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SUPPLEMENTATION OF DATE PALM (*Phoenix dactylifera*) SEED AS FEED ADDITIVE FOR IMPROVED PERFORMANCE OF JUVENILE AFRICAN CATFISH (*Clarias gariepinus*) (Burchell, 1822).

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ABSTRACT
The present study examined the utilization potential of date palm seed (DPS) as feed additive in the diet of African catfish *Clarias gariepinus* for efficient feed utilization. A total of 150 African catfish (weight ranged from 81.13 to 91.35 g) were divided into five experimental groups. The experiment was conducted for 70 days. Experimental diets were identical in all composition except for the variation in DPS level. Non-inclusion of DPS was used at 0% (control) and other levels are 0.5% (diet 2), 1.0% (diet 3), 1.5% (diet 4) and 2.0% (diet 5). The growth performance and nutrient utilization of African catfish including Weight Gain (WG), Specific Growth Rate (SGR), Protein Efficiency Ratio (PER) and Protein Productive Value (PPV) were significantly ($p<0.05$) higher than those of fish fed control diets, where DPS supplementation at 1.5% appears to be generally higher than other levels. In the same trend fish fed diets with DPS supplementation produced improved fish carcass values and the haematology of fish was generally better from diets in all DPS fed fish. Overall production performance and subsequent fish quality assessments indicated that the diets containing 1.5% DPS level recorded the best performance in African catfish compared to other levels including the control diet. Based on the result of the present study, it is concluded that date palm seed supplementation positively influenced growth performance and feed utilization of African catfish as well as ensuring their healthy status

Key words: Date palm seed, *Clarias gariepinus*, Supplementation, Feed additive, Diet

INTRODUCTION

African catfish, *Clarias gariepinus* (Burchell, 1822), is one of the most highly valued and widely cultured finfish species in Africa (Egypt, Ethiopia, Ghana, Mali and Nigeria) and Asia (China, Indonesia, Malaysia, Philippines and Thailand) as described by Haylor (1993). African catfish is considered one of the best suitable alternatives to tilapia for subsistence fish farming in Africa. Yields of catfish from ponds could be higher than those of tilapia while using low grade feed composed of local agricultural by-products (Hogendoorn, 1983; Sule & Sotolu, 2010). Catfish is cultured for its high growth rate, disease resistance and its tolerance in wide range of temperature, dissolved oxygen and high salinity levels (Huisman & Richter, 1987; Haylor, 1993). Recently, *Clarias gariepinus* has been introduced in some European and Latin American countries and its culture up scaling (FAO, 1997; Ali, 2001). The superior performance of *Clarias gariepinus* compared to other *Clarias* species in terms of growth rate has probably contributed to the fact that *C. gariepinus* has been widely introduced to areas outside its natural range (Verreth, Eding, Rao, Huskens & Segner, 1993). Antibiotics had been frequently used to enhance growth and or resistance to diseases in aquaculture but in hatchery operations and growth out systems. However, the excess use of antibiotics to prevent bacterial diseases in aquaculture, have led to an increase in antibiotic-resistant bacteria (Teuber, 2001). In recent years, there has been increase in the use of organic-based plant by-products like *Azadirachta indica* leaf extract as component of fish diet in order to fortify fish health status (Harikrishnan, Rani & Balasundaram, 2003). Some of the advantages of using such plant material are due to their antibiotics properties for therapeutic remedy and they are relatively available impacting non-residual effects on the fish thereby ensuring human food safety (Teuber, 2001). The present study was conducted to determine the effect of date palm seed (DPS) as nutrient additive in the utilization and performance of *Clarias gariepinus* (African catfish) for healthy fish production.

