USE OF FERMENTED SESAME SEED MEAL AS A FISH MEAL SUBSTITUTE IN A PRACTICAL DIET FOR *CLARIAS GARIEPINUS*

BY

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CERTIFICATION

This is to certify that the experiment reported here was conducted by:

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The report has been read and approved having met the requirements of the department of fisheries and aquaculture, faculty of Agriculture, federal university oye Ekiti, for the award of Bachelor of fisheries and aquaculture degree (B. fisheries).

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DEDICATION

This project is dedicated to Almighty God, my parents, my siblings and my friends.

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All glory, honour and praise for Jesus Christ for his wonderful and marvelous care, protection, provision and guidance during the course of my study in the university.

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ABSTRACT

The study evaluated the effects partial and total replacement of fish meal protein fermented sesame seed meal (SSM) protein in practical diets on growth performance, feed utilization and hematology of African catfish *(Clarias gariepinus).* Fish of an average initial weight of 5.18g were stocked in 15 plastic aquaria of about 25litres capacity each at a rate of 15 fish per aquarium. Fish meal protein (18% of the diet) was used as the sole source of animal protein in the control diet. Percent replacements of fish meal by sesame seed meal on the basis of crude protein were as follows: 0 % (control diet 1), 25 % (diet 2), 50 % (diet 3), 75 % (diet 4) and 100 % (diet 5). African catfish *(Clarias gariepinus) Clarias gariepinus* juvenile were fed at 5% body weight per day in glass tanks with four iso-nitrogenous diets containing different levels of fermented sesame seed meal (25, 50, 75,100%) for a period of eight weeks. Growth performance and hematological indicators such as weight gain, relative weight gain, specific growth rate, feed conversion ratio, protein intake, protein efficiency ratio, percentage mortality, pack cell volume (PCV), haemoglobin (Hb), red blood cell, white blood cells, mean corpuscular volume (MCV, Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC) were used to assess the effects of the diet on the fish. The results of this study revealed that, fish fed on diet 1 (100 % FM) had the highest average body weight , Diet 3, 4 and 5 are significantly different from each other in specific growth rate. Fish fed with SSM at 25% inclusion level gave the best Specific growth rate (1.57) which was marginal difference from those fed on fish meal without sesame seed meal (1.06). Feed conversion ratio (FCR) (0.75), and protein efficiency ratio (PER) (1.31), but the lowest was obtained with fish fed diet E (100 % SSM). specific growth rate (SGR)(0.33g) feed conversion ratio (FCR)(0.2.77), and protein efficiency ratio (PER)(0.42), The same parameters of fish fed on diets 2 (25 % SSM) and diet 3

(50 %SSM) were not significantly different ($P > 0.05$) from those of fish fed on the control diet 1.Fish fed on diet 1 (100 % FM) had the pack cell volume(PCV) (38.67), haemoglobin(5.87), red blood cell(6.06), white blood cells(35.00),mean corpuscular volume(MCV)(6.38), Mean corpuscular haemoglobin(MCH)(0.97) and Mean corpuscular haemoglobin concentration(MCHC)(0.16).

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CHAPTER ONE

1.0 INTRODUCTION

The exponential growth of the aquaculture sector during the past two decades is a result of the progressive intensification of production systems and use of quality feeds, which meet the nutritional requirements of cultured fish (Food Agricultural organization, 2006).

Stimulated by higher global demand for fish, world fisheries and aquaculture production reached 157million tons in 2012 and is projected to reach about 172 million tons in 2021, with most of the growth coming from aquaculture (FAO, 2013). This increase of aquaculture production must be supported by a corresponding increase in the production of designed diets for the cultured aquatic animals (Rahman *et al.,* 2013). The development of sustainable aquaculture depends on the establishment of alternative feedstuffs to Fish meal (Olukayode and Emmanuel, 2012).

African catfish (*Clarias gariepinus)* is a typical air-breathing fish, scale less, bony elongated body with long dorsal and anal fins, and a helmet like head. Colour varies dorsally from dark to light brown and is often mottled with shades of olive and grey while the underside is a pale cream to white (Skelton, 2001).

The native range of *C. gariepinus* is widely distributed in most of the African countries, with the exception of Maghreb, Upper and Lower Guinea, and the Cape provinces of South Africa (Picker and Griffiths, 2011).

C. gariepinus is widely tolerant of many different habitats, even the upper reaches of estuaries, but is considered to be a freshwater species. It favours floodplains, slow flowing rivers, lakes and dams (Skelton, 2001).

The main protein source in the production of fish feed is fish meal, and the high cost of fish meal has been a major factor affecting the growth of fish culture in Nigeria. To reduce the cost of fish production and use of fish meal in fish diet some plants and animal protein sources have been investigated (Eyo and Olatunde, 1996; Fagbenro and Fasakin, 1996; *Absalom et al., 1999, Fasakin et al., 2000).* The evaluation of alternative protein sources to fishmeal is therefore a research priority.

Sesame (*Sesamum indicum L.*) seed is one of the most important oil seed crops in the world. Sesame is an oilseed plant in the genus Sesamum and the family Pedaliceae. It is an annual plant growing 50 to 100 cm in height, with opposite leaves 4 to 14 cm long with an entire margin; they are broad lanceolate, to 5 cm broad, at the base of the plant, narrowing to just 1 cm broad on the flowering stem. The flowers are white to purple, tubular, 3 to 5 cm long, with a four-lobed mouth (Salunkhe *et al.*, 1992).

Its primary marketable products are the whole seeds, seed oil and meal. While sesame seeds have been grown in tropical regions throughout the world since prehistoric times, traditional myths hold that their origins go back even further. It is not clear where the seed originated, but it is speculated that it was native to tropical Africa. Moreover, the importance of sesame in the economies of several African countries justifies the African continent to be the center of its origin. It has since travelled all over the world and is now extensively grown in many tropical and warm temperate regions (Biabani and Pakniyat 2008). The genus *Sesamum* belongs to the family Pedaliaceae, which contains more than 30 species, of which only *S. indicum* L. is cultivated (Padma *et al.*, 2011). In 2009, the world production of sesame seed was 3,976,968 tons, and the major production areas were Asia (2,489,518 tons) and Africa (1,316,690 tons), constituting about 62.6% and 33.1% of the total world production (FAO, 2011).

