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Use of fluted pumpkin (*Telfairia occidentalis*) leaf powder as feed additive in African catfish (*Clarias gariepinus*) fingerlings

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ABSTRACT

The effect of dietary *Telfairia occidentalis* leaf meal on growth, heamatological profile and body composition was investigated in African catfish *Clarias gariepinus*. Fingerlings of about 5 g were fed diets supplemented with four concentrations (5, 10, 15 and 20 g kg^{-1}) of *T. occidentalis* leaf powder for eight weeks. Fish fed supplemented diets showed significantly improved growth performance, haematological parameters and feed utilization over the control (0 g kg^{-1} *T. occidentalis* leaf meal) treatment. The highest specific growth rate (7.33 ± 2.37% day^{-1}) and best food conversion ratio (0.86 ± 0.06) were obtained in the 5 g kg^{-1} *T. occidentalis* leaf meal diet treatment. Protein efficiency ratio was higher in fish fed with *T. occidentalis* leaf meal and lowest in the control. No differences occurred in fish carcass moisture, protein or ash content among the treatments ($p > 0.05$). The results suggest that dietary supplementation with *T. occidentalis* leaf powder improved growth; feed utilization and survival of *C. gariepinus* fingerlings.

Keywords: Medicinal plant, growth performance, blood parameters, carcass composition, mudfish.

INTRODUCTION

The African catfish, *Clarias gariepinus* is the most extensively cultured food-fish species in Nigeria. High feed costs have forced producers to examine ways to reduce production costs. One of the ways to reduce costs is through the use of dietary supplements that include plant-based additives.

The use of plant-based additives in aquaculture is one of the methods used to improve weight gain, feed efficiency, and/or disease resistance in cultured fish. Therefore, several kinds of plant-based additives for aquafeed that are used to improve the performance of fish have been studied: plant products such as *Astragalus radix* and *Scutellaria radix* (Yin et al., 2006); *Allium sativum* (Sahu et al., 2007); Mango (Awad and Austin, 2010) and *Nigella sativa* (Awad et al., 2013) .These plants have been examined in fish to replace antibiotic growth promoters. For example, phytogenic feed additives have been examined in African catfish (Dada and Oviawe, 2011) and Tilapia (Dada, 2012) and results show improvements in weight gain, food conversion ratio (FCR), and blood indices.

Telfairia occidentalis (fluted pumpkin) is cultivated in various parts of southern Nigeria. The darkish green leafy vegetable is used as food and herbal medicine. The leaf is

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a rich source of protein, oil, vitamins and minerals but low in crude fiber and also rich source of folic acid, calcium, zinc, potassium, cobalt, copper, iron, vitamins A, C and K (Ajibade et al., 2006).

Telfairia occidentalis leaf contains active ingredients such as bioflavonoid, an active chemical, a plant growth promoter, which promotes growth in birds (Fasuyi and Nonyerem, 2007), and may have similar effects in fish. The leafy vegetables possess anti-microbial and antiviral properties (Nwozo et al., 2004; Olorunfemi et al., 2005). Aqueous extract of *T. occidentalis* is also reported to increase hematological parameters (Alada, 2000). The objective of this study was to investigate the effects of dietary concentration of *T. occidentalis* leaf meal on growth, body composition and haematological parameters of *C. gariepinus* fingerlings.

MATERIALS AND METHODS

Four diets based on a formulation of 41.3% protein, 9.08% lipid and 5.35% Ash were prepared to contain different levels of fluted pumpkin (*T. occidentalis*) leaf powder. Fluted pumpkin leaf powder was incorporated into the diets as follows: no Fluted pumpkin (control), 0.5 (group 1), 1.0 (group 2), 1.5 (group 3) and 2.0 $g100^{-1}$ feed (group 4). Fluted pumpkin at different incorporation was added to pelletized feed and then feed were separately dried at ambient temperature (27–30 °C). Pellets were packed and stored in tightly sealed plastic bags at 8-10°C until they were used in the feeding experiments.

