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Digestibility and growth performance in fingerlings of tilapia *Oreochromis niloticus* fed with diet containing high-carbohydrate ingredients

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Abstract
Apparent digestibility Coefficients (ADCs) of 4 raw materials (Corn, Millet, Sorghum and Fonio meals) and growth performance were determined for juvenile tilapia *Oreochromis niloticus*. A reference diet and 4 test diets (Corn, Millet, Sorghum and Fonio) each including 70 % of reference diet and 30 % of the raw material to be tested were formulated. After 7 weeks of feeding in hatchery of glass tanks (containing 50-L of water), faeces were sample for proximate analyses. Body weights of fish from each tank were recorded weekly. The highest ADCs of carbohydrate (96.15%) and energy (93.64 %) were obtained in fish fed with fonio. Maximum growth and feed utilization were observed in fish fed with control diet and corn. Among studied test ingredients, fonio presented the best digestible energy.

Keywords: *Oreochromis niloticus*, Digestibilities, energy, carbohydrates, growth performance.

1. Introduction
Global fish production is constantly increasing in relation to the increase in population and the satisfaction of their needs for protein of fish origin. From 1960 to 2012, fish consumption per capita increased from 9.9 kg to 19.2 kg (FAO, 2014) [1]. Fish is known as a premium food due to its high protein content of high biological value and its exceptional richness in omega 3 polyunsaturated long fatty acids, specific vitamins, minerals and trace elements (Médale, 2008) [2]. Several aquaculture production systems ranging from extensive to intensive are practiced in the world according to the geographical and socio-economic contexts of the populations. But, whatever the production system, food is one of the important factors of production. In semi-intensive and intensive fish farming, fish feeding could represent up to 50% of the cost of production (Siddhuraju and Becker, 2003) [3].
Among the ingredients used in the formulation of fish foods, fish meal remains the best source of protein. However, fish meal is still expensive and its supply contributes in part to the depletion of natural fish stocks. Plant-based ingredients are increasingly explored and their ability to meet the nutritional needs of fish is studied (Kaushik *et al*., 1995; Shiau, 1997) [4, 5].
In Côte d'Ivoire, cereals such as rice, maize, sorghum and millet are the staple foods of major part of population. These products are also used in animal feed. Agricultural by-products are mainly used as fish feed in semi-intensive and extensive fish farming (Kimou, 2016) [6]. Studies on the nutritional potential of plant ingredients or agro-industrial by-products produced locally have shown that these materials can be used in fish feed as a substitute for fish meal (Otchoumou *et al*., 2012; Adjanké *et al*., 2012) [7, 8].
Tilapia *Oreochromis niloticus* is among the most important warm water fish used for aquaculture production (Charo-Karisa *et al*., 2006) [9]. In Côte d'Ivoire, this fish is the main species of aquaculture. It is raised in monoculture or in association with other species and adapts to several environments. To support sustainable aquaculture production in different regions of the country, the use of low-cost and good quality food-stuff made from locally available raw materials is one of the major issues. This study evaluates the nutritive potential of different cereals as carbohydrate source for Nile Tilapia *Oreochromis niloticus* fingerlings.

2. Materials and Methods

2.1 Experimental design and fish

This study was carried out in the hatchery of the Oceanological Research Center (CRO). The device used were constituted of 40 glass tanks (containing 50 L of water) in a closed circuit system. An electric motor pump ensured a constant flow of well-aerated tap water. Water was filtered by settling and a 30% daily exchange of water. Every day, at 7h and 30 min, before feeding, temperature (28.1 – 28.4°C), dissolved oxygen (6.2 – 6.9 mg/L) and pH (7.2 – 7.3) were measured with a multi parameter (BANTE 900P).

