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Growth performance, haematological and biochemical study of *Clarias gariepinus* (Burshell) fingerlings fed formlulated *Rana galamensis* (Galam) meals

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Abstract

A 56-day feeding experiment involving Clarias gariepinus fingerlings was conducted to investigate the growth response, serum and biochemical parameters of the fingerlings to formulated feed from Rana galamensis at 0%, 10%, 20%, 30% and 40% inclusion levels. 180 fingerlings were divided into 5 treatment groups containing 36 fingerlings and each group was further replicated 3 times. A significant difference (P<0.05) was established in the growth parameters determined in the C. gariepinus fingerlings fed with the four frog formulations, namely, final weight, total weight, and mean feed gain, mean weight gain and feed conversion ratio The 40% R. galamensis ration elicited higher growth values than all other rations. Similarly, significance difference (P<0.05) was established in the serum (Total protein, Albumin and Cholestreol) and biochemical (Na^+. K^+ and HCO_3^-) parameters determined in the blood samples collected from the fingerlings fed with the four frog formulations. The increasing values observed in the growth factors, serum and biochemical parameters of the fingerlings when increasing level of R. galamensis protein was incorporated into the fish diet is an indication that the formulated feed can serve as growth promoter as well as rich sources of mineral supply to the fish; hence, a potential protein source for aquacultural practices.

Keywords: *Rana galamensis*, *Clarias gariepinus*, Haematological , Biochemical parameters

Introduction

The burgeoning human population in the developing countries of Africa, Asia, and South America has not only presented developmental challenges, it has also led to increasing demands on all agricultural sectors of their economies. This fact is alluded to by the growing demand for fish as food. Aquaculture as a subsector of agriculture has risen to this trend by rising so rapidly to becoming the fastest growing food-producing industry in the world. So promising is the growth that it is now estimated that by 2030, over half of the fish consumed by the world's people will be produced by aquaculture (FAO,2000).

In Nigeria, African catfish, *Clarias gariepinus* has grown to be one of the most cultured catfishes. Some of its aquacultural importance lie in its fast grow rate, very good organoleptic properties of the flesh, and the appreciation by consumers for the quality of its meat whether smoked or used as soups (Oladosu, 1994; Pruszynski, 2003).

Despite this encouraging picture in the growing profile of aquaculture, it is still faced with technical and economic drawbacks. Feed as one of the major inputs in aquaculture production with its high cost has been cited as one of the problems hampering aquacultural development. It had been reported that feed accounts for almost 60% of the total cost of fish production in Africa and it is more or less the determinant factor of viability and profitability of fish farming as a productive enterprise (Jamiu and Ayinla,2003).

Sourcing for non-conventional feed of animal origin comparable with the conventional types has been suggested as one of the ways of addressing high cost of feeds in aquacultural practice. Examples of such explorable alternatives are tadpole meal, maggots, earthworm meal, house fly larva, etc (Anunne, 1990; Faturoti *et al*., 1998; Ugwumba and Abumoye, 1998; Ugwumba *et al*., 2001; Akinwande *et al*., 2002; Ibiyo and Olowosegun, 2004).

In Nigeria, *Rana galamensis* is found in many states such as Lagos, Ogun, Oyo, Kwara, Osun, Ondo, Ekiti, Kaduna and Benin City (Walker, 1967). On a sub-regional note, the consumption of *R. galamensis* had been reported in various West Africa countries (Mohneke *et al.,* 2010). The high acceptability and commercial status of this animal protein is not surprising, since the meat has an appreciable large size and is available in large quantities. Its appreciable content of amino acids serve as key intermediates in cellular metabolism (Murray *et al.,* 2000).

Although, blood variables most known to be influenced by the diet type are the red blood cell, RBC, packed cell volume, PCV, PP and glucose levels, the nutritional state of a fish has also been documented to influence the animal health and possibly way they deal with stress (Ologhobo,. 1992; Hemre *et al*. , 1996). Hence, the aim of this study was to investigate the growth performance, serum and biochemical parameters of *C. gariepinus* fingerlings fed with different inclusions levels of meals formulated from *R* . *galamensis*.