MATERIALS AND METHODS

Experimental Fish and Culture Technique

The present study was conducted at the Nasarawa State University, Fish Farm Lafia campus. African Catfish (*Clarias gariepinus*) were obtained from the farm's hatchery section. The average initial body weight of experimental catfish ranged from 86.50 to 91.40g. A total of 150 African catfish juveniles were equally divided in 5 treatments with triplicate groups and carefully stocked into 15 concrete tanks of 1.44m^3 each. The fish were adapted to experimental conditions for 2 weeks prior to the commencement of the experiment and were fed a commercial diet 5% of their body weight that was split into three feeding regimes per day. Following this was the 10-week feeding trial proper during which the tanks were supplied with freshwater from well to maintain good culture media. Water level was reduced in all tanks by one-third and replaced by fresh water in order to achieve recommended acceptable limits for good fish culture according to APHA (1989). During the feeding trial, water temperature and Dissolved Oxygen (DO) were measured using a pH meter (Orion pH meter, USA) and an oxygen meter (YSI model 56 Yellow Springs Instrument, Yellow Springs, OH, USA) daily while ammonia and alkalinity were measured on weekly intervals. Dietary treatments were administered in triplicates at of 3% of body weight for 10 weeks and fish

were fed in three regimes daily at 0900, 1200 and 1500hrs. Changes in the weight of fish randomly carried out per treatment forthnightly and the amount of daily dietary intake was adjusted accordingly.

Experimental Diets:Table 1 presents the five isonitrogenous and isocaloric diets that were formulated for the purpose of the experiment. The control diet had no date palm seed while the other four diets contained DPA at 0.5, 1.0, 1.5 and 2.0% levels, respectively. The fishmeal, soybean meal (SBM), wheat bran and indomie waste were use in the preparation of the diets. Date palm was obtained from Lafia central market and was processed to get dried seed separately from the whole fruit as described by Sotolu, Kigbu & Oshinowo (2011). The diets were processed by blending the dry ingredients into a homogenous mixture and then passing the mixed feed through a laboratory pellet mill (California Pellet Mill Co., San Francisco, CA). The proximate composition of diets as analyzed using the methods of AOAC (2000) is also shown in Table 1.

Table 1: Formulation and proximate analysis of experiment diets (% dry matter)

Ingredients (%)	Control	0.5% DPS	1.0% DPS	1.5% DPS	2.0% DPS
Fish meal	19.38	19.38	19.38	19.38	19.38
Soya bean meal	28.76	28.76	28.76	28.76	28.76
Wheat bran	26.21	25.71	25.21	24.71	24.21
Indomie waste	20.65	20.65	20.65	20.65	20.65
Fish oil	2.00	2.00	2.00	2.00	2.00
Di-calcium Phosphate	1.00	1.00	1.00	1.00	1.00
*Vit. and mineral premix	2.00	2.00	2.00	2.00	2.00
DPF	-	0.50	1.00	1.50	2.00
Analysed composition (dry matter basis)					
Crude protein (%)	39.60	39.58	39.46	39.55	39.52
Lipid (%)	5.75	5.82	5.72	5.48	5.86
Ash (%)	13.03	13.57	12.04	13.59	13.55
Crude fibre (%)	3.71	4.21	3.00	3.15	3.46
Ether extract (%)	30.52	31.71	30.60	30.08	30.77
Gross energy Kcal/Kg	2963.53	2988.40	2980.28	2994.11	2987.05

Biomix fish vitamin/mineral providing per kg of diet at 5kg per tonne inclusion: 20,000 iu, vitamin A, 200 i.u, α-tocopherol acetate 400mg, Ascorbic acid 100mg, Vit. D3, 200 mg Vit E, 8 mg Vit k3, 20mg Vit B1, 30 mg Vit B2, 12 mg Vit B6, 50 mg Pantothenic acid, 0.8 mg Biotin, 150 mg Niacin, 0.05mg Vit B12, 4.0mg Cobalt, 40 mg Iron, 5.0 mg Iodine, 30 mg Manganese, 4 mg Copper, 40 mg Zinc, 0.2 mg Selenium, 100 mg Lysine, 100 mg Methionine, 100 mg Anti-oxidant.

Data Collection and Analysis:A sample of 10 fish was taken for determination of the proximate composition (initial carcass sample) of the experimental fish since all were from the same source. Five (5) fish were sampled for the final carcass composition at the end of the experiment. Fish were killed with an excess concentration of anesthetic (t-amyl alcohol) and then weighed. The fish were then pooled per treatment for the analyses. The pooled fish samples were sundried for five days on treatment basis and finely ground in a Thomas Wiley miller. Diets and fish carcass samples were analyzed for crude fiber, ash, crude protein, lipid and gross energy content.

