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Haematological Characteristics of *Clarias gariepinus* (Buchell, 1822) Juveniles Fed with Poultry Hatchery Waste

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Abstract: A twelve week feeding trial was carried out in order to assess the effect of feeding poultry hatchery waste on haematological parameters of *Clarias gariepinus* juveniles as a bioindicator of their health status. A control experiment was set up with fish fed with 37% crude protein. Fish fed with the poultry hatchery waste showed a slight decrease in haematological values of Parked Cell Volume (PCV,0.29±0.01), Haemoglobin (HB,98.66 ±5.13), Red blood cell (RBC,1.83±0.05), Mean Corpuscular Volume (MCV,161.33±9.291), Mean Corpuscular Haemoglobin (MCH,58.83±2.96), Mean Corpuscular Haemoglobin Concentration MCHC, (33.57±0.65), White Blood Cell (WBC,23.66±2.5) compared to the values of fish fed the control diet, PCV (0.37±0.01), HB (12.33±5.03), RBC (1.9±0.1), MCV (19.66±16.44), MCH (66.14±5.37), MCHC (33.74±1.016) and WBC (22.33±2.51). It is concluded that using poultry hatchery waste as supplementary feed on *Clarias gariepinus* showed a slight decrease in the haematological parameters but it has no negative impact on the health status of the specie. Therefore direct use of poultry hatchery waste as sole supplementary feed should be encouraged.

Key words: Haematology % *Clarias gariepinus* % Poultry hatchery waste % Fish feed % Bioindicator

INTRODUCTION

Fish is very important to humans because it contains protein of very high quality and also has sufficient amounts of all the essential amino acids required by the body for growth and maintenance of lean muscle tissue. The protein in fish, as well as similar foods of animal origin, makes up complete protein sources in many people's diets around the world. High quality proteins, such as the protein in most fresh fish, can be used to maintain an active metabolism. Low quality protein does not contain all essential amino acids required for use in protein synthesis and means the protein must either be used for energy or converted to fat [1]. Fish is one of our most valuable sources of protein; about 25% of animal protein is obtained from fish and shell fish. About 35% of all fish is eaten fresh, chilled or frozen. It is also cured or canned (16%) or made into oil and fish meal (32%). Fishes are used as medicine, ground into vitamins, or processed into cosmetics and perfumes, lubricants, varnishes, soap and margarine. Whales, seals and oysters are valued for many of the above uses. Scientists often use fish, especially goldfish, for experiments and medical research [1]. Fishes are rich in Omega-3 fatty acids which plays very important role for normal growth particularly for the blood vessels and the nerves as well as keeping our skin and other tissues youthful. Research studies have revealed that in populations that consume large quantities of fish, with a high utilization of Omega3s, there is a reduced risk of heart disease. Fish is important in the diets and livelihoods of many poor people suffering from vitamin and mineral deficiencies [2].

The national fish demand is about 1.85 million tonnes while the local production is only 0.51 million tonnes, based on a population figure of 140 million people. Nigeria currently imports 0.7 million tonnes of frozen fish annually making it the highest importer of frozen fish in the world with annual foreign exchange drain of N35.0 billion. The challenge therefore is to bridge the wide gap between fish demand and supply.

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In order to meet the growing demand of fish in Nigeria aquaculture industry is growing. Tilapia and catfish are the primary species produced at domestic fish farms, but it will be quite some time before production can match consumer demand [3]. Aquaculture alone has the potential to supply the national requirement for fish if properly harnessed. The act of fish culture has been existing for long and for over two decades, domestic supply of fish has been inadequate, hence animal protein in the diet of Nigerian's is affected from its normal recommended 40% protein level [4]. Aquaculture is the least expl oited, the only option left is to practice aquaculture (fish farming). But one of the major problems faced by aquaculturist today is the provision of nutritive and cheap feed to reduce cost of production.

Nigerian aquaculture industry is currently faced with the problem of inadequate supply and prohibitive cost of quality fish feeds. [1, 5] reported increasing attempt to develop practical diets for farmed fish in Nigeria. However most fish farmers particularly in the rural areas still depend on agricultural wastes including poultry litters for feeding fish [6]. It was noted that Nigeria produces large quantities of agricultural and agro-industrial by products, which serves as alternative feed sources to conventional feed. The nutrition requirements of fish are similar to those of animals. For growth, reproduction and other normal physiological functions, they need to consume protein, minerals, vitamins and growth factor and energy sources [7]. It was also stated that a deficiency of one or more of the essential nutrients may results in a reduced rate of performance, disease or even death. These nutrients may come from artificial or prepared diet or from natural aquatic organisms.