225 *Clarias gariepinus* fingerlings (mean body weight: 5.17 ± 0.06 g) were obtained from a commercial fish farm in Akure, Nigeria and transferred to the department of fisheries and aquaculture technology hatchery at Federal University of Technology, Akure, Ondo State. Fish were randomly allocated between 15 tanks in triplicate at a density of 15 fish per 52 litres rectangular plastic troughs and maintained in continuously aerated dechlorinated fresh water. During the experiment, the following conditions were maintained: water temperature 26.70 ± 0.85 ˚C, dissolved oxygen concentration 5.20 ± 0.48 mg/l and pH 7.01 ± 0.07. After 14 days adaptation, fish in each group were fed one of the four different diets at a total daily rate of 3% body weight in three equal meals, every five hours between 08:00 and 18:00 for eight weeks. All fishes were weighed and counted fortnightly and feeding rates were adjusted accordingly.

At the termination of the feeding trial, 5 fish in each tank were individually weighed 24 h after the last feeding. Specific growth rates (SGR), feed conversion ratio (FCR) and protein efficiency ratio were calculated as indicators for growth performance (Heidarieh et al., 2012).

FCR = total feed given/total weight gain

SGR = 100 × [(ln Wf – ln Wi)/days],

Where Wf is mean final weight and Wi is mean initial weight

PER = Live body weight gained (g) / Protein intake (g)

Where: Protein Intake (g) = Protein (%) in feed × Feed given (g)/ 100

The diets were analyzed for proximate composition, including crude protein, crude lipid, crude fibre, ash and moisture (Table 1).

Water temperature, pH and dissolved oxygen concentration were routinely monitored in all tanks. At the beginning and end of the feeding trial, pooled samples of 15 fingerlings were analyzed for carcass composition using AOAC (2005) procedures. Twelve fish (four fish per replicate) were used for blood analysis and 5 ml blood samples from each treatment were collected by cardiac puncture using 5 ml disposable syringes, into treated Bijou bottles. The blood was stored at −40 °C prior to analysis. The blood analysis followed the methods described by Svobodova et al. (1991).

Statistical analysis

Analysis of variance (ANOVA) was used at 95% significance level to test for significant differences between the various treatment means obtained for the growth, feed utilization, carcass composition and haematological parameters. Tukey's multiple range tests was used to determine which pairs of the treatment means differed significantly (Zar, 1996).

RESULTS

Mean water quality parameters during the experiment were: dissolved oxygen 5.20 ± 0.48 mg l^{-1}, pH 7.00 ± 0.07 and 26.7 ± 0.85 °C. Water quality parameters were not significantly different between treatments and were within the recommended ranges for the culture of *C. gariepinus*

There were improvements in the growth responses of fish fed on *T. occidentalis* leaf meal. The best growth responses were obtained in the fish fed on diet D2 (0.5 g 100^{-1} of *T. occidentalis* leaf powder) while the slowest growth was obtained in the fish fed the control diet D1 (Table 2). However, there were no significant differences in growth performance across the different *T. occidentalis* leaf concentrations. The results suggest that dietary *T. occidentalis* leaf at all concentrations promoted the growth of *C. gariepinus* fingerlings. This is a first report regarding the potential of fluted pumpkin leaf powder as a growth-promoting agent in *C. gariepinus*.

There were greater improvements in the feed conversion ratio (FCR) of fish fed on *T. occidentalis* leaf meal than the control fish. The average FCRs were 0.86, 1.32, 1.44 and 1.33 for diets D2, D3, D4 and D5 respectively. The PER was 0.07, 0.04, 0.03 and 0.04 for the fish fed diets D2, D3, D4 and D5 respectively. Fish fed on *T. occidentalis* meal had a significantly ($p < 0.05$) higher protein content than fish fed the control diet. The body composition values are given in Table 3. These results showed that the *T. occidentalis* leaf meal treatment enhances nutrient utilization, which is reflected in improved weight gain, FCR, PER and SGR. Generally, better feed conversion ratio values were obtained in all treatments, but the poorest occurred in D1 (0 g kg^{-1} *T. occidentalis* leaf meal) (Table 2). Although better FCR values were obtained in the *T. occidentalis* dietary treatments compared to the control, differences among the treatment means were not significant ($p > 0.05$).

Table 1: Ingredient composition (kg) and proximate composition (% DM) of basal diet.

