

Journal of Agriculture and Ecology Research International 11(2): 1-7, 2017; Article no.JAERI.31639 ISSN: 2394-1073



SCIENCEDOMAIN international www.sciencedomain.org

# Growth Performance of African Catfish Fed Diet Supplemented with *Gossypium herbaceum* Powder as Dietary Feed Additive

# Olarinke Victoria Adeniyi<sup>1\*</sup> and Abdulkabir Oluwatosin Lawal<sup>1</sup>

<sup>1</sup>Department of Animal Production, Fisheries and Aquaculture, Kwara State University, Malete, Nigeria.

# Authors' contributions

This work was carried out in collaboration between the two authors. Author OVA designed the study, wrote the protocol and the first draft of the manuscript. Authors OVA and AOL managed the analyses of the study. Author OVA managed the literature searches. Both authors read and approved the final manuscript.

# Article Information

DOI: 10.9734/JAERI/2017/31639 <u>Editor(s):</u> (1) Ahmed Esmat Abdel Moneim, Department of Zoology, Helwan University, Egypt And Institute of Biomedical Research Center, University of Granada, Spain. (1) Magdy Mohamed Aly Gaber, National Institute of Oceanography and Fisheries, Cairo, Egypt. (2) Carlos Henrique dos Anjos dos Santos, Instituto Nacional de Pesquisas da Amazônia, Brazil. (3) Kwasi Adu Obirikorang, KNUST, Ghana. Complete Peer review History: <u>http://www.sciencedomain.org/review-history/18191</u>

**Original Research Article** 

Received 17<sup>th</sup> January 2017 Accepted 27<sup>th</sup> February 2017 Published 15<sup>th</sup> March 2017

# ABSTRACT

Aim: To assess the growth performance of *Clarias gariepinus* fed diets containing cotton leaf powder and the control diets.

Study Design: Completely Randomized Design (CRD).

**Place and Duration of the Study:** Department of Animal Production, Fisheries and Aquaculture, Kwara State University, Malete, Nigeria, between January and April 2015.

**Methodology:** Two hundred and twenty five (225) *Clarias gariepinus* fingerlings of average initial body weight  $3.56 \pm 0.03$  g were randomly distributed into the 50 litre tanks filled with borehole water up to  $3/4^{\text{th}}$  in a static renewal system. The fish were fed diets supplemented with 0.5%, 1.0% and 1.5% air-dried cotton leaf, 0.03% oxytetracycline (positive control) and 0.0% (negative control) diets in triplicates for 12 weeks at 5% body weight. Weight Gain (WG), Relative Growth Rate

\*Corresponding author: E-mail: adeniyiovic@yahoo.com, olarinke.adeniyi@kwasu.edu.ng;

(RGR), Feed Conversion Ratio (FCR), Protein Efficiency Ratio (PER), and Specific Growth Rate (SGR), Condition Factor (CF), Hepatosomatic Index (HI), Gonadosomatic Index (GI) and Speensomatic Index (SI) were calculated. Data were analyzed using ANOVA at P = .05.

**Results:** *C. gariepinus* fed diets containing cotton leaf exhibited better WG, RGR, SGR, FCR and PER than the control diets. Fish fed diets containing 1.0-1.5% air-dried cotton leaf powder had significantly higher (P < .05) WG and lower FCR than those fed the negative control diet but did not differ significantly from oxytetracycline-treated group. SGR and PER showed the same pattern among the dietary treatments as that of the WG. However, the SGR, CF and HI were not significantly affected by the five experimental treatments. Supplementing the diets of *C. gariepinus* with cotton leaf did not affect CF, HI, GI and SI significantly (P < .05) when compared to the negative control diet.

**Conclusion:** Supplementing the diets of *Clarias gariepinus* with 1.0-1.5% air-dried cotton leaf powder improved the growth performance and nutrient utilization as oxytetracycline used in this study.

Keywords: Clarias gariepinus; oxytetracycline; cotton leaf powder; growth performance; organ indices.

# 1. INTRODUCTION

In aquaculture and livestock industries synthetic antibiotics are used worldwide to improve production, prevent or treat diseases with desired results. However, the use of synthetic antibiotics in these sectors is under serious scrutiny globally. The indiscriminate use of synthetic antibiotics have been reported to promote development of resistant pathogen strains, suppression of the immune system of the animal, destabilization of beneficial gut microflora, environmental pollution and risks of chemical residues in fish food [1,2,3]. The most common route for the delivery of antibiotics to fish occurs through mixing the drugs with specially formulated feed. The European Union has banned some of these drugs due to its adverse effects. Oxytetracycline is one of the few antibiotics that are authorized in United States of America for use in aquaculture [4].