They are tiny, flat ovals, measuring about 3 mm (1/8 in) long. The protein of sesame seed, though deficient in lysine, is rich in sulphur amino acids such as methionine and cystine which make it an appropriate supplement to diets based on groundnut, soybean and certain cereals, all of which tend to be deficient in the sulphur amino acids (Elkafi *et al.*, 1991). Not only as a source of edible oil, but also the seeds provide nutritious elements for human and livestock. With adequate heat involvement and extraction of oil, reduction in seed toxicants is achieved and nutritive values also improved. Sesame seed have more appropriate nutritional quality as compared to other oil seeds proteins including soybean and other traditional cereal seeds. And their potential as a source of nutritional protein in animal foods is well known.

Sesame seed cake is rich in methionine, arginine, high nitrogen free extract (NFE), some essential fatty acids and its apparent digestibility in fish is very high (Mukhopadhyay and Ray, 1999). The seeds are especially rich in mono-unsaturated fatty acid, *oleic acid*, which comprises of up to 50% of fatty acids in them. Oleic acid helps lower LDL or "bad cholesterol" and increases HDL or "good cholesterol" in the blood. Research studies suggest that Mediterranean diet which is rich in mono-unsaturated fats may help prevent coronary artery disease, and stroke by favoring healthy serum lipid profile.

The seeds are also very valuable sources of dietary protein with fine quality amino acids that are essential for growth, especially in children. Just 100 g of seeds provide about 18 g of protein (32% of daily-recommended values).

Sesame seeds contain many health benefiting compounds such as sesamol (3, 4-methylene-dioxyphenol), sesaminol, furyl-methanthiol, guajacol (2-methoxyphenol), phenylethanthiol and furaneol,vinylguacol, and decadienal. *Sesamol* and *sesaminol* are phenolic anti-oxidants. Together, these compounds help stave off harmful free radicals from the human body.

Considering the quality of sesame seed meal (SSM), it is a promising feed ingredient in fish feed. However, there is a paucity of information on the incorporation of fermented sesame seed meal in a diet of catfish.

1.1 OBECTIVES

The general objective of this research is evaluate the effects of fermented sesame seed meal on growth performance of African catfish. Towards this, the specific objectives are to:

- determine the effects of fermented sesame seed meal as a fish meal substitute in a practical diet on the growth performance of *Clarias gariepinus*
- determine the changes in the hematological parameters of *Clarias gariepinus* fed the experimental diet.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Composition and Nutritive Value of Fermented Sesame Seed

Sesame seed contains ω3 and ω6 and is a rich source of nutrients like vitamins A, E, B, D; protein, lecithin; minerals such as copper, magnesium, zinc, calcium and phosphorous. Its calcium content is much higher than milk. Sesame contains amino acids of methionine, niacin, tryptophan and cysteine (FAO, 2012). Also, they have desirable physiological effects including antioxidant activity, blood pressure and serum lipid lowering potential as proven in experimental animals and humans *(Sirato-Yasumoto et al., 2001)*. The major protein fraction (globulin) in sesame contains about 95% of 13S globulin and seems to be a simple, salt soluble, very susceptible to heat denaturation and similar in subunit structure to soybean 11S globulin with more hydrophobic properties.

2.2 Factors affecting use of sesame seed

The anti-nutritional factors (ANFs) may be defined as those substances generated in natural feed stuffs by the normal metabolism of species and by different mechanisms (e.g., inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed) which exert effects contrary to optimum nutrition.

The presence of anti-nutritional factors in sesame seed has limited their widespread usage and direct incorporation into animal feeds *(Ogunji et al 2005)*.

2.2.1 Phytate

Phytate, myo-inositol hexakisphosphate, is the salt of phytic acid and is widely distributed in all seeds and possibly all cells of plants. It serves as a storage of phosphorous and minerals and accounts for 60-90% of the phosphorous in the plant. It is not digested by monogastric animals, so it does not provide monogastrics with sufficient phosphorus and minerals and it leads to phosphorus runoff causing P pollution of ground water from animal wastes *(Bilyeu et al., 2008).*

In oil seeds such as sunflower seeds, soybeans, sesame seeds, linseeds and rape seeds the phytic acid content ranges from 1- 5.4% (dmb).Phytate is the principal storage form of phosphorus in many plant tissues, especially bran and seeds. Phytate has been shown to block the absorption of not only phosphorus but also other minerals, particularly calcium, magnesium, iron, and zinc. Phytate has also been shown to negatively affect the absorption of lipids and proteins.

Animals do not produce the enzyme phytase needed to break down phytate. As a result, diets with high levels of phytate have reduced nutrient availability. At least 75% of the total phosphorus in corn, for example, is in the form of phytate-phosphorus.

Phytate is found in many grains and legumes. The level of phytate will depend on the feedstuff and the conditions under which it was grown. Cooler temperatures during the growing season produce lentils with reduced phytate content.

Phytase is now available as an additive to animal feeds to help with the breakdown of phytate. Breeders are also looking to produce low-phytate grains and legumes.

During the germination of seeds, phytate is hydrolysed, and phosphorous along with minerals such as calcium, magnesium and iron are liberated, becoming available for germination and development of the seedlings.

A high content of phytate in sesame seed has been considered a factor for limiting mineral bioavailability.

2.2.2 Oxalates

Oxalic acid is one of a number of anti-nutrients found in forage. It can bind with dietary calcium (Ca) or magnesium (Mg) to form insoluble Ca or Mg oxalate, which then may lead to low serum Ca or Mg levels as well as to renal failure because of precipitation of these salts in the kidneys.

Despite the sesame seed associated benefits, just like other legumes it contains oxalate. Oxalic acid is one of the most highly oxidized organic compounds widely distributed in plants, which occurs in two forms: soluble and insoluble oxalates. Soluble oxalate usually forms with sodium (Na^+), potassium (K^+) and ammonium (NH_4^+) ions, and insoluble oxalate forms with calcium (Ca^{2+}), magnesium (Mg^{2+}) and iron (Fe^{2+}) ions (Holloway et al. 1989; Savage et al. 2000).

