO = \frac{1.28 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.54 mg/l$$

WEEK III

FM0% (CEBM 15%)

$$DO = \frac{1.34 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.82 mg/l$$

FM 5% (CEBM 10%)

$$DO = \frac{1.23 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.25 mg/l$$

FM10% (CEBM 5%)

$$DO = \frac{1.31 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.70 mg/l$$

FM15% (CEBM 0%)

$$DO = \frac{1.21 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.18 mg/l$$

WEEK IV

FM 0% (CEBM15%)

$$DO = \frac{1.18 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.60 mg/l$$

FM5% (CEBM10%)

$$DO = \frac{1.29 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.60 mg/l$$

FM10% (CEBM5%)

$$DO = \frac{1.28 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.55 mg/l$$

FM15% (CEBM0%)

$$DO = \frac{1.33 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.80 mg/l$$

WEEK V

FM0% (CEBM15%)

$$DO = \frac{1.23 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.30 mg/l$$

FM5% (CEBM10%)

$$DO = \frac{1.25 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.26 mg/l$$

FM10% (CEBM5%)

$$DO = \frac{1.23 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.40 mg/l$$

FM15% (CEBM0%)

$$DO = \frac{1.22 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}}$$

250 = 6.20mg/l

WEEK VI

FM0% (CEBM15%)

$$DO = \frac{1.25 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.37mg/l$$

FM5% (CEBM10%)

$$DO = \frac{1.16 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 5.90mg/l$$

FM10% (CEBM5%)

$$DO = \frac{1.25 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.45mg/l$$

FM15% (CEBM0%)

$$DO = \frac{1.32 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.71mg/l$$

WEEK VII

FM0% (CEBM15%)

$$DO = \frac{1.23 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.26mg/l$$

FM5% (CEBM10%)

$$DO = \frac{1.20 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.15mg/l$$

FM10% (CEBM5%)

$$DO = \frac{1.28 \times 0.025 \times 8 \times 1000}{100 \times \frac{100 - 2}{250}} = 6.58mg/l$$

FM15% (CEBM0%)

$$DO = \frac{1.29 \times 0.025 \times 8 \times 1000}{}$$

$$100 \text{ X } \frac{100 - 2}{250} = 6.60\text{mg/l.}$$

WEEK VIII

FM0% (CEBM15%)

$$\text{DO} = \frac{1.25 \text{ X } 0.025 \text{ X } 8 \text{ X } 1000}{100 \text{ X } \frac{100 - 2}{250}} = 6.38\text{mg/l}$$

FM5% (CEBM10%)

$$\text{DO} = \frac{1.18 \text{ X } 0.025 \text{ X } 8 \text{ X } 1000}{100 \text{ X } \frac{100 - 2}{250}} = 6.00\text{mg/l}$$

FM10% (CEBM5%)

$$\text{DO} = \frac{1.23 \text{ X } 0.025 \text{ X } 8 \text{ X } 1000}{100 \text{ X } \frac{100 - 2}{250}} = 6.40\text{mg/l}$$

FM15% (CEBM0%)

$$\text{DO} = \frac{1.31 \text{ X } 0.025 \text{ X } 8 \text{ X } 1000}{100 \text{ X } \frac{100 - 2}{250}} = 6.70\text{mg/l.}$$

The formular used for the calculation of standard error of water quality parameters as recommended by Boyd(1979).

A) Sample variance or $S2 = \sum^{n}_{1=1} \frac{(x1-\bar{x})^2}{n-1}$

where xi = value for the observations

$\bar{x}$ = sample means

n= number of observation

b) standard deviation or $s = \sqrt{S^2}$

c) standard deviation of the mean or standard error $SE = \sqrt{SE^2} = S/\sqrt{n}$

Table 2. Temperature Determination

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
26	25.75	0.25	0.0625
26	25.75	0.25	0.0625
26	25.75	0.25	0.0625
25	25.75	-0.75	0.5625
26	25.75	0.25	0.0625
26	25.75	0.25	0.0625
25	25.75	0.75	0.5625
26	25.75	0.25	0.0625
			$\sum i(xi-x^-)^2$ =1.5