It is therefore of necessity to search for alternatives to the synthetic antibiotics. One of such alternatives is phytobiotics, which are specially prepared parts of plants or their extracts containing active ingredients. Natural products, especially of organic origin, are a continuing source of novel active metabolites with immense impact in modern medicine globally. Plant derived substances have recently become of great interest in aquaculture and livestock industries due to their versatile application. Medicinal plants are the richest bio-resource of drugs of traditional system of medicine, modern medicines, nutriceuticals, food supplements, and folk medicines, pharmaceutical intermediate and chemical entities for synthetic drugs [5].

A number of studies have revealed the beneficial effects of using phytobiotics in fish production. It has been demonstrated that phytogenics act as growth promoters, immunostimulants as well as chemotherapeutic agents in fish production [6-12]. Cotton is a tropical plant which belongs to the family Malvaceae and the genus Gossypium. The genus comprises of about 52 species of which four are cultivated: These include G. hirsutum. G. barbadense. G. arboretum and G. herbaceum [13]. G. herbaceum is widely cultivated in Nigeria for its cotton lint and medicinal purpose. Cotton leaf is widely used by Nigerians for the treatment of diarrheal diseases. Cotton seed meal is used in live stock production, especially in ruminants. Antibacterial property of cotton leaf extracts against some human pathogens was reported [14]. The use of cotton leaf in traditional medicine, owing to its antimicrobial property, makes it a possible ingredient to be tested in vivo in animal production. There is paucity of information on the use of cotton leaf in live stocks and fish production. Therefore, this study was carried out to assess the growth performance of Clarias gariepinus diets supplemented with air-dried cotton leaf powder compared to oxytetracycline.

# 2. MATERIALS AND METHODS

# 2.1 Fish and Experimental Condition

African catfish, *Clarias gariepinus*, fingerlings (initial body weight 3.55±0.03 g) were purchased from a reputable hatchery in Ilorin, Kwara state, Nigeria. The fish were transported to Teaching and Research Farm of College of Agriculture, Kwara State University, Malete, Nigeria, where they were acclimatized for two weeks. They were fed with commercial diet during acclimatization. Thereafter, 225 fingerlings were sorted, randomly selected, weighed, and stocked into the 50 litre tanks filled with borehole water up to 3/4<sup>th</sup> in a static renewal system. The culture water was completely renewed every 3 days. Each treatment is assigned fifteen [15] fingerlings per tank in triplicates. The fish were not fed 24 hours before been placed on the experimental diet to empty their gut and increase their appetite and reception for the experimental diets.

# 2.2 Preparation of Experimental Diet and Feeding

Fresh leaves of cotton was obtained from llorin, cleaned, air-dried and ground into fine powder with blender. 40% CP isonitrogenous and isocaloric basal diets were formulated using fish meal, soybean meal, groundnut cake meal, corn meal as main ingredients as shown in Table 1.

Diet 1 (negative control) did not contain any of the test ingredients while Diet 2 (positive oxytetracycline contained 0.03% control) antibiotic); (synthetic Diet 3-5 were supplemented with 0.5%, 1.0% and 1.5% airdried cotton leaf powder respectively. All the main ingredients and feed additives were weighed using ohaus scale (scout pro, 400 g maximum capacity and 0.01 g resolution, Ohaus Corporation, USA), homogenized and bind with a mixture of starch and hot water. The feeds were then pelletized using 1 mm die, sundried, packed in polythene bag, labelled and stored in dry place during usage. The fish were fed daily at 5% body weight; The daily feed ration was divided into two equal portions, half of which was offered in the morning hours (between 08.30 h - 09.00 h) and the second half in the evening hours (between 16.30 h and 17.00 h). Four fish were randomly sampled from each replicate and weighed fortnightly to assess fish growth and adjust the feed rations.

