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Research Article
Growth Performance of *Clarias gariepinus* Fed Crude Extracts of *Allium cepa*, *Moringa oliefera* and *Vernonia amygdalina*

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Abstract

Background and Objective: The need for food security has led to the prohibition of antibiotics used as growth promoter, to replace their effects, phytoadditives obtained from plant could be an alternative. Thus, this study was carried out in order to assess the growth performance of *Allium cepa*, *Moringa oliefera* and *Vernonia amygdalina* extracts on *Clarias gariepinus*. **Materials and Methods:** Leaves were shade-dried, ground and sieved, while the fresh *A. cepa* bulbs were ground. About 100 g of each plant material was soaked in 500 mL ethanol for 24 h with constant shaking at intervals, afterward it was filtered. A total of 300 *Clarias gariepinus* fingerlings of mean weight ranging between 3.73±0.35-4.63±0.32 g were obtained from a hatchery. Fingerlings were divided into 10 groups representing the treatments (Control, A1, A2, A3, M1, M2, M3, V1, V2 and V3). Each group consist of 30 fishes. Fingerlings were stocked in outdoor concrete tanks (measuring 2×2×1.5 m). Water parameters were monitored twice in a week. **Results:** The highest final mean weight and mean weight gained were observed in group of fish fed with 0.5 g kg^{-1} of *M. oliefera* extract (M1) with values of 44.22±3.89 and 39.72±3.30 g, respectively, while the least for both parameters were observed in group of fish fed with 1.5 g kg^{-1} of *A. cepa* (A3) with values of 29.44±4.26 and 25.48±1.93 g, respectively. At higher concentrations the 3 plant extracts exerted low growth response on *C. gariepinus*, while at low concentrations *A. cepa* and *M. oliefera* extracts exerted positive growth response on *C. gariepinus*. Statistical analysis showed significant difference when the highest mean weight gained was compared with the control and other groups. **Conclusion:** This study infers that, at low concentration *Allium cepa*, *Moringa oliefera* and *Vernonia amygdalina* has growth promoting effects, furthermore *Moringa oliefera* leaf and *Allium cepa* bulb had superior performance over the control diet. The use of phytoadditives to replace antibiotics has just begun, it is hoped that, further research on their long duration use in fish culture will further establish their potency.

Key words: *Clarias gariepinus* fingerlings, *Allium cepa* bulb, phytoadditives, *Moringa oliefera* leaf, plant extracts, fish, feed

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Hormones, antibiotics, vitamins and several synthetic products have been used as growth promoters, anti-bacterials and other purposes in mariculture, though they have been reported to have positive effects on fish and shrimps it has been observed that they cannot be recommended in commercial culture operations due to their residual effects in the muscles of fish and shrimps[1]. Furthermore, the growing tendency for food safety has led to the ban of antibiotics which has widely been used to enhanced growth. To replace their activity on growth there is need to investigate for natural, cheap and biodegradable alternatives. Phytoadditives are folder additive obtained from plant extract. Plant materials serve as storehouses for safer, cheaper and biodegradable chemicals[2]. It has also been found to contain an antioxidant a type of molecules that neutralizes harmful compounds called free radicals that damage living cells and spoil food[3-6].

Ethanolic extract of *Garcinia kola* (bitter kola) was reported to enhance the growth of African catfish, *Clarias gariepinus* with significant variations in the growth parameters and food conversion ratio[7]. Improved feed conversion rates were observed when garlic was added to grower-finisher pigs diet at the levels of 1 or 10 g kg^{-1} diet[8]. *Yucca schidigera* plant extracts has been found to improve growth, feed efficiency and health in ruminants[9]. Significant increase in weight was reported when *Allium cepa* was fed to broiler chicken[10,11]. Also *Vernonia amydalina* leaves has been used to enhance growth of broiler chicken[12].

Allium cepa, *Moringa oleifera* and *Vernonia amydalina* plant extracts have been reported to enhance growth in broiler chicken and tilapia. There are fewer work on their effects on the growth response of *Clarias gariepinus*. *Moringa oleifera* leaf meal and not the extract was used as substitute for fish meal authors reported poor growth response with the control group having the highest mean weight gained[13,14]. However, earlier work in 2016 reported appreciable growth response when *Moringa oleifera* leaf meal was fed to fish14 but no significant difference was recorded between the highest growth and the control group. Most work on *Allium cepa*, *Moringa oleifera* and *Vernonia amydalina* was at replacing fishmeal with the plant material, thus the need to study the effects of these phytoadditives (not as a substitute) on growth response of *Clarias gariepinus*.