The fingerlings male of tilapia *O. niloticus* (100 ± 2.04 g) used for the experiments were from the aquaculture experimental station (Layo, CRO). The fish were acclimatized to experimental conditions for two weeks during which they were fed with commercial diet (35 % protein) twice daily (8 am and 4 pm) before the beginning of the experiment (Nwanna, 2003) [10]. Before the feeding trial, the fish were randomly distributed in triplicate groups of 10 fish per tank. Each triplicate tanks were assigned to each experimental diet. The digestibility test was performed over 15 days. Body weights of fish from each tank were recorded weekly and tanks were cleaned. The monitoring of growth parameters lasted 50 days.

2.2 Feed ingredients and diet preparation

The feed ingredients tested were maize, millet, sorghum and fonio (*digitaria exilis*) meals available in local markets. The proximate composition is given in Table 1. A control diet is formulated on the basis of the nutritional requirements of *O. niloticus*. 4 experimental diets, each containing 70% of the control diet and 30% of the test ingredient (maize, millet, sorghum and fonio meals on dry weight basis) were formulated and prepared as described by Cho and al. (1982) [11]. The control diet contained 1% chromic oxide (Cr_2O_3) as the inert marker. They were prepared using a pellet press with a 2-mm diameter. The composition of the diet is reported in Table 2.

Table 1: Proximate composition of ingredients (% dry weight)

Ingredients	Proteins	Lipids	Ash	Fiber
Fish meal	45.23	8.75	20.35	-
Soybean meal	45.25	4.30	5.95	5.22
Wheat bran	18.81	3.89	5.86	10.16
Corn meal	9.17	3.20	1.08	6.29
Millet meal	11.38	3.80	1.57	10.80
Sorghum meal	10,68	3,98	1.63	10.18
Fonio meal	9.31	2.10	1.08	6.36

Table 2: Formulation and composition of the experimental diets (% dry weight)

Ingredients	Reference diet		Experimental diets		
(g/ 100 g)		Corn	Millet	Sorghum	Fonio
Fish meal	29	20.3	20.3	20.3	20.3
Soybean meal	53	37.1	37.1	37.1	37.1
bran	6	4.2	4.2	4.2	4.2
Corn meal	-	30	-	-	-
Millet meal	-	-	30	-	-
Sorghum meal	-	-	-	30	-
Fonio meal	-	-	-	-	30
Cassava starch	3	2.1	2.1	2.1	2.1
Fish oil	5	3.5	3.5	3.5	3.5
Vitamin mixture[1]	1.5	1.05	1,05	1,05	1,05
Mineral mixture[2]	1.5	1.05	1.05	1.05	1.05
Chromic oxide	1	0.7	0.7	0.7	0.7
Total	100	100	100	100	100
Proximate analysis					
Crude protein (%DM)	38.23	29.51	30.17	29.96	29.55
Total fat (% DM)	10,05	7.99	8.17	8.23	7.66
Ash (% DM)	9.41	7.06	7,03	7,07	6,91
Crude fiber (% DM)	3.38	5,45	9,60	9,42	5,47
Nitrogen free extract (% DM)	30.79	39.95	39.21	39.13	40.16
[3]Digestible energy					
(Kj/ g)	14.45	13.31	13.42	13.39	12.98
[4]P/E					
(mg protein/Kj ED)	26.45	22.17	22.48	22.37	22.76

DM : Dry matter

[1]Per kg premix: cobalt 20 mg, iron 17 600 mg, iodine 2 000 mg, copper 1 600 mg, zinc 60 000 mg, manganese 10 000 mg, selenium 40 mg

[2]Per kg premix: vitamin A1 760 000 IU, vitamin D3 880 000 IU, vitamin E 22 000 mg, vitamin B1 4 400 mg, vitamin B2 5 280 mg, vitamin B6 4 400 mg, vitamin B 1 236 mg, vitamin C 151 000 mg, vitamin K 4 400 mg, vitamin P 35 200 mg, folic acid 880 mg, choline chloride 220 000 mg, pantothenic acid D 14 080 mg

[3]Digestible energy = 18,8 x protein + 37,7 x lipid content + 11,3 x carbohydrate content (Smith, 1971; Page et Andrews, 1973) [12, 13].