2.3 Dietary protein requirements of Catfish

Protein comprises about 70% of the dry weight of fish muscle. A continual supply of protein is needed throughout life for maintenance and growth. Catfish, like other animals, actually do not have a protein requirement, but they require a source of nonspecific nitrogen and indispensable amino acids. Catfish requires about 40-45% crude protein. Proteins are large, complex molecules made up of various amino acids that are essential components in the structure and functioning of all living organisms (NRC, 1983). Protein Comprises about 15-20% of the dry weight of fish muscle (Eyo, 2002).

Ingested proteins are hydrolyzed to release amino acids that may be used for synthesis of tissue proteins or, if in excess, used for energy. Use of protein for energy is expensive; thus, catfish feeds should be balanced to assure that adequate level of nonspecific nitrogen, amino acids, and non-protein energy are supplied in proper proportions.

2.3.1 Amino acid requirements of catfish

Nutritionally, amino acids may be classified as either indispensable (essential) or dispensable (nonessential). An indispensable amino acid is one that the animal cannot synthesize or cannot synthesize in quantities sufficient for body needs; thus, they must be supplied in the diet. A dispensable amino acids one that can be synthesized by the animal in quantities sufficient for maximal growth. There are differences in amino acid requirements among the various species of fish and other animals, but that is expected since the physiological needs for certain amino acids and the relative proportion of structural proteins may vary among species. Dispensable amino acids can be synthesized by catfish, but there are certain advantages if they are provided in the diet. For example, if these amino acids are in the diet, energy is saved in their synthesis, and some dispensable amino acids can partially replace some indispensable amino acids (cystine can replace about 60% of the methionine, and tyrosine can replace about 50% of the phenylalanine). Practical catfish feeds contain liberal amounts of dispensable amino acids inherent in the proteins of various feedstuffs. In a practical feed, amino acid requirements are best met by feeding a mixture of feedstuffs or by using a mixture of feedstuffs supplemented with amino acids.

Table1: Nutrient composition of sesame seeds

Nutrient	Quantity (%)
Moisture	04.0-05.3
Protein	18.3-25.4
Oil	43.3-44.3
Saturated Fatty Acids (% in oil)	14.0
Monounsaturated Fatty Acids (% in oil)	39.0
Polyunsaturated Fatty acids (% in oil)	46.0
Ash	05.2-06.2
Glucose	03.2
Fructose	02.6
Sucrose	0.2
Phytosterols	0.4

Source: Agricultural Conspectus Scientificus (2010)

Table 2: Amino acid and fatty acid composition in sesame seeds

Nutrient	Quantity
Amino acid (g/16g N)	
Threonine	3.1-3.7
Valine	3.9-4.6
Cysteine + methionine	2.8-4.8
Isoleucine	4.0-4.2
Phenylalanine + tyrosine	6.4-9.6
Histidine	2.7
Tryptophan	1.3-1.5
Lysine	2.6-2.7
Argenine	12.0
Fatty acid (%)	
Palmitic acid (16:0)	11.7
Stearic acid (18:0)	05.2
Oleic acid (18:1)	41.4
Linoleic acid (18:2)	39.4
Linolenic acid (18:3)	00.4
Arachidic acid (20:0)	00.4
Behenic acid (22:0)	00.6

Source: Agricultural Conspectus Scientificus (2010)

2.4 Sesame seed and oil composition

Sesame oil meal has been found to have a value similar to that of soybean meal for carnivorous fish, and has successfully used as a fish meal protein substitute without negatively affecting their growth. About 50% of fish meal could be replaced with sesame oil meal in rainbow trouts (*Oncorhynchus mykiss*) *(Nang Thu et al., 2011)* and European sturgeons (*Huso huso*) (Jahanbakhshi et al., 2012

Sesame seed has higher oil content (around 50%) than most of the known oil-seeds although its production is far less than the major oilseeds such as soybean or rapeseed due to labor-intensive harvesting of the seeds. Sesame oil is generally regarded as high-priced and high-quality oil. Sesame ranks eighth in the world production of edible oil seeds. The total annual production of sesame seeds is around 3 million metric tons (MT) worldwide from 2000 - 2002.

It is one of the most stable edible oil despite its high degree of unsaturation. The presence of lignin type of natural antioxidants accounts for both the superior stability of sesame oil and the beneficial physiological effects of sesame. When sesame capsules are mature, they are fragile and will burst open easily, scattering the seeds on the ground and thus difficult to collect. Harvesting of sesame seeds is usually performed by cutting the plant stalks and stacking them vertically under the sun with the cut-ends downward in the threshing yard. Each dried stalk is then shaken or beaten over a cloth to catch the seeds that fly out from the dried capsules.

Sesame seed is a rich source of edible oil. It contains more oil than the major oil-seeds, such as soybean, rapeseed-canola, sunflower seed, and cotton seed. The oil content of sesame seed varies with the variety of sesame; it may range from 28% to 59% .The wild seeds contain less oil (around 30%) than the cultivated seeds because the oil content is an important criterion for seed selection in agriculture practice. In general, the cultivated seed has around 50% oil, whereas the color of the seed coat exhibits slight influence on the oil content.

Sesame oil belongs to the oleic-linoleic acid group. It has less than 20% saturated fatty acid, mainly palmitic (7.9-12%) and stearic (4.8-6.1%) acids. Oleic acid and linoleic acid constitute more than 80% of the total fatty acids in sesame oil. Unlike other vegetable oils in this group, the percentages of oleic acid (35.9–42.3%) and linoleic acid (41.5–47.9%) in the total fatty acids of sesame oil are close.

CHAPTER THREE

3.0 MATERIALS AND METHODS

The experiment was carried out at the wet laboratory of the Department of Fisheries and Aquaculture, Federal UniversityEkiti, Ekiti State.

3.1 Source and treatment of samples

Sesame (*Sesamum indicum L.*) seed used for this experiment were bought from Ilorin market, Kwara state.

It was cooked for about two hours and soaked in warm water for about five hour in order to get rid of the anti-nutritional factor present, and then placed in a plastic container with tight cover so as to create anaerobic condition and to provide efficient fermentation for seven days. After fermentation of the seeds, it was sun dried for three days, ground and incorporated into the test diets formulation.

3.2 Source of experimental fish

One hundred and fifty (150) juvenile of *Clarias gariepinus* were purchased from private fish farm Ikole Ekiti, Ekiti state.

