Growth and Haematological Studies of African Catfish (*Clarias gariepinus*) Juveniles Fed with Housefly Larva (*Musca dometica*) as Feed Supplement

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Abstract
An investigation was carried out to determine the effect of supplementing conventional fish feed (coppens), with Musca domestic maggots on the growth and haematological performance of Clarias gariepinus. Fifteen juvenile Clarias girepinus were stocked each in three treatment tanks and twenty in a control experiment. They were cultured in mini plastic water baths of 30 litres capacity at 24.5-26.5°c temprature. Initial mean weight and length recorded in the four treatment tanks were 6.81± 0.3g, 9.53± 0.9g, 10.71±1.0g and 7.52±0.4g, and 10.1±0.9cm, 9.5±0.08cm, 8.26±0.05cm, 5.4±0.03cm for T1, T2, T3, and T4 (control Tank) respectively. The percentage of the conventional feed to maggot inclusions were 70% to 30%, 55% to 45%, 35% to 65%, and 100% conventional feed for T1, T2, T3, and T4 respectively. The juveniles were fed at 3% body weight for 12 weeks and the final mean weights recorded were 54.72±.1.0g, 74.67± 1.5g, 31.17±0.8g and 26.45±0.6g for T1, T2, T3, and control(T4) respectively. The highest mean weight of 61.6g with specific growth rate of 1.8775, food conversion ratio of 24.28 and survival rate of 92% was observed in treatment 2 (T2). The result of this study shows that a combination of 50% compounded ration and 45% maggot gives the best growth performance. The juveniles fed with maggot showed significant variation ($p<0.05$) in the haematological values recorded. In T2 the haematological records were Haemoglobin count (HB) (9.5 ± 2.4^{a}), Pack cell volume (PCV) (28.0 ± 6.6^{a}),Total red blood cell (TRBC) (5.0 ± 2.5^{a}) Total white blood cell (TWBC) (3.7 ± 2.12^{a}), Mean corpuscular volume (MCV) (7.0 ± 2.0^{b}), Mean corpuscular haemoglobin (MCH) (2.33 ± 0.72^{b}), Mean corpuscular haemoglobin concentration (MCVC) (3.3 ± 0.08^{a}). The control experiment recorded the following values control ($HB5.7\pm0.50^{a}$), PCV (16.7 ± 1.5^{b}), TRBC (1.4 ± 0.20^{a}), TWBC (2.4 ± 0.32^{a}), MCV (11.9 ± 0.66^{b}), MCH (4.0 ± 0.25^{a}), MCHC (3.4 ± 0.02^{a}). It can hence be concluded that using housefly maggot directly as supplementary feed for Clarias girepinus at appropriate ration will enhance its growth and haematological performance.

Keywords: *Growth; Haematological: African catfish (Clarias gariepinus); Housefly larva (Musca dometica); Feed.*

Introduction

The African catfish is a chordate animal and belongs to Class Osteichthytes (bony fishes), Family Clariidae. It is a dominant freshwater fish. It can grow up to 1.4 and 2 m long and can weigh anything from 8 kg to 59 kg. The South African angling record is 35 kg; however a 58.9 kg specimen was caught in the Vaal River (FAO, 2012). It is popular specie grown in many manmade ponds because of high survival ability.

The increasing cost of fish feed has been at an alarming rate and this has affected the development and expansion of aquaculture in African countries particularly Nigeria (Sogbesan *et al.*, 2006).The conventional Fish meal (Coppens) has been the major protein source in the fish diet, hence the high demand and exponential rice in its price.

The supply for fish meal has been constrained and not affordable to poor subsistent farmers. The need for more research for vital protein augments to make affordable fishmeal and thus increase the production of catfish becomes eminent.

The research for suitable and cost-effective alternative protein sources for use in industrial aqua feeds will be the most critical factor in the development of intensive aquaculture in Nigeria (Sogbesan *et al.*, 2006).

Several attempts have been made to augment fish meal with other animal protein sources such as earth worm, shrimp waste, poultry waste, insect and plant protein sources such as sun flower, rape seed, soy bean meal and cottonseed meal (Fagbenro, 2013; Ayoola, 2010; Omitoyin, 2006). Insects and vertebrate animals has been employed for the production of fish feed; the developmental stages of most insects are important in the production of fish, poultry and pig feeds (Griffin *et al.*, 1994).