Poultry litter has been considered to have some nutritional values containing about 25.75% crude protein [8]. It has been noted that the concept of utilizing poultry litter is highly desirable since it will not only eliminate the problem of waste disposal but also provide cheap fish feed at litter cost. Different species of fish have been fed with poultry litters and other form of non protein nitrogen with different results. However, the adverse effect of feeding fish with poultry droppings particularly on haematological parameters is very scanty. Blood is a good indicator to determine the health of an organism [9]. It also acts as pathological reflector of the whole body; hence hematological parameters are important in diagnosing the functional status of exposed animal to toxicants [10]. Poultry hatchery waste is an unconventional feed that is now widely used in commercial freshwater aquaculture in order to reduce cost [3]. However, the health implication of the use of poultry hatchery waste has not been investigated, therefore, there is need to know the merit and demerit effect of this waste on the fish. The aims of the study is to investigate the haematological changes in the juvenile catfish *C. gariepinus* fed with hatchery poultry waste for the duration of 12 weeks and to determine the ability of the fish to adapt to poultry hatchery waste as food and to observe the growth pattern.

MATERIALS AND METHODS

One hundred and fifty juveniles *Clarias gariepinus* were bought from Ibafo Fish Farm in Ogun State. The fishes were transported in an open 25l plastic container to the Marine Sciences Laboratory of University of Lagos and acclimated for 14 days in the laboratory in fifteen plastic aquaria specification (70x 95 x 70cm^3). The tanks were already disinfected and filled with dechlorinated tap water. Ten fishes were randomly distributed into each aquarium and fed with 37% crude protein commercial feed prior to the commencement of the experiment.

In the laboratory, *C. gariepinus* were distributed into 15 plastics tanks already disinfected and filled with dechlorinated tap water, the water was and filled to 2/3 of the volume of each tank (50 litres).The test animals were then put in the plastic tanks and after 14 days of acclimatization, the tanks were labeled T_0, T_1, T_2, T_3 and T_4 in replicates of 3 and 10 juveniles of *C. gariepinus* were transferred into each of the tanks using a scoop net. The physico-chemical parameters in this experiment were not measured but suitable conditions were maintained by cleaning the tanks and constant changing of the water which took place every day.

Preparation of Feed: The feeds used for this experiment were categorized as follows with 100% poultry hatchery waste for tank T_4, tank T_3 was fed 25% compounded feed and 75% poultry waste, tank T_2 was fed 50% compound feed and 50% poultry waste, tank T_1 was fed 75% compound feed and 25% poultry waste while tank T_0 was fed with 100% formulated feed.

`Poultry hatchery waste was collected from Ajanla fish farm, along Lagos - Ibadan expressway in Oyo state, it was processed, parboiled then sun-dried for two days before it was grinded and small quantity was taken to the Department of animal science, University of Ibadan for proximate analysis after which the rest was then pelleted into 2mm sizes to enable the fishes feed on them,

the fishes were then fed with it while the control group were fed with 37% crude protein commercial feed at 5% of their body weight.

The compounded feed had compositions which include Maize, Wheat offal, Indomie, Groundnut cake, Fish meal, Vitamin premix, Mineral premix, Soya meal. The ingredients were mixed together, grinded into powdery form and pelleted into 2mm sizes to enable the fishes feed on them.

Experimental Procedure: The mean average weight of the fish in each tank was determined at the beginning of the experiment and at every 1 week. The weight of the fishes was determined using weighing scale (OHAUS MODEL Cs 5000, CAPACITY 5000×2g). This was done by placing a container on the scale and the balance adjusted to zero, after which the fishes (10) in each tank were collected by the use of a scoop net into the container and measurement taken.

Haematological Studies: After 12 weeks haematological studies was carried out on the fishes.

The fishes were taken out individually using a small hand net and placed belly upward on a table. Blood samples of about 4milliliters was collected from the caudal peduncle [11] with the aid of a $2cm^3$ plastic syringe,1ml of the blood was dispensed into ethylene diamine tetra-acetic acid (EDTA) anticoagulant for haematological studies while 3ml was transferred into a tube containing lithium heparin anticoagulant to obtain plasma for biochemical analysis. The use of plastic syringe is a necessary precaution with fish blood because contact with glass result in decreased coagulation time. The plasma obtained by centrifugation from the lithium heparinised samples was stored at 20^0C until analysed.

Haematological values were measured following standard methods [9, 12, 13]. Packed cell volume (haematocrit method) and haemoglobin (Hb) concentration (cyanmethaemoglobin method) were analysed within two hours after collection. Red blood cells (RBC) and White blood cells (WBC) were counted by Neubauer's improved haematocytometer using Hyem's and Turk's solution as a diluting fluid respectively, Packed cell volume (PCV), Mean corpuscular haemoglobin (MCH) and Mean cell volume (MCV) were calculated respectively using standard formula described by [14, 15].