3.3 Formulation of diets

Five isonitrogenous and isoenergetic diets were prepared (40% crude protein). The control diet (Diet A) contained no fermented sesame seed meal (FSSM) and 20% fishmeal. In the other diets (diets B, C, D and E) fishmeal protein contribution in the control diet was substituted at 25, 50, 75 and 100% with FSSM respectively. In each diet, chromic oxide was added at 0.05% of the diet as a non-digestible marker for nutrient digestibility determination.

TABLE 3

Gross percentage composition of Experimental diets for African catfish (*Clarias gariepinus*) **with varying inclusion levels of Sesame seed meal.**

INGREDIENTS	A	B	C	D	E
Fish meal (72%)	25	18.75	12.5	6.25	0
Soybean meal (44%)	24	21.9	21.8	22.4	21.7
Sesame seed meal (57.75%)	0	7.8	15.6	23.4	31.2
Groundnut cake (44%)	20	22	22.7	22.3	23.9
Maize	14.3	12.4	10.8	8.8	9.6
wheat offal	10	10.55	10	10.25	7
Starch	1.80	1.80	1.80	1.80	1.80
Vegetable oil	2.00	1.90	1.90	1.90	1.90
Salt	0.15	0.15	0.15	0.15	0.15
Methionine	1.00	1.00	1.00	1.00	1.00
Vitamin/mineral premix	1.00	1.00	1.00	1.00	1.00
Vitamin C	0.25	0.25	0.25	0.25	0.25
Chromic oxide	0.50	0.50	0.50	0.50	0.50
TOTAL	100	100	100	100	100

3.4 Experimental design

The experiment was conducted using 150 juvenile African catfish (*Clarias gariepinus*), obtained from private fish farms Ikole Ekiti, Ekiti state.

Fishes were acclimated for one week to experimental conditions and fed with commercial diet prior to the commencement of the experiment that lasted for seven weeks.

Fishes were distributed randomly into fifteen plastic aquaria there are three replicates per treatment and ten fish per replicate. Fish were weighted once in a week by taking the body weight, of the fishes in order to determine the increment in body weight and body length of fish.

15 plastic aquaria were used; each aquarium of about 25litres capacity was filled with fresh water from nearby tap water. Each aquarium was individually aerated while dissolved oxygen, pH and water temperature will be continuously monitored.

3.5 Statistical analysis

The statistical software programme SPSS (Version 16) was used to compare differences among the test groups. The experimental design is factorial in Complete Randomized Design. A two-way ANOVA was employed to determine whether there were significant differences in the variables measured among the experimental groups. When a difference was detected ($P<0.05$), Duncan Multiple Range Test was applied.

3.6 Data collection

3.6.1 Body weight Gain (g)

The body weight gain for each was obtained by taking the difference between the body weight for the given week and the body weight for the preceding week. Body weight gain per day was obtained by dividing body weight gain per week by seven.

I. Mean weight gain

Final weight- Initial weight.

II. Feed conversion efficiency

From the weight gained and feed consumed by fish in different treatments, the feed efficiency was computed using the following expression:

$$FCR = \frac{\text{Average feed intake per day}}{\text{Body weight gain per day}}$$

III.Protein Efficiency Ratio (PER)

This was calculated using data obtained feed intake and weight gain:

$$PER = \frac{\text{Body weight gain (g)}}{\text{Protein intake (g)}}$$

IV. Specific growth rate

$$\frac{\textbf{Log final weight-log initial weight} \times \mathbf{100}}{\textbf{Days of experiment}}$$

3.6.2. Hematological Parameter sample collection

At the end of the feeding trial, fish were tranquilized with 150mgl-1 solution of methane sulphonate (MS22) (Wagner et al; 1997) for blood collection. Blood samples were obtained from caudal vein of fish from each tank.one ML blood samples were collected into the bottles containing 0.05ml EDTA as anticoagulant.

Immediately after sampling, blood smears were prepared, red blood and white blood cell counts were carried out using standard hematological techniques (Dacie and Lews2001).

50ul haematocrit tube was filled with blood samples, after centrifugation (7200rpm for 10 minutes) of each blood sample, packed cell volume (PCV) was determined by the win tube and westergreen method as described by(Blaxhall and Daisley 1973).

Haemoglobin levels (Hb in grams per deciliters) were obtained by the cyanomet haemoglobin spectrophotometric method (Dorafshan et al; 2008).The blood indices including mean corpuscular volume (MCV in femtoliters). Mean corpuscular haemoglobin (MCH in pictograms per cell) and Mean corpuscular haemoglobin concentration (MCHC in grams per deciliter) were calculated according to the following formulas:

$$MCV\ (fl) = \frac{PCV\ (\%)}{RBC\ (10^6\mu l^{-1})} \quad \text{......equation 1}$$

$$MCH\ (pg) = \frac{(Hb\ (gdl^{-1}))}{RBC\ (10^6\mu l^{-1})} \quad \text{......equation 2}$$

$$MCHC\ (gdl^{-1}) = \frac{(Hb\ (gdl^{-1}))}{PCV\ (\%)} \quad \text{......equation 3}$$

CHAPTER FOUR

4.0 RESULTS AND DISCUSION

4.1 Growth Performance of African Catfish

Results of the effects of fermented sesame seed meal in diet of *Clarias gariepinus* is presented on Table 1. It reveals that diet1, 2, 3, 4 and 5 were significantly different ($p < 0.05$) from one other.

Fish fed with SSM at 25% inclusion level gave the best mean weight gain (7.40) which showed marginal difference from those fed fish meal without sesame seed meal. On the other hand fish fed 100% SSM (Diet 5) resulted in the lowest (2.26) mean weight gain. Feed intake were similar ($p > 0.05$) in fish fed control (16.39), 50% (15.88), 75 %(17.61) and 100 % (16.51) sesame seed meal inclusion but were significantly different ($P <0.05$) from catfish fed diet containing 25% fermented sesame seed meal.

Diets containing 75% and 100% inclusion levels of sesame seed meal showed similarity ($p>0.05$) on their effects on the fish. However, fish fed diets with 25 % (1.90), 50% (2.25) and control had different ($p < 0.05$) feed conversion ratio. Fish fed with SSM at 25% inclusion level gave the best feed conversion ratio which was marginal difference from those fed on fish meal without sesame seed meal.