Materials and methods

Experimental design

The experiment which lasted for 56 days was carried out in the research laboratory of the Fishery Unit of Department of Pure and Applied Biology Department, Ladoke Akintola University of Technology, Ogbomoso (8^0 8′N - $8^0$14 and $4^0$17′E), Oyo State, Nigeria. Fingerlings of *Clarias gariepinus* were procured from the Fisheries unit of the Ministry of Agriculture and Natural resources, Ogbomoso, Oyo State, Nigeria. The fingerlings were transported in well aerated polythene bags to the laboratory where they were allowed to acclimatize to laboratory conditions for 2 weeks; during the period of acclimatization, the fingerlings were fed with commercial floating pellets at 10% of body weight. Five bowls, each of $0.05m^3$ capacity was used for the experiment. Prior to usage, each bowl was washed with 5% acetic acid to remove possible toxic materials. Unconsumed food materials were removed and water replenished regularly at 24 hour intervals.

In line with Oyelese and Faturoti (1995), water temperature and water pH were measured every 24 hours. Each of the five bowls was stocked with 20 fingerlings and fed with the formulated feeds for 56 days in line with recommended standard (FAO, 1986). Aeration was facilitated with the use of aerator pumps for constant supply of oxygen. The main ingredients used for

formulation were: Dried processed *Rana galamensis*, *Zea mays*, Pure fish meal, *Arachis hypogea,* and *Glycine max.* Pearson's square method (1988) was used to prepare 40% crude protein of *R. galamensis* and from this four inclusion levels (10 %, 20 %, 30 % and 40 %) were produced. Feeds were made into pellets, crushed into smaller sizes and slowly spread on the water surface. The fingerlings were fed twice daily at 0700hrs and 1800hrs respectively. 5 % of the body weight was the equivalent amount of feed used to feed the fingerlings.

Initial weights of *C. gariepinus* fingerlings were determined before the commencement of the experiment. The growth response of the fingerlings to each feed formulation was taken on a weekly basis using Sautex weighing balance (Model 400). The weights taken in gram (g) was done every 7 days. The temperature of the water was determined using the mercury glass thermometer .The temperature of the water was determined on daily basis, while weekly and mean weekly temperature were also recorded. Similarly, the pH of the bore hole water used was taken on a daily basis and recorded using pH meter.

For hematological analysis, blood samples were taken from the fish through the caudal circulation with the aid of heparinized 2 cm^3 disposable syringe and a 27 guage disposable hypodermic needles. Standard laboratory assay procedures were used in determining both the haematological and biochemical parameters namely namely Glucose, Total protein, Albumin, Cholesterol. Na+, k+, Cl- and HCO_3^-. Total Plasma Protein was determined using cyanmetheamoglobin method (Darcie and Lewis, 1984). The cholesterol concentration was analysed with a modified method of Hogendoom (1979) while the Albumin concentration was analysed using the Bromocresol green method according to Doumas *et al* (1971). The mineral elements namely Na+, k+, Cl- and HCO_3^-. determined by first by dissolving 0.5g of the R. *galamensi* ash in 100ml of Aqua regia and this was placed on hot plate for 2 hours to digest until clear solution was obtained. This solution was filtered through an acid-washed Whatman No 1 filter paper and the filtrate was made up to 100ml with distilled water. The samples from the stock were taken and analysed for the presence Na+. K+ and HCO_3^- using Lead using Elmer Pelkin 3100 atomic absorption spectrophotometer

As for biochemical components, Data obtained were subjected to analysis of variance (Steel and Torrie, 1980) and the significance between means were separated using Duncan's Multiple Range Test at 5 % level of probability.

Results and discussion

The physico-chemical parameters of the water used during the investigation and the temperature condition obtained are shown in Table 1. The mean temperature obtained was 24.8^0C while the mean pH was 7.3. Both conditions were within the range recommended for effective fish culture (Viveen *et al*., 1986).

The growth response of the fingerlings of *Clarias gariepinus* fed with the four feed formulations prepared from *Rana galamensis* is presented in Table 1. Significant difference ($P<0.05$) was established in each of the six growth parameters measured. With the exception of average weight gain, the noted trend was increase in the value of the growth parameters upward from 10% to 40%. It is evident from the work that 40% *R.*

galamensis ration elicited higher growth values than the remaining lower rations. Furthermore on a comparative note, 40% *R. galamensis* ration posted higher growth value than the conventional feed (control). No significant difference was obtained in the Feed Convertion Ratio (FCR) by *C. gariepinus* fingerling fed with the four formulated feeds; however, the (FCR) of the conventional feed was greater than that of any of the formulated feeds.