The plasma was analysed for Triglyceride [16], Urea and Creatinine [11], Alkaline phosphate (ALP), Cholesterol [17] and Total protein [18]. The data obtained were statically evaluated using the Randox kits for each parameter respectively.

Statistical Analysis: All the results were subjected to analysis of variance (ANOVA). Duncan multiple range test [19] was further used to evaluate the mean differences at 0.05 significant levels.

RESULTS

Table 1 shows the composition of formulated feed used for the experiment; it includes Maize, Wheat offal, Indomie, Vitamin premix, Soya meal, Groundnut cake, Fish meal. While the weekly mean average weight of the fishes are shown in Table 2. Growth and nutrient parameters of *Clarias gariepinus* fed with compounded ration and poultry hatchery waste are shown in table 3. Table 4 is the proximate analysis of the poultry hatchery waste in

Table 1: Composition of formulated feed used as control

Ingredient	(%) Composition
Maize	10.54
Wheat offal	10.54
Fish meal	21.96
Indomie	21.96
G.N.C	21.96
Premix	2.5
Soya meal	10.54
Total %	100%

Table 2: The Weekly Mean Average Weight of the Fishes

Sample	Week 0	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9	Week 10	Week 11	Week 12
T_0	10	14.8	19.6	23.5	17.8	22.6	22.6	24.4	26.0	27.1	28.5	30.1	30.9
T_1	10	16.7	21.2	24.2	26.7	30.0	30.0	32.7	33.5	34.5	35.5	36.3	36.9
T_2	10	17.5	20.4	21.3	26.3	28.3	30.0	30.5	31.0	31.9	32.7	33.4	34.0
T_3	10	14.3	21.1	20.9	22.6	26.9	28.7	29.2	29.7	30.1	31.3	31.9	28.7
T_4	10	19.8	22.7	22.6	25.7	27.5	30.3	31.1	31.6	32.2	33.4	34.7	35.4

Table 3: Growth and nutrient parameters of *Clarias gariepinus* fed with compounded ration and poultry hatchery waste.

Parameters	T0	T1	T2	T3	T4
MWG	20.9	22.7	19.8	16.2	21.1
% MWGT	209	159.9	139.4	129.6	147.6
%WG/Wk	17.4	13.3	11.6	10.8	12.3
SGR	44	45.3	43.3	40.3	44
FCR	3.657	3.711	4.159	5.257	4.214
DRF	0.0224	0.2498	0.2441	0.2720	0.2564
DRG	0.0105	0.0090	0.0085	0.0073	0.0086
GEFC	0.4688	0.0360	0.0348	0.0268	0.0335
PER	0.153	0.151	0.135	0.106	0.133
PI	136.6	150.5	147.1	152.2	158.9

Table 4: Proximate analysis of poultry hatchery waste

% Crude protein	21.44
% Ash	50.00
% Ester extract	12.00
% Crude fibre	2.00
% Dry matter	97.57
% Moisture content	2.43

Table 5: The Mean Plasma concentration of the test fish and control fish fed with poultry hatchery waste for 12weeks.

Mean	T_0	T_1	T_2	T_3	T_4
Urea mg/dl	31.70±3.15	17.96±1.51	13.45±1.01	14.16±1.09	22.05±3.27
Total protein g/l	37.58±2.52	38.38±5.48	39.26±2.41	29.98±4.00	28.16±3.31
Creatinine mg/dl	1.18±0.01	0.50±0.01	0.58±0.01	0.84±0.01	1.03±0.01
Triglyceride mg/dl	101.40±15.30	129.03±11.43	163.90±11.39	180.46±13.24	131.18±11.63
Alkaline phosphatase mg/dl	57.22±1.38	80.55±1.12	82.13±0.90	67.44±0.92	128.31±0.98
Cholesterol mg/dl	237.68±7.78	261.63±11.06	279.74±10.58	245.48±11.29	278.39±13.83

Table 6: Haematological parameters of the test fish and control fish fed with poultry hatchery waste for 12 weeks

Sample	Hbg/l	PCV g/l	RBC $x10^{12}$(T)	WBC $x10^{9}$(G)	MCH $x10^{-12}$(pg)	MCV$x10^{-15}$(Fe)	MCHC g/l
T_0	125.33±5.03	0.37±0.01	1.9±0.1	22.33±2.52	66.14±5.38	198.66±16.44	332.74±1.02
T_1	108.66±18.58	0.32±0.05	1.7±0.4	20±2	65.09±10.59	195±31.22	332.56±1.32
T_2	105±11.53	0.31±0.03	1.5±0.3	24±2.64	72.44±18.81	218±0.57	331.60±0.47
T_3	94±19.28	0.28±0.05	1.8±0.2	18.33±3.51	51.92±5.37	156.33±16.44	331.83±1.33
T_4	98.66±5.13	0.29±0.01	1.83±0.05	23.66±2.51	53.83±2.96	161.33±9.29	332.57±0.65

percentages, it comprises of Crude protein (21.44), Ash (50.00), Moisture content (2.43), Ester extract (12.00), Dry matter (97.57) and Crude fibre (2.00). Table 5 Shows the Mean Plasma concentration of the test fish and control fish fed with poultry hatchery waste for 12weeks.While Table 6 shows the haematological concentration of the test fishes and control fishes fed with hatchery poultry waste.