Ingredients	Diets (% inclusion levels)						
-	Diet 1 (0.0)	Diet 2 (0.03 OTC)	Diet 3 (0.5)	Diet 4 (1.0)	Diet 5 (1.5)		
Fish meal	24.20	24.20	24.20	24.20	24.20		
Ground nut cake	24.30	24.30	24.30	24.03	24.30		
Soybean meal	24.00	24.00	24.00	24.00	24.00		
Corn meal	19.00	19.00	18.50	18.00	17.50		
Soybean oil	1.50	1.50	1.50	1.50	1.50		
Dicalcium	1.00	1.00	1.00	1.00	1.00		
phosphate							
Premix*	2.00	2.00	2.00	2.00	2.00		
Table salt	0.50	0.50	0.50	0.50	0.50		
Starch	3.50	3.47	3.50	3.50	3.50		
Oxytetrcycline	-	0.03	-	-	-		
Cotton leaf**	-	-	0.50	1.00	1.50		
Total	100.00	100.00	100.00	100.00	100.00		
Proximate compo	sition						
Moisture	6.53	6.55	6.53	6.56	6.53		
Crude protein	40.63	40.56	40.76	40.88	40.96		
Ether extract	6.63	6.62	6.66	6.69	6.71		
Crude fibre	10.09	10.09	10.11	10.23	10.36		
Ash	11.54	11.53	11.52	11.61	11.76		
Nitrogen free	24.58	24.65	24.42	24.03	23.68		
ovtract							

 Table 1. Ingredients and proximate composition (% dry matter) of experimental diets fed to

 Clarias gariepinus for 12 weeks

\*Vitamin and Minerals: Vitamin A –10,000,000 I.U.; D3- 2,000,000 I.U.; E –23,000mg; K3 – 2,000 mg; B1 – 3,000 mg; B2-6,000 mg; Niacin– 50,000 mg; Calcium Pathonate – 10,000 mg; B6 – 5,000 mg; B12- 25.0 mg; Folic acid

1,000 mg; Biotin- 50.0 mg; Choline chloride – 400,000 mg; Manganese – 120,000 mg; Iron- 100,000 mg;

Copper– 8,500 mg; lodine – 1,500 mg; Cobalt-300 mg; Selenium-120 mg; Antioxidant 120,000 mg. \*\*Moisture content – 9.90; Crude protein – 19.15; Ether extract – 11.64; Crude fibre – 8.6, Ash – 10.00;

Nitrogen free extract – 40.71; OTC = Oxytetracycline

#### 2.3 Chemical Analyses

The proximate composition of cotton leaves and experimental diets were analyzed using the standard method of AOAC [15]. Qualitative screening of some phytochemical components of cotton leaves aqueous and ethanol extracts was also determined [16,17].

#### 2.4 Fish Performance

At the end of the feeding trial the following growth performance and nutrient utilization indices were calculated:

Weight Gain (WG) = Final body weight – Initial body weight.

Relative Growth Rate (RGR) = 100 x WG / Mean initial weight.

Feed Conversion Ratio (FCR) = Feed intake / Weight gain.

Protein Efficiency Ratio (PER) = WG / Protein fed.

SGR = 100 x Log (mean final weight) – Log (mean initial weight) (g) / Time of experiment (days).

Where SGR = Specific Growth Rate.

#### 2.5 Determination of Biological Indices

At the end of the feeding trial, the weight and standard length of four fish sampled for final weight were taken and then sacrificed on ice. The abdominal cavities of the fish were opened to remove the liver, gonad and the spleen, which were weighed to determine the following indices:

Condition Factor (CF) =  $100 \times \text{Wean}$  final weight / Mean final standard lenght<sup>3</sup>.

Hepatosomatic Index (HI) = Weight of liver / Weight of fish.

Gonadosomatic Index (GI) = Weight of gonad / Weight of fish.

Speen-somatic Index (SI) = Weight of spleen / Weight of fish.

#### 2.6 Statistical Analysis

One-way Analysis of Variance (ANOVA) was used to analyze the data and Duncan multiple

range tests was used to compare differences among means at 5% probability level using statistical software SAS (Statistical Analysis System, 2010).

# 3. RESULTS AND DISCUSSION

#### 3.1 Phytochemical Components of Cotton Leaf

The results on the qualitative screening of phytochemicals in both the aqueous and ethanol extracts of cotton leaf showed presence of saponin, tannin, terpenoids and flavonoids.