Fish fed with SSM at 100% inclusion level gave the poorest feed conversion ratio.

Diet 3, 4 and 5 are significantly different from each other. Fish fed with SSM at 25% inclusion level gave the best Specific growth rate (1.57) which was marginal difference from those fed on fish meal without sesame seed meal (1.06). Diet 1, 3,4 and 5 are not significantly different from one another but significantly different from diet 2 in protein intake.

Protein efficiency ratio of fish fed with diet 4 and 5 are not significantly different from each other but are significantly different from diet 1 so also diet 2 and 3 are not significantly different from each other but are significantly different from diet 1.

The unsuitability of complete substitution was further reflected by the observed lack of response of the fish to the test diet at feeding times.

The present results exhibited that sesame seed meal protein can replace fish meal protein up to (25 %) in practical diets of African catfish (*Clarias gariepinus*). Similar research was done by (Deyab *et al.* 2009) who replaced fish meal protein by sesame seed meal (SSM) protein in commercial diets on growth performance, feed utilization and body composition of juvenile mono sex Nile tilapia, *Oreochromis niloticus* (L.) at 25% , 50 %,75 and 100 % and concluded that 50% inclusion of sesame seed meal has the best growth performance on mono sex Nile tilapia compare to inclusion at 75% and 100%.

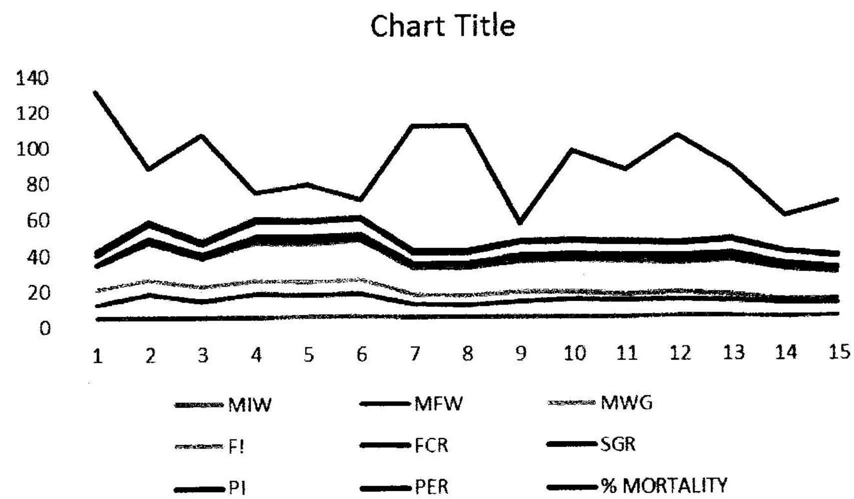
In the present study, however, catfish which has an accelerated growth rate in much higher water temperatures have shown very significant growth depression with sesame seed meal protein over 25% % of dietary protein (diets 3, 4 and 5). This growth depression was further reported by the lack of response of the fish to the test diets at feeding times. This is in agreement with the results of (Hashim *et al.*, 1994) who reported that winged bean seed meal cannot be used as a sole protein source for *O. niloticus* fry. Fish fed winged bean seed meal based diets exhibited low growth performance, feed conversion ratio (FCR) and SGR. El-Saidy & Gaber (2001) and El-Saidy *et al* (2005) reported the same results with the Nile tilapia and the common carp, respectively.

TABLE 4: **Growth Performance of African Catfish of African cat fish (*Clarias gariepinus*) fed with fermented sesame seed meal at varying inclusion level.**

PARAMETERS	DIET1	DIET2	DIET3	DIET4	DIET5
MIW	5.18 ± 0.04^{a}	5.25 ± 0.05^{a}	5.18 ± 0.035^{a}	5.24 ± 0.03^{a}	5.25 ± 0.02^{a}
MFW	9.68 ± 3.16^{a}	12.68 ± 0.30^{b}	7.30 ± 1.04^{a}	9.19 ± 0.43^{a}	8.01 ± 0.71^{a}
MWG	8.31 ± 0.29^{e}	7.40 ± 0.29^{d}	5.30 ± 0.10^{c}	3.94 ± 0.40^{b}	2.76 ± 0.69^{a}
FI	16.39 ± 3.65^{a}	21.62 ± 0.17^{b}	15.88 ± 1.36^{a}	17.61 ± 0.99^{a}	16.51 ± 2.41^{a}
FCR	0.75 ± 0.15^{a}	1.90 ± 0.10^{b}	2.25 ± 0.15^{c}	2.62 ± 0.02^{d}	2.77 ± 0.02^{d}
SGR	1.06 ± 0.57^{ab}	1.57 ± 0.04^{b}	0.60 ± 0.26^{a}	1.00 ± 0.08^{a}	0.75 ± 0.15^{a}
PI	6.55 ± 1.46^{a}	8.65 ± 0.07^{b}	6.35 ± 0.54^{a}	7.05 ± 0.39^{a}	6.60 ± 0.96^{a}
PER	1.31 ± 0.30^{c}	0.85 ± 0.02^{b}	0.84 ± 0.06^{b}	0.56 ± 0.08^{a}	0.42 ± 0.05^{a}
% MORTALITY	60.00 ± 30.0^{b}	15.00 ± 5.00^{a}	50.00 ± 34.64^{ab}	50.00 ± 10.00^{ab}	30.00 ± 10.00^{ab}

Different superscripts down a row indicate a significance ($p < 0.05$) difference.

FIGURE 1: **Growth Performance of African Catfish** of African cat fish (*Clarias gariepinus*)



4.2 Hematological analysis

The mean hematological values obtained from fish fed with five isonitrogenous and isoenergetic diets for *Clarias gariepinus* from the study are as presented in Table.

The Red Blood Cell counts in *Clarias gariepinus* has a range of 5.19 x 10^6mm^3 to 6.06 $\times 10^6mm^3$

The Packed cell volume (PCV) ranges 27.67 to 38.67%.Heamoglobin concentration ranges between 5.20 to 6.60g/dl. The White blood cells counts ranges between 20880 to $70670mm^3$

Mean corpuscular hemoglobin (MCH) range from 0.86 to 1.12pg.