However, the inclusion of adult insects in fish diet can only replace fish meal partially due to the presence of chitin in their exoskeleton. Ng *et al.*, (2001) reported that chitin found in the exoskeletons of adult insects is a polymer of glucosamine insoluble in common solvents and its presence leads to reduced growth performance and protein utilization in catfish fed with high levels of these insects. In the same vein, problem of anti- nutritional factor in tropical legumes have limited their usage and direct incorporation into animal feeds (Ogunji and Wirth, 2001).

Edible insects contain high quality protein, vitamins and amino acids (Finke, 2002). Raumpold and Schlüter (2013) compiled nutrient compositions for 236 edible insects, as published in the literature (based on dry matter). Bukkens (2009) reported that a whole range of insects contain thiamine (also known as vitamin B1, an essential vitamin that acts principally as a co-enzyme to metabolize carbohydrate into energy). The thiamine content ranged from 0.1 mg to 4 mg per 100 g of dry matter.

Womeni *et al.* (2009) investigated the content and composition of oils extracted from several insects and thus opined that edible insects are a good source of fat.. Their oils are rich in polyunsaturated fatty acids and frequently contain the essential linoleic and α-linolenic acids.

Insects have a high food conversion rate e.g. crickets need six times less feed than cattle, four times less than sheep, and twice less than pigs and broiler chickens to produce the same amount of protein and they emit less greenhouse gases and ammonia than conventional livestock (Adesina *et al.*, 2011). Insects can be grown on organic waste. Therefore, insects are a potential source for conventional production (mini-livestock) of protein, either for direct human consumption, or indirectly in recomposed foods (with extracted protein from insects) (Bukkens *et al.*, 2009).

There are more than 1900 edible insect species and the most important ones are in the orders Coleoptera (beetles), Lepidoptera (butterfly and moths), Hymenoptera (bees, wasps and ants), Orthoptera (grasshoppers and crickets), Isoptera (termites), Hemiptera (true bugs), and Diptera (flies) (FAO, 2012).

Musca domestica is of the order Diptera. The order Diptera includes insects that are commonly called true flies or two-winged flies, and examples include mosquitoes, black flies, midges, fruit flies and house flies e.t.c. The common house fly, *M. domestica*, belongs to the Muscidae family and can be found almost anywhere on earth including garbage heaps, faecal matter, decaying matter and discharges from wounds and sores (Resh and Omoregie, 2003). *M. domestica* undergoes complete metamorphosis - the egg, larva (maggot), pupa and adult stages. It takes only 12-14 days to complete its life cycle. Matured instar larva are 3 to 9 mm long, typical creamy whitish in color, cylindrical but tapering toward the head. The head contains one pair of dark hooks. The posterior spiracles are slightly raised and the spiracular openings are sinuous slits which are completely surrounded by an oval black border. The legless maggot emerges from the egg in warm weather within eight to 20 hours after laying. Maggots immediately begin feeding on and developing in the material in which the egg was laid. The housefly larva has shown to be of great benefits as a potential protein source in livestock nutrition.

This study is hence aimed at determining the growth performance of *Clarias gariepinus* fed with maggot (the larva stage of the housefly, *Musca domestica*) as feed supplement; analysing the effect of the maggot supplement on the hematology of *Clarias gariepinus;* and in turn proving if *M.domestica maggot* will successfully supplement the conventional fish feed, with a goal of providing a cost effective protein diet for poor farmers.

MATERIALS AND METHOD

The research was carried out in the biology laboratory of the Department of Zoology and Environmental Biology, Micheal Okpara University Of Agriculture Umudike, Umudike Umuahia, Abia State in eastern Nigeria. The Area has a tropical climate and total annual rainfall range of 1800-2100 mm and a daily air temperature range of 21°-32°C. The average relative humidity is about 82-85% especially during the rainy season.