#### 3.2 Effects of the Experimental Diets on Growth Performance and Nutrient Utilization of *C. gariepinus*

Weight Gain (WG), Feed Intake (FI), FCR, Protein Intake (PI), Protein Efficiency Ratio (PER), Relative Growth rate (RGR) and Specific Growth Rate (SGR) are shown in Table 2. C. gariepinus fed Diets 4 and 5 containing air-dried cotton leaf powder  $\geq$  1.0% had significantly higher (P < .05) WG than those fed Diets 1 (negative control) without cotton leaf. The values of the WG of 1.0 and 1.5% groups were also significantly better than fish fed Diets 2 and 3, containing 0.03% oxytetracycline (OTC) and 0.5% cotton leaf powder respectively. The FI, PI and PER showed the same pattern among the dietary treatments as that of the WG. Fish fed 1cotton leaf-treated diets exhibited 1.5% significantly lower (P < .05) FCR than 0.0% diet, but the FCR did not differ significantly from the OTC-treated diet. However, the SGR were not significantly affected by the five experimental treatments. There was progressive increase in the RGR of the fish as the rate of inclusion of cotton leaf powder increases, though there was slight reduction with fish fed diet 5 but without any significant difference.

# 3.3 Effects of the Experimental Diets on Biological Indices of *C. gariepinus*

Table 3 shows Condition Factor (CF), Hepatosomatic Index (HI), Gonadosomatic Index (GI) and Speen-somatic Index (SI) of *C. gariepinus* fed different dietary levels of air-dried cotton leaf powder compared to control diets. Supplementing the diets of *C. gariepinus* with the air-dried cotton leaf powder did not affect the CF, HI and GI of the fish significantly (P > .05) compared to the control groups. Increasing the

Parameters	Diets (% inclusion levels) S					
	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	
	1(0.0)	(0.03 OTC)	(0.5CL)	(1.0CL)	(1.5CL)	
IBW (g)	3.55	3.57	3.57	3.57	3.57	0.03
FBW (g)	33.00 <sup>b</sup>	36.51 <sup>ab</sup>	37.03 <sup>ab</sup>	41.19 <sup>a</sup>	39.82 <sup>a</sup>	1.59
WG (g)	29.47 <sup>b</sup>	33.00 <sup>ab</sup>	33.47 <sup>ab</sup>	37.59 <sup>a</sup>	36.26 <sup>a</sup>	1.58
RGR (%)	834.49 <sup>b</sup>	925.00 <sup>ab</sup>	938.00 <sup>ab</sup>	1054.40 <sup>a</sup>	1015.30 <sup>a</sup>	44.62
FI (g)	47.66 <sup>b</sup>	51.54 <sup>ab</sup>	50.27 <sup>ab</sup>	52.85 <sup>a</sup>	51.97 <sup>a</sup>	1.23
FCR	1.62 <sup>a</sup>	1.54 <sup>ab</sup>	1.51 <sup>ab</sup>	1.41 <sup>b</sup>	1.44 <sup>b</sup>	0.05
PI (g/fish)	19.03 <sup>b</sup>	20.29 <sup>ab</sup>	20.11 <sup>ab</sup>	21.14 <sup>a</sup>	20.76 <sup>a</sup>	0.48
PER	1.54 <sup>b</sup>	1.63 <sup>ab</sup>	1.66 <sup>ab</sup>	1.78 <sup>a</sup>	1.74 <sup>a</sup>	0.05
SGR (%/day)	1.14	1.15	1.14	1.26	1.24	0.05

Table 2. Growth performance and nutrient utilization of *Clarias gariepinus* fed diets supplemented with cotton leaves compared to oxytetracycline for 12 weeks

Means along the same row with the same superscript are not significantly different (P < .05) OTC = Oxytetracycline; CL = Cotton Leaf; IBW = Initial Body Weight; FBW = Final body Weight; WG = Weight Gain; RGR = Relative Growth Rate; FI = Feed Intake; FCR = Feed Conversion Ratio; PER = Protein Efficiency Ratio; SGR= Specific Growth Rate

 Table 3. Biological indices of C. gariepinus fed varying levels of cotton leaf powder and the control diets

Diets (%)	CF	HI	GI	SI
1 (0)	1.0073 <sup>a</sup>	0.0127 <sup>a</sup>	0.0067 <sup>ab</sup>	0.0017 <sup>ab</sup>
2 (0.03 OTC)	1.0897 <sup>a</sup>	0.0080 <sup>a</sup>	0.0063 <sup>ab</sup>	0.0027 <sup>a</sup>
3 (0.5 CL)	0.9735 <sup>a</sup>	0.0100 <sup>a</sup>	0.0077 <sup>a</sup>	0.0023 <sup>a</sup>
4 (1.0 CL)	1.0910 <sup>a</sup>	0.0103 <sup>a</sup>	0.0073 <sup>ab</sup>	0.0017 <sup>ab</sup>
5 (1.5 CL)	1.1357 <sup>a</sup>	0.0150 <sup>a</sup>	0.0043 <sup>b</sup>	0.0010 <sup>b</sup>
SEM	0.1551	0.0023	0.0009	0.0003