MATERIALS AND METHODS

Study location: This study was carried out between the month of July 22nd to August 19th 2017 at the Fisheries and Hydrobiology Research unit, Department of Biological Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria. It lies on the geographical coordinates of 12°16'41"N, 4°27'6"E.

Acquisition and preparation of plant materials: The fresh bulb of *Allium cepa* were purchased from Aliero market. Fresh leaves of *Moringa oliefera* and *Vernonia amydalina*, were obtained within the University and Shade-dried. The dried leaves and bulb were ground with an electric blender. About 100 g of each ground plant materials were soaked in 500 mL ethanol for 24 h with constant shaking at intervals[15]. Afterwards each were filtered using Whatman No. 1 filter paper, the filtrate was concentrated using a water bath to obtain jelly-like extract.

Experimental design: A total of 300 *Clarias gariepinus* fingerlings of mean weight ranging between 3.73±0.35-4.63±0.32 g were obtained from the hatchery. Fingerlings were divided into 10 groups representing the treatments (Control, A1, A2, A3, M1, M2, M3, V1, V2 and V3). Each group consists of 30 fishes. Fingerlings were stocked in outdoor concrete tanks (measuring 2×2×1.5 m), they were fed 3% of their body weight. The feeding trails lasted for 28 days.

Preparation of experimental diets: A basal diet containing 40% crude protein was formulated with the following ingredients; Yellow maize, Groundnut cake, Soybean meal, Fish meal, Blood meal, Cassava starch (binder), Methionine and Vitamin/Mineral premix. All ingredients and the inclusion levels (0.5, 1.5 and 2.0 g kg^{-1} of diet) of the plant extracts were weighted mixed and pelleted using a pelleting machine. Each of the 4 experimental diets (per plant extract) was pelletized with a die of 1 mm in diameter, air dried, kept in labeled cellophane bags and stored in the refrigerator until when needed.

Determination of some physico-chemical parameters of cultures: The physico-chemical parameters which include; temperature, pH, alkalinity and dissolved oxygen were determined bi-weekly during study.

Determination of growth/survival rate: Average daily growth rate (ADG):

$$ADG = \frac{Wt\text{-}Wi}{t}$$

Specific growth rate (SGR %/day):

$$SGR = \frac{InW_f - InW_i}{t} \times 100$$

Where:

W_f = Final average weight at the end of the experiment
W_i = Initial weight at the beginning of the experiment
t = Culture period in days

Food conversion ratio (FCR): Leftover feeds were collected at interval of 2 weeks, dried in an oven at 50°C and weighed:

$$FCR = \frac{\text{Dry weight of diet (g)}}{\text{Total wet weight gain by fish (g)}}$$

Survival rate (%):

$$SR\ (\%) = \frac{N_i}{N_o} \times 100$$

Where:

N_i = Total number of fish at the end of the experiment
N_o = Total number of fish stocked at the beginning of the experiment

Determination of nutrient utilization

Protein efficiency ratio (PER):

$$\text{Protein efficiency ratio (PER)} = \frac{\text{Gain of test fish}}{\text{Protein consumed}}$$

Protein consumed is to be calculated as the difference between the quantity of feed fed and the leftover on dry matter basis.

Data collection: Weights gained, leftover feed and water parameters were taken weekly using the meter scale and meter rule.

Statistical analysis: The data obtained were analyzed using SPSS 18.0 a statistical software package for mean, standard deviation and one-way ANOVA to test for significant difference within the mean and student's t-test to test between two independent observations.

RESULTS AND DISCUSSION

Physico-chemical parameters of water used in culture: Data in Table 1 showed the physico-chemical parameters of water in each experimental tank. The water temperature in the experimental concrete tanks ranged between 25.00±1.0-27.00±1.0°C. There was no significant difference ($p \geq 0.05$) when the highest value was compared with the lowest value. The pH of the water used for the culture ranged between 6.63±0.32-7.04±.0.17. There was no significant difference ($p \geq 0.05$) across the groups. Dissolved oxygen content of water used for the culture ranged between 4.77±0.25-5.73±0.25 mg L^{-1}. Highest value was observed in tank containing fish fed with 0.5 g kg^{-1} of *V. amygdalina*, while the lowest value was observed in tank containing fish fed with 1.0 g kg^{-1} of *A. cepa*. There was variation ($p \leq 0.05$) when the highest value was compared with the lowest. However, other groups showed no variation when compared with the highest and lowest values obtained. Physico-chemical parameters of water used for the culture were within the range of values requiring for optimum fish production in tropical region[16].