$S^2 = \frac{1.5}{7} = 0.21$

$S=\sqrt{0.21}=0.4582575$

SE = 0.452575/2.8264271 = 0.16

Table 3.pH Determinations

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
8.00	7.43	0.57	0.3249
7.45	7.43	0.02	0.0004
6.97	7.43	-0.46	0.2116
6.49	7.43	-0.94	0.8836
7.50	7.43	0.07	0.0049
8.15	7.43	0.72	0.5184
7.26	7.43	-0.17	0.0289
7.61	7.43	0.18	0.0324
			$\sum i(xi-x^-)^2$ =2.0051

$S^2 = \frac{2.0051}{7}$

$S = \sqrt{0.29} = 0.5385$

$SE = \frac{0.5385}{2.8284271}$

=0.19

X_1	$\bar{x}$	$X_1-\bar{x}$	$(X_1-\bar{x})^2$
7.38	7.48	-0.1	0.01
7.47	7.48	-0.01	0.0001
7.15	7.48	-0.33	0.1089
8.32	7.48	0.84	0.7056
6.42	7.48	-1.06	1.1236
7.57	7.48	0.09	0.0081
7.26	7.48	0.01	0.0001
8.07	7.48	0.59	0.3481
			$\sum i(xi-\bar{x})^2$ =2.3045

S^2 = 2.3045
7
= 0.33
S = √0.33
SE = 0.5745
2.8284271
= 0.20

X_1	$\bar{x}$	$X_1-\bar{x}$	$(X_1-\bar{x})^2$
7.29	7.54	-0.25	0.0625
7.78	7.54	0.24	0.0576
8.10	7.54	-0.56	0.3136
7.67	7.54	0.13	0.0169
6.87	7.54	-0.67	0.4489
7.61	7.54	0.07	0.0049
7.26	7.54	-0.28	0.0784
7.71	7.54	0.2	0.04
			$\sum i(xi-\bar{x})^2$ =1.0228

S^2 = 1.0228
7
= 0.15
S= √0.15
= 0.3873
SE=0.3873
2.8284271

= 0.14

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
6.95	7.54	-0.59	0.3481
7.45	7.54	-0.09	0.0081
7.65	7.54	0.11	0.0121
7.28	7.54	-0.26	0.0676
8.35	7.54	0.81	0.6561
7.67	7.54	0.13	0.0169
7.38	7.54	-0.16	0.0256
7.56	7.54	0.02	0.004
			$\sum i(xi-x^-)^2$ =1.1349

$S^2 = \frac{1.1349}{7}$

= 0.16

S= √0.16

SE= $\frac{0.4}{2.8284271}$

= 0.14

Table 4.Do Determination

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
6.38	6.27	0.11	0.0121
5.50	6.27	-0.77	0.5929
6.82	6.27	0.55	0.3025
6.00	6.27	-0.27	0.0729
6.30	6.27	0.03	0.0009
6.37	6.27	0.1	0.01
6.26	6.27	-0.01	0.0001
6.38	6.27	0.11	0.0121
			$\sum i(xi-x^-)^2$ =1.0035

$S^2 = \frac{1.0035}{7}$

= 0.14

S =√0.14

SE=$\frac{0.3742}{2.8284271}$ = 0.13

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
6.01	6.06	-0.05	0.0025
5.34	6.06	-0.72	0.5184
6.25	6.06	0.19	0.0361
6.60	6.06	0.54	0.2916
6.26	6.06	0.2	0.04
5.90	6.06	-0.16	0.0256
6.15	6.06	0.09	0.0081
6.00	6.06	-0.06	0.0036
			$\sum i(xi-x^-)^2$ =0.9259

$S^2 = \frac{0.9259}{7}$

$= 0.13$

$S = \sqrt{0.13}$

$= 0.3606$

$SE = \frac{0.3606}{2.8284271}$

$= 0.13$

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
5.80	6.41	-0.61	0.3721
6.45	6.41	0.04	0.0016
6.70	6.41	0.29	0.0841
6.55	6.41	0.14	0.0196
6.40	6.41	-0.01	0.0001
6.45	6.41	0.04	0.0016
6.58	6.41	0.17	0.0289
6.40	6.41	-0.01	0.0001
			$\sum i(xi-x^-)^2$ =0.5081