Means along the same column with the same superscript are not significantly different (P < .05) OTC = Oxytetracycline; CL = Cotton Leaf; CF = Condition Factor; HI = Hepatosomatic Index; GI = Gonadosomatic Index; SI = Speen-somatic Index (SI)

dietary levels of the cotton leaf powder resulted in reduction of GI and SI with lowest significant (P < .05) value at 1.5% compared to 0.5% inclusion level. However, the lowest value of GI obtained did not differ significantly from the control groups while the value for SI differ significantly (P < 0.05) from OTC-treated group.

#### 3.4 Discussion

The results from this study demonstrated better growth performance and nutrient utilization of *Clarias gariepinus* fed the air-dried cotton leaf supplemented diets compared to the control groups. The efficiency of fish in converting feeds to flesh is usually assessed in livestock and fish nutritional study by determining the FCR and PER; The lower the value of FCR and higher the PER, the better for the farmer. The improved growth performance and nutrient utilization observed in this study could be attributed to presence of growth stimulants or phytochemical constituents (flavonoid, saponin, terpenoids) present in cotton leaf. These phytochemicals might have enhanced feed digestibility, nutrient absorption and utilization resulting to the increase in growth performance of the fish. Phytoadditives have been shown to enhance digestion as a result of increase production of bile acid, secretion of digestive enzymes and increase in villi absorption area [18,19]. The results of this study coincided with results of some other researchers demonstrating the beneficial effects of supplementing fish diets with phytobiotics. Significant increase in growth performance of C. gariepinus broodstocks fed diets supplemented with ethanolic extract of Garcinia kola at the rate of 0.25-2.0 g/kg inclusion levels have been reported [6]. The authors [6] similarly reported reduction in the growth performance of the experimental fish at 2.0 k/kg, the value of which was still better than the control diet. On the contrary, [7] did not observe significant differences (P<0.05) in weight gain, feed conversion ratio and specific growth rate of Oreochromis niloticus fed varying

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inclusion of Chinese chive, Allium teberosum, oil compared to 0.01% (100 mg/kg diet) oxytetracycline; although the author fed the fish for only two weeks. The body weight gain of Oreochromis niloticus fed diets supplemented with Echinacea purpurea and Allium sativum was significantly higher than those fed with diets treated with oxytetracycline and the negative control [8]. Better growth performance of C. gariepinus fed with diets supplemented with residue of walnut leaf and onion bulb [9], roselle [10], date palm seed [11] and fluted pumpkin leaf powder [12] were also reported.

The improved growth performance and lower FCR, though non-significant, of the fish fed with OTC-treated diet compared to the negative control diet coincided with the significant weight gain of *O. niloticus* fed with OTC-treated diets [20]. However, better growth performance obtained at 1.0-1.5% CL inclusion levels compared to the OTC group is an indication of suitability of cotton leaf at this level as alternative organic growth promoter in place of OTC.

Organ-somatic indices and condition factors are indicators of health status of fish [21] and so it could be used to predict health status of fish. The non-significant values obtained for biological indices and condition factor of fish fed diets treated with cotton leaf and negative control is an indication that cotton leaf used in this study is safe and might not have negative effect on the health status of the experimental fish. Similarly, there were insignificant differences in HI of Nile tilapia fed diets treated with garlic compared to the containing groups fed with diet chloramphenicol and negative control groups [1]. Similar observation on HI of sea bream fed with mixture of Massa mediate, Crataegi fructus, Artemisia capillaries and Cnidium officinale [22] and basil leaf and seed [23] were reported. Also there were no significant differences among the organ indices of fish fed residue of walnut leaf and onion bulb compare to those fed the control diet [24].

# 4. CONCLUSION

It could be concluded that cotton leafsupplemented diets enhanced the growth performance and nutrient utilization of *Clarias gariepinus* used in this study. Therefore, 1.0% cotton leaf powder is recommended for dietary additive in aquafeed. Further study could be carried out on the haematological and histopathological effects dietary cotton leaf on fish in order to evaluate its safety in aquaculture use.

# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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