[4]P/E: Protein / Energy ratio

Nitrogen-free extract = 100 – (% protein + % lipid + % moisture + % ash + % fiber)

2.3 Feeding and feaces collection

Diet were provided *ad libitum* twice daily. To quantify the exact feed intake, refused feed was siphoned out immediately, dried, and weighed. The tanks were then cleaned for faeces collection that was made the following day before the first feeding. The collection of faeces began 3 days after the beginning of feeding trial by siphoning (Choubert, 1999; Windell and al., 1978) [14, 15].

2.4 Analytical methods

The feaces sample per replicate were oven dried at 37 °C, during 24 h and frozen at -20 °C until they were analyzed.

Proximate compositions of diets and feaces were determined as follows: dry matter after drying at 105 °C for 24 h; fat by petroleum ether extraction (Soxtherm, Gerhardt, Germany); protein content (N × 6.25) by the Kjeldahl method after acid digestion; ash by combustion at 550°C in a muffle furnace to a constant weight; and crude fiber by acid/alkali digestion. Gross energy contents were calculated according to 38.9 kJ/g for crude fat, 22.2 kJ/g for crude protein, and 17.2 kJ/g for carbohydrates (Luquet and Moreau, 1989) [16]. The proportioning of chromic oxide was done according to the method of Bolin and al. (1952) [17].

2.5 Zootechnical and feed utilization parameters

Apparent digestibility coefficients (ADCs) of each nutrient or energy in the experimental diet (ACD N or E_{diet}) and ingredients (ADC N or $E_{ingredient}$) were determined as follows:

- In the diet according to Maynard and Loosly (1969) [18]:
  ADC DM_{diet} (%) = 100 – [100 (Ii / If)]
  ADC N_{diet} (%) = 100 – [100 (Ii / If) x (N ou Ef / N ou Ei)]
- In the ingredient according to Sugiura *et al*. (1998) [19]:
  ADC $DM_{ingredient}$ (%) = [(ADC of the diet – 0.7 x ADC of the reference diet)] / 0.3
  ADC N or $E_{ingredient}$ (%) = [(N or E in test diet x ADC N or E of the test diet) – (0.7 x N or E in reference diet x ADC N or E of the reference diet)] / (0.3 x N or E in ingredient).
  Where, I = % indicator, I = ingesta, f = feces, N = % nutrient, E = % energy, DM = dry matter

Growth performances were calculated as follows

Survival rate (SR) = N_2 / N_1

Daily weight gain (DWG) = $(W_2 – W_1) / t$

Specific growth rate = $(LnW_2 – LnW_1) (t \times 100)$

Where, W_1 is the initial weight W_2 the final body weight, t the duration of experiment,

N_1 is the initial number, N_2 the final number of fish.

Feed conversion ratio (FCR) = feed intake (g) / dry weight gain (g)

Protein efficiency ratio (PER0 = weight gain (g) / protein gain (g)

2.6 Statistical analysis

The statistical analysis was done using software STATISTICA 7.1 software. All data (percentages, weight, ADCs) were treated by using the analysis of variance (ANOVA) with a factor, and the multiple comparisons of averages were made at 5 % significance level by Duncan test (Duncan, 1955) [20].

3. Results

Apparent digestibility coefficients (ADCs) of dry matter, protein, carbohydrates and energy of compound diets are shown in Table 3. ADCs for dry matters varied from 72% to 78% with a slight variation between diets. Proteins ADCs were generally high and ranged between 89% and 93%. The values were over 90% in fish fed for Corn, Millet and Fonio based diets. Carbohydrates ADCs varied from 75% to 90% with the highest values found in fish fed Fonio based diet. Significant differences were observed between fish fed on cereal-based diets and those fed on the control diet (P> 0.05).