MATERIALS

70 juvenile of *clarias gariepinus* were purchased from Oriental Integrated Farms Limited, located at K/M 4ovom Street by Ikot-Ekpene, Aba, Abia Sate. The fishes were conveyed to the biology laboratory in a portable well-aerated white polythene bag containing water from the fish farm. They were cultured in a mini plastic water bath of 30 litres capacity at 24.5-26.5°c and acclimatized for 24hours in `dichlorinated municipal water. Water was changed once in three days. The fishes were accepted as well adapted to laboratory condition when less than 3% death was recorded in 14 days period. The initial length and weight of the fishes were recorded.

FORMATION OF EXPERIMENTAL GROUP

Two weeks after the purchase of the fishes, they were divided into 4 groups and placed in separate tanks; Tank 1, 2, 3, 4 groups according to their weight and length. There were 15 fishes per treatment tank and 20 in the control tank 4.

MAGGOT PRODUCTION

Poultry droppings were collected from the poultry farm of the National Root Crop Research Institute Umudike, an adjacent neighbour to the University. The poultry droppings were placed in an open bucket and constantly wet with water to make it moist to attract flies to lay eggs in it. Maggots were generated from the third to fourth day. Portions of the maggot infested droppings were put in another bucket and enough water was added, the maggot were sieved from the water surface and washed severally. After washing the maggot, they were oven dried at about 35-45°C (Adesini *et al.*, 2011).Part of the maggot infested droppings were also left for the maggot to pupate and mature into adults, in order to determine the fly species.

FEEDING

The fishes were fed 3% of their body weight with 1mm pelleted diet containing 35% crude protein twice per day within the first week of culturing. The maggot was introduced to the fishes together with the conventional feed at different ratios. 0% in Control, 30% in T1, 45% in T2, 65% in T3 manually throughout the 12 weeks i.e in ratios 30:70, 45:55, 65:35, and 0:100 respectively. Feeding was done twice daily in the morning and evening. Data was collected in every two weeks throughout the twelve weeks of the experimental period to determine the weight of fish. The quantity of feed fed to the fishes was adjusted as their weight increased.

GROWTH ASSESSMENT

Fish sampling was done once in two weeks; five fishes were randomly picked from each tank using a hand net sieve. They were weighed using a weighing scale (OHAUS MODEL Cs CAPACITY 5000X2g) and their length taken using a meter rule. The Specific Growth Rate (SGR), Feed Conversion Ratio (FCR), Mean weight gain (MWG), Mean length gain (MLG), and survival rate (SR) were calculated from the values obtained from the weight and length measurement as follows,

Specific growth rate (SGR%/DAY)

SGR% / Day= $\frac{(in\ w2 - ln\ W1)}{t} \times \frac{(100)}{1}$

Were ln w2 = natural log of final average weight at the end of the experiment.

In w1 =natural log of initial average weight at the beginning of the experiment.

t = culture period in days

(Kolkovski *et al*., 1995)

Feed conversion ratio (FCR)

FCR = Amount of Feed/Weight gain (g)

Survival rate (SR%)

SR% = $\frac{N1}{N0} \times \frac{(100)}{1}$

Were N_1 = total no of fish alive at the end of the experiment

N_0 = Total no of fish stocked at the beginning of the experiment

(Kolkovski *et al*., 1995)

Mean Weight Gain(MWG)

Mw_2-Mw_1

Were Mw_2 = final mean weight after experiment

Mw_1= initial mean weight at beginning of experiment.

Mean length gain(MLG)

Ml_2-Ml_1

Were Ml_2 = final mean length after experiment

Ml_1= initial mean lenght at beginning of experiment.

(Kolkovski *et al*., 1995)

HAEMATOLOGICAL ANALYSIS

At the end of the experiment, three fishes from each tank were randomly taken for quick blood sampling. Blood (1-2ml, depending on fish size) was collected from the vertebral blood vessel towards the caudal peduncle of each fish. 1-2 ml blood samples were collected from cardiac puncture using 2mL disposable heparinised syringe treated with EDTA as anti-coagulant. The blood and plasma produced after centrifugation were analyzed for Haematocrit Packed cell volume (PCV), whole blood haemoglobin count(HBC), Total red blood cell (TRBC) and white blood cell (TWBC).