There were significant difference in PCV of fish fed with diet1, 2 and 3.fish feed with diet 2 and 4 are not significantly different from each other, while diet 3 and 5 are not significantly different from each.

From statistical analysis, there was no significant difference in the Hb of fish fed with diet 1,2,3,4 and 5 at P>0.05.

There was no significant difference in the RBC of fish fed with diet 1,2,3,4 and 5 at P>0.05.

White blood cell

(WBC) is a defensive cell of the body. According to Douglas and Jane 2010, its amount has implication in immune responses and the ability of the animal to fight infection. Fish with higher value of WBC will be able to fight infection more than the one with lower value of WBC. In this study the mean value of circulating white blood cells in *Clarias gariepinus* fed with diet 1,2,3,5 and 5 ranges from 20880 to 46200mm^3

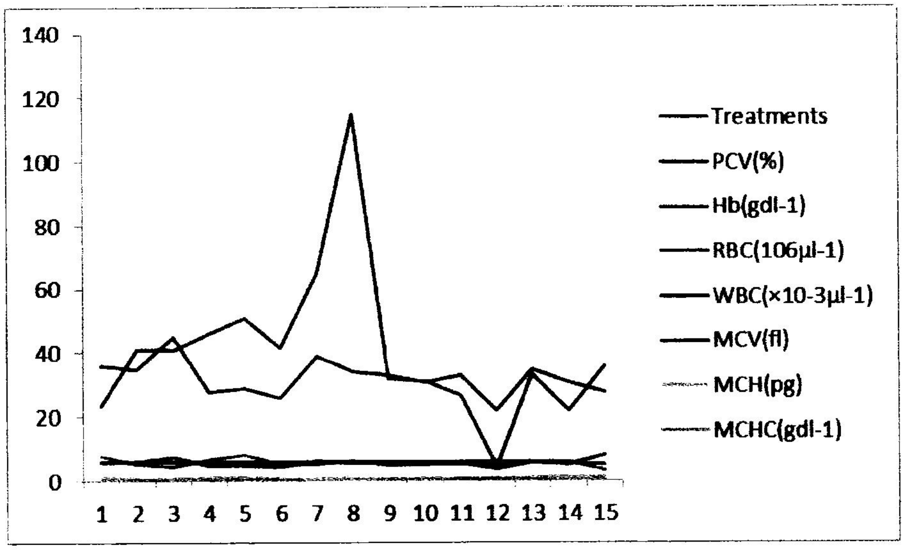
The physiological state resulting from different levels of SSM in diets was clearly reflected by the absence of significant differences ($p>0.05$) in the red blood cell count (RBC) and packed cell volume (PCV) observed in this study. This is in line with the observation of Obasa *et al.* (2013), feeding the African catfish with fermented African breadfruit (T. africana) seed meal based diets. Likewise, Brucka-Jastrzebska and Protasowicki (2005) subjected common carp *(Cyprinus carpio)* to cadmium and nickel exposure for a prolonged period.

TABLE 5: Hematological parameters of African cat fish (*Clarias gariepinus*) fed with fermented sesame seed meal at varying inclusion level.

PARAMETERS	DIET 1	DIET 2	DIET 3	DIET 4	DIET 5
PCV (%)	38.67^{b}	27.67^{a}	35.33^{ab}	28.67^{a}	31.33^{ab}
Hb(gdl^{-1})	5.87^{a}	6.60^{a}	5.37^{a}	5.20^{a}	5.50^{a}
RBC($10^{6}\mu l^{-1}$)$\times 10^{6}$	6.06^{a}	6.06^{a}	6.06^{a}	6.06^{a}	5.19^{a}
WBC$\times 10^{3}$	35.00^{ab}	46.20^{ab}	70.67^{b}	20.88^{a}	30.45^{ab}
MCV(fl)	6.38^{a}	4.56^{a}	5.83^{a}	4.73^{a}	6.33^{a}
MCH(pg)	0.97^{a}	1.09^{a}	0.88^{a}	0.86^{a}	1.120^{a}
MCHC(gdl^{-1})	0.16^{a}	0.24^{b}	0.15^{a}	0.19^{ab}	0.18^{ab}

Figures in each row with different superscript are significantly different at $p<0.05$. Figures in rows without superscripts are not significantly different at $p>0.05$. Hb: Haemoglobin content, PCV: Packed cell volume, WBC: White blood count, RBC: Red blood count, MCHC: Mean corpuscular haemoglobin concentration, MCV: Mean corpuscular volume, MCH: Mean corpuscular, haemoglobin

Figure 2: Hematological parameters of African cat fish (*Clarias gariepinus*) fed with fermented sesame seed meal at varying inclusion level.



CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATION

This present study revealed that sesame seed meal protein can replace up to 25 % of fish meal protein in practical diet of African catfish (*Clarias gariepinus*. In addition, sesame seed meal used in the present study is available at much lower prices (₦200/kg) than fishmeal (₦750kg) in Nigeria. Shortage in world production of fish meal, the conventional protein feed source, coupled with increased demand for fish meal in feeds for livestock and poultry is likely to reduce the dependence on fish meal as a single protein source in aqua-feeds. Throughout the world the efficiency of various alternative protein sources as partial or complete dietary replacements for fish meal has been evaluated in fish diets (Wee and Wang, 1987; Abdel-Fattah and El-Sayed, 1999; Ali *et. al.*, 2003). Sesame seed meal has greater potentials as promising plant protein source than others because of the high protein, coupled with abundant methionine and arginine (EAAs) (Eyo and Olatunde, 1996; Eyo et al., 2004) which are deficient in most plants protein feedstuff.

It is recommended that more work be conducted on sesame seed processing techniques to achieve the best and maximize the inclusion of sesame cake to fish meal. In order to cheapen cost of producing fish feeds and fish proteins in Africa, more people should go for mass production of sesame seed.