$S^2 = \frac{0.5081}{7}$

$= 0.073$

$S = \sqrt{0.07}$

$= 0.2646$

SE= 0.2646
2.8284271 = 0.09

X_1	x^-	X_1-x^-	$(X_1-x^-)^2$
6.40	6.53	-0.13	0.0169
6.54	6.53	0.01	0.0001
6.18	6.53	-0.35	0.1225
6.80	6.53	0.27	0.0729
6.20	6.53	-0.33	0.1089
6.71	6.53	0.18	0.0324
6.60	6.53	0.07	0.0049
6.70	6.53	0.17	0.0289
			$\sum i(xi-x^-)^2$ =0.3875

S^2= 0.3875
7
=0.05
S=√0.06
SE=0.245
2.8284271
=0.09

Calculation Proximate Composition of the Samples

FM0% (CEBM 15%)

1. % Moisture $\frac{w_1 - w_2}{w_1} \times \frac{100}{1}$

$\frac{5 - 4.5}{5} \times \frac{100}{1}$ = 10%

2. % Ash $\frac{\text{wgt of Ash}}{\text{Wgt of sample}} \times \frac{100}{1} = \frac{0.06}{0.5} \times \frac{100}{1}$ = 12%

3. % Fat = $\frac{\text{wgt of Oil}}{\text{Wgt of sample}} = \frac{4.3}{10} \times \frac{100}{1}$ = 43%

4. % protein = % Nitrogen x 6.25

= $\frac{1.2 \times 0.1 \times 0.014 \times 20}{1g} \times \frac{100}{1} = 3.36$

3.36 x 6.25 = 21%

5. Crude fibre

$\frac{(66.5 + 0.5) - (66.5 + 0.0025)}{2} \times \frac{100}{1} = 23.7\%$

6. Carbohydrate = 100-(%protein+% ash+% moisture+ % fat)

= 100 – (21.+10+12+43) = 14%

7. Carbohydrate value

(% protein x 4) + (% fat x 9) + (% carbohydrate x 4)

= (21 x 4 + 43 x 9 + 14 x 4) = 527Kcal/g

FM5% (CEBM10%) PROXIMATE RESULT

1. % Moisture $\frac{w_1 - w_2}{w_1} \times \frac{100}{1}$

$\frac{5 - 4.5}{551} = \frac{0.5}{10\%} \times \frac{100}{1}$

2. % Ash $\frac{\text{wgt of Ash}}{\text{Wgt of sample}} \times \frac{100}{1} = \frac{0.018}{0.5} \times \frac{100}{1} = 3.6\%$

3. % Fat = $\frac{\text{wgt of Oil}}{\text{Wgt of sample}} = \frac{3.1}{10} \times \frac{100}{1} = 31\%$

4. % protein = % Nitrogen x 6.25

= $\frac{1.4 \times 0.1 \times 0.014 \times 20}{1} \times \frac{100}{1} = 3.92 \times 6.25 = 2.45\%$

5. Crude fibre content:

$\frac{66.5 + 0.7 - 66.5 + 0.05}{2} \times \frac{100}{1} = \frac{67.20 - 66.55}{2}$

$\frac{0.65}{} \times \frac{100}{}$

2 1 = 33%

6. Carbohydrate = 100-(%protein+% ash+% Moisture+ % fat)
= 100 – (24.5+3.6+10.0+31.0) = 100-69.1 = 30.9%
7. Carbohydrate value
(% protein x 4) + (% fat x 9) + (% carbohydrate x 4)
= 501Kcal/g

FM10% (CEBM5%) PROXIMATE RESULT

1. % Moisture= $\frac{w_1 - w_2}{W_1} \times \frac{100}{1}$ $\frac{5-4.5}{5} = \frac{0.5}{1} \times \frac{100}{1}$ = 10%