Blood cell count: Haemocytometer was used in blood cell count through the Neubauer chamber. The blood diluting fluid was prepared as described by Svobodova *et al.*, (1991). The blood cells were counted on the counting chamber of haemocytometer with the aid of compound microscope:

TRBC = No of cells counted x3x10x200 (10^6 mm 3)

TWBC = No of cells counted x0x25x10x20 (10^4 mm 3)

Haemoglobin estimation: Haemoglobinometer was used for haemoglobin estimation based on acid haematin method (SAHLI):

Packed cell volume (PCV): The packed cell volume was measured after placing sealed micro-haematocrit tube in a centrifuge at 10,500 rpm using micro-haematocrit reader and expressed as percentage.

Other parameters were determined according to the method described by Svobodova *et al.* (1991).

Mean corpuscular volume (MCV); this is the mean volume of the erythrocyte counted in the sample and it was calculated using the standard formula

MCV= $\frac{hematocrit \text{ x } 1000}{red\ blood\ cell\ count}$

Mean corpuscular haemoglobin (MCH); this represents the mean mass of haemoglobin in the red blood cell (RBC) and it is calculated thus

MCH= $\frac{hemaglobin \text{ x } 10}{red\ blood\ cell\ count}$

Mean corpuscular haemoglobin concentration (MCHC); this is the mean concentration of haemoglobin in the red blood cell, it is gotten by haemoglobin MCHC= $\frac{hemaglobin}{hematocrit}$

STATISTICAL ANALYSIS

The data obtained were statistically evaluated using Randox kits for each parameter. All the results were subjected to analysis of variance (ANOVA). Duncan multiple range test was used to evaluate the mean difference at 0.05 significant levels.

RESULTS

GROWTH PERFORMANCE

Specific Growth Rate

The Specific Growth Rate (SGR) of the fishes fed with different levels of maggot inclusion differed respectively, the highest growth rate of 1.88 was observed in T2, as shown in Table 1, Specific growth rate of the *C. gariepinus* juveniles.

Table 1, Specific growth rate of the *C. gariepinus* Juveniles.

EXPERIMENTAL TREATMENT	SPECIFIC GROWTH RATE
T1(30% maggot)	0.9370
T2 (45% maggot)	1.8775
T3 (65% maggot)	0.7090
T4 (control)	0.0702

T- Treatment

3.1.2 **Mean Weight Gain**

The highest mean weight gain in all the treatments (1- 4) was observed in T2 with weight of 61.6± 2.0, as in Table 2, Mean weight gain of the *C. gariepinus* juveniles.

Table 2, Mean weight gain of the *C. gariepinus* Juveniles.

EXPERIMENTAL Treatments	Mean Weight gain(g)
T1	44.46
T2	61.62
T3	24.04
T4	19.70

T- Treatment

3.1.3 **Mean length gain**

The highest mean weight gain was observed in T2 with the value 17.0 cm, as shown in Table 3, Mean length gain of the *Clarias gariepinus* juveniles.

Table 3, Mean length gain of the *Clarias gariepinus* juveniles.

EXPERIMENTAL Treatments	Mean length gain(cm)
T1	12.6
T2	17.0
T3	11.54
T4	10.84

T-Treatment

Figure 1, Mean weight and Length gain of the *Clarias gariepinus* Juveniles, shows the mean weight and length gained by the C.gariepinus juveniles throughout the period of study.