Table 2 shows the biochemical composition of the blood of *Clarias gariepinus* fed with increasing level of high *R. galamensis* meal. From the table, it can be seen that the mineral composition values obtained showed increasing values from 10% to 40% rations. In each of the four biochemical components determined (Na^+, K^+ , Cl^- and HCO_3^-), significant difference ($P<0.05$) was established in each of the four feed formulations investigated.

Table 3 shows the haematological profile of four parameters taken from the blood of *Clarias gariepinus* fed with four compounded meals from *R. galamensis*. From the table, siqnificant difference ($P<0.05$) was established in the glucose level of the blood of the fingerlings fed with 10% - 40% *R. galamensis* meals. The level of glucose in the blood of the fingerlings fed with lower levels of the formulated meals 10% and 20% were significantly lower than the levels found in 30% and 40%. There was a significant difference ($P<0.05\%$) in the total protein content of the blood of *C. gariepinus* fed with four *R. galamensis* meals. The total protein content was increasing with increasing level of *R. galamensis* in the food rations. It is instructive to note that the total protein content in the blood of the fingerlings fed with *R. galamensis* were in all cases greater than that of the control meal. No definite trend was obtained in the albumin level in the blood of the fingerlings fed with *R. galamensis* meals. However, albumin level in the blood of *C. gariepinus* fed with the conventional feed was lower than the values obtained from blood of fingerlings fed with the compounded meals. The cholesterol levels of blood of *C. gariepinus* fed with 10% - 30% compounded feeds of *R. galamensis* were not significantly different ($P>0.05$); however, the three meals elicited cholesterol level significantly lower ($P<0.05$) than the value obtained under 40% *R. galamensis* inclusion. The cholesterol value found in the fingerlings fed with the conventional meal was significantly lower ($P<0.05$) than each of the values obtained under 10%-40% *R. galamensis* inclusions.

In five out of six parameters used to measure growth performance of *C. gariepinus* fingerlings fed with the compounded feeds from *R. galamensis* protein source, 40% *R. galamensis* inclusion posted the highest growth values. That protein component of 40% *R. galamensis* inclusion was more efficiently utilized by *C. gariepinus* fingerlings at 40% inclusion level than other levels of inclusions is in tandem with results obtained by Degani *et al*., (1989) on *C. lazera*. The noticeable increase in growth factors with increasing protein levels of *R. galamensis* inclusion noted in this work is comparable to the observations of Faturoti *et al*., (1986) on *C. lazera* fingerlings, Obasa and Faturoti (2000) on *Chrysichthys walkeri* fingerlings and Erondu *et al*., (2006) on *C. nigrodigitatus*.

The low food conversion ratio values obtained in all the compounded meals form *R. galamensis* as compared to the conventional feed is indicative of the capability of *C. gariepinus* fingerlings to

accept and utilize the compounded meals at all levels of inclusion (Fagbenro *et al*., (1986).

Significance difference ($P<0.05$) was obtained in both the biochemical and haematological parameters determined in the blood samples of *C. gariepinus* fed with four formulated meals of *R. galamensis*. Comparable values of the concentration of Na+ was found in the blood samples collected from *C. gariepinus* fingerlings fed with three out of the four formulated feeds (10%, 20% and 40%)all of which posted significantly higher values than the level found in 30% food ration. Similarly, significance difference ($P<0.05$) was established in the K+ level found in the blood samples of fingerlings fed with the four formulated meals; it was observed that, K+ level in the blood samples of *C. gariepinus* fingerlings fed with 30% and 40% *R. galamensis* were higher than in other food rations and also than the value of the control.

Similar trend to the above concentrations (Na^+ and K^+) was repeated in Cl^- and HCO_3^- parameters; higher concentrations of food formulation of *R. galamensis* posting higher levels of the biochemical parameters that were found to be even higher than the levels found in the control diet.

Significance difference ($P<0.05$) were also established in each of the four haematological parameters measured in the blood samples collected from *C. gariepinus* fingerlings fed with increasing level of *R. galamensis* meal inclusion. Although, the levels of glucose in the blood samples of *C. gariepinus* fingerlings fed with both 30% and 40% formulated meals of *R. galamensis* were not significantly different ($P>0.05$), each of the parameters posted glucose concentrations were significantly higher than the values obtained in 10%, 20% and also in the control. Similar trend to the above was observed in other three haematological parameters (Total protein, Albumin and Cholestreol) determined in the blood samples collected from *C. gariepinus* fingerlings fed with four formulated *R. galamensis* meal diets.