Ingredients	**g/kg diet**
Menhaden fish meal	250
Corn meal	150
Soybean meal	350
Blood meal	100
Cod liver oil	60
Vegetable oil	40
Vitamin-mineral premix	30
Corn starch	20
Proximate composition (%)	
Crude protein	41.3
Crude lipid	9.08
Ash	5.35
Gross energy (Kcal/g)	4.65

Vitamin premix – A Pfizer livestock product containing the following per kg of feed: A = 4500 I. U, D = 11252 I.U, E = 71 I.U, K_3 = 2 mg, B_{12} = 0.015 mg, panthothenic acid = 5 mg, nicotinic acid = 14 mg, folic acid = 0.4 mg, biotin = 0.04 mg, choline = 150 mg, cobalt = 0.2 mg, copper = 4.5 mg, iron = 21 mg, manganese = 20 mg, iodine = 0.6 mg, selenium= 2.2 mg, zinc = 20 mg, antioxidant = 2 mg.

Table 2: Mean growth performance and feed utilisation of *C. gariepinus* fingerlings fed experimental diets for 56 days.

Parameter	Dietary treatment				
	D1 (Control)	**D2**	**D3**	**D4**	**D5**
Initial mean weight (g)	5.11(0.08)	5.12(0.07)	5.21(0.07)	5.27(0.06)	5.13(0.02)
Final mean weight (g)	6.76 (0.09)^{a}	9.23 (0.24)$^{a\ b}$	7.26 (0.51)$^{b\ c}$	7.04 (0.15)^{c}	7.55 (0.62)^{d}
Weight gain (g)	1.622 (0.09)^{a}	4.11 (0.33)$^{a\ b}$	2.05 (0.33)$^{a\ b}$	1.78 (0.06)^{b}	2.42 (0.94)^{c}
PER	0.03 (0.00)^{a}	0.07 (0.02)$^{b\ c}$	0.04 (0.01)$^{a\ b}$	0.03 (0.00)^{c}	0.04 (0.01)^{c}
SGR (% day^{-1})	2.89 (0.16)^{a}	7.33 (0.37)^{a}	3.65 (0.60)^{a}	3.17 (0.11)^{b}	4.32 (0.67)^{b}
FCR	1.53(0.14)^{a}	0.86 (0.60)$^{b\ c}$	1.32 (0.40)$^{a\ b}$	1.44 (0.51)^{c}	1.33 (0.59)^{c}
FI (g/d)	2.48 (0.08)^{a}	2.65 (0.09)$^{b\ c}$	2.56 (0.00)$^{a\ b}$	2.57 (0.05)^{c}	2.58 (0.14)^{c}

Values in parentheses are standard errors of means. Means in a given row with the same superscript letter were not significantly different at $p < 0.05$.
SGR = specific growth rate, PER = protein efficiency ratio, FCR = feed conversion ratio, FI = feed Intake.

Table 3: Chemical composition of whole body of *C. gariepinus* fingerlings fed experimental (wet basis).

Composition (%)	Dietary treatment				
	D1 (control)	**D2**	**D3**	**D4**	**D5**
Moisture	4.83 (0.42)$^{a\ b}$	4.31 (0.31)^{b}	4.73 (0.40)^{a}	4.59 (0.07)$^{a\ b}$	4.78 (0.42)$^{a\ b}$
Crude protein	60.76 (0.00)^{a}	61.75 (0.28)^{a}	61.13 (0.56)^{a}	61.46 (0.00)^{a}	61.42 (0.10)^{a}
Crude lipid	5.00 (0.20)^{a}	4.51 (007)^{a}	4.88 (0.03)^{a}	3.96 (0.69)^{a}	5.05 (0.06)^{a}
Ash	13.62 (0.20)^{a}	13.49 (0.33)^{a}	11.89 (0.51)^{a}	13.20 (0.91)^{a}	12.85 (0.00)^{a}

Means in a given row with the same superscript letter were not significantly different at $p < 0.05$. Values in parentheses are standard errors of means.

Table 4: Some haematological characteristics of *C. gariepinus* fed the experimental diets.