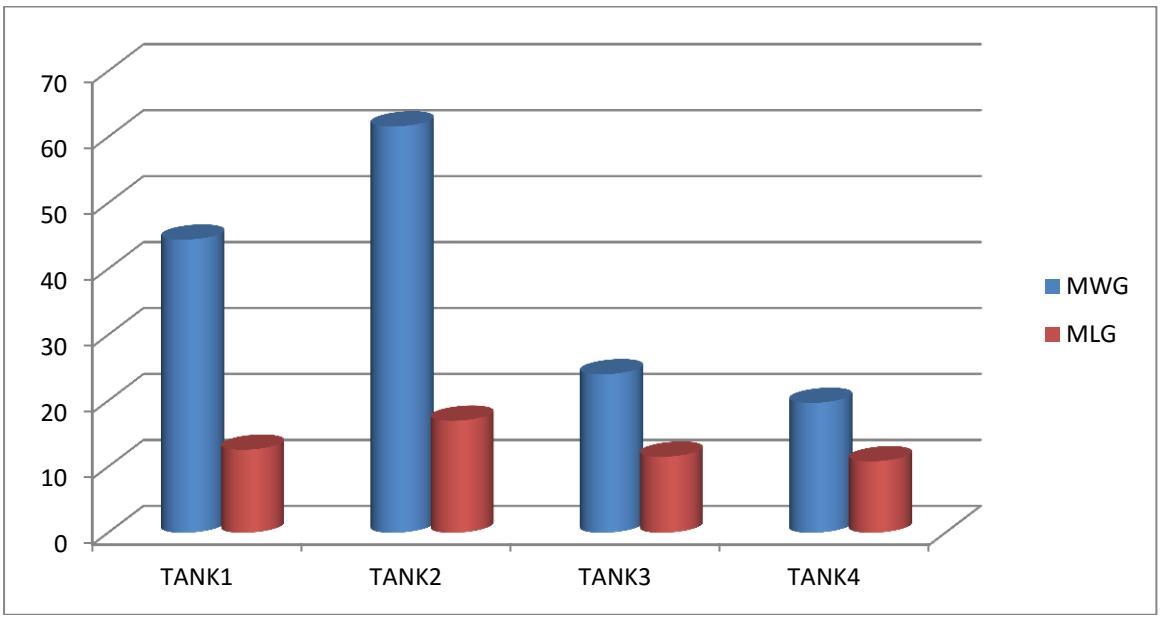

Figure 1, Mean weight and Length gain of the *Clarias gariepinus* Juveniles.

FOOD UTILIZATION BY THE JUVENILES

Food Conversion Ratio (FCR)

The highest food conversion ratio was observed in Tank 4 at the rate of 33.07, this is displayed in Table 4, Feed conversion ratio of the *C.gariepinus* Juveniles in the various treatment Tanks.

Table 4, Feed conversion ratio of the *C.gariepinus* Juveniles in the various treatment Tanks.

Treatments	Food conversion ratio (FCR)
T1	24.07
T2	22.7
T3	26.07
T4	33.07

T-Treatment

SURVIVAL RATE

At the end of the experiment, the highest survival was observed in T2 with percentage survival rate of 92%, this can be seen in Table 5, Percentage survival rate of the *C.gariepinus* Juveniles.

Table 5, Percentage survival rate of the *C.gariepinus* Juveniles.

Treatments	Percentage Survival rate (%)
T1	86%
T2	92%
T3	80%
T4	80%

T-Treatment

HAEMATOLOGY RESULT

There was significant increase (>0.05) in the haematological parameters recorded in all the treatments as seen in Table 6, Haematological record for the *C. gariepinus* fed with supplement of *M. domestica* maggot. The increases observed in these values are also displayed in Figures 2, the haematological parameters.

Table 6, Haematological record for the *C. gariepinus* fed with supplement of *M. domestica* maggot.

Treatments	HBC	PCV	TRBC	TWBC	MCV	MCH	MCHC
T1	9.5±2.4[a]	28.0±6.64[a]	3.3±1.3[ab]	3.7±1.7[a]	8.33±2.51[a] b	2.2±0.63[b]	3.3±0.08[a]
T2	9.5±2.54[a]	28.0±6.62[a]	5.0±2.5[a]	3.7±2.12[a]	7.0±2.01[b]	2.3±0.72[b]	3.3±0.08[a]
T3	8.2±.90[a]	24.3±5.5[ab]	2.7±1.15[a] b	3.8±1.73[a]	9.5±2.17[ab]	3.2±0.85[ab]	3.3±0.05[a]
T4	5.7±0.50[a]	16.7±1.54[b]	1.4±0.20[b]	2.4±0.32[a]	11.9±0.66[b]	4.0±0,25[b]	3.4±0.02[a]