2. % Ash $\frac{\text{wgt of Ash}}{\text{Wgt of sampl}} \times \frac{100}{1}$

= $\frac{0.04}{0.5} \times \frac{100}{1}$ = 8%

3. % Fat = $\frac{\text{wgt of Oil}}{\text{Wgt of sampl}} \times \frac{100}{1}$ $\frac{2.5 \text{ X } 100}{10 \text{ x } 1}$ = 25%

4. % Protein = % Nitrogen x 6.25

Where % Nitrogen = $\frac{cm^s \text{ (acid) x M x 0.014 x 20 x 100}}{\text{Wgt of sample}} \times \frac{}{1}$

$\frac{1.9 \text{ x } 0.1 \text{ x } 0.014 \text{ x } 20}{1g} \times \frac{100}{1}$ = 5.32%

:. % Protein = 5.32 x 6.25 = 33.3%

5. Crude fibre

$\frac{\text{Wgt of crucible x dried residue – wgt of crucible + Ash}}{\text{Wgt of sample}} \times \frac{100}{1}$

$\frac{66.5 + 0.6 – 66.5 + 0.02}{2} \times \frac{100}{1}$ = $\frac{67.10 – 66.52}{2} \times \frac{100}{1}$

= $\frac{0.58}{2} \times \frac{100}{1}$ = 29%

6. Carbohydrate = 100–(%protein+% ash+% moisture+% fat)
100 – (33.3+8+10+25) = 23.7%

7. Calorific value

(% protein x 4)+(% fat x 9) + (% carbohydrate x 4)
= (33.3 x 4) + (25 x 9) + (23.7 x 4) = 453Kcal/g

FM 15(CEOBM0%) PROXIMATE RESULTS

1. % Moisture $\frac{w_1 - w_2}{w_1} \times \frac{100}{1}$

Where w_1 = wgt of sample before drying
W_2 = wgt of sample after drying

$= \frac{5 - 4.5}{5} = \frac{0.5}{5} \times \frac{100}{1} = 10\%$

2. % Ash $\frac{\text{wgt of Ash}}{\text{Wgt of sample}} \times \frac{100}{1} = \frac{0.02}{0.5} \times \frac{100}{1} = 4\%$

3. % Fat = $\frac{\text{wgt of Oil}}{\text{Wgt of sample}} = \frac{1.9}{10} \times \frac{100}{1} = 19\%$

4. % protein = % Nitrogen x 6.25

Where % Nitrogen = $\frac{cm^s \text{ (acid)} \times M \times 0.014 \times 20}{\text{Wgt of sample}} \times \frac{100}{1}$

Where cm^3 acid = ml titre value
M = Molarity of acid
0.014 = constant
20 = dilution factor

$= \frac{2.5 \times 0.1 \times 0.014 \times 20}{1g} \times \frac{100}{1} = 7\%$

:. 7 x 6.25 = 43.8%

5. Crude fibre

$$\frac{\text{Wgt of crucible + dried residue – wgt of crucible + Ash}}{\text{Wgt of sample}} \times \frac{100}{1}$$

$$\frac{66.5 + 0.8 - 66.5 + 0.03}{2} \times \frac{100}{1} = \frac{67.30 - 66.53}{2}$$

$$\frac{0.77}{2} \times \frac{100}{1} = 38.5\%$$

6. Carbohydrate

= 100-(%protein+% ash+% moisture + % fat)

= 100 – (43.8+4+10+19)

100 – 76.8 = 23.2%

7. Carbohydrate value

(% protein x 4) + (% fat x 9) + (% carbohydrate x 4)

= (43.8 x 4) + (19 x 9) + (23.2 x 4)

= 175.2 + 171 + 92.8

= 439Kcal/g

PROXIMATE RESULT OF CHICKEN ENTRAILS

1. % Moisture = $\frac{5 - 4.1}{5} \times \frac{100}{1} = 18\%$

2. % Ash $\frac{\text{wgt of Ash}}{\text{Wgt of sample}} \times \frac{100}{1} = \frac{0.03}{0.5} \times \frac{100}{1} = 6\%$