Determination of Fish Performance:Actual mean weight of fish per treatment was determined from triplicates group and data generated on forthnight basis was used alongside feed intake. Weight Gain (WG), Specific Growth Rate (SGR), Feed Conversion Ratio (FCR), Protein Efficiency Ratio (PER) and Protein Productive Value (PPV) was calculated using the following equations:WG = Final body weight (g) - initial body weight (g);

SGR = [(ln FBW - ln IBW) x100] period day^{-1}, where FBW= Final body weight (g), IBW= Initial body weight (g) and ln= Natural logarithm;

FCR was calculated as: feed intake (g)/wet weight gain (g);

PER was calculated as: Wet weight gain (g)/protein intake (g)

Protein intake= Feed fed x crude protein of the feed;

PPV as (Retained protein/protein intake) x 100;

Retained protein=Final carcass protein-Initial carcass protein;

Haematological study:Haematological analysis of fish was carried out using recommended procedures described by Svobodova, Fravda & Palakova, (1991), and Wagner, Jeisen, Arndt, Routledge & Breddwisch, (1997). Initial blood samples were taken prior to the commencement of the feeding and final blood samples were taken from fish in triplicates on the last day of the experiment. The haematological indices of mean cell haemoglobin concentration (MCHC), mean cell volume (MCV), mean cell haemoglotan (MCH) were calculated using the total red blood cell count (RBC), haemoglobin concentration (Hb) and haematocrit (Hct) (Dalcie & Lewis, 2001).

Statistical Analysis:Fish growth performance, feed utilization efficiency and carcass chemical composition were statistically compared using a one-way ANOVA ($p \leq 0.05$) and significant differences among means were identified using the SPSS version 10.0 for windows as described by Steel, Torrie & Dickey (1997).

RESULTS

Fish exhibited 100% survival rate as no mortality was observed in all treatment during the 70-day feeding trial of dietaryt Date Palm Seed (DPS) supplementation. All the water quality parameters were within the acceptable range for the culture of African catfish. The water temperature ranged from 26.8 to 29.5°C. Dissolved Oxygen (DO) from 5.0 to 6.5 mg L^{-1} and pH from 6.4 to 8.0. Data in Table 2 shows that African catfish fed diets containing 2.0% DPS had a significantly higher ($p \leq 0.05$) weight gain (81.67g) than control and 0.5% DPS fed fish. Specific growth rate (SGR=0.93% day^{-1}) was highest in diet supplemented with 1.5% DPS and least in fish fed control diet. Values of feed conversion ratio (FCR) and protein efficiency ratio (PER) were highest in fish fed 1.5% DPS supplemented diet (0.84 and 2.12) but least in control (0.65 and 1.63) respectively. Productive protein value (2.51) was significantly ($p<0.05$) higher in fish fed 1.0% DPS supplementation than other treatments which was followed by fish fed 1.5 and 2.0% DPS. Table 3 showed that dietary date palm supplementation had no significant ($p>0.05$) influence on the values of moisture and Nitrogen free extract (NFE) from proximate analysis of fish carcass but significant variations were observed in the values of crude protein, lipids and ash. The highest values of fish carcass protein, lipids and ash are 63.60, 5.93 and 11.60 while the least values are in the control respectively. The table further revealed that fish heamatology was significantly affected by dietary date palm supplementation (treatment) as significant variations occurred between initial and final values of blood parameters assessed except values of heamoglobin which ranged between 5.27 g/dl (control) and 5.74 g/dl (1.5% DPS).

Table 2: Nutrient utilization assessment of African catfish (*Clarias gariepinus*) after 70 days feeding with dietary Date palm seed (*Phoenix dactylifera*) (DPS) supplementation as Feed Additive.