T-Treatment

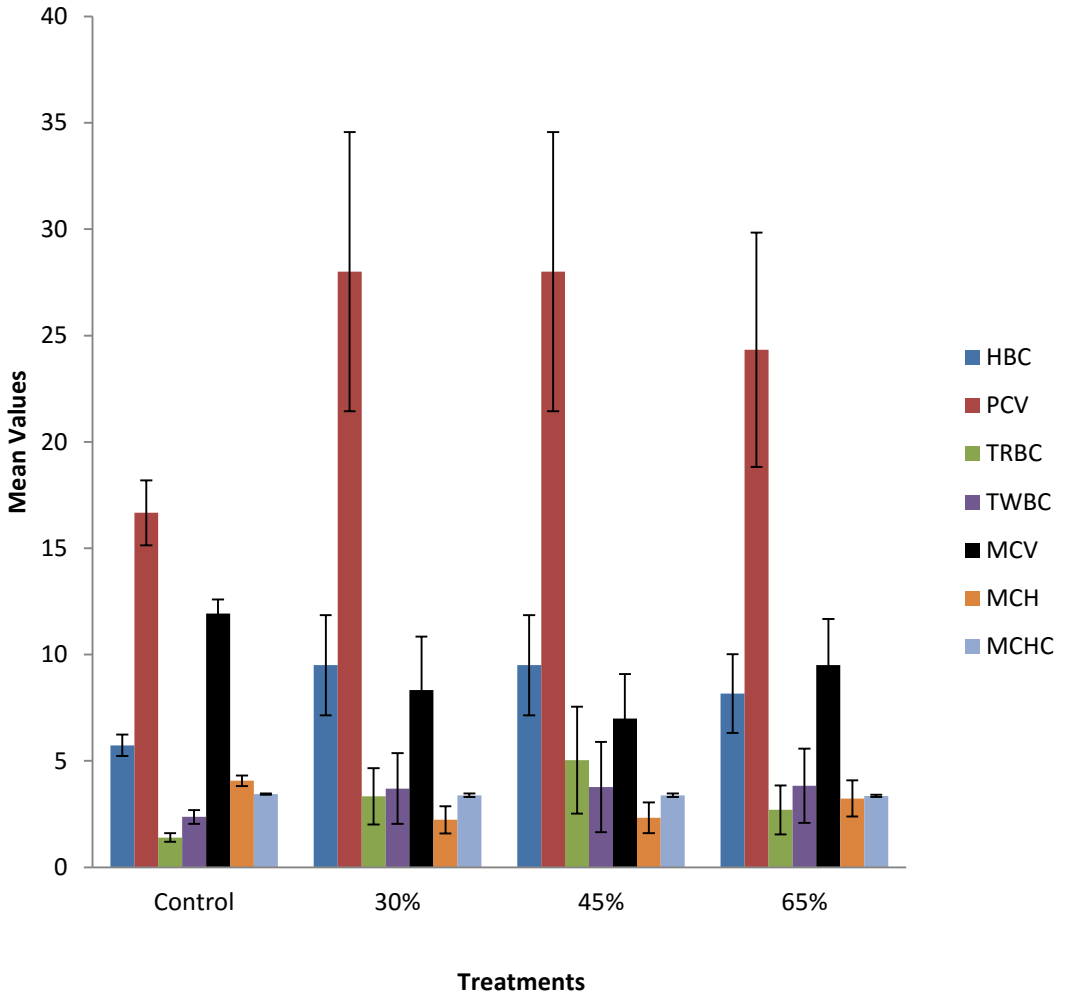

Fig 2, tl

WBC - whole blood haemoglobin count.
PCV-Packed cell volume.
TRBC -Total red blood cell.
TWBC - Total white blood cell .
MCH- Mean corpusclar haemoglobin.
MCV- Mean corpusclar volume.
MCHC- Mean corpuscular hemoglobin concentration