Blood parameter	Experimental diets				
	D1	**D2**	**D3**	**D4**	**D5**
PCV (%)	37.5 (2.50)^{d}	37.5 (1.50)$^{b\,c}$	29.0 (3.00)^{a}	30.0 (1.00)$^{a\,b}$	32.0 (1.00)$^{c\,d}$
Hb (g/100ml)	12.50 (0.08)^{c}	12.60 (0.50)^{b}	9.65 (0.95)^{a}	10.00 (0.30)^{b}	10.75 (0.35)^{b}
WBC ($\times 10^3$/µl)	5150 (95.0)^{a}	5550 (25.0)^{c}	8050 (12.50)^{e}	8050 (45.0)^{d}	6750 (10.50)^{b}
RBC ($\times 10^6$/µl)	1.18 (0.28)^{d}	1.45 (0.15)$^{b\,c}$	3.20 (0.35)^{a}	3.30 (0.10)$^{a\,b}$	3.58 (0.18)$^{c\,d}$

Means in a given row with the same letter were not significantly different at $p < 0.05$. Values in parentheses are standard errors of means. PCV = packed cell volume, H b = haemoglobin estimation, WBC = white blood cell count, RBC = red blood cell count.

DISCUSSION

T. occidentalis leaf powder in diets promoted growth and feed conversion efficiency in birds (Fasuyi and Nonyerem, 2007). Similar results were reported by Turan (2006) who used the medical herb red clover *Trifolium pratense* as a growth-promoting agent for tilapia *Oreochromis aureus.* Diab et al. (2002) also reported that Nile tilapia *O. niloticus* fingerlings fed on diets supplemented by medicinal plants exhibited faster growth than those fed with the control diet. Similar results were reported for using medicinal plants as growth-promoting agents for common carp *Cyprinus carpio* (Yilmaz et al., 2006), guppy *Poecilia reticulata* (Cek et al., 2007a), the cichlid *Cryptoheros nigrofasciatus* (Cek et al., 2007b), tilapia *Oreochromis niloticus* (Metwally, 2009) and African catfish *C. gariepinus* (Dada and Oviawe, 2011). Fallahpour et al. (2014) suggested that unknown factors in various medicinal herbs led to favourable results in fish growth trials.

The present findings may indicate that the presence of bioflavonoids in *T. occidentalis* stimulate growth in fish. In addition, bioflavonoid are plant chemicals with estrogenic activity, and studies have shown that estrogen promotes growth in common carp (Kocour et al., 2005). Therefore, the *T. occidentalis* leaf powder that promotes growth performance in the African catfish should be tested for its efficacy to induce efficient and economical propagation in other fish. The best feed utilization values observed with diets supplemented with *T. occidentalis* leaf meal suggested that the addition of *T. occidentalis* leaf meal improved feed utilization.

The haematological parameters of *C. gariepinus* (Table 4) showed no significant differences in mean cell volume, mean corpuscular haemoglobin concentration, mean cell haemoglobin or pack cell volume in all the treatments. However, there was a significant difference in the white blood cell count of fish among the treatments. Fish fed on diets supplemented with *T. occidentalis* leaf meal had significantly higher white blood cell counts.

White blood cell counts were significantly higher in fish fed diets including 0.5, 1.0, 1.5 and 2.0 g/100 of *T. occidentalis* leaf powder. The haematological values obtained in the present study are similar to those obtained by Dada and Oviawe (2011) who used *G. kola* as a growth-promoting agent in *C .gariepinus* fingerlings. Differences in blood parameters of fish in this study could therefore be ascribed to differences in the dietary inclusions of *T. occidentalis* leaf meal in the diets.

The body composition values obtained in this study were similar to those reported by Diab et al. (2002), Lara-Flores et al. (2003), Hamid and Mohamed (2008) and Fallahpour et al. (2014).

Conclusion

Results from the aforementioned study indicate promising potential and reliable method for propagating fingerlings production and rearing strategy. The use of medicinal plants, especially of fluted pumpkin in catfish, will be an efficient tool to achieve sustainable, economical, and safe fish production. Future research should focus on the improvement of rearing technologies for different species of fish reared using *T. occidentalis* leaf powder as a feed supplement.

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