The test cereals ingredients (Corn, Millet, Sorghum and Fonio) ADCs are presented in Table 4. Dry matters ADCs for these different cereals meals varies between 62% for millet meal and 83% for corn meal. Proteins ADCs value ranged between 52% and 70%., with high value for millet meal and low value for sorghum meal. Carbohydrate ADC was highest at 96% for fonio meal and lowest for corn meal at 76%. The ADCs of Carbohydrate and energy in fonio meal were highest than that of corn, millet and sorghum meal. It should be noted that digestible proteins content (8 $mg.g^{-1}$ dw) were better in millet meal and the energy digestible (14 kJ/g).was highest in the fonio meal. Data on growth performance are reported in Table 5. The survival rate during the experience, ranged from 70 to 100 %. Final body weight (FBW) ranged from 125.44 to 113.8 g, with the highest values in fish fed with reference diet and the lowest values in fish fed with Sorghum based diet. No significant different (P > 0.05) was observed between the diets at specific growth rate (SGR). In contrast, fish fed with Corn based diet had a better feed conversion ratio (FCR) and the highest final body weight (FBW).

Table 3: Apparent digestibility coefficients (ADCs) of dry matter, protein, carbohydrate and energy in *Oreochromis niloticus* fed with experimental diets based on corn, millet, sorghum and fonio.

Parameters	ADCs (%)				
	Reference diet	Corn diet	Millet diet	Sorghum diet	Fonio diet
Dry matter	76.42 ± 0.20^{b}	78.46 ± 0.09^{c}	72.22 ± 0.39^{a}	76.82 ± 0.33^{b}	78.19 ± 0.63^{c}
Proteins	93.89 ± 0.05^{d}	91.64 ± 0.04^{c}	90.94 ± 0.13^{b}	89.44 ± 0,15^{a}	91.09 ± 0.26^{ab}
Carbohydrate	75.25 ± 0.13^{a}	79.40 ± 0.04^{b}	80.59 ± 0.16^{c}	80.87 ± 0.13^{c}	89.64 ± 0.16^{d}
Energy	88.03 ± 0.1^{b}	86.89 ± 0.06^{a}	86.73 ± 0.19^{a}	86.97 ± 0.18^{a}	91.44 ± 0.25^{c}

Table 4: Apparent digestibility coefficients (ADCs) of dry matter, protein, carbohydrate and energy and digestible protein and energy content in test cereal meal (corn, millet, sorghum and fonio) in

Ingredients	ADCs (%)				Digestible protein ($mg.g^{-1}dw$)	Digestible energy ($kJ.g^{-1}$)
	Dry matter	Protein	Carbohydrate	Energy		
Corn	83.24± 0.31^{d}	69.83±0.38^{c}	76.49 ± 0.38^{c}	76.85 ±0.21^{a}	6.40 ± 0.04^{b}	11.54± 0.03^{a}
Millet	62.44± 1.30^{a}	67.86±1.12^{bc}	79.50 ± 1.80^{b}	76.56 ±0.67^{a}	7.72 ± 0.13^{c}	11.71 ± 0.1^{a}
Sorghum	77.77± 1.08^{b}	52.32±1.39^{a}	79.28 ± 0.31^{b}	77.28 ±0.67^{a}	5.59 ± 0.15^{a}	11.73 ± 0.1^{a}
Fonio	82.34± 2.07^{c}	64.26±2.69^{b}	96.15 ± 0.36^{a}	93.64 ±0.89^{b}	5.98 ± 0.25^{ab}	13.8 ± 0.13^{b}