Growth response of *C. gariepinus* fed with *A. cepa*, *M. oliefera* and *V. amygdalina* extract: Table 2 presented the growth response of *C. gariepinus* fed with *A. cepa*,

Table 1: Physico-chemical parameters of water used in culture

Groups	Temperature (°C)	pH	DO (mg L^{-1})
Control	25.50±0.50^{ab}	6.74±0.49^{a}	5.13±0.32^{ab}
A1 (0.5 g kg^{-1})	27.00±1.00^{b}	6.85±0.25^{a}	5.30±0.30^{ab}
A2 (1.0 g kg^{-1})	26.77±1.12^{ab}	6.77±0.35^{a}	4.77±0.25^{a}
A3 (1.5 g kg^{-1})	26.83±0.76^{ab}	7.03±0.35^{a}	5.43±0.32^{ab}
M1 (0.5 g kg^{-1})	25.00±1.00^{a}	7.03±0.25^{a}	5.17±0.25^{ab}
M2 (1.0 g kg^{-1})	25.17±-0.76^{ab}	6.83±0.31^{a}	5.50±0.40^{ab}
M3 (1.5 g kg^{-1})	26.00±0.50^{ab}	6.63±0.32^{a}	5.13±0.49^{ab}
V1 (0.5 g kg^{-1})	26.17±0.76^{ab}	6.90±0.46^{a}	5.73±0.25^{b}
V2 (1.0 g kg^{-1})	26.33±1.52^{ab}	7.04±0.17^{a}	4.90±0.75^{ab}
V3 (1.5 g kg^{-1})	26.50±1.04^{ab}	6.87±0.35^{a}	5.43±0.81^{ab}

A1, A2 and A3: *A. cepa* bulb extract. M1, M2 and M3: *M. oliefera* leaf extract, V1, V2 and V3: *V. amygdalina* leaf extract

Table 2: Growth response of *C. gariepinus* fed with *A. cepa*, *M. oliefera* and *V. amygdalina* extract

		Allium cepa			*Moringa oliefera*			*Vernonia amygdalina*		
Parameters	Control	A1 (0.5 g kg^{-1})	A2 (1.0 g kg^{-1})	A3 (1.5 g kg^{-1})	M1(0.5 g kg^{-1})	M2(1.0 g kg^{-1})	M3 (1.5 g kg^{-1})	V1 (0.5 g kg^{-1})	V2 (1.0 g kg^{-1})	V3 (1.5 g kg^{-1})
Initial mean wt (g)	4.63±0.32^{b}	3.77±0.21^{a}	3.97±0.40^{ab}	3.73±0.35^{a}	4.50±0.60^{b}	4.17±0.21^{ab}	4.17±0.20^{ab}	4.07±0.06^{ab}	4.53±0.55^{b}	4.10±0.26^{ab}
Final mean wt (g)	36.27±2.64^{bcd}	36.45±3.72^{bcd}	38.33±2.52^{d}	29.44±4.26^{a}	44.22±3.89^{e}	37.00±1.00^{cd}	36.89±3.72^{bcd}	35.78±1.69^{bcd}	34.00±2.18^{bc}	32.89±1.84^{ab}
Mean wt gain (g)	31.63±1.32^{bcd}	32.68±3.5^{cd}	34.60±2.13^{d}	25.48±1.93^{a}	39.72±3.30^{e}	32.83±1.80^{cd}	32.72±1.68^{cd}	31.71±2.64^{bcd}	29.47±2.07^{bc}	28.79±1.61^{b}
ADG	1.16±0.06^{b}	1.13±0.02^{b}	1.05±0.08^{ab}	1.12±0.22^{b}	1.42±0.12^{c}	1.17±0.03^{b}	1.17±0.03^{b}	1.17±0.13^{b}	1.24±0.08^{b}	0.91±0.03^{a}
SGR	7.36±0.19^{ab}	7.77±0.02^{ab}	7.44±0.15^{ab}	7.21±0.41^{a}	8.17±0.17^{c}	7.80±0.09^{b}	7.62±0.11^{b}	7.66±0.16^{b}	7.42±0.18^{ab}	7.17±0.18^{a}
FCR	1.55±0.08^{b}	1.57±0.07^{b}	1.50±0.30^{b}	1.65±0.09^{bc}	1.40±0.3^{a}	1.51±0.08^{b}	1.50±0.04^{b}	1.55±o.17^{a}	1.73±0.12^{c}	1.63±0.13^{bc}
PER	3.83±0.76^{abc}	3.93±0.40^{b}	3.70±0.60^{ab}	3.60±0.23^{a}	5.77±0.25^{b}	5.37±0.35^{b}	5.23±0.25^{b}	4.60±1.05^{bc}	3.00±0.50^{a}	3.40±0.27^{c}
Survival (%)	96.67±5.77^{ab}	93.33±5.77^{ab}	96.67±5.77^{ab}	90.00±0.00^{a}	100.00±0.00^{b}	96.67±5.77^{ab}	100.00±0.00^{b}	96.67±5.77^{ab}	90.00±0.00^{a}	96.67±5.77^{ab}