It is evident from this work that increasing level of *R. galamensis* inclusion as a protein source in the diets of *C. gariepinus* fingerlings promoted better growth response which is shown by higher growth values elicited in the fish. Consequently, in five out of six growth parameters measured, 40% feed inclusion recorded higher values (even when control feed was counted). This result is a confirmation of the acceptability of *R. galamensis* as a possible protein source for feed compounding in aquacultural practices. This finding is in tandem with works done on the dietary potentials of *R. galamensis* which had been confirmed to include low fat, high protein and mineral sources (Oloyede and Fowomola, 2003).

Table 1: Weekly mean values of some selected physical parameters

Week/Parameter	1st week	2nd week	3rd week	4th week	5th week	6th week	7th week	8th week	Mean
Temperature	24^0 C	24^0 C	25^0 C	25^{0-} C	25^0 C	25^0 C	25^0 C	25^0 C	24.8^0 C
pH	7.4	7.4	7.3	7.3	7.3	7.3	7.3	7.3	7.3

Table 2: Growth performance of *Clarias gariepinus* fingerlings fed with formulated diets of *Rana galamensis*

PARAMETERS	10%	20%	30%	40%	0%	SEM
INITIAL WEIGHT	46.68^{b}	44.65^{b}	45.60^{a}	47.90^{a}	47.05^{ab}	0.45
Final weight	119.25^{b}	192.50^{ab}	193.65^{ab}	231.08^{a}	135.25^{b}	15.60
Total weight gain	71.65^{c}	146.60^{a}	148.50^{b}	183.10^{a}	36.20^{d}	18.15
Average feed gain	12.75^{c}	20.90^{b}	21.50^{b}	25.65^{a}	8.95^{d}	2.05
Average weight initiated	24.40^{c}	39.05^{b}	40.90^{b}	47.95^{a}	22.20^{a}	3.33
Feed conversion ratio	1.91^{b}	1.87^{b}	1.87^{b}	1.90^{b}	2.48^{cb}	0.08

^{a,b,c,d} *Means along the same row with different superscripts differ significantly ($p<0.05$)*

Table 3. Some biochemical parameters of *Clarias gariepinus* fingerlings fed with formulated diets of *Rana galamensis*

PARAMETERS	10%	20%	30%	40%	0%	SEM
Na^{+} (mmol/dL)	140.50^{bc}	140.50^{bc}	142.50^{a}	144.50^{c}	139.00^{c}	2.12
K^{+} (mmol/dL)	7.00^{c}	7.25^{c}	7.55^{bc}	7.75^{a}	6.95^{c}	0.34
CL^{-} (mmol/dL)	106.50^{b}	106.50^{b}	111.50^{a}	117.50^{a}	105.00^{b}	5.57
HCO_3^{-} (mmol/dL)	21.50^{ab}	20.50^{bc}	22.50^{ab}	23.50^{a}	19.00^{c}	21.40

^{a,b,c,d} *Means along the same row with different superscripts differ significantly ($p<0.05$)*

Table 4. Four serum parameters of *Clarias gariepinus* fingerlings fed with formulated diets of *Rana galamensis*

PARAMETERS	10%	20%	30%	40%	0%	SEM
GLUCOSE (g/dL)	23.50^{b}	26.00^{b}	29.00^{a}	30.50^{a}	20.50^{c}	3.90
TOTAL PROTEIN(g/dL)	3.90^{cd}	4.15^{bc}	4.40^{b}	4.85^{a}	3.85^{d}	0.39
ALBUMIN (g/dL)	2.55^{a}	2.20^{b}	2.30^{ab}	2.55^{a}	2.05^{b}	0.22
CHOLESTROL (mg/dL)	96.00^{b}	95.00^{b}	97.50^{b}	101.50^{a}	91.00^{c}	3.77

^{a,b,c,d} *Means along the same row with different superscripts differ significantly ($p>0.05$)*

Conclusion

Haematological and biochemical profiles of blood are known used to be useful in providing important information on health conditions of the organisms like fish (Hrubec *et al*., 2000). Consequent upon this, these two parameters are affected by such factors like species, the environment, diet, age, nutrition, maturation, etc (Regost *et al*.,2001). The increasing values of these two parameters in the blood of *C. gariepinus* with increasing level of *R. galamensis* diet is a pointer to the fact that the proteinous formulated feed can adequately serve as rich sources of mineral supply to the fish and is therefore a potential source for promoting aquacultural practices.

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