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APPENDIX

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
MV	1	3	5.1833	.04041	.02333	5.0829	5.2837	5.14	5.22
	2	3	5.2533	.04726	.02728	5.1359	5.3707	5.20	5.29
	3	3	5.1833	3512	.02028	5.0961	5.2706	5.15	5.22
	4	3	5.2433	.03215	.01856	5.1635	5.3232	5.22	5.28
	5	3	5.2467	.02309	.01333	5.1893	5.3040	5.22	5.26
	Total	15	5.2220	.04507	.01164	5.1970	5.2470	5.14	5.29
MFW	1	3	9.6767	3.15826	1.82342	1.8311	17.5222	7.10	13.20
	2	3	12.6800	.30000	.17321	11.9348	13.4252	12.38	12.98
	3	3	7.3000	1.0	.60302	4.7054	9.8946	6.37	8.43
	4	3	9.1867	.42724	.24667	8.1253	10.2480	8.70	9.50
	5	3	8.0067	.70550	.40732	6.2541	9.7592	7.56	8.82
	Total	15	9.3700	2.32000	.59902	8.0852	10.6548	6.37	13.20
MWG	1	3	8.3133	.29092	.16796	7.5907	9.0360	8.01	8.59
	2	3	7.4000	.29000	.16743	6.6796	8.1204	7.11	7.69
	3	3	5.3000	.10000	.05774	5.0516	5.5484	5.20	5.40
	4	3	3.9433	.40377	.23312	2.9403	4.9464	3.48	4.22
	5	3	2.7600	.69311	.40017	1.0382	4.4818	2.34	3.56
	Total	15	5.5433	2.17294	.56105	4.3400	6.7467	2.34	8.59
FI	1	3	16.3900	3.65956	2.11285	7.2991	25.4809	13.21	20.39
	2	3	21.6200	.17000	.09815	21.1977	22.0423	21.45	21.79
	3	3	15.8867	1.36001	.78520	12.5082	19.2651	14.91	17.44
	4	3	17.6167	.99203	.57275	15.1523	20.0810	16.59	18.57
	5	3	16.5100	2.41752	1.39576	10.5045	22.5155	14.33	19.11
	Total	15	17.6047	2.79596	.72191	16.0563	19.1530	13.21	21.79
FCR	1	3	.7500	.15000	.08660	.3774	1.1226	.60	.90
	2	3	1.9000	.10000	.05774	1.6516	2.1484	1.80	2.00
	3	3	2.2500	.15000	.08660	1.8774	2.6226	2.10	2.40

	4	3	2.6267	.02517	.01453	2.5642	2.6892	2.60	2.65
	5	3	2.7767	.02517	.01453	2.7142	2.8392	2.75	2.80
	Total	15	2.0607	.75357	.19457	1.6434	2.4780	.60	2.80
SGR	1	3	1.0567	.56757	.32769	-.3533	2.4666	.55	1.67
	2	3	1.5733	.03786	.02186	1.4793	1.6674	1.53	1.60
	3	3	.6000	.25632	.14799	-.0367	1.2367	.36	.87
	4	3	1.0000	.07810	.04509	.8060	1.1940	.91	1.05
	5	3	.7500	.14731	.08505	.3841	1.1159	.66	.92
	Total	15	.9960	.42234	.10905	.7621	1.2299	.36	1.67
PI	1	3	6.5547	1.46556	.84614	2.9140	10.1953	5.28	8.16
	2	3	8.6493	.07001	.04042	8.4754	8.8232	8.58	8.72
	3	3	6.3547	.54773	.31623	4.9940	7.7153	5.96	6.98
	4	3	7.0500	.39585	.22855	6.0666	8.0334	6.64	7.43
	5	3	6.6033	.96542	.55738	4.2051	9.0016	5.73	7.64
	Total	15	7.0424	1.11934	.28901	6.4225	7.6623	5.28	8.72
PER	1	3	1.3128	.30479	.17597	.5557	2.0699	.98	1.58
	2	3	.8562	.02587	.01494	.7920	.9205	.83	.88
	3	3	.8367	.06658	.03844	.6713	1.0021	.76	.88
	4	3	.5633	.08622	.04978	.3492	.7775	.47	.64
	5	3	.4167	.05508	.03180	.2799	.5535	.36	.47
	Total	15	.7971	.34101	.08805	.6083	.9860	.36	1.58
MORTALITY	1	3	60.0000	30.00000	17.32051	-14.5241	134.5241	30.00	90.00
	2	3	15.0000	5.00000	2.88675	2.5793	27.4207	10.00	20.00
	3	3	50.0000	34.64102	20.00000	-36.0531	136.0531	10.00	70.00
	4	3	50.0000	10.00000	5.77350	25.1586	74.8414	40.00	60.00
	5	3	30.0000	10.00000	5.77350	5.1586	54.8414	20.00	40.00
	Total	15	41.0000	24.79919	6.40312	27.2667	54.7333	10.00	90.00

MIW

	VAR00 010	N	Subset for alpha = 0.05
			1
Duncan[a]	1	3	5.1833
	3	3	5.1833
	4	3	5.2433
	5	3	5.2467
	2	3	5.2533
	Sig.		.056

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

MFW

	VAR00 010	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	3	3	7.3000	
	5	3	8.0067	
	4	3	9.1867	
	1	3	9.6767	
	2	3		12.6800
	Sig.		.108	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

MWG

	VAR00010	N	Subset for alpha = 0.05				
			1	2	3	4	5
Duncan[a]	5	3	2.7600				
	4	3		3.9433			
	3	3			5.3000		
	2	3				7.4000	
	1	3					8.3133
	Sig.		1.000	1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

FI

	VAR00010	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	3	3	15.8867	
	1	3	16.3900	
	5	3	16.5100	
	4	3	17.6167	
	2	3		21.6200
	Sig.		.369	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

FCR

	VAR00 010	N	Subset for alpha = 0.05			
			1	2	3	4
Duncan[a]	1	3	.7500			
	2	3		1.9000		
	3	3			2.2500	
	4	3				2.6267
	5	3				2.7767
	Sig.		1.000	1.000	1.000	.114

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

SGR

	VAR00 010	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	3	3	.6000	
	5	3	.7500	
	4	3	1.0000	
	1	3	1.0567	1.0567
	2	3		1.5733
	Sig.		.101	.053

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

PI

	VAR00 010	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	3	3	6.3547	
	1	3	6.5547	
	5	3	6.6033	
	4	3	7.0500	
	2	3		8.6493
	Sig.		.367	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

PER

	VAR00 010	N	Subset for alpha = 0.05		
			1	2	3
Duncan[a]	5	3	.4167		
	4	3	.5633		
	3	3		.8367	
	2	3		.8562	
	1	3			1.3128
	Sig.		.251	.874	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