DISCUSSION

The feeding trials revealed that *Clarias gariepinus* responded to all the diets, irrespective of their composition. *Clarias gariepinus* was able to effectively utilize the poultry hatchery waste for growth. It is interesting to note that better growth and nutrient utilization were achieved at relatively low inclusion level of poultry hatchery waste compared to high incorporation of the test ingredient.

The result of the study showed that there was a slight decrease in the values of haematological parameters of the *Clarias gariepinus* fed with poultry hatchery waste compared to those fed with compounded feed. This is in agreement with the reported effect of toxicant on blood parameters in freshwater teleost fish *Clarias batrachus* [15]. Haemoglobin and parked cell volume (PCV) have been suggested as tests that can be carried out on routine basis in fish hatchery as a check on health status [20]. The increase that was observed in the haematological parameters observed in the control fish fed with normal diet is in collaboration with the findings of [9] that survival of fish can be correlated with increase in antibody production which helps in the survival and recovery. The reduction that was observed in the haematological parameters is indicative of blood loss from fish fed with poultry hatchery waste compared to fish fed with normal feed. The values obtained in this report for fish fed with poultry hatchery

waste are lower than those reported in the literature for the African catfish [21, 22].

Haematological characteristics have been widely used in clinical diagnosis of diseases and pathologies of human and domestic animals. The applications of haematological techniques have proved valuable for fishery biologists in assessing the health of fish [1] and monitoring stress response [23]. Some of the values are slightly low due to the condition under which the fishes were kept, the condition based on the fact that the fishes are not in their natural habitat and also because of the small sizes of the fishes, values such as the erythrocyte values, RBC values and Haemoglobin values. In a stress situation, erythrocyte count is one of the first parameters that is affected.

Increase in WBC (leucopomia) as observed in the fish fed with compounded feed is attributed to increase in production of leucocytes in the haematopoietic tissue of the kidney and perhaps the spleen. Lymphocytes are the most numerous cells comprising the leucocytes, which function in the production of antibodies and chemical substances serving as defense against infection. The primary consequence of observed changes in leucocyte count in stressed fish is suppression of the immune system and increased susceptibility to disease [24].

Decrease in RBC and haemoglobin values in *Clarias garipienus* fed with poultry hatchery waste in this study is similar to the observations conducted by Joshi *et al.*; Gill and Pant [10, 25] in *Clarias batrachus* exposed to different toxicants. Sampathy *et al.* [26] also reported a decrease in haematological parameters of *Oreochromis mossambrius* exposed to copper and zinc. The anemia condition of fish fed poultry hatchery waste in this study may however be due to its protein inadequacy to meet the fish nutrient requirements, which might have inhibited erythrocyte production or increase rate of destruction. Low haemoglobin level according to Joshi *et al.* [10] might decrease the ability of fish to enhance its activity in order to meet occasional demands.

Plasma enzyme activities in fishes are considered to be a significant factor to assess the state of the liver and some other organs in fish [27]. In this study, attention was focused on alkaline phophatase which were observed by Verma and his coworkers [28] to promote gluconeogenesis from amino acids. The cholesterol and triglyceride values of *C. gariepinus* in this study were higher and almost in accordance with the observation by Omitoyin [5]. Further observed by McCarthy *et al.* [29] the value of blood cholesterol also changes depending on fish species.

Total protein values in this study are similar to those reported for *L. cephalus* by Yasar *et al.* [30]. The values of plasma excretory products of urea were similar to those reported by Faleye [31] for *C. mrigala* but slightly lower than those reported values in the literature [5]. This difference may be due to environmental condition of rearing facilities and handling.

The result of this present study therefore provides values for some haematological and plasma biochemical parameters for *C. gariepinus* fed with poultry hatchery waste which according to Klinger and his coworker [20] can be used to assess fish health.

CONCLUSION

In conclusion the present study revealed that feeding poultry hatchery waste as a supplementary feed to *Clarias gariepinus* created a slight decrease in the haematological paramaters of the specie.

Poultry hatchery waste has no negative impact on the health status of *Clarias gariepinus.* Therefore direct use of poultry hatchery waste as sole supplementary feed should be encouraged.

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