A1, A2 and A3: *A. cepa* bulb extract. M1, M2 and M3: *M. oliefera* leaf extract. V1, V2 and V3: *V. amygdalina* leaf extract

M. oliefera and *V. amygdalina* extract. The highest final mean weight and mean weight gain were observed in the group of fish fed 0.5 g kg^{-1} of *M. oliefera* extract (M1) with mean values of 44.22±3.89 and 39.72±3.30 g, respectively, while the least for both parameters were observed in group of fish fed 1.5 g kg^{-1} of *A. cepa* (A3) with mean values of 29.44±4.26 and 25.48±1.93 g, respectively. At higher concentrations the three plant extracts showed low growth response, while at low concentrations *A. cepa* and *M. oliefera* extract showed good growth response. The mean weight gained in decreasing order is as follows; M1>A2>M2>M3>A1 >V1>Control>V2>V3>A3. Statistical analysis showed significant difference ($p \leq 0.05$) across all the groups.

Fish fed 0.5 g kg^{-1} of *M. oliefera* had the best average daily growth (ADG), specific growth rate (SGR), food conversion ratio (FCR) and protein efficiency ratio (PER) with values of 1.42±0.12, 8.17±0.17, 1.40±0.30 and 5.77±0.25, respectively. The least average daily growth (ADG), specific growth rate (SGR), food conversion ratio (FCR) and protein efficiency ratio (PER) were observed in groups; V3 (0.91±0.03), V3 (7.17±0.18), V2 (1.73±0.12) and V2 (3.00±0.50), respectively. Analysis of variance showed significant difference ($p \leq 0.05$) across all the parameters when compared with the control group. Hundred percent survival rate was recorded in groups of fish fed 0.5 g kg^{-1} (M1) and 1.5 g kg^{-1} (M3) *M. oliefera* extract, while the least (90.00±0.00%) was recorded in group of fish fed 1.5 g kg^{-1} *A. cepa* (A3) and 1.0 g kg^{-1} *V. amygdalina* (V2) leaf extracts. Significant difference ($p \leq 0.05$) was observed when the highest value for survival rate was compared with the lowest survival rate.

This is the first research to report on the growth promoting effect of ethanolic extract of *Moringa oliefera* leaf and *Allium cepa* bulb on of *C. gaiepinus*, with high significant difference when compared with the control. Earlier studies reported poor growth even at low inclusion level[13], this could be because plant materials were used as a substitute in replacing fishmeal and not as phytoadditives.

At low concentrations, *Moringa oliefera* and *Allium cepa* significantly increase the growth parameters of *Clarias gariepinus*, while increase in the concentration of the plant extract led to decrease in growth rates of the same fish species. Similar observation was made in earlier studies on other plant extracts, authors reported increase in growth rate at low concentrations[7,17-20].

At high concentration *A. cepa* and *V. amygdalina* retarded the growth of *C. gariepinus*, in contrast to some researchers who observed increased in growth rate with increase in the concentration of plant extract fed to broiler chicken[10,21] this could be as a result of differences in the

physiology of this two organisms, chicken being a warm blooded (Poikilotherms) while fish is cold blooded (Homeotherms) organism.

CONCLUSION

Moringa oliefera and *Allium cepa* showed promising growth promoting effect on *Clarias gariepinus*. In the growing tendency for food safety which had led to the prohibition of synthetic hormones as growth promoter, Phytoadditives like ethanolic extract of *Moringa oliefera* leaf and *Allium cepa* bulb, could serve as alternative growth promoters.

SIGNIFICANCE STATEMENT

With the current growing world population there is need to meet up with the demand for animal protein. Hormones, antibiotics, vitamins and several synthetic products have been used as growth promoters in aquaculture, however the raising apprehension as to their safety to human and the environment led to the prohibition of their use. In a bit to replace their effect led to the search for natural alternatives, this alternative to replace synthetic growth promoter is the phytoadditives. Phytoadditives are additives obtained from plants extracts, they are safe, cheap and biodegradable. The use of phytoadditives to replace antibiotics has just begun, it is hoped that, further research on its long during use in fish culture will further establish its potency.

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