Parameters	Control	0.5% DPF	1.0% DPF	1.5% DPF	2.0% DPF	SEM
Initial body wt. (g fish-1)	91.40	88.40	86.50	89.20	91.20	-
Final body wt. (g fish-1)	154.11a	154.30a	164.48b	170.53b	172.87b	8.62
Weight gain ((g fish-1)	62.71 a	75.90b	77.98b	81.33c	81.67c	1.30
Total Feed intake (g)	97.12a	99.74a	96.00a	97.15a	99.17a	3.70
SGR (% day-1)	0.75a	0.80b	0.92c	0.93c	0.91c	0.21
FCR	0.65a	0.76b	0.81c	0.84c	0.82c	0. 12
PER (x10-2)	1.63a	1.92b	2.08c	2.12c	2.08c	0.04
PPV (x10-3)	2.15a	2.02a	2.51c	2.35b	2.24b	0.11
Survival (%)	100	100	100	100	100	-

Mean in the same row sharing the same superscript are not significantly different ($p \geq 0.05$)

Table 3: Carcass and haematology composition of of African catfish (*Clarias gariepinus)* fed dietary Date palm seed (*Phoenix dactylifera*)(DPS) supplementation for 70 days.

Parameters (%)	Initial Value	Final fish carcass value at different percent of DPS supplementation					SE Mean
		Control	0.5%	1.0%	1.5%	2.0%	
Crude protein	54.16^{a}	62.44^{b}	62.13^{b}	63.60^{c}	63.19^{c}	62.92^{c}	0.24
Lipid	4.27^{a}	5.93^{c}	4.22^{a}	5.98^{c}	4.83^{b}	4.96^{b}	0.19
Ash	11.14^{b}	11.02^{b}	10.94^{a}	10.86^{a}	11.60^{b}	10.80^{a}	0.50
Crude fiber	ND	ND	ND	ND	ND	ND	-
Moisture	5.93^{a}	5.58^{a}	5.73^{a}	5.59^{a}	5.87^{a}	5.59^{a}	0.14
NFE	24.50^{b}	15.03^{a}	16.98^{a}	13.97^{a}	14.51^{a}	15.73^{a}	3.59
Haematocrits							
PCV (%)	22.21^{a}	24.44^{b}	25.26^{b}	25.28^{b}	25.78^{b}	25.50^{b}	1.35
Hb (g/dl)	5.27^{a}	5.42^{a}	5.66^{a}	5.65^{a}	5.74^{a}	5.71^{a}	0.07
Rbc ($x10^{12}$ /l)	1.53^{a}	1.63^{b}	1.64^{b}	1.66^{c}	1.64^{b}	1.66^{c}	3.20
Wbc ($x10^{9}$/l)	130.37^{a}	131.12^{a}	132.57^{b}	135.85^{c}	136.19^{c}	136.35^{c}	0.06
MCV (fl)	145.16^{a}	144.77^{a}	148.20^{b}	149.26^{b}	150.20^{b}	148.10^{b}	3.82

ND – Not Detected.

Means with the same superscript in the same row are not significantly different ($p \geq 0.05$)