MORTALITY

	VAR00 010	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	2	3	15.0000	
	5	3	30.0000	30.0000
	3	3	50.0000	50.0000
	4	3	50.0000	50.0000
	1	3		60.0000
	Sig.		.093	.143

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
PCV	DIET1	3	38.6667	5.50757	3.17980	24.9851	52.3482	35.00	45.00
	DIET2	3	27.6667	1.52753	.88192	23.8721	31.4612	26.00	29.00
	DIET3	3	35.3333	3.21455	1.85592	27.3479	43.3187	33.00	39.00
	DIET4	3	28.6667	5.85947	3.38296	14.1109	43.2224	22.00	33.00
	DIET5	3	31.3333	3.51188	2.02759	22.6093	40.0573	28.00	35.00
	Total	15	32.3333	5.57631	1.43980	29.2453	35.4214	22.00	45.00
Hb	DIET1	3	5.8667	1.77858	1.02686	1.4484	10.2849	4.60	7.90
	DIET2	3	6.6000	1.50000	.86603	2.8738	10.3262	5.10	8.10
	DIET3	3	5.3667	.81445	.47022	3.3435	7.3899	4.80	6.30
	DIET4	3	5.2000	.10000	.05774	4.9516	5.4484	5.10	5.30
	DIET5	3	5.5000	.45826	.26458	4.3616	6.6384	5.00	5.90
	Total	15	5.7067	1.07933	.27868	5.1090	6.3044	4.60	8.10
RBC	DIET1	3	6.0625	.00000	.00000	6.0625	6.0625	6.06	6.06
	DIET2	3	6.0625	.00000	.00000	6.0625	6.0625	6.06	6.06
	DIET3	3	6.0625	.00000	.00000	6.0625	6.0625	6.06	6.06
	DIET4	3	6.0625	.00000	.00000	6.0625	6.0625	6.06	6.06
	DIET5	3	5.1917	1.50833	.87083	1.4448	8.9386	3.45	6.06
	Total	15	5.8883	.67454	.17417	5.5148	6.2619	3.45	6.06
WBC	DIET1	3	35.0000	10.04639	5.80029	10.0434	59.9566	23.40	40.90
	DIET2	3	46.2000	4.70000	2.71355	34.5246	57.8754	41.50	50.90
	DIET3	3	70.6667	41.78915	24.12698	-33.1433	174.4767	32.00	115.00
	DIET4	3	20.8833	14.36961	8.29630	-14.8128	56.5794	4.50	31.35
	DIET5	3	30.4500	7.60740	4.39213	11.5522	49.3478	21.80	36.10
	Total	15	40.6400	24.84650	6.41534	26.8805	54.3995	4.50	115.00
MCV	DIET1	3	6.3780	.90875	.52467	4.1205	8.6355	5.77	7.42
	DIET2	3	4.5640	.25204	.14552	3.9379	5.1901	4.29	4.78
	DIET3	3	5.8280	.53040	.30623	4.5104	7.1456	5.44	6.43
	DIET4	3	4.7283	.96624	.55786	2.3281	7.1286	3.63	5.44
	DIET5	3	6.3340	1.57815	.91114	2.4137	10.2543	5.11	8.12
	Total	15	5.5665	1.14206	.29488	4.9340	6.1989	3.63	8.12
MCH	DIET1	3	.9677	.29337	.16938	.2389	1.6965	.76	1.30
	DIET2	3	1.0887	.24742	.14285	.4740	1.7033	.84	1.34

	DIET3	3	.8852	.13435	.07756	.5515	1.2190	.79	1.04
	DIET4	3	.8577	.01650	.00952	.8168	.8987	.84	.87
	DIET5	3	1.1154	.29021	.16755	.3945	1.8363	.92	1.45
	Total	15	.9829	.21748	.05615	.8625	1.1034	.76	1.45
HC	DIET1	3	.1558	.05926	.03421	.0086	.3030	.10	.22
	DIET2	3	.2371	.04159	.02401	.1337	.3404	.20	.28
	DIET3	3	.1530	.02928	.01690	.0802	.2257	.13	.19
	DIET4	3	.1877	.04652	.02686	.0722	.3033	.15	.24
	DIET5	3	.1759	.00646	.00373	.1599	.1920	.17	.18
	Total	15	.1819	.04667	.01205	.1561	.2077	.10	.28

PCV

	VAR00008	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	DIET2	3	27.6667	
	DIET4	3	28.6667	
	DIET5	3	31.3333	31.3333
	DIET3	3	35.3333	35.3333
	DIET1	3		38.6667
	Sig.		.065	.070

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Hb

	VAR00008	N	Subset for alpha = 0.05
			1
Duncan[a]	DIET4	3	5.2000
	DIET3	3	5.3667
	DIET5	3	5.5000
	DIET1	3	5.8667
	DIET2	3	6.6000
	Sig.		.190

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

RBC

	VAR00008	N	Subset for alpha = 0.05
			1
Duncan[a]	DIET5	3	5.1917
	DIET1	3	6.0625
	DIET2	3	6.0625
	DIET3	3	6.0625
	DIET4	3	6.0625
	Sig.		.176

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

WBC

	VAR00008	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	DIET4	3	20.8833	
	DIET5	3	30.4500	30.4500
	DIET1	3	35.0000	35.0000
	DIET2	3	46.2000	46.2000
	DIET3	3		70.6667
	Sig.		.192	.050

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

MCV

	VAR00008	N	Subset for alpha = 0.05
			1
Duncan[a]	DIET2	3	4.5640
	DIET4	3	4.7283
	DIET3	3	5.8280
	DIET5	3	6.3340
	DIET1	3	6.3780
	Sig.		.059

MCH

	VAR00008	N	Subset for alpha = 0.05
			1
Duncan[a]	DIET4	3	.8577
	DIET3	3	.8852
	DIET1	3	.9677
	DIET2	3	1.0887
	DIET5	3	1.1154
	Sig.		.222

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

MCHC

	VAR00008	N	Subset for alpha = 0.05	
			1	2
Duncan[a]	DIET3	3	.1530	
	DIET1	3	.1558	
	DIET5	3	.1759	.1759
	DIET4	3	.1877	.1877
	DIET2	3		.2371
	Sig.		.353	.110

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.