3. % Fat = $= \frac{3.7}{10} \times \frac{100}{1} = 3.7\%$

4. Crude fibre

$$\frac{(66.5 + 0.3) - (66.5 + 0.026)}{2} \times \frac{100}{1} = 13.7\%$$

$$\frac{0.77}{2} \times \frac{100}{1} = 38.5\%$$

5. % protein = % Nitrogen x 6.25

$$= \frac{1.6 \times 0.1 \times 0.014 \times 20}{1g} \times \frac{100}{1} = 4.48$$

$4.48 \times 6.25 = 28\%$

6. Carbohydrate

$100 - (28 + 6 + 18 + 37)$

$100 - 89 = 11\%$

7. Carbohydrate value

(% protein x 4) + (% fat x 9) + (% carbohydrate x 4)

$= (28 \times 4) + (37 \times 9) + (11 \times 4)$

$= 112 + 333 + 44$

$= 489$Kcal

PROXIMATE RESULT OF OX-BLOOD

1. % Moisture= $\frac{5\text{-}4.0}{5} = \frac{1}{5} \times \frac{100}{1} = 20\%$

2. % Ash = $\frac{0.04}{0.5} \times \frac{100}{1} = 8\%$

3. % Fat = $\frac{2.5 \times 100}{10 \quad 1} = 12\%$

4. % Protein = % Nitrogen x 6.25

$$\frac{1.75 \times 0.1 \times 0.014 \times 20}{1g} \times \frac{100}{1} = 4.9 \times 6.25 = 30\%$$

5. Crude fibre

$$\frac{(66.5 + 0.2) - (66.5 + 0.028)}{2} \times \frac{100}{1} = 8.6\%$$

6. Carbohydrate = 100–(30+8+20+12) = 100 – 70 = 30%

7. Calorific value

= (30 x 4) + (12 x 9) + (30 x 4) = 120+108+120

= 348Kcal

Table 5 Weekly Weight Gain of ***Clarias gariepinus*** Fingerlings Fed with Various Percentage Level of Chicken Entrails And Blood Meal for Weeks

Weekly Weights of Fish (g)											WEIGHT GAIN±SE
TREATMENTS	**REPLICATES**										
		W0	W1	W2	W3	W4	W5	W6	W7	W8	
CONTROL(0%)	A	29.10	34.90	43.40	52.70	68.30	84.70	108.10	130.50	146.60	117.50
	B	29.70	34.40	36.30	42.50	55.30	70.00	91.20	119.90	132.60	102.90
	C	29.40	34.70	45.50	54.60	69.00	86.70	114.20	130.50	146.80	117.40
	TOTAL	88.20	104.00	125.20	149.80	192.60	241.40	313.50	380.90	426.00	338.80
	MEAN ± SE	29.40±0.12	34.67±0.04	41.73±2.78	49.93±4.59	64.20±4.45	80.47±5.26	104.50±6.88	126.97±3.53	142.00±4.70	112.60±4.85
CONTROL(5%)	A	29.20	32.30	38.40	47.30	63.60	77.20	96.30	121.80	138.30	109.10
	B	29.20	34.10	42.90	52.40	65.80	79.30	95.30	120.20	143.00	113.20
	C	29.10	37.10	42.80	54.80	60.10	77.20	98.00	114.70	133.20	117.40
	TOTAL	87.50	103.90	124.10	154.50	189.50	233.70	289.60	356.70	414.50	327.00
	MEAN ± SE	29.17±0.03	34.64±1.52	41.37±1.45	51.50±2.21	63.17±1.66	77.90±0.71	96.53±0.79	118.90±2.15	138.17±2.83	109.00±2.80
CONTROL(10%)	A	29.40	35.00	38.30	41.60	47.00	61.50	77.90	84.90	96.10	66.70
	B	29.10	36.20	40.70	45.00	56.10	67.50	78.10	92.40	116.90	87.80
	C	29.30	33.10	36.40	38.70	46.30	50.60	66.40	81.30	103.20	73.90
	TOTAL	87.80	104.30	115.40	125.30	149.40	179.60	222.40	258.60	316.20	228.40
	MEAN ± SE	29.27±0.09	34.77±0.90	38.47±1.24	41.77±1.82	49.80±3.16	59.87±4.95	74.13±3.87	86.20±3.27	105.40±6.10	76.13±6.19
CONTROL(15%)	A	29.40	38.70	40.30	43.50	47.60	49.40	57.70	75.70	95.70	66.30
	B	29.10	37.20	39.50	43.40	51.10	69.50	80.40	95.30	104.20	75.10
	C	29.20	39.20	44.10	50.70	58.20	68.00	81.60	98.00	113.40	84.20
	TOTAL	87.70	115.10	123.90	137.60	156.90	186.90	219.70	269.00	313.30	181.30
	MEAN ± SE	29.23±0.09	38.37±1.20	41.30±18.02	45.87±2.42	52.30±3.12	62.30±6.46	73.23±7.77	89.67±7.03	104.43±5.11	60.44±11.65