DISCUSSION

All the diets supplemented with DPS resulted in improved feed utilization than the control, suggesting that the addition of date palm seed enhanced the growth performance and feed utilization of African catfish *Clarias gariepinus*. It seem date palm seed had no detrimental effect on fish and may be reason for general positive growth rate and fish performance across all treatment. The positive trend in the nutrient utilization efficiency demonstrated by fish on DPS supplementation could be an indication that date palm seed possess the ability to aid digestion of feed for effective utilization. Nutrient utilization efficiency had been attributed to the ability to digest feed well which in turn creates positive influence on growth rate in juvenile carp *Cyprinus carpio* L. (Noh, Han, Won & Choi, 1994; Bogut, Milakovic, Bukvic, Brkic & Zimmer, 1998). The observations are therefore in agreement with the findings of Wang and Zirong (2006), who found that the addition of different bacterial strain (photosynthetic bacteria (PSB) and lyophilized *Bacillus* sp. (B) and their mix) in common carp basal diets improved growth performances, feed utilization and digestive enzyme activities compare to the control diet. The mechanism of feed additives positive influence on nutrient utilization efficiency by fish had been linked to increase in population of beneficial micro-organism, microbial enzyme activity, improved intestinal microbial balance and hence improvement in digestibility and absorption of feed and its utilization (Bomba *et al.*, 2002). The work of Mostafa and Ahmad (2009) on the use of dried fenugreek seed as feed additives in Nile tilapia diets further revealed the potential of plant-based material as feed additives for aquaculture purpose which is in line with the earlier submission of Magi & Sahk (2003). Catfish showed improved growth and nutrient utilization in the present study which could be due to the presence of a range of relevant digestive enzymes which include amylase, protease and phytase in date palm seed (Pascual, Fernández, Díaz, Garcés & Rubert-Alemán, 2000). These enzymes would enhance growth performance as a result of higher nutrient digestibility and effectiveness of gastrointestinal activities (Al-Qarawi, Ali, Al-Mougy & Mousa, 2003), which could explain the better growth and feed efficiency seen with the DPS supplemented diets. In contrast, Gildberg, Johansen & Bogwald, (1995) and Efthimiou (1996) found that there was no effect of probiotics supplementation on growth performance in Atlantic salmon fry and dentex, respectively. Also, Shariff, Yusoff, Devaraja & Rao (2001) and McIntosh *et al.* (2000) found that treatment of commercial Bacillus probiotic supplementation did not significantly increase ($p \geq 0.05$) either survival or growth rate in *P. monodon* and *Litopenaeus vannamei*. From practical point of view of observations made in the present study, it means that date palm seed supplementation impart positively on fish growth and increase the feed utilization efficiency causing decrease the amount of feed necessary for optimum growth, which could result in reducing production cost in aquaculture. With respect to survival, African catfish survival rate was 100% after 120 days of feeding diets with and without date palm seed supplementation; this may be due to the fulfillment of dietary requirement and healthy fish used in this experiment and favourable experimental conditions. African catfish *Clarias gariepinus* is an omnivore that is capable of efficiently digesting and utilizing diverse feeds and ingredients of both plants and animal origin (Sotolu & Faturoti, 2011). African catfish *Clarias gariepinus* is an omnivore that is capable of efficiently digesting and utilizing diverse feeds and ingredients of both plants and animal origin (Sotolu & Faturoti, 2011). The positive effect of DPS supplementation in the final fish carcass value followed the trend in fish SGR, FCR, PER and PPV since nutrient utilization consequent to high digestibility value has been directly linked with final fish carcass composition (Sotolu & Sule, 2011). The present study suggests that date palm seed supplementation is practically good in fish diet formulation for healthy fish production. These results may be supported by the postulation from Lara-Flores, Olvera-Novoa, Guzman-Mendez & Lopez-Madrid (2003) who found that the Protein Efficiency Ratio and Apparent Protein Utilization recorded the best values with probiotic-supplemented diets in fish as feed additive. Referring to the haematological composition of fish in the present study, DPS supplemented diets yielded superior blood parameters in fish than the control indicating that date palm seed had no adverse effect on fish. Fish health status can be affected as a result of the composition or presence of one or more ingredients in a diet that possess threat to the performance and health condition of the fish. Haematology indices are index and reflection of the effects of dietary treatments on the animal in terms of the type, quality and amounts of the feed ingested and available for the animal to meet its physiological, biochemical and metabolic necessities (Ewuola *et al.*, 2004). It can also serve as an index of fish health to detected physiological changes due to unfavourable condition such as exposure to disease and metal pollutants (Duthie & Tort, 1985) or due to poor chemical composition of diet (Adeyemo, 2005). The observed improvement in the values of most blood parameters assessed (PCV, Hb, Rbc and Wbc) although non-significant difference between them, it could serve as indication that it can be included in fish diet to prevent anaemic condition. This observation is similar to the reports of Al-Maiman (2005) that observed favourable effects of fibers of date palm seed (*Phoenix dactylifera*) on plasma lipids in rats and Harikrishnan *et al.*, (2003) who observed improved hematological and biochemical parameters in common carp, *Cyprinus carpio*, following herbal treatment. In the study by Mostafa & Ahmad (2009) it was further ascertained that use of plant-based feed additive (fenugreek seed meal) is capable of improving blood parameters

which is attributed to a shift of water from the plasma to the muscle cells, thereby increasing the haematocrit concentration.

CONCLUSION

The present study revealed that the addition of date palm seed as a feed additive is capable of improving fish growth performance and nutrient utilization as well as ensuring production of healthy fish on sustainable basis. Supplementation of DPS at 1.5% level as feed additive is recommended in catfish feeding for improved fish performance.

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