dw: dry weight

Table 5: Growth performances of *Oreochromis niloticus*

Parameters	Reference diet	Test diets			
		Corn	Millet	Sorghum	Fonio
IBW (g)	100.4 ± 0.04[a]	100.7± 0.05[a]	100.6 ± 0.05[a]	100.1 ± 0.05[a]	100.2 ± 0.03[a]
FBW (g)	125.44 ± 0.03[e]	122,68 ±0.04[d]	116.5 ± 0.06[c]	113.8 ± 0.03[a]	115.2 ± 0.04[b]
SR (%)	90 ± 28.28[a]	90 ± 28.28[a]	80 ± 28.38[b]	70 ± 14.14[b]	100 ± 0.00[a]
DWG (g/j)	1.21 ± 0.01[b]	1.09 ± 0.3[ab]	0.79 ± 0.23[ab]	0.66 ± 0.22[a]	0.7± 0.1[ab]
SGR (%/j)	0.87 ± 0.08[a]	0.89 ± 0.37[a]	0.63 ± 0.21[a]	0.51± 0.00[a]	0.44± 0.04[a]
FCR	1.4 ± 0.43[a]	1.9 ±0.58[ab]	2.65 ± 0.23[b]	3.14± 0.55[c]	2.06 ± 0.15[abc]
PER	1.97 ± 0.61[a]	1.87 ± 0.88[a]	1.26 ± 0.11[a]	1.09 ± 0,19[a]	1.65 ± 0.16[a]

IBW: Initial body weight; FBW: Final body weight; SR: Survival rate; DWG: Daily weight gain;
SGR: Specific growth rate; FCR: Feed conversion ratio; PER: Protein efficiency ratio

4. Discussion

Digestibilities of ingredients in feed are considered as one of the most important factors affecting the growth of fish (De silva *et al*., 1996) [21]. In this study, digestibilities of 4 feed ingredients (corn, milet, sorghum and fonio) available on the local market were determined in fingerlings of tilapia *O. niloticus*. Variation of the dry matter digestibilities of the different diets shows that digestibility depends on the nature and the proximal composition of the ingredients even if diets protein content (30%) were similar.

The ingredients used in our study were cereal grains rich in carbohydrates. Utilization of carbohydrates in fish usually depends on their molecular and physical status and the dietary carbohydrate level (Fu, 2005) [22]. In this study, maximum growth and feed utilization were observed in fish fed with reference diet and corn. Corn is the most common cereal used in many parts of the world and has a high coefficient of digestibility of amino acids (Vasan *et al*., 2008) [23]. In contrary, growth and food conversion efficiency were low in fish fed diets based on Millet and Sorghum. It should be noted that these two feed ingredients content 9.6% and 9.4% of crude fiber that were higher than those of the other ingredients. The high fiber in diet could have been the cause of the low growth performance in this group. Similar observations were made by Anderson *et al*. (1984) [24] and Ali and Jauncey (2004) [25]. They reported that high dietary fiber levels can reduce utilization of other nutrients.

Although the content of available amino acid in diets have not been determined for *Oreochromis niloticus* in this study, it is possible that amino acids deficiencies in diet would explain the differences in growth observed in fish. This was also reported in tilapia *Oreochromis aureus* fed with poultry manure meal (Sadiku and Jauncey, 1995) [26]. The food conversion efficiency which is relatively low in fish fed with diet based on Sorghum and Fonio suggests the requirements for other nutrients may not have been provided, reducing the feed intake (Sadiku and Jauncey, 1995) [26]. For these two ingredients, protein digestibility ranged from 52% to 64% and remained low compared to the values obtained in hybrid tilapia *O. niloticus x Oreochromis aureus* (Sklan and Lupatsch, 2004) [27]. For The soghum, the low digestibility presented by fish also may be linked to the presence of a limiting factor as the tannins that contribute to poor digestibility of starch in some varieties of sorghum (Dreher and Berry, 1984) [28]. Condensed tannins have been reported as responsible for the inhibition of the enzymatic activity of the cellulase, pectinase, amylase, lipase, proteolytic enzymes and the alpha-galactosidase (Chung *et al*., 1998) [29]. However, other studies have shown that tannin in sorghum has no effect on protein digestibility and fish growth (Pinto *et al*., 2001, Aiura and De Carvalho, 2007) [30, 31].

5. Conclusion

This study shown the ability of Nile tilapia, *Oreochromis niloticus* to utilize varieties of diets based on cereals. Among studied test ingredients Fonio presented the best digestible energy. A more adequate treatment of the feed ingredients would improve carbohydrate utilization by fish as a source of energy. Processing (Cooking, extruding or gelatinization) the carbohydrate-rich ingredients significantly improves digestibility.

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