w = Weeks SE=standard error ; Mean ±; CEBM 0% = chicken entrails and blood meal 0%; CEBM 5% = chicken entrails and blood meal 5%; CEBM 10% = Chicken entrails and blood meal 10%; CEBM 15% = chicken entrails and blood meal 15%.

Table 6: Analysis of Valiance for the Weekly Mean Weight Gain of *Clarias gariepinus* Fingerlings Fed with Various Percentage of Chicken Entrails and Blood Meal for 8 Weeks.

ANOVA

Percentage Weight gain

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	43095.344	3	14365.115	15.489	.001
Within Groups	7419.625	8	927.453		
Total	50514.969	11			

Post Hoc Tests

Multiple comparisons

Dependent Variable: Percentage Weight gain

LSD

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
control	CEOBM 5%	9.49000	24.86568	.713	-47.8504	66.8304
	CEOBM 10%	122.91667*	24.86568	.001	65.5763	180.2570
	CEOBM 15%	125.87000*	24.86568	.001	68.5296	183.2104
	Control	-9.49000	24.86568	.713	-66.8304	47.8504
CEOBM 5%	CEOBM 10%	113.42667*	24.86568	.002	56.0863	170.7670
	CEOBM 15%	116.38000*	24.86568	.002	59.0396	173.7204
CEOBM 10%	Control	-122.91667*	24.86568	.001	-180.2570	-65.5763

	CEOBM 5%	-113.42667*	24.86568	.002	-170.7670	-56.0863
	CEOBM 15%	2.95333	24.86568	.908	-54.3870	60.2937
	Control	-125.87000*	24.86568	.001	-183.2104	-68.5296
CEOBM 15%	CEOBM 5%	-116.38000*	24.86568	.002	-173.7204	-59.0396
	CEOBM 10%	-2.95333	24.86568	.908	-60.2937	54.3870

*. The mean difference is significant at the 0.05 level.

Table 7.Analysis of Variance for the Percentage Weight Gain of *Clarias ganepinus* Fingerlings Fed With Varies Percentage of Chicken Entrails and Blood Meal for 8 Weeks.

ANOVA

Percentage Weight gain

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	43095.344	3	14365.115	15.489	.001
Within Groups	7419.625	8	927.453		
Total	50514.969	11			

Post Hoc Tests

Multiple Comparisons

Dependent Variable: Percentage Weight gain

LSD

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
control	CEOBM 5%	9.49000	24.86568	.713	-47.8504	66.8304
	CEOBM 10%	122.91667*	24.86568	.001	65.5763	180.2570
	CEOBM 15%	125.87000*	24.86568	.001	68.5296	183.2104
	Control	-9.49000	24.86568	.713	-66.8304	47.8504
CEOBM 5%	CEOBM 10%	113.42667*	24.86568	.002	56.0863	170.7670
	CEOBM 15%	116.38000*	24.86568	.002	59.0396	173.7204
	Control	-122.91667*	24.86568	.001	-180.2570	-65.5763
CEOBM 10%	CEOBM 5%	-113.42667*	24.86568	.002	-170.7670	-56.0863
	CEOBM 15%	2.95333	24.86568	.908	-54.3870	60.2937
	Control	-125.87000*	24.86568	.001	-183.2104	-68.5296
CEOBM 15%	CEOBM 5%	-116.38000*	24.86568	.002	-173.7204	-59.0396
	CEOBM 10%	-2.95333	24.86568	.908	-60.2937	54.3870

*. The mean difference is significant at the 0.05 level.