DISCUSSION

The effect of *M. domestica* maggot as feed supplement on the growth performance of *Clarias girepinus* juveniles as shown in tables 1, 2 and 3, indicate that they were well utilised by *C. greipinus* juveniles as evidenced in the good performance recorded in their weight and length. The non significant difference of the evaluated parameters on growth and nutrient utilization among the three (3) experimental treatments implies that the maggot meal can successfully replace fish meal in fish diets. This result agree with the report of other authors who have observed a better performance of fish fed diets containing maggot meal over those solely fed on fish meal diets meal. Thus, this is a reflection of the nutritive quality and acceptance of this biomaterial (Ogunji *et al.,* 2001). The result also corroborates previous observation that maggot meal, like other animal protein sources is well accepted and utilised by fish (Alegbeleye *et al*., 2003). Amongst the treatment tanks, the Food Conversion Ratio (FCR) recorded; T2 (22.7), T1 (24.07), T3 (26.07) and T4(33.07) indicates that the maggot meal was most utilized by the fishes in T2 and T1. This result corresponds with Steffens (1989) findings and this confirms that the higher the Protein Efficiency Ratio (PER) value, the more efficient the protein utilization. Idowu *et al.,* (2003), also reported a PER values of 1.07–2.05 for *Clarias gariepinus* fed diets containing maggot meal. Adesulu and Mustapha (2000) also indicated that maggot meal may be superior to other protein sources in fish diets. They suggested that the tender easily digested nature of maggots may contribute to this. Spinelli *et al.,* (2001), have reported the amino acid profile of maggot meal and indicated that it contains the same number and outstanding level of amino acids found in fish meal including all the essential ones. Table 5 shows the survival rate amongst the treatments, the best survival rate was recorded in T2 with 92%, followed by T1(86%), T3(80%) and T4(75%).The percentage survival showed that there was less mortality recorded during the period of study and this might be as a result of stress, water difference. This results on the survival rate correlates with the observation of Faturoti and Ifili (2007), who indicated that the feeding of *C. gariepinus* fingerlings on maggot diets made for high survival. The haematology records shown in table 6 revealed that there was significant ($p>0.05$) variation in the haematological parameters of all the treatments T1- T4 except for the MCHC in T3 and T4 where there was no significant ($p<0.05$) variation. There was also significant ($p>0.05$) increase between treatments fed with maggot meal rations and the control in their HBC, PCV,TRBC,TWBC, MCV and MCH counts respectively. The highest HBC and PCV were observed in T2 followed by T1, T3 and T4. The highest TRBC was recorded in T2, ($5.0x\ 10^9\ mm^3$) and TWBC in T3 ($3.8x10^4\ mm^3$). T4 gave the highest MCV and MCH values. The observed presence of significant difference in the haematological parameters of *C. gariepinus* in this study probably indicates that those parameters were significantly affected by the diets. This is similar to that observed by Akintayo *et al.,* (2008) on *C. gariepinus* fed toasted sunflower seed meal based diets. El-baraasi and Farma (2004) reported that differences in blood parameters of fish could be ascribed to differences in diet composition. However, the trend observed in this study had a beneficial effect on *C. gariepinus*, given that the values are above the normal range recorded for African catfish (Erondu *et al.,* 1993). Akinwande *et al.,* (2004) opined that a measurable increase in white blood count of fish or any animal is a function of immunity and animals' resistance to some vulnerable illness or disease. This increase might indicate that the fish under study had high immunity or resistance to disease. George *et al.,* (2007) observed that when 45 - 50% fish meal was replaced by soybean meal in the diet for *Clarias gariepinus*, there was increase in PCV, HB and RBC of the fish fed the diet. High value of Erythrocyte count recorded in this study was also observed by Akintayo *et al*. (2008) which indicates high oxygen absorption and transportation capacity of the cells of the fish under study.

CONCLUSION

The inclusion of maggot meal at high level of (30%-45%) in the diet of *Clarias gariepinus*, had significant increase in the growth parameters. There was also increase in the health condition of *Clarias gariepinus* when fed at this level of inclusion. This ratio is hence best advocated for. Thus, Fish meal can to a great extent be replaced by maggot meal, as this will reduce cost and meet growth standards in *Clarias gariepinus.*

RECOMMENDATION

Musca domeatica larva from this study is proved suitable for use in aquaculture. It is therefore recommended that it should be used in the formulation of fish feed to obtain maximum growth as regards to fish meal which is very expensive. Emphasis should be made on the culture and availability of the insect larva since its demand is low and can be used as a substitute to fishmeal.

Finally *M. domestica* larvae is the hope and solution to the high cost of fish meal. Its application will promote sustained aquaculture business. These research diets are promising if developed. Further researches would help to fortify and improve on the diets for better performances of fishes as this will reduce the high consumption of beef-meat which can then be fully supplemented with fishes with high protein content.

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