Table 8 Analysis of Variance of the Specific Growth Rate of *Clarias gariepinus*Fingerlings Fed with Chicken Entrails and Blood Meal for 8 Weeks.

ANOVA

Specific growth rate

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	.150	3	.050	14.181	.001
Within Groups	.028	8	.004		
Total	.178	11			

Post Hoc Tests

Multiple Comparisons

Dependent Variable: Specific growth rate

LSD

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Control	CEOBM 5%	.01333	.04848	.790	-.0985	.1251
	CEOBM 10%	.22667*	.04848	.002	.1149	.3385
	CEOBM 15%	.23333*	.04848	.001	.1215	.3451
CEOBM 5%	Control	-.01333	.04848	.790	-.1251	.0985
	CEOBM 10%	.21333*	.04848	.002	.1015	.3251
	CEOBM 15%	.22000*	.04848	.002	.1082	.3318
CEOBM 10%	Control	-.22667*	.04848	.002	-.3385	-.1149
	CEOBM 5%	-.21333*	.04848	.002	-.3251	-.1015
	CEOBM 15%	.00667	.04848	.894	-.1051	.1185
CEOBM 15%	Control	-.23333*	.04848	.001	-.3451	-.1215
	CEOBM 5%	-.22000*	.04848	.002	-.3318	-.1082
	CEOBM 10%	-.00667	.04848	.894	-.1185	.1051

*. The mean difference is significant at the 0.05 level.

Table 9: Analysis of Variance for the Food Conversion Ratio of *Clarias gariepinus* Fingerlings Fed with Chicken Entrails and Blood Meal for

ANOVA

Food Conversion Ratio

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.332	3	.111	5.862	.020
Within Groups	.151	8	.019		
Total	.482	11			

Post Hoc Test

Multiple Comparisons

Dependent Variable: Food Conversion Ratio

LSD

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Control	CEOBM 5%	-.03000	.11213	.796	-.2886	.2286
	CEOBM 10%	-.34000*	.11213	.016	-.5986	-.0814
	CEOBM 15%	-.35333*	.11213	.014	-.6119	-.0948
CEOBM 5%	Control	.03000	.11213	.796	-.2286	.2886
	CEOBM 10%	-.31000*	.11213	.024	-.5686	-.0514
	CEOBM 15%	-.32333*	.11213	.020	-.5819	-.0648

	Control	.34000*	.11213	.016	.0814	.5986
CEOBM 10%	CEOBM 5%	.31000*	.11213	.024	.0514	.5686
	CEOBM 15%	-.01333	.11213	.908	-.2719	.2452
	Control	.35333*	.11213	.014	.0948	.6119
CEOBM 15%	CEOBM 5%	.32333*	.11213	.020	.0648	.5819
	CEOBM 10%	.01333	.11213	.908	-.2452	.2719

Table 10. Analysis of Variance for the Protein Efficiency Ratio of *Clarias gariepinus* Fingerlings Fed with Chicken Entrails and Blood Meal for 8 Weeks

Source	Df	Sum of square	Mean Square	F-cal	Ftab 0.05
Trial	11	2.186367		21.33	
			4.07		
Treatments	3	1.9267	0.6422333		
Error	8	0.259667	0.03245		

Therefore there was significant difference on protein efficiency ratio

Inference. Fcal ≥ Ftab ($p \leq 0.05$)

Treatment	**1**	**2**	**3**	**£x**	**X**
T0%	1.71	1.62	1.58	4.91	1.64
T5%	2.13	2.18	2.04	6.35	2.12
T10%	2.02	2.68	2.46	7.16	2.39
T15%	2.73	2.65	2.82	8.2	2.73
	8.59	9.13